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A Study to Determine the Proper Dose of Gamma Radiation for Inducing Beneficial Genetic Variability in Bread Wheat (*Triticum aestivum* L.)

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Abstract: Pure dry seeds of three wheat cultivars viz. Pirsabak-91 (P-91), Khyber-87 (K-87) and Tarnab-78 (T-78) were treated with 100, 200,300 and 400 Gy doses of gamma rays from ⁶⁰Co at Nuclear Institute for Food and Agriculture (NIFA) Tarnab, Peshawar. The influence of radiation was examined on some morphological characteristics like germination percentage(%), survival percentage (%), number of tillers plant⁻¹, days to heading, spike length, number of grains spike⁻¹ and grain yield (kg ha⁻¹). A gradual decrease in the mean values for all the parameters was observed with the increase in the radiation intensity except number of tillers plant⁻¹ which showed significant increase in case of all the cultivars. In general, adverse effects on most of the parameters were produced as a result of higher doses (300 and 400 Gy) of gamma rays.

Key words: Gamma rays, wheat cultivars, morphological characteristics

Introduction

Wheat (Triticum aestivum L.) is one of the world primer crops in regard to its antiquity, acreage and importance as an international item of food. The major constraint in the evolution of improved varieties is the limited genetic variability among the existing wheat genotypes. However, Muller (1927) and Stadler (1928) opened a new era in the field of crop improvement and now mutation induction has become an established tool in the field of plant breeding that can supplement the existing germplasm and can improve cultivars in certain specific traits as well. The beneficial genetic variability induced in crop plants through the use of radiation can either be utilized directly or the mutated genes may be incorporated into otherwise good commercial cultivars through hybridization. The natural mutation rate is too low to collect naturally originating positive mutants significantly. On the other hand, applying radiation, the mutation rate can be raised several thousand times in order to maximize the frequency of certain rare type of mutants of special nature to a level where they can be utilized by the plant breeder. Thus the use of radiation results in minimizing the time, labour and experimental area required for the collection of beneficial mutants and helps as well to achieve the results that would not be possible to be accomplished so easily by other means. Mutation breeding experiments have been conducted on almost all crop species including wheat. Sen and Joshi (1958) reported several desirable high yielding M2 mutants in wheat with reduced plant height and increase number of tillers. Siddiqui et al. (1979)

selected an $\rm M_2$ mutant (M-115) with highest grain yield superiority over the parent and standard varieties. Reddy and Viswanathan (1999) induced rust resistance in hexaploid wheat variety "WH147" by using gamma rays and EMS.

In view of these encouraging results, the present study was made to determine the proper dose of gamma radiation for inducing beneficial genetic variability in wheat.

Materials and Methods

Pure dry seeds of Pirsabak-91, Khyber-87 and Tarnab-78 were irradiated with 100, 200, 300 and 400 Gy doses of gamma irradiation from 60Co source at Nuclear Institute for Food and Agriculture (NIFA), Tarnab, Peshawar. Unradiated seeds of each cultivar were used as control. Following performing preliminary radio sensitivity test in the laboratory, the field experiment was conducted at Agricultural Research Station, Sarai Naurang, Bannu. The M₁ generation was raised in four replications on a plot of size 360 m² (net) under split plot design. Each replication (15x6 m²) was divided into three plots (5x6 m²) and each plot in turn was subdivided into five sub-plots for Randomization of the cultivars and doses respectively. Each sub-plot (1x6 m²) was consisted of four rows with fifty seeds row-1. Plant-to-plant and row-to-row space was 10 and 25 cm respectively. The data were recorded for germination percentage, survival percentage, days to heading, number of tillers plant⁻¹, spike length, number of spikelets spike⁻¹, number of grains spike⁻¹ grain yield (kg ha⁻¹).

Germination percentage was determined by counting the seedlings emerged in each sub-plot (fifteen days after sowing) per total number of seeds sown, multiplied by hundred. For survival percentage, fifty seedlings were selected in each sub-plot twenty days after germination and percentage was recorded by applying the following formula.

For data collection of the remaining parameters, ten guarded plants in each treatment were selected at random and the recorded data were statistically analyzed (Steel and Torrie, 1980) for all the levels of significance. For significant F ratios, treatment means were compared using New Duncan's Multiple Range Test (DMRT) (Leclarg *et al.*, 1983.). The percent increase/decrease values were calculated by dividing the difference between the highest and lowest value over the highest value multiplied by hundred.

Results and Discussion

Germination percentage: Highly Significant differences in the mean values for germination percentage were observed under the influence of different radiation doses (Table 1). The mean values for germination percentage ranged from 19.7-90.2%. Maximum reduction of 76.56% was observed in T-78 over control under 400-Gy dose of gamma irradiation (Table 1). The present results coincide with those of Chaudary (1983) and Davies (1970) who also reported adverse effects of radiation on germination. In general, germination of all the cultivars was reduced with the increase in the radiation intensity. Highly significant differences in the mean values were observed due to different gamma rays doses. Non-significant differences in the mean values were observed due to cultivars as well as for interaction between cultivars and doses.

Survival percentage: The mean values for survival percentage differed significantly under the influence of different gamma rays doses. However, non-significant differences were observed in the mean values under the same dose for all the cultivars (Table 1). The mean values ranged in descending order between 84.4-54.7, 81.30-53.1 and 82.4-54.2% for P-91, K-87 and T-78 respectively. The most drastic decrease in percentage survival for all the cultivars was observed as a result of 400 Gy dose of gamma rays. The percent decrease in percentage survival over control was 43.8, 44.3 and 41.2% for P-91, Khyber-87 and T-78 respectively. Highly significant differences in the mean values were observed in response to different

radiation doses as well as due to interaction between cultivars and doses respectively. However, the differences in the mean values in response to cultivars effect were non-significant (Table 2). In general, all the doses of gamma rays adversely affected the survival. The present results are in agreement to those of Matsumura (1959), Khan and Bari (1971) and Chaudary (1983) who advocated that increase in the radiation intensity was associated with the decrease in the survival.

Days to heading: The mean values regarding days to heading in response to different gamma rays doses were significantly different except for 100 GY dose in comparison to control. The mean values ranged between 111.0-126.0 and the values for P-91, Khyber-87 and T-78 ranged between 111.5-117.3, 111.0-118.0 and 112.8-126.8 respectively (Table 1). T-78 took 11.59% more days to heading as compared to P-91 (5.20%) and K-87 (6.01%) in comparison to their respective control (Table 1). K-87 and T-78 with respect to P-91 observed little tendency towards earliness in the character in response to 100-Gy dose of gamma radiation as evident from the percent decrease values of days to heading (Table 1). Non-significant differences in the mean values were observed due to cultivars effect as well as for interaction between doses and cultivars (Table 2). The differences in the mean values in response to different doses were highly significant. Generally, it was found that increase in the radiation dose resulted in the delay of heading. Mansoor et al. (1981), Khalil et al. (1986), Hassan et al. (1988) and Zhu et al., (1991) have already reported similar results.

Number of tillers plant⁻¹: All the doses of gamma radiation produced a stimulatory effect on the number of tillers plant⁻¹ on the cultivars except 100 GY dose in case of K-87 which produced slight decrease in the character. The differences in the mean values for the character under consideration in response to different gamma rays doses were highly significant (Table 1). The mean values ranged between 5.7-11.7 and the range between the mean values for P-91, K-87 and T-78 was 5.7-10.8, 7.6-11.7 and 7.10-1.85 respectively. The (%) increase in the number of tillers plant⁻¹ over control for P-91, K-87 and T-78 was computed as 112.74, 36.84 and 53.9% respectively (Table 1). The (%) decrease in case of K-87 over control was recorded as 11.11% in response to 100 GY gamma rays dose. Highly significant differences in the mean values were observed for interaction between cultivars and doses whereas, the difference in the mean values due to cultivars effect as well as doses were non-significant (Table 2). Konzak et al. (1969), Davies (1970), Ghafoor and Siddiqui (1976) and Khamankar (1989) have also

Table 1: Effects of various doses of gamma rays on germination percentage(A), survival percentage(B), days to heading(C), number of tillers plant⁻¹(D), spike length (cm) (E), number of spikelets spike⁻¹(F), number of grains spike⁻¹ (G) and grain yield (Kg ha⁻¹)(H) on three wheat cultivars

| Characters | Cultivars | 0-Gy | 100-Gy | 200-Gy | 300-Gy | 400-Gy | Mean | Range | %increase | %decrease |
|------------------------|-----------|----------|---------|----------|---------|----------|--------|-------------|-----------|-----------|
| (A) Germination | P-91 | 93.0a | 78.5c | 59.8e | 39.5g | 19.3I | 58.0C | 19.3-93.0 | 0.00 | 79.24 |
| %age | K-87 | 91.5a | 79.5c | 62.0ef | 41.2gh | 19.7I | 58.8C | 19.7-91.5 | 0.00 | 78.48 |
| | T-78 | 86.2ab | 79.3ef | 53.5e | 39.7g | 20.2I | 55.8C | 20.2-86.2 | 0.00 | 76.56 |
| | Mean | 90.2A | 79.2B | 58.4C | 40.16D | 19.7E | | | | |
| (B) Survival | P-91 | 97.72a | 84.78b | 69.20c | 57.28d | 35.78e | 68.95A | 97.72-35.78 | 0.00 | 63.38 |
| %age | K-87 | 95.25a | 78.28b | 66.03c | 58.58d | 32.67ef | 66.16B | 95.25-32.67 | 0.00 | 65.28 |
| | T-78 | 93.00a | 78.50b | 65.50c | 53.83d | 27.65f | 63.69B | 93.00-27.65 | 0.00 | 70.26 |
| | Mean | 95.33A | 80.52B | 66.91C | 56.56D | 32.03E | | | | |
| © Days to | P-91 | 111.5f | 117.5df | 114.3def | 115.3ce | 117.3bc | 115.4B | 115.3-117.3 | 5.2 | 0.00 |
| heading | K-87 | 111.3f | 110.0f | 115.5cde | 117.0bc | 118.0bc | 114.6C | 111.0-118.0 | 6.01 | 0.26 |
| | T-78 | 113.8def | 112.8cd | 113.0df | 121.8ab | 126.0a | 117.4C | 112.8-126.8 | 11.59 | 0.87 |
| | Mean | 112.2D | 112.4D | 114.3C | 119.7B | 120.4A | | | | |
| (D) | P-91 | 5.10a | 8.15c | 6.04b | 9.35cd | 10.85e | 7.97A | 5.10-10.85 | 112.74 | 0.00 |
| Number of | K-87 | 8.55b | 7.6b | 8.9c | 8.55c | 11.7f | 9.06A | 7.6-11.7 | 36.84 | 11.11 |
| tillers | T-78 | 7.10b | 9.10cd | 9.55cd | 10.85e | 8.00c | 9.51A | 7.10-10.85 | 53.9 | 0.00 |
| plant ⁻¹ | Mean | 7.9C | 8.25B | 8.98B | 9.59A | 9.51A | | | | |
| (E) | P-91 | 9.9abcd | 9.6abcd | 9.4bcd | 9.3cd | 8.9d | 9.5B | 8.9-9.3 | 0.00 | 4.49 |
| Spike | K-87 | 10.7a | 10.5ab | 9.6abcd | 9.3cd | 8.8d | 9.8B | 8.8-10.7 | 0.00 | 21.59 |
| length | T-78 | 10.6a. | 10.3ab | 9.7abcd | 9.1cd | 8.6d | 9.6B | 8.6-10.6 | 0.00 | 23.25 |
| (cm) | Mean | 10.4A | 10.1A | 9.5B | 9.2B | 8.7C | | | | |
| (F) | P-91 | 44.8 cde | 43.4de | 33.5g | 31.8h | 29.3I | 36.5E | 29.3-44.8 | 0.00 | 52.9 |
| Number of | K-87 | 50.5ab | 49.2bc | 47.5cd | 36.0ef | 34.3g | 43.44B | 34.3-50.5 | 0.00 | 47.23 |
| spikelets | T-78 | 58.3a | 51.9ab | 55.3bc | 47.6cd | 43.44b | 50.8A | 41.1-58.3 | 0.00 | 41.84 |
| spike ^{−1} | Mean | 51.2A | 48.1B | 45.4C | 38.4D | 34.9E | | | | |
| (G) | P-91 | 44.8 cde | 43.45de | 33.5 b | 37.15ef | 37.6 efg | 39.30C | 44.8-33.5 | 0.00 | 25.22 |
| Number | K-87 | 50.55abc | 50.25bc | 47.5 bcd | 36.00fg | 44.30cde | 45.72B | 50.55-36.00 | 0.00 | 28.78 |
| of grains | T-78 | 58.35a | 51.9 ab | 55.3 ab | 46.60cd | 48.5bcd | 52.04A | 58.35-46.6 | 0.00 | 20.13 |
| spike ^{−1} | Mean | 51.23A | 48.53B | 45.43C | 39.92E | 43.32D | | | | |
| (H) | P-91 | 3818ab | 3810ab | 3800ab | 3645bc | 2760e | 3567A | 3818-2760 | 0.00 | 27.71 |
| Grain | K-87 | 3728abc | 3691a | 3440cd | 2933e | 2680e | 3294B | 3728-2680 | 0.00 | 28.11 |
| yield | T-78 | 3665abc | 3652ab | 3363d | 2850e | 2680e | 3242C | 3665-2680 | 0.00 | 26.87 |
| (Kg ha ⁻¹) | Mean | 3737B | 3717A | 3577C | 3142D | 2707E | | | | |

Means sharing same alphabets are not significantly different according to DMRT. Capital alphabets represent significance at 5% level of probability

Table 2: Mean square values of analysis of variance (ANOVA) for various plant characters in wheat cultivars affected by different gamma irradiation doses

| Table 2. Mean square values of analysis of variance (1110 v11) for various plant characters in wheat cultivals affected by different gaining infadiation doses. | | | | | | | | | | |
|---|-----|------------------------|-----------------------|----------------------|---------------------|------------------|-------------------------|-------------------------|-------------------------|--|
| | | Germination | Survival | Days to | Tillers | Spike | Spikelets | Grains | Grain yield | |
| SOV | D.F | % age | %age | heading | plant ⁻¹ | length | spike ⁻¹ | spike ⁻¹ | (Kg ha ⁻¹) | |
| Repeats | 3 | 4.12 | 92.65 | 5.88 | 2.96 | 0.12 | 3412.25 | 3015.36 | 31104.44 | |
| Cultivars | 2 | $11.47^{\mathrm{N.S}}$ | 138.25 ^{N.S} | 44.87 ^{N.S} | 12.54^* | $1.92^{\rm N.S}$ | 28937.69 ^{N.S} | 25811.56** | 647251. ^{67**} | |
| Error (a) | 6 | 13.32 | 12.70 | 21.27 | 4.13 | 1.27 | 1589.37 | 1471.68 | 32196.11 | |
| Doses (D) | 4 | 7808.25** | 6942.0^{**} | 187.57** | 26.65** | 1.71^* | 49378.75* | 40233.53** | 2733010.83** | |
| (DxV) | 8 | 4.62 N.S | 11.48** | 24.49 ^{N.S} | 6.84 ^{N,S} | $0.38^{\rm N.S}$ | 39537.52 ^{N.S} | 31047.29 ^{N.S} | 146824.58* | |
| Error (b) | 36 | 7.48 | 23.23 | 11.47 | 3.24 | 0.44 | 23752.92 | 73325.09 | 58554.44 | |

^{* =} Significant at 0.05% level of probability

suggested similar results who advocated that increase in the number of tillers is associated with the increase in the radiation intensity. However, quite opposite results have been reported by Abrams (1957) and Arain and Siddiqui (1976) who observed an adverse effect of radiation on the number of tillers plant⁴. Such type of deviation might be due to different material utilized and the agro-climatic conditions under which the experiments were conducted. Highly significant differences in the mean values were observed because of different gamma rays doses. Significant differences in the mean values were observed due to cultivars effect while the interaction of cultivars with doses showed non-significant differences (Table 2).

Spike length (cm): A gradual decrease in spike length was observed in all the cultivars in response to increase

in the doses of gamma irradiation. Maximum decrease of 4.49, 21.59 and 28.25% was observed in response to 400-Gy dose for P-91, K-87 and T-78 respectively over their controls. The range between the mean values for P-91, K-87 and T-78 was 8.9-9.3, 8.8-10.7 and 8.6-10.6 cm respectively (Table 1). As evident from the study of Table 1, the cultivars mean value under 100-Gy dose (10.1A) was not significantly different from that of the control (10.4A). Differences in the mean values were significant as a result of different gamma rays doses (Table 2). Non-significant differences in the mean values were observed due to cultivars effect as well as for interaction between cultivars and doses respectively (Table 2). Generally, the increase in the radiation intensity was associated with the gradual decrease in the spike length. As the decrease in spike length is directly

^{** =} Significant at 0.01% level of probability

N.S = Non-significant

proportional to the decrease in plant height, so the present findings are in agreement to those of Abrams (1957), Sen and Joshi (1958) Matsumura (1959), Bhatia and Swaminathan (1962), Molle (1965) and Khalil *et al.* (1986).

Number of spikelets spike⁻¹: The differences in the mean values for number of spikelets spike-1 in response to different gamma irradiation doses were significant and the values computed in Table 1 ranged from 15.6-20.3. The highest mean values were recorded in control for all the cultivars while the lowest mean value (15.6) was recorded under 400-Gy dose for P-91 (Table 1). A gradual reduction in the mean values was observed with the increase in the radiation intensity. The percent decrease in the spikelets spike⁻¹ under 400-Gy dose in comparison to control was computed as 18.58, 22.43 and 17.34% for P-91, K-87 and T-78 respectively (Table 1). The cultivars mean values showed significant differences with respect to those of the control under 200, 300 and 400-Gy doses. It is evident from the study of Table 1 that the cultivars mean values under 200 and 300-Gy were not significantly different from each other. Non-significant differences in the mean values were observed due to different cultivars as well as for interaction between cultivars and doses (Table 2). However, the differences in the mean values as a result of different radiation doses were significant for all the cultivars (Table 2). Generally, it was observed that the number of spikelets spike-1 was decreased with the increase in radiation intensity. The present results coincide to those reported by Galal et al. (1975) who also observed adverse effects of radiation on number of spikelets spike⁻¹.

Number of Grains spike⁻¹: The mean values recorded for number of grains spike⁻¹ were significantly different under the influence of increase doses of gamma irradiation (Table 1). Maximum reduction in the mean values occurred at 200 and 300-Gy doses of gamma irradiation for all the cultivars (Table 1). A slight increase in the number of grains spike⁻¹ occurred under 100 and 400 Gy dose but it was still less than that of the control. Maximum reduction (33.50) in the character under consideration was observed under 200-Gy for P-91. While for K-87 and T-78, it was 36 and 46 respectively under 400-Gy dose. The mean values for the character ranged between 48.80-53.5, 50.55-36 and 58.35-46.8 for P-91, K-87 and T78 respectively. The (%) decrease in the character over control was recorded as 25.22, 28.78 and 20.13 for P-91, K-87 and T-78 respectively. No increase over control in the number of grains spike⁻¹ was observed under the influence of gamma irradiation (Table 1). The present results coincide to those reported

by Galal *et al.* (1975). However, the findings of Muhammad *et al.* (1985), don't agree with the present findings. This type of contradiction might be due to either difference in the genetic material used or difference in the agro-climatic condition under which the experiment was conducted. The differences in the mean values for number of grains spike⁻¹ due to the interaction between cultivars and doses were non significant (Table 2). Whereas, highly significant differences were observed because of the cultivars effect as well as radiation doses.

Grain yield (kg ha⁻¹): The study of Table 1 reveals that all the gamma rays doses adversely affected the grain yield (kg ha⁻¹). Gradual decreases appeared in the yield with an increase in the radiation intensity. The cultivars mean values differed significantly under the influence of all the gamma rays doses. The range between the mean values was recorded as 3818-2760, 3728-2680 and 3665-2680 for persabak-91, k-87 and Tarnab-78 respectively. No cultivar showed increase in the yield kg ha⁻¹ under any gamma rays dose. The (%) decrease under 400-Gy dose over control was computed as 27.71, 28.11 and 26.87% for P-91, K-87 and T-78 respectively (Table 1). Significant differences in the mean values were observed due to interaction between cultivars and doses (Table 2). Highly significant differences in the mean values were observed due to cultivars effect as well as due to the effect of different gamma rays doses (Table 2). Generally, negative effect on yield kg ha⁻¹ was produced on all the cultivars in response to gamma radiation. The present findings are in agreement to those of Horvat (1961), while working on rice (Oriza sativa) he claimed that yield under higher doses of gamma radiation was reduced significantly. Similar results have also been reported by Penic and Jelemic. (1963), Konzak. et al. (1969), Davies. (1970), Burton et al. (1975) and Khalil et al. (1986). However, findings of Fowler and Mac queen. (1972), Ram (1974), Carvahlo (1977) and Song et al. (1992) opposed these findings. These types of contradictions might be due to agro-climatically-changed conditions under which the experiment was conducted or due to different genetic background of the material used.

The lowest dose of gamma irradiation (100 Gy) caused a stimulatory effect on a number of ear bearing tillers. However, reduction in the mean values for all the parameters was observed in response to all the gamma rays doses. The beneficial gamma rays dose in the present research study was determined to be 100 Gy because of its stimulatory effect on the number of tillers per plant. The study recommends that 100 Gy and even low doses can induce positive mutations in bread wheat (*Ttriticum aestivum* L.).

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