Effect of Gamma Rays on Seed Germination, Seedling Height, Survival Percentage and Tiller Production in Some Rice Varieties Cultivated in Sierra Leone

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ABSTRACT

Thirteen rice varieties cultivated in Sierra Leone were used to examine varietal differences in radiosensitivity to gamma radiation during the wet season of 2006 in the lowland ecology. Dry seeds of rice varieties were exposed to gamma radiation ranging from 50 to 800 Gy to determine their responses to radiation and the effective radiation dose for mutation breeding. Percentage germination, percentage survival (field condition), seedling height and tiller production were the traits measured on the M₁ generation. The results indicated that increasing doses of gamma irradiation had no effect on germination for the first seven days under laboratory conditions. Percentage survival of germinated seedlings from the 8th to 14th day under laboratory conditions decreased significantly with increase in radiation dose up to 600 Gy. With increase in radiation above 300 Gy a reduction in seedling height and percentage survival under field conditions was observed in irradiated plants of M₁ generation. Increase in gamma ray doses from 50 to 300 Gy had little or no effect on tiller production as there were no significant differences in tiller number of irradiated seeds and non-irradiated (control) for all the varieties evaluated. The LD₅₀ values determined from regression analysis based on percentage field survival ranged from 345 Gy for ROK18 to 423 Gy for ROK22. These ranges of LD₅₀ values determined for the different rice varieties could be useful in rice varietal improvement programmes in Sierra Leone.

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ABBRVIATIONS

\( \text{Gy} \): Gamma ray; \( \text{DAT} \): days after transplanting; \( \text{LD}_{50} \): lethal dose at 50%; \( \text{St Dev} \): standard deviation; \( \text{CV} \): coefficient of variation; \( \text{DAG} \): days after germination.

1. INTRODUCTION

Crop improvement at the Rokupr Agricultural Research Centre (RARC), (formerly Rice Research Station, Rokupr) in Sierra Leone commenced in the 1930’s with collection and screening of several exotic accessions from India, Sri Lanka, Burma and China, as well as local landraces which were mostly glaberrimas. Hybridization and selection of desirable phenotypes commenced in the 1950’s. In order to facilitate crop improvement at the centre, there is need to broaden methods and activities to meet the challenges of increasing crop yields. While the traditional and conventional approaches of screening, hybridization and selection of germplasm have resulted in the release of several high yielding and adaptable varieties, there was need to complement these approaches to include modern breeding and crop improvement methods.

Mutation has been successfully employed in breeding of several food crop varieties, ornamentals and export crops (Mohamad et al., 2005). Past research activities in mutation breeding have reported seedling height, survival rate and tiller production as important traits to be improved upon for optimum yields (Katoch et al., 1992; Mohamad et al., 2002a). Similar studies undertaken by Harding and Mohamad (2009), Cheema and Atta (2003) and Narasimba and Bhalla (1998) revealed similar findings in the cultivars: Roselle, Aruna and Pigeon Pea, respectively. Mutation breeding activities on rice in Sierra Leone has in the past resulted in the release of rice varieties ROK11 and ROK12 for the lowlands and ROK18 for the upland (RRS, 1995).

This study was therefore carried out on some Sierra Leone released rice varieties to determine the optimum mutagen doses and their effect on physiological traits which could be useful in rice varietal improvement programmes.

2. MATERIALS AND METHODS

Two hundred and fifty (250) seeds of thirteen rice varieties were subjected to gamma rays from 60Co source using doses from 50 to 800 Gy (Tables 1 and 2). Seeds received a pre-treatment by moistening and equilibrated in a 60% glycerol in a desiccator before exposure to cobalt radiation. Irradiation was undertaken at the Sieberadorf Laboratory of the International Atomic Energy Agency (IAEA) in Austria in April 2006. Non irradiated seeds were established as controls.

2.1 Germination Evaluation

Fifty (50) seeds by doses and varieties (Table 3) of irradiated and non-irradiated rice varieties were sown in five replications per treatment on blotting paper in 9 cm Petri dishes soaked with distilled water to observe germination under laboratory conditions (Temperature
27 ± 2°C) on the 10th June, 2006. Filter paper was maintained moist with distilled water on need basis. The seeds were then observed daily for a period of fourteen (14) days after plating. Percentage germination was recorded for each variety for the first seven days and percentage survival of germinated seedling at the fourteenth (14th) day after plating (24th June, 2006).

2.2 Field Experiment

The experiment was established at the lowland of Mawir Inland Valley Swamp on the 24th June, 2006 using germinated seedlings that survived after 14 days (irradiated and non-irradiated seedling). Seedlings were transplanted in rows in the field on a plot size of 18m² each per treatment using an intra-row spacing of 10cm with one seedling per hill and a row spacing of 20cm.

Planting of each variety commenced with seeding of the control (non-irradiated) followed by their radiated seeds of the variety and ending with the control for ease of comparison. The recommended practice of fertilization of N60-P40-K40 in the form of Urea, Single Superphosphate and Muriate of potash was applied in two splits at two and six weeks after transplanting. Weeding was also performed before each fertilizer, application.

Percentage field survival, seedling height and tiller produced were recorded to measure the physiological effect and to evaluate their usefulness in estimating the optimum doses required for mutation breeding. Plant height was taken from the base of the plant at soil level to the tip of the tallest leaf blade at 28 days after transplanting in the field whilst the total number of tillers produced was done by counting the number of tillers per square meter at 42 days after transplanting. At 14 DAG (days after germination), the total number of germinated seedlings that was transplanted and the total number of seedlings that survived in the field 28 DAT were recorded. These series of information were then used to calculate percentage field survival as follows:

\[
\text{Percentage field survival} = \left\{ \frac{\text{Number of survived seedlings 28 DAT}}{\text{Total number of seedlings transplanted at 14 DAG}} \right\} \times 100
\]

Linear regression analysis from MSTAC software package was used to estimate the optimum LD₅₀ doses for the different rice varieties using percentage survival in the field as a standard measure of physical effect. The determination of the optimum doses (Table 3) was based on Gaul, Osborne and Lunden (Mohamed et al., 2005).

3. RESULTS AND DISCUSSION

3.1 Germination

Table 1 shows that germination percentages decreased after gamma irradiation. Increase in doses of gamma irradiation had no significant effect on seed germination from 0-7 days. The decrease in germination was not directly proportional to the increase in dosage nor was a definite pattern observed in all the thirteen rice varieties. Similar results have been reported in rice by Sareen and Koul (1999), Sanjeev et al. (1998), Sarawgi and Soni (1993) and Cheema and Atta (2003). In figure 1, however, the effectiveness of gamma irradiation on percentage survival of germinated seedling 14 days after germination was evident at 300 and 400 (LD₅₀) and reduces significantly at 500 and 600 Gy. Beyond 600 Gy, none of the seedlings for any of the varieties survived.
Table 1. The effect of gamma irradiation on the mean percentage germination of rice varieties 0-7 days

| Gy  | ROK3 | ROK5 | ROK10 | ROK11 | ROK14 | ROK16 | ROK18 | ROK21 | ROK22 | ROK24 | ROK25 | ROK29 | ROK30 |
|-----|------|------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| 0   | 94   | 95   | 95    | 96    | 95    | 93    | 93    | 98    | 99    | 99    | 98    | 94    | 99    |
| 50  | 92   | 94   | 94    | 96    | 94    | 92    | 92    | 98    | 97    | 97    | 97    | 97    | 99    |
| 100 | 92   | 93   | 94    | 96    | 93    | 91    | 92    | 97    | 96    | 97    | 96    | 93    | 100   |
| 200 | 92   | 93   | 94    | 94    | 91    | 93    | 93    | 94    | 96    | 97    | 96    | 96    | 100   |
| 300 | 93   | 94   | 93    | 93    | 92    | 91    | 98    | 96    | 98    | 96    | 96    | 93    | 98    |
| 400 | 92   | 94   | 92    | 94    | 91    | 91    | 96    | 96    | 98    | 98    | 94    | 94    | 95    |
| 500 | 94   | 93   | 94    | 95    | 93    | 90    | 90    | 97    | 97    | 98    | 96    | 93    | 95    |
| 600 | 93   | 94   | 93    | 92    | 93    | 92    | 95    | 98    | 98    | 95    | 90    | 96    |
| 800 | 94   | 94   | 94    | 92    | 92    | 92    | 95    | 95    | 97    | 97    | 90    | 96    |
| Mean| 93   | 94   | 94    | 93    | 92    | 92    | 92    | 97    | 97    | 98    | 97    | 93    | 98    |
| St  | 0.9  | 0.7  | 0.9   | 1.8   | 1.0   | 1.0   | 1.0   | 1.2   | 1.2   | 0.7   | 1.0   | 2.3   | 2.1   |
| Dev |      |      |       |       |       |       |       |       |       |       |       |       |       |
3.1.1 Seedling height

The result of this study demonstrated that increasing doses from 0 – 300 Gy had no effect on seedling height (Table 2) even though there were slight decrease in heights with increase in dosage but the decrease was not proportional to the increase in dosage for all rice varieties. This indicated that the varieties did not differ in radio sensitivity with respect to seedling height as the dose of 300 Gy causing 50% seedling height reductions was same in all thirteen varieties. However, at 400 Gy and above the effect became significant. Similar studies on rice have been documented by Cheema and Atta (2003) and Konzak et al. (1972). However, a linear dependency of seedling height of rice on the dosage of physical and chemical mutagens have been reported by Katoch et al. (1992) and Wang et al. (1995) on Roselle by Harding and Mohamad (2009) and on Pigeon pea by Sinha and Chowdhury (1991).

Table 2. The effect of gamma irradiation on seedling heights (cm) 28DAT on rice varieties

| Variety | Dose (Gy) | 0  | 50  | 100 | 200 | 300 | 400 | 500 |
|---------|-----------|----|-----|-----|-----|-----|-----|-----|
| ROK3    | 24.4      | 23.8| 23.4| 23.4| 23.4| 0   | 0   |     |
| ROK5    | 32.4      | 31.6| 30.2| 29.4| 29.9| 26.2| 0   |     |
| ROK10   | 31.4      | 30.0| 29.8| 29.8| 29.5| 0   | 0   |     |
| ROK11   | 30.0      | 29.2| 28.8| 28.4| 28.4| 0   | 0   |     |
| ROK14   | 29.0      | 28.2| 27.6| 27.5| 27.3| 18.8| 0   |     |
| ROK16   | 24.8      | 23.0| 22.4| 22.2| 20.0| 18.6| 0   |     |
| ROK18   | 25.2      | 25.0| 25.0| 25.0| 25.0| 0   | 0   |     |
| ROK21   | 26.8      | 26.6| 26.6| 26.4| 26.0| 0   | 0   |     |
| ROK22   | 31.8      | 29.8| 29.4| 29.4| 29.2| 23.6| 0   |     |
| ROK24   | 30.8      | 30.4| 30.2| 29.9| 29.7| 0   | 0   |     |
| ROK25   | 30.2      | 29.4| 29.4| 29.2| 29.2| 25.6| 0   |     |
| ROK29   | 31.0      | 29.4| 29.4| 29.0| 29.0| 0   | 0   |     |
| ROK30   | 37.6      | 36.8| 36.6| 35.9| 35.5| 24.6| 0   |     |
| CV (%)  | 12.5      | 10.8| 15.6| 20.6| 8.4 | 23.3| -   |     |
| St Dev  | 3.7       | 3.6 | 3.6 | 3.4 | 3.7 | 12.1|     |     |

3.1.1.1 Percentage field survival and tiller production

Figures 2 and 3 show the effect of physical treatment of rice varieties on percentage seedling survivals in the field 28 DAT (days after transplanting) and tiller produced 42 DAT, respectively. The results showed that increasing dosage from 50 to 500 Gys reduced field survival (Figure 2). At above 500 Gy, physiological damage on seedling height became more severe as none of the varieties survived. However, for tiller production (Figure 3), it was observed that the number of tillers produced were not significantly affected with increased in dosage from 50 up to 300 Gy but became reduced thereafter with increase in gamma radiation above 300 Gy. Similarly at above 500 Gy, none of the varieties survived.

The decrease in the percentage survival with increase in gamma radiation is in agreement with results of Cheema and Atta (2003) who worked on Basmati rice varieties and reported decrease in percentage field survival of M1 plants with increase in radiation dose.
Fig. 1. The effect of gamma irradiation on percentage survival of germinated seedlings (8-14 days after germination) on rice varieties

Fig. 2. The effect of gamma irradiation on percentage field survival of seedlings 28 DAT on rice varieties
Fig. 3. The effect of gamma irradiation on tiller produced 42 DAT on rice varieties

$LD_{50}$ values (Table 3) determined based on percentage field survival ranged from 345 for ROK18 to 423 for ROK22. The varieties ROK18, ROK11, ROK29, ROK10, ROK16, and ROK3 had highest radiosensitivity whilst ROK14 and ROK22 were most tolerant.

Table 3. $LD_{50}$ values for percentage field survival

| Variety | Regression Equation | $LD_{50}$ |
|---------|---------------------|-----------|
| ROK3    | $Y = 85.09 - 0.120x$ | 355       |
| ROK5    | $Y = 92.21 - 0.120x$ | 384       |
| ROK10   | $Y = 94.60 - 0.128x$ | 370       |
| ROK11   | $Y = 101.4 - 0.143x$ | 355       |
| ROK14   | $Y = 90.07 - 0.110x$ | 409       |
| ROK16   | $Y = 93.92 - 0.126x$ | 373       |
| ROK18   | $Y = 68.32 - 0.099x$ | 345       |
| ROK21   | $Y = 80.85 - 0.103x$ | 392       |
| ROK22   | $Y = 82.87 - 0.098x$ | 423       |
| ROK24   | $Y = 78.82 - 0.100x$ | 394       |
| ROK25   | $Y = 71.42 - 0.091x$ | 392       |
| ROK29   | $Y = 80.95 - 0.112x$ | 361       |
| ROK30   | $Y = 80.26 - 0.102x$ | 393       |
| Mean    |                     | 380.5     |
| St Dev  |                     | 23.0      |
4. CONCLUSION

Increasing doses of gamma irradiation above 300 Gy caused severe physiological damage on seedling height, % field survival and tiller production but no effect on germination. The optimum dose determined for improving the efficiency of the rice varieties based on % field survival ranged from 345 Gy for ROK18 to 423 Gy for ROK22. These optimum mutagen doses determined for the different rice varieties could be useful in rice varietal improvement programmes in Sierra Leone.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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