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Growth and Production of *Brachiaria brizantha* cv. MG5 in Three Difference Regrowth Phase Treated by Gamma Radiation Dose

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ABSTRACT

This study was aimed to determine the effect of dose of gamma radiation of seeds on plant growth and plant production of *Brachiaria brizantha* cv. MG5 during 3 different growth phases. The variables measured were plant growth parameters (height and length of plant, number of leaves and tillers) and production (dry matter and organic matter productions). Data were analyzed in a 5 x 3 factorial design. The first factor was dose of gamma radiation consisted of 5 levels i.e., 0, 100, 200, 300, and 400 Gy. The second factor was regrowth phase consisted of 3 levels i.e., growth phase 1, growth phase 2, and growth phase 3. The difference between means was analyzed using Duncan's Multiple Range Test (DMRT). The results showed that dose of gamma radiation influenced the plant growth and production (P<0.05) on different growth phases. The dose of 100 Gy gamma radiation showed the highest height and length, as well as dry matter and organic matter productions. The dose of 200 Gy gamma radiation showed the highest number of leaves and tillers. During regrowth phase 2, the grasses showed the highest of height, number of leaves and tillers, and organic matter production, while during regrowth phase 3, the grasses showed the highest length (P<0.05). There was an interaction between the dose of gamma radiation of seeds and regrowth phases on the growth and production of *B. brizantha* cv. MG5 (P<0.05). The dose of 100 Gy radiation of seeds and regrowth phase 2 gave the best interaction. In conclusion, the dose of 100 Gy radiation of seeds gave the best growth and production of *B. brizantha* cv. MG5 during regrowth phase 2.

***Keywords:** Brachiaria brizantha cv. MG5, gamma radiation, plant growth and production*

INTRODUCTION

Brachiaria is a perennial grass which can be used as animal feed resources or cover crop. *Brachiaria brizantha* produces more dry matters (10.67 ton/ha) than *Brachiaria decumbens* (10.63 ton/ha), and *Brachiaria ruziziensis* (9.42 ton/ha) (Miles *et al*., 1996). This grass is tolerant to drought condition. *B. brizantha* has low palatability because the leaves have a coarse texture and are slightly sharp. High production and less palatable physical features lead to the effort to improve the extensive use of *B. brizantha* as a forage crop. Umami *et al*. (2018) states that the cutting management of *B. brizantha* was applied at the right time to get good qualities of the nutrients and lower anti-nutrient composition of grass.

The production of this forage may be increased by gamma radiation mutation technique. Mutation technique for plant breeding has been applied in some countries for improving the quality of crops; however, this technique is rarely developed for forage crop. Plant breeding mainly aims to enhance reproduction of the plants to produce more superior varieties. Mutation can be induced by utilizing some mutagenic agents such as radiation, non-radiation, and chemical materials. Sutapa & Kasmawan (2016) state that mutation induction is an effective method for increasing plant diversity. Gene mutations occur as a result of changes in genes. Mutations have important roles for development of plant varieties. The most common radiation sources for mutation techniques are X-ray, gamma-ray, and ultraviolet (Jusuf, 2001). The experiment in this research used gamma-ray radiation. Gamma-ray radiation is an ionizing radiation that passes very high penetrating emission power. Therefore, it can penetrate deeply into the plant cells and possibly creates changes both at the levels of gene and chromosome. Based on research on several plants such as *Triticum aestivum* L. (Borzouei *et al*., 2010), rice (Haris *et al*., 2013), soybeans (Hanafiah *et al*., 2010), Korean lawngrass (Lee *et al*., 2016), Ciherang rice (Sulistyo *et al*., 2016), *Zea mays* L. (Rafiuddin *et al*., 2013), *Glicine ma*x L. (Mudibu *et al*., 2012), and sorghum (Surya & Soeranto, 2009) showed that gamma radiation at doses of 100 to 300 Gy had an effect on the genetic variety. Genetic veriety of mutant lines can be measured using morphological and molecular characterizations. The effect of gamma radiation improving plant growth, seed quality, and physiological processes is highly related to the level of doses use (Mudibu *et al*., 2011; Lima *et al*., 2011). These changes eventually modify the characteristic of the plant, which is expected to improve the genetic variety of *B. brizantha* cv. MG5. Genetic varieties consist of the number of tillers and the height, length, and weight of seed as well as crop production that influence crop productivity.

Regrowth occurs as the result of plant metabolism (photosynthesis and respiration) after the plant undergoes defoliation. This process influences plant productivity. Plant persistence after defoliation becomes crucial characteristics since *B. brizantha* cv. MG5 as the forage has multiple harvesting times. The speed of regrowth depends on the supply of food reserves available in the soil. Rapid defoliation affects the regrowth process. In this study, defoliation was performed 60 d after planting. Mutimura *et al*. (2017) showed that defoliation of *Brachiaria* 60 d after planting increased dry matters while defoliation 90 d and 120 d after planting decreased dry matters of plant. So this reseacrh was conducted to determine the effect of doses of gamma radiation of seeds on plant growth and production of *Brachiaria brizantha* cv. MG5 during different regrowth phases.

MATERIALS AND METHODS

This research was conducted in greenhouse and Laboratory of Pasture and Forage Science, Animal Science Faculty, Universitas Gadjah Mada. Gamma irradiation was applied using ^{60}Co gamma source at the Laboratory of Research and Development of Isotopes and Radiation Technology, National Nuclear Energy Agency (BATAN), Pasar Jumat, South Jakarta. The seed of *B. brizantha* cv. MG5 was irradiated with 0, 100, 200, 300, and 400 Gy. This research process consisted of some steps after seed radiation: germination, land preparation, planting, watering, weeding, fertilizing, and harvesting. The seed of *B. brizantha* cv. MG5 was germinated in two weeks. The seed of *B. brizantha* cv. MG5 were put in a polybag. One polybag contained one seed for germination. Each treatment (0, 100, 200, 300, and 400 Gy) was repeated in 15 replications. The addition of fertilizer was conducted 30 d after planting. Defoliation was conducted 60 d after planting. Watering was conducted every day in the morning and afternoon. Weeding was conducted every week. Defoliation was done by cutting *B. brizantha* cv. MG5 at the point of 10-15 cm above the ground. The regrowth phase 1 was completed 60 d after the first defoliation. The defoliation of regrowth phase 1 was performed 60 d after the first defoliation. The regrowth phase 2 was completed 60 d after the defoliation of regrowth phase 1. The defoliation of regrowth phase 2 was performed 60 d after the defoliation of regrowth phase 1. The regrowth phase 3 was completed 60 d after the defoliation of regrowth phase 2. The defoliation of regrowth phase 3 was conducted 60 d after defoliation of regrowth phase 2. The data were collected before defoliation. The data collected were plant growth (the height and plant length, leaves number and tillers) and plant production (dry matter and organic matter productions). Analysis of dry matter and organic matter was conducted according to method of AOAC (2005).

Plant height was measured individually starting from the ground to the highest point of each plant. Plant lenght was also measured from the ground to the tip of the longest on each plant. While leaf number was determined by counting the green leaves from each plant. Tiller number was determined by counting the tiller growing in each plant.

The collected data was analyzed by analysis of variance (ANOVA) based on a completely radomized design in a 5 x 3 factorial arrangement. The first factor was dose of gamma radiation consisted of 5 levels i.e., 0, 100, 200, 300, and 400 Gy. The second factor was regrowth phase consisted of 3 levels i.e., regrowth phase 1, regrowth phase 2, and regrowth phase 3. The difference between means was analyzed using Duncan's Multiple Range Test (DMRT).

RESULTS

The Plant Growth of *Brachiaria brizantha* cv. MG5

Measurement of plant growth of *B. brizantha* cv. MG5 was recorded by measuring the height and plant length, the leaves number and tillers. The plant growth characteristics of *B. brizantha* cv. MG5 with different doses of gamma radiation at various regrowth phases are shown in Table 1.

Plant height. Plant height was significantly influenced by the dose of gamma radiation and regrowth phase ($P<0.05$). The highest plant height of *B. brizantha* cv. MG5 was found during the regrowth phase 2 of grasses germinated from seeds radiated with gamma radiation at doses of 0, 100, 200, 300, and 400 Gy. The highest plant height was produced at grasses germinated from seed radiated with gamma radiation at doses 100 during regrowth phase 2 (117.67 cm). However, the heights of *B. brizantha* cv. MG5 decreased during the regrowth phase 3 in all grasses germinated from seeds radiated with all doses of gamma radiation.

Plant length. The plant lenght of *B. brizantha* cv. MG5 are shown in Table 1. Plant length was influenced by dose of gamma radiation and regrowth phase ($P<0.05$). The length of *B. brizantha* cv. MG5 increased during the regrowth phase. The best height was found in grasses germinated from seeds radiated with gamma radiation at a dose of 100 Gy during the regrowth phase 2 (153.46 cm).

Leaves number. The interaction between doses of gamma radiation and regrowth phases had significant affect ($P<0.05$) on the leaves number of *B. brizantha* cv. MG5. Table 1 showed that the highest interaction between dose of gamma radiation and regrowth phases was the dose of 200 Gy and regrowth phase 2 with the average number of leaves of 112.93. The data indicated that grasses germinated from seeds radiated with gamma radiation at a dose 200 Gy produced the highest leaves number during the regrowth phase 2.

Tiller number. The tiller number are presented in Table 1. This study shown that tiller number of *B. brizantha* cv. MG5 was affected by the interaction between doses of gamma radiation and regrowth phase (P<0.05). The results showed that the number of tillers in grasses germinated from seed radiated with gamma radiation at doses 0, 100, 200, 300, and 400 Gy significantly increased during the regrowth phase 1 and phase 2, but decreased during the regrowth phase 3. The highest tillers number was produced by *B. brizantha* cv. MG5 with dose gamma radiation 200 Gy (26.73) while the lowest *B. brizantha* cv. MG5 with dose gamma radiation 100 Gy (9.47).

The Plant Production of *Brachiaria brizantha* cv. MG5

The plant production of *B. brizantha* cv. MG5 was evaluated by measuring the dry matter production and organic matter production. The plant production of *B. brizantha* cv. MG5 with different doses of gamma radiation at various regrowth phases are shown in Table 2.

Dry matter production. Dry matter production of *B. brizantha* cv. MG5 was significantly affected (P<0.05) by the dose of gamma radiation and regrowth phase. The highest dry matter production of *B. brizantha* cv. MG5 was found in grasses germinated from seed radiated with gamma radiation at a dose of 100 Gy during the regrowth phase 2 (2.55 ton/ha).

Organic matter production. The result showed that an interaction between the dose of gamma radiation and regrowth phase influenced the organic matter production of *B. brizantha* cv. MG5 (P<0.05). The highest organic matter production was obtained in *B. brizantha* cv. MG5 germinated from seeds radiated with gamma radiation at a dose of 100 Gy during the regrowth phase 2 (2.22 ton/ha) and the lowest at a dose of 200 Gy during the regrowth phase 1 (0.42 ton/ha).

DISCUSSION

The Plant Growth of *B. brizantha* cv. MG5

Table 1 shows that the dose of gamma radiation of the seeds significantly affected the plant growth of *B. brizantha* cv. MG5. These results indicate that the higher doses of gamma radiation of seeds cause the decrease in the growth of *B. brizantha* cv. MG5 germinated from the seeds. The research of Makhziah *et al*. (2017) stated that the increase in the dose of gamma radiation caused the decreased characteristic of the plant. Previous study by Hanafiah *et al*. (2010) showed that the most top soybean plant height found in soybean plants germinated from seeds radiated with gamma radiation at doses of 50 Gy and 100 Gy. On the contrary, radiation of the seeds at higher doses i.e., 150 Gy and 200 Gy, produced soybean plants with decreased heights. Refer to the research of Emrani *et al.* (2013), the increase of the dose of gamma radiation from 200 until 600 Gray caused the decrease of plant growth and the number of Zea mays seeds. The viability of soybean seeds (Satpute & Fultambkar, 2012)

Table 1. The plant growth characteristics of *Brachiaria brizantha* cv. MG5 with different doses of gamma radiation at various regrowth phases

Variables	Radiation (Gy)	Regrowth phase		
		1	2	3
Plant height (cm)	0	84.00± 9.52^{de}	114.28± 6.46^{a}	73.58±10.62^{def}
	100	64.54±14.15^{f}	117.67±31.13^{a}	100.06±20.43^{bc}
	200	74.00±12.36^{def}	98.20±25.26^{bc}	78.60±15.72^{def}
	300	82.86±12.52^{de}	105.54±24.74^{ab}	88.13±13.15^{cd}
	400	79.43±12.21^{de}	108.46±21.74^{ab}	72.86±20.73^{ef}
Plant length (cm)	0	118.02± 6.72^{cde}	94.11±12.34^{f}	131.40±13.08^{bc}
	100	112.41±10.75^{e}	153.46±23.87^{a}	139.93±16.43^{b}
	200	114.41±11.19^{de}	119.59±25.02^{cde}	119.26± 7.89^{cde}
	300	116.75± 9.81^{de}	124.71±26.84^{cde}	124.87±12.73^{ced}
	400	115.41± 8.18^{de}	130.99±23.42^{bc}	127.87±12.64^{bcd}
Leaves number	0	48.44± 6.61^{gh}	63.56±15.88^{fg}	65.44±21.55^{efg}
	100	31.00±13.23^{h}	105.40±45.28^{ab}	76.40±32.31^{def}
	200	50.20±15.03^{gh}	112.93±36.09^{a}	86.27±25.56^{bcde}
	300	51.47±13.87^{gh}	98.80±38.8^{abc}	83.33±28.93^{cdef}
	400	39.93± 9.23^{h}	94.80±26.6^{abcd}	84.60±30.60^{bcdef}
Tiller number	0	15.80± 1.78d^{ef}	19.27± 2.81^{bcd}	14.07± 4.73^{efg}
	100	9.47± 3.72^{h}	22.33± 8.13^{b}	13.40± 5.38^{fgh}
	200	14.33± 4.68^{efg}	26.73± 6.92^{a}	18.13± 5.42^{cde}
	300	16.33± 4.45^{def}	22.47± 4.94^{b}	17.27± 5.78^{def}
	400	11.07± 2.54^{gh}	21.67± 5.24^{bc}	17.80± 7.42^{cde}

Note: Means with different superscripts differ significantly (P<0.05)

Table 2. The plant production of *Brachiaria brizantha* cv. MG5 with different doses of gamma radiation at various regrowth phases (ton/ha)

Variable	Radiation (Gy)	Regrowth phase		
		1	2	3
Dry matter production	0	1.04±0.26^{de}	0.69±0.11^{ef}	0.84±0.11^{ef}
	100	0.49±0.11^{f}	2.55±0.58^{e}	0.98±0.28^{de}
	200	0.88±0.25^{ef}	1.78 ±0.52^{bc}	0.87±0.18^{ef}
	300	1.11±0.09^{de}	1.47±0.20^{cd}	0.82±0.13^{ef}
	400	0.63±0.04^{ef}	2.09±0.43^{ab}	1.07±0.22^{de}
Organic matter production	0	0.91±0.23^{de}	0.61±0.09^{ef}	0.74±0.10^{ef}
	100	0.42±0.09^{f}	2.22±0.47^{a}	0.86±0.26^{def}
	200	0.75±0.22^{ef}	1.56±0.45^{bc}	0.77±0.16^{ef}
	300	0.95±0.07^{de}	1.29±0.18^{cd}	0.72±0.12^{ef}
	400	0.54±0.03^{ef}	1.84±0.38^{ab}	0.94±0.21^{de}

Note: Means with different superscripts differ significantly (P<0.05).

and black bean seeds (Lal *et al*., 2009) also decreased during the increase of the dose of gamma radiation.

In this research, the plant growth of *B. brizantha* cv. MG5 with radiation gamma was higher than control. According to Degwy (2013), the dose of gamma radiation produced the highest number of plant with significant difference with the control. Haris *et al*. (2013) stated that the highest number of tillers was found in plants germinated from paddy seeds radiated with gamma radiation at the doses of 200 Gy (22.43) and 300 Gy (22.58) in a variety of rice. The number of tillers is highly dependent on the number of node and the length of the stolon, therefore the pattern of the increasing number of tillers follows the dynamic pattern of the two variables (Abdullah, 2009). Dose of gamma radiation in this research caused genetic varieties in *B. brizantha* cv. MG5, such as the increased of plant height, plant lenght, leaves number, and tillers number. This research is inline with Puteri *et al.* (2015) stated that genetic difference could influence the difference in sorghum variety, such as in the number of leaves and leaf blade.

The ^{60}Co gamma-ray radiation emits high energy which eventually damages the chemical bond of a new compound when given to seeds, plant buds, pollen, apical shoots, tissues, and cells. ^{60}Co gamma rays emitted to seeds can cause cell nuclei to have genome mutations, chromosome mutations, gene mutations or mutations outside the nucleus such as in plastids and the mitochondria. Genomic mutations change the chromosomes numbers such as the addition or subtraction of the entire chromosome. Therefore, this process alters the properties and morphology of plants, such as plant height, the leaves number, and the number of tillers per clump (Wiryosimin, 1995). Warid *et al*. (2017) explains that the higher the radiation dose the higher the reduction of plants' survival ability, particularly at a dose of 400 Gy.

In this research, the regrowth phase significantly affects the height and plant length, the leaves number, and tillers of *B. brizantha* cv. MG5. The grass will undergo regrowth depending on the condition during defoliation. One crucial factor that determines the ability of grass to grow and produce after cutting is the availability of nutrients in the soil (Roni *et al*., 2016). Gadner *et al*. (2008) suggests that differences in growth rates and plant size are influenced by the environmental factors and the genetic properties of the plant. Preussa & Britta (2003) suggest that plant stems become longer and more significant. Cell mutation due to radiation impedes cell division during the process of cell division; therefore anaphase will not occur. Radiation disturbs the processes of forming cell walls, so the cells are unable to undergo cell division. Eventually, the optimal length of the plant will not be achieved.

The defoliation of regrowth phase was performed 60 d after the first defoliation. The study of Monteiro *et al*. (1997) showed that the tiller number of *B. brizantha* increased with the harvesting, in the second harvesting the grass significantly increased the number of tillers. Those value are the same with this research, there were increased number of tillers *B. brizantha* cv. MG5 in the regrowth phase. Ferraro & Oesterheld (2005) also reports that repetitive defoliation has either negative or positive effects due to several factors such as the availability of nutrients in the soil, meristem, the length of growing season, the frequency and intensity of defoliation.

The result showed that there were interaction between the dose of gamma radiation and regrowth phase. The interaction between the dose of gamma radiation and regrowth phase affected the height and plant length, as well as the leaves number and tillers of *B. brizantha* cv. MG5. It is expected that the appropriate dose of gamma radiation increases plant height at various regrowth phases. Having considerable height, plants could increase the weight of their biomasses. Previous study by Sriagtula *et al.* (2016) showed that there was significant interaction between the sorgum mutant lines and harvest time on the sorghum dry matter production. Guenni *et al*. (2005) investigate that *Brachiaria* biomass has increased during the regrowth phase. Purbajanti (2013) strengthen that some environmental factors, such as temperature, humidity, solar radiation, soil, soil reaction, biotic factors, and nutrient supply, also affect plant growth. Telleng *et al*. (2016) state that soil microorganisms have an important role in maintaining soil function involved in mineralization and mobilization of nutrients required for plant growth.

The Plant Production of *Brachiaria brizantha* cv. MG5

The average plant production *B. brizantha* cv. MG5 presented in Table 2. Gamma radiation dose significantly affected dry matter and organic matter production in *B. brizantha* cv. MG5. There was interaction between dose of gamma radiation and regrowth phase. The higher the dry matter production, the higher the organic matter production. Meliala & Soegianto (2016) suggest that radiation causes plants to mutate, but plant genetic changes due to mutations were not leaded in the same direction, even in one treatment dose can produce different genetic diversities depending on the genetic of the crop.

Crop production is determined by plant organs, age, environmental conditions, cultivation, and climate. The amount of crop production is also influended by the vegetative and generative phases. Refer to the research of Sriagtula *et al*. (2017), BMR sorghum mutant lines produced higher digestibility compared to non-BMR sorghum mutant lines. Koten *et al*. (2012) states that the production of dry matter increased with the age of cutting. Several factors affecting the dry matter content of plants were plant species, growing phase, cutting time, soil fertility, and water availability (Purbajanti, 2013).

CONCLUSION

B. brizantha cv. MG5 germinated from seeds radiated with gamma radiation at a doses of 100 Gy have the best growth and production during the regrowth phase 2. *B. brizantha* cv. MG5 germinated from seeds radiated with gamma radiation has a potential to produce high quality and quantity of grass.

CONFLICT OF INTEREST

There is no conflict of interest with any financial, personal, or other relationships with other people or organization related to the material discussed in this manuscript.

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