

Unique Flowers Produced from West Indian Lemongrass, *Cymbopogon citratus* (DC.) Stapf. Through Induced Mutation

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Abstract: West Indian Lemongrass, *Cymbopogon citratus*, is hardly seen to flower which contributes the major obstacle for hybridization. Induced mutation with gamma irradiation has been suggested as the solution to this problem. The objective of this study is to analyse the effect of gamma irradiation dosage on the survival rate of lemongrass prior to mutation. Vegetative stalks of lemongrass were exposed to different doses of gamma rays at doses of 0, 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120 Gy. Results showed that the practical ranges for induced mutation were 40, 60 and 80 Gy with mutation rates were 25.8%, 36.4% and 69.2% respectively. Dose 80 Gy was identified as the dose for LD₅₀. Irradiation caused plants to produce long above ground stem (not stalk) with obvious appearance of nodes and internodes together with unique production of flowers. This phenomenon has created an astonishing opportunity for future studies in this flower of West Indian Lemongrass mutant as another potential source of Halal traditional medicine.

Key words: gamma irradiation • induced flowering • *Cymbopogon citratus* • Lemongrass • Halal medicine

INTRODUCTION

West Indian Lemongrass, *Cymbopogon citratus* is one of the well known Halal medicinal plants in Malaysia. It is a perennial aromatic grass which has been cultivated for several centuries in South and South-East Asia as industrial and garden crop for its essential oil and as the main ingredient in cooking [1]. This type is a native to Southern India, Ceylon [2, 3] and Malaysia [4]. There is another type of lemongrass, East Indian Lemongrass (*Cymbopogon flexuosus*) which is a native to India, Cambodia, Sri Lanka, Burma and Thailand. Both species of lemongrass (East Indian and West Indian lemongrasses) produce up to 75-85% citral in their essential oils [5].

Citral, a monoterpene that gives characteristic of lemon aroma in lemongrass has been reported to have antifungal activity against plant and human pathogens [6, 7], together with bactericidal properties [8, 9] insecticidal properties [10], antifungal properties [11], inhibits seed germination [12] and recently has been found to poses anti-Leishmania properties [13].

These have made citral as an important raw material used in the pharmaceutical, perfumery and cosmetics industries, especially for synthesis of Vitamin A and ionones; which are generally synthesized from synthetic citral that derived from conifer turpentine [14].

As of to date, the information on conventional breeding program for *C. citratus* is limited. This is due to the fact that it flowers very rarely or not at all, which is the main hindrance to do hybridization in West Indian Lemongrass [1, 15]. Generally, *C. citratus* is vegetative propagated, while *C. flexuosus* is commercially propagated by seed as well as vegetative by ramets [16]. As to generate variability in West Indian Lemongrass, induction mutation is one of the possible approaches to widen the genetic variations [17].

Gamma irradiation is an ionizing radiation using γ -rays which are typically categorized as low light emission transfer (LET) radiation. Unlike high LET radiation (such as α -particles and heavy ions particles) which give relatively high energy per unit length of the particle's path and transfer energy to very limited

regions, low-LET radiation gives relatively low energy per unit length of the particle's path and transfers energy evenly to the irradiated area. This will lead to different types of damage to chromosomal DNA [18]. This irradiation can be used to induce mutations and thereby generate genetic variations from which desired mutants may be selected. Furthermore, mutation induction has become a proven way of creating variations within a crop variety. It offers the possibility of inducing desired attributes that either cannot be expressed in nature or have been lost during evolution [19].

Since the lemongrass plants rarely flower or set seed, induced mutation would be the ideal and attractive method which allows the development of new and valuable alterations in plant characters. These can significantly contribute to the increase in the lemongrass potential for either essential oil yield production or beneficial characters in agronomy perspectives. Therefore, this study was carried out to determine the effective dosage (LD_{50}) of gamma irradiation on vegetative stalk of West Indian Lemongrass and to observe the physical alterations made from this mutation treatment.

MATERIALS AND METHODS

Experiment was conducted by irradiating the lemongrass stalks with different doses of gamma rays at doses of 0, 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120 Gy. The gamma rays were from cobalt (^{60}Co) source on 40 Gy Fricke dosimeter and below 400 Gy using Perspex gamma slope. Immediately after the irradiation, 60 vegetative stalks per dose were planted in the glasshouse in Universiti Kebangsaan Malaysia (UKM), Bandar Baru Bangi, Selangor. Plant survival and alteration of the plant characters were observed on three month- old plants and data were taken based on LD_{50} (lethal dose at 50% of the plant population) values. Mutant lines produced at LD_{50} dose were chosen for further analysis. A total of 20 stalks from mutant lines of LD_{50} dose were then planted in open field at USIM Fertigation House, Universiti Sains Islam Malaysia, Bandar Baru Nilai for further physical observation in the duration of 12 months.

RESULTS AND DISCUSSION

Comparing the different doses of gamma irradiated vegetative stalk have shown that an increment of gamma ray dosage would kill the growing point at vegetative stem and from 90 Gy dose and above would cause the decreasing quality in germination percentage of first generation, V_1 .

Only three doses (40, 60 and 80 Gy) produced mutants (Table 1). Initially, dose 40 Gy was found to be the minimum amount of death percentage (0%) that would not affect the plant survival compared to doses 60 Gy (8.3%) and 80 Gy (40%). Among the survived plants, dose 80 Gy had the highest percentage of mutants (69.2% mutants) followed by dose 60 Gy (36.4% mutants) and dose 40 Gy (25.8% mutants). Dose 80 Gy was thus identified as the suitable LD_{50} due to the percentage of successful survival plants of 69.2%. These survival plants were the mutated plants. Hence, this dose was recognized to produce the highest percentage of mutants.

Preliminary studies on the physical observation of the mutant lines (80 Gy) showed some alterations, such as the unique surfacing of stems with obvious appearance of nodes and internodes emerging above ground and needle-like lemongrass leaves in first generation, V_1 (Figure 1). This unique long stem can be exploited for the mechanical harvesting of lemongrass. Harvesting lemongrass manually is a main problem since its leaves have long, sharp-edged blades containing traces of silica (a glass ingredient) that give their rough edged blades with slicing power. By having this unique character, lemongrass could be harvested mechanically, probably with a machine such as a lemongrass reaper.

Interestingly, further analysis on physical observation of the mutant lines (80 Gy) during 12 months in open field have shown that flowering was initiated when plant reached maturity (6 months) (Figure 2). These flowers were found to reach full bloom in two weeks with almost 30% of the total inflorescence in the lemongrass clump were flowers. Besides that, cutting the flower inflorescence would initiate new inflorescence and flowering in the next two week, continuously.

Table 1: Number of plant survived and mutation percentage at different doses

Dose	Number of plant		Dead	Death Percentage (%)	Mutation Percentage (%)
	Normal	Mutant			
40 □	43	17	0	0.0	25.8
60 □	35	20	5	8.3	36.4
80 □	18	18	24	40.0	69.2



Fig. 1: Picture of nodes and internodes of lemongrass mutant line of 80 Gy. Left: The emergence of stem above the ground. Right: Lengthening of the stems with obvious appearance of nodes and internodes.



Fig. 2: Picture of flower from lemongrass mutant line 80 Gy. Left: Flowers of mutant line in open field growing. Right: The entire plant of lemongrass mutant with height more than 1.5m.

This frequent flowering behaviour of West Indian Lemongrass mutant line has definitely created a lot of remarkably potentials for further exploitation and new findings. As previously noted, West Indian Lemongrass is hardly seen to flower and thus difficult to do crossing [1, 15]. Perhaps, this important finding could give the chances to the plant breeders to initiate a plant breeding program for West Indian Lemongrass, *C. citratus* in the future.

CONCLUSION

In conclusion, this study had successfully identified the LD₅₀ for vegetative lemongrass *i.e.*, dose 80 Gy which gave an amazing new characteristics in West Indian Lemongrass. Mutant characteristic identified in V₁ generation was the unique surfacing of long above ground stem (not stalk) with obvious appearance of nodes and internodes together with frequent flowering behaviour. However, further evaluation on the essential oil yield production from this mutant line, particularly the flowers, should be analyzed to recognize other possible changes.

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