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# EVALUATION OF SEQUENTIAL APPLICATION OF SLASHING AND GLYPHOSATE FOR DROUGHT GRASS (*ISCHAEMUM MUTICUM* L.) CONTROL IN COCONUT PLANTATION

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## ABSTRACT

This study aimed to determine the effectiveness of sequential application of slashing and glyphosate for drought grass control under glasshouse and field conditions. Sequential application of slashing and glyphosate at 1.83 kg a.i. $ha^{-1}$ provided at least 70% reduction of node production, sprouting of nodes, stolon number, stolon length and viable stolon of drought grass under glasshouse conditions. In field experiment, slashing followed by two sequential application of glyphosate at 1.83 kg a.i. $ha^{-1}$ reduced total density and dry weight of drought grass by 58 and 54% respectively, as compared with untreated plots at three months after treatments. These results suggest that the sequential application of slashing and glyphosate is effective to provide drought grass control in coconut plantation.

**Keywords:** *Ischaemummuticum*, glyphosate, coconut and weed control.

## INTRODUCTION

Physical, mechanical and chemical methods are the options for weed management in plantations (Singh *et al*., 2014). Chemical method employing post emergence herbicide, however, becomes popular and increase dramatically because it is one of the most practical, effective and economical means of managing the weed problem (Wibawa*et al*., 2009). In Malaysia, application of post emergence herbicides has been the main weed management tools in young coconut plantations with 12 rounds per year. Glyphosate has been widely used as post applied herbicide to control weeds in the coconut plantations of Malaysia. Glyphosate is a foliar-applied, non-selective and systemic herbicide that controls many annual and perennial grasses and broadleaf weeds (Schilling *et al*., 2006). It acts by inhibiting 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) in the shikimate pathway. Upon application, glyphosate will be translocated throughout the plant and highly effective on actively growing parts (Schilling *et al*., 2006). EPSPS is an important enzyme in catalyzing the formation of the aromatic amino acid tyrosine, phenylalanine, and tryptophan (Mosanto and Mosanto, 2010).

Generally, there are a few possible responses when weeds are subjected to a post emergence herbicide. These include complete plant death, reduced growth rate, temporary cessation of plant growth, death of one or more shoot meristems, or no phytotoxicity (Mager *et al*., 2006). Incomplete weed control is defined as weed regrowth following a post emergence herbicide application and this may result in lower crop yield and reduced harvest efficiency (Sosnoskie *et al*., 2014). Survival of weeds following herbicide application may lead to seed production, thus replenishing the weed seed bank (Hartzler and Battles, 2001).

Gower *et al*. (2003) reported that incomplete control of weedswas observed with single post emergence application herbicides in corn fields.As a result, many growers have suggested that a sequential application of post emergence herbicide is required (Higgins *et al*., 1988). Advantages of sequential application include enhanced control of weeds (Schilling *et al*., 2006), season long suppression of weeds and it is able to give higher crop yield through reduction of crop competition (Bradley and Hagood, 2002; Hutchinson *et al*., 2003;Akin and Shaw, 2004). Control of weed species which undergo vegetative reproduction like stolon, rhizome, and/or tuber with a sequential applications of glyphosate have been reported in a number of studies (Bradley and Hagood, 2002; Akin and Shaw, 2004; Hart *et al*., 2005). Stoloniferous and/or rhizomatous grassy weeds such asmugwort (*Armetisia vulgaris*) (Bradley and Hagood, 2002), creeping rivergrass (*Echinochloapolystachya)* (Bottoms *et al*., 2011), Bermuda grass (*Cynodon*spp.) (Webster *et al*., 2003; Ferrel*et al*., 2005) and sourgrass (*Digitariainsularis*) (Correia *et al*., 2010; Giancotti *et al*. 2013) controlled by single or sequential applications of glyphosate have been demonstrated by many researchers in few countries.For example, Bradley and Hagood (2002) reported that three sequential applications of glyphosate at 4.5 kg a.i. $ha^{-1}$

provided at least 70% control of rhizomatous perennial weed, mugwort (*Artemisia vulgaris*) in Virginia.

Besides, slashing or mowing has been shown to be effective when integrated with post emergence herbicide on difficult-to-control perennial weed species (Renz and DiTomaso, 2006; Johnson and Davies, 2014). Slashing or mowingbefore applying a post emergence herbicide may improve herbicide contact on lower leaves or vegetative parts of the weed (Hunter,1996). Drought grass (*Ischaemummuticum*), a stoloniferous and perennial grassy weed that found in coconut plantation have been reported as an aggressive colonizer of open and disturbed habitats which can develop into huge thickets in drainage channels and ditches (Ipor and Baki, 1992). Furthermore, this weed species is able to scramble several meters high among and over shrubs, capable of spreading prolifically from rooting at the nodes when they touch the soil with numerous joints and also its stolon can grow up to 5 m or more (Ipor and Baki, 1992).Barnes (1990) has documented that glyphosate, sodium chloride, paraquat and MSMA in combination with diuron, can be used to control drought grass. However, to date, information on effectiveness ofsequential applications of slashing and glyphosate on the control of drought grass is still scanty.This study aimed to evaluate whether the sequential applications of slashing and glyphosate is effective to provide good control of drought grass.

## MATERIALS AND METHODS

**Experimental site:**Two experiments were conducted in a glasshouse (5.24°N, 103.05°E) located in School of Food Science and Technology, University of Malaysia Terengganu and field experiment located at Agricultural Research and Development Institute of Malaysia (MARDI), Hilir Perak Station, TelukIntan, Malaysia (3.53°N 100.51°E).

**Herbicide and plant materials:** Glyphosateiso propylamine (Sentry, 41.0% (w/v) was purchased as standardfrom CP manufacturing Pvt. Ltd., Klang, Selangor, Malaysia. Drought grass (*Ischaemummuticum)* was chosen as bioassay species in this study because it is one of the dominant stoloniferous weed species found at a coconut plantation of Malaysia. Stolons of drought grass were collected from a coconut plantation of Hilir Perak (3̊ 53’ 42”N; 100̊ 52’ 0”E). Then, the stolons were propagated in a glasshouse, University ofMalaysia Terengganu (UMT), Terengganu and maintained at a relative humidity of 78-80% and a temperature of 28-30ºC with 12h photoperiod and photosynthetic photon flux density (PPFI) of 800 ± 200 mol $m^{-2}s^{-1}$.

**Phytotoxicity of glyphosate under glasshouse conditions:** Stolons of drought grass were harvested from mature plantsgrowing in the glasshouse and cut to the three node segments with approximately 20 cm length. Two segments were planted in each tray (40 by 30 by 5 cm) containing 1500g silt loam soil(76.4% silt, 22.96% clay, 0.64% sand, pH 4.9, 10.4% organic carbon) at a depth of 2 cm and placed under glasshouse conditions. After transplanting, the stoloniferous plants were left for 2 weeks for acclimation. Water was applied using a watering can as needed. All the drought grass plants wereslashed to a height of 10 cm, leaving cut stems with no foliage. Then, a series rate of glyphosate was applied at 0, 0.46, 0.92 or 1.83kg a.i. $ha^{-1}$ where the recommended rate is 1.83 kga.i. $ha^{-1}$ and each glyphosate application was carried approximately two weeks after slashing. Meanwhile, untreated stolons grown in soil moistened with water were served as control. Glyphosate application was made using a compression sprayer (Matabi Style 7; Goizper, Bergara, Spain) with a flat-fan nozzle, delivering a spraying volume of 1100 L $ha^{-1}$ at 200 kPa. One month after the treatments,the plants were removed from the trays and the roots were washed with tap water. Numbers of node production, sprouting, and stolon production were counted and stolon length were measured. After measurement, the plants were transferred to new trays and left for four weeks before the second slashing was carried out.Two weeks after slashing, asecond application of glyphosate at 1.83kg a.i. $ha^{-1}$ was done as described previously. Visual injury of drought grass, where 0% was no plant injury and 100% was plant death (Frans *et al*. 1986) were performed while the number of node production, sprouting production, stolon production, viable of stolon and stolon length of drought grass were then recorded one month after the treatments.

**Phytotoxicity of glyphosate under field conditions:** Each plot measuring 20 $m^2$ (4 m width x 5 m length) of an area was established in December 2013. A total of two treatments including untreated plots were carried out at the inter-row areas of coconut trees as follows: T1: Weed checks (Control), where weeds were not treated at the inter-row areas. T2: Weeds at the inter-row areas were managed with sequential mowing and glyphosate applications at 1.83 kg a.i. $ha^{-1}$. Weedy plants were mowed at 5-10 cm height using a tractor (New Holland SE37 0095) and brush cutter (Mitsubishi TL33 engine) and left for two weeks prior to the first glyphosate application. The weedy plants were sprayed with glyphosate twice at one month interval. Glyphosate was applied using a knapsack sprayer (Dofra, Malaysia) at 200 kPa with a flat-fan nozzle, delivering a spraying volume of 1100 L $ha^{-1}$ in the morning.Weed density and dry weight were evaluated at two months after the first glyphosate treatment by placing two quadrates (1m x 1m each) randomly in each plot. Drought grass appeared in the quadrats were harvested and then counted to obtain density. The dry weight was determined after drying the samples at the glasshouse for three weeks.

**Statistical Analysis:** All experiments were arranged in completely randomized design with five replications. The data of stolon length, node segments, visual score and percentage data of node segments, stolon, stolon length, and viable of stolon were checked for homogeneity of variance before being subjected to one-way analysis of variance. Means were separated using the Tukey test at 5% of significant level. Meanwhile, the data of sprouting, stolon production and the percentage data of sprouting werecompared using the non-parametric Kruskall-wallis test before subjected to Dunn's Multiple Comparison Test. For the field experimental data, the independent T-test was used to compare the mean between treated and non-treated plots. Differences were regarded as significant when the p-values were equal or less than 0.05 ($P \leq 0.05$).

## RESULT

**Phytotoxicity of glyphosate under glasshouse conditions:** Effects of sequential applicationof slashing and glyphosate on stolon length, node production, sprouting of nodes and stolon production of drought grass are presented in Table 1. The assessment was carried out based on the initial growth parameters to determine regrowth capacity of treated stoloniferous plants after sequential application of slashing and glyphosate.Second glyphosate application at 1.83 kg a.i. $ha^{-1}$ was able to reduce stolon length significantly ($p \leq 0.05$) regardless of any initial rate of glyphosate except for drought grass plants subjected to first glyphosate application at 0.92 kg a.i. $ha^{-1}$. Surprisingly, the initial stolon length was reduced from 40 cm per plant to 5 cm per plant after two sequential glyphosate applications at 1.83 kg a.i. $ha^{-1}$.

Similarly, node production of drought grass decreased significantly ($p \leq 0.05$) with second glyphosate application at 1.83 kg a.i. $ha^{-1}$ regardless of any initial rate of glyphosate. It is interesting to note that the initial node production of non-treated plant was 5 nodes per plant, it was reduced to only one node per plant after two sequential glyphosate applications at 1.83 kg a.i. $ha^{-1}$.Unlike node production, second glyphosate application at 1.83 kg a.i. $ha^{-1}$ had no significant effect on sprouting of nodes and stolon production irrespectively of any initial rate of glyphosate. However, zero sprouting of nodes was observed on drought grass plants subjected to two sequential glyphosate applications at 1.83 kg a.i. $ha^{-1}$. Furthermore, stolon production of the treated drought grass plants was less than the initial stolon number of untreated plants (1 stolon per plant) (Table 1).

The treatments were compared with non-treated control in order to determinethe percentage of reduction in selected growth parameters. Two sequential applications of glyphosate at 1.83 kg a.i. $ha^{-1}$ gave excellent drought grass control (Table 2). This treatment reduced node production, sprouting of nodes, stolon production and stolon length by 93 - 100% of non-treated plants. In comparison, sequential applications of glyphosate at 0.46 or 0.92 followed by 1.83 kg a.i. $ha^{-1}$ provided poor to moderate control of drought grass. Node production, sprouting of nodes, stolon production and stolon length of drought grass plants were reduced by 0 - 72% when subjected to these treatments. However, no significant differences were observed between both treatments in these growth parameters. Sequential applications of glyphosate at 0.46 or 0.92 followed by 1.83 kg a.i. $ha^{-1}$ reduced total viable stolon by 30 to 40 %. On the other hand, total viable stolon was reduced by 70% with two sequential applications of glyphosate at 1.83 kg a.i. $ha^{-1}$. Nevertheless, no significant differences were observed among these treatments in totalviablestolon.

**Table 1. Effects of sequential slashing and glyphosate applications on stolon length, node production, sprouting and stolon production of treated drought grass under glasshouse conditions.*** 

| Glyphosate application Time | Rate (kg a.i. $ha^{-1}$)** | Stolon length (cm) | Nodes (no.) | Sprouting (no.) | Stolon (no.) |
|---|---|---|---|---|---|
| 1 | 0.46 | 67.1 ± 10 c*** | 13 ± 2 d | 2.6 ± 0.5 bc | 1.6 ± 0.5 b |
| 2 | 1.83 | 48.6 ± 3 b | 6.7 ± 0.6 bc | 2 ± 0.8 ab | 1 ± 0.4 ab |
| 1 | 0.92 | 48.4 ± 9 b | 9.2 ± 1 c | 2.2 ± 1 bc | 1.2 ± 0 ab |
| 2 | 1.83 | 43.2 ± 1 b | 5.2 ± 0.5 b | 1.5 ± 1 ab | 0.8 ± 0.5 ab |
| 1 | 1.83 | 47.2 ±3 b | 6 ± 1 b | 1.7 ± 0.8 ab | 1 ± 0.4 ab |
| 2 | 1.83 | 5.1 ± 11 a | 0.8 ± 2 a | 0 ± 0 a | 0.2 ± 0.4 a |

Abbreviations: *Initial growth parameter values of non-treated plants: stolon length, 40 cm $plant^{-1}$; node, 5 $plant^{-1}$; sprouting, 0 $plant^{-1}$; stolon, 1 $plant^{-1}$. **Drought grass plants were slashed at 10 cm height and left for two weeks prior to each glyphosate application. Drought grass plants were sprayed with glyphosate for two times. Assessment was carried out one month after the first glyphosate application. Then, it was followed by second slashing and glyphosate application at 1.83 kg a.i. $ha^{-1}$. Assessment was then done one month after the treatment.***Mean within the same columns followed by the same lowercase letter indicates no significant difference at $P \leq 0.05$, as determined by a Tukey test.

**Phytotoxicity of glyphosate under field conditions:** In the inter-row areas, mowing followed by sequential applications of glyphosate at recommendation rate was examined. This treatment is slightly different as compared to the treatment in glasshouse experiment. This is because weedy plants appeared to be similar in size with height ranging from 10-15 cm one month after first mowing followed by glyphosate application treatment under field conditions. As a result, second mowing was not carried out. Table 3 shows the effectiveness of sequential mowing and glyphosate applications on reduction of weed density and weed dry weight three months after the first glyphosate application at the inter-row of coconut plantations. The results clearly show that sequential mowing and glyphosate applications at 1.83 kg a.i. $ha^{-1}$ reduced density and dry weight of drought grass significantly. It is noted that density and dry weight of drought grass was reduced by 58 and 54% respectively, as compared with non-treated plots.

**Table 2: Effects of sequential slashing and glyphosate applications on visual injury, node production, sprouting of nodes, stolon production, stolon length and viability of drought grass under glasshouse conditions.**

| Herbicides | Rate (kg a.i. $ha^{-1}$)* | Visual injury (%)** | Nodes | Sprouting | Stolon | Stolon length | Viable of stolon |
|---|---|---|---|---|---|---|---|
| | | | ---% reduction--- | | | | |
| Glyphosate | 0.46 fb 1.83 | 82 ± 2 a*** | 61 ± 4 b | 0 b | 72 ± 5 b | 47 ± 9 b | 30 ± 27 a |
| | 0.92 fb 1.83 | 83 ± 4 a | 68 ± 1 b | 29 ± 24 b | 72 ± 5 b | 58 ± 6 b | 40 ± 42 a |
| | 1.83 fb 1.83 | 98 ± 4 b | 95 ± 12 a | 100 a | 93 ± 15 a | 95 ± 12a | 70 ± 27 a |
| Non-treated control | 0 | 0 | 0 | 0 | 0 | 0 | 0 |

Abbreviations: fb, followed by. *Drought grass plants were sprayed with glyphosate for two times. The plants were slashed at 10 cm height and left for two weeks prior to each glyphosate application. Assessment was done one month after the second glyphosate treatment. **Drought grass visual injury was rated on 0 to 100% scale; 0% equals no plant response and 100% equals plant death. ***Mean within the same columns followed by the same lowercase letter indicates no significant difference at $P \le 0.05$, as determined by a Tukey test.

**Table 3: Effects of sequential mowing and glyphosate applications on reduction of weed density and weed dry weight at the inter-row areas of coconut plantations.**

| Treatment | Weed density (no $m^{-2}$) | Weed dry weight (g $m^{-2}$) |
|---|---|---|
| SLSH fb PE glyphosate fb PE glyphosate* | 19 ± 13 a** | 61 ± 15 a |
| Weedy (non-treated) | 46 ±16 b | 133 ± 12 b |

Abbreviations: *Weedy plants were mowed at 10 cm height using tractor and brush cutter and left to 2 weeks before first post emergence application of glyphosate at 1.83 kg a.i. $ha^{-1}$. Then, it was followed by (fb) second glyphosate application at 1.83 kg a.i. $ha^{-1}$, one month after the first glyphosate application. Assessment was carried out one month after the second glyphosate application. **Mean within the same columns followed by the same lowercase letter indicates no significant difference at $P \le 0.05$.

## DISCUSSION

Stolon plays an important role in the propagation of drought grass. Thus, best long term control of this grass could be achieved by reducing stolon number, stolon length, sprouting production, and viability of stolon.The effective initial rate of glyphosate for control of drought grass is important to determine the effectiveness of sequential glyphosate applications. It is found that first glyphosate application at a recommended rate of 1.83 kg a.i. $ha^{-1}$ is more effective than glyphosate application at 0.46 or 0.92 kg a.i. $ha^{-1}$ in slowing down the vegetative growth of drought grass. Interestingly, sequential application of glyphosate at 1.83 kg a.i. $ha^{-1}$ led to drastic reduction of drought grass growth. In contrast, when the first glyphosate application rate was lower than the recommended rate, it failed to reduce stolon length, node sprouting and stolon production of drought grass even though second application of glyphosate at the recommended rate was carried out (Table 1). Although visual injury of drought grass was more than 80% after being subjected to two sequential glyphosate applications, reduction of node production, stolon production, stolon length, sprouting of nodes and viable of stolon ranged from 1 – 100% depending on initial rate of glyphosate application (Table 1 &2). These findings imply that visual injury measurement was not a good indicator of overall effectiveness of sequential glyphosate application on control of drought grass.

The results of the present study have indicated that sequential mowing followed by glyphosate applications at a recommended rate of 1.83 kg a.i. $ha^{-1}$ provided good control of drought grass. Similarly, Bradley and Hagood (2002) reported that two sequential mowing conducted before an application of 1.7 kg a.i. $ha^{-1}$glufosinate enhanced the control of mugwort, a rhizomatous weed by 74% compared with either un-mowed mugwort (65%) or mugwort mowed once (48%) before an application of 1.7 kg a.i. $ha^{-1}$ glufosinate. Likewise, Renz and DiTomaso (1998) have demonstrated that mowing followed by single application of glyphosate at 3.33 kg a.i. $ha^{-1}$ to re-sprouting tissue increased effectiveness control of perennial pepper weed (*Lepidiumlatifolium*) compared with mowing or glyphosate application alone. These previous studies suggest that integration of slashed or mowed and herbicide application has potential to improve control of weed which reproduces through vegetative growth.

A few mechanisms have been proposed to explain enhanced control of weed through slashing or mowing. It is suggested that mowing can alter physiological and morphological processes in plants (Renz and DiTomaso, 2006) which may improve weed control. Hunter (1996) stated the enhanced control of Canada thistle (*Cirsiumarvense*) after mowing occurred due to canopy architecture which alter herbicide deposition pattern. This will increase the accumulation of herbicide at below ground tissues, thus providing long term control (Hunter, 1996). This hypothesis has been supported by Renz and DiTomaso (2004) who reported that integrated mowed and glyphosate enhanced control of perennial pepper weed due to the changes in the canopy structure of pepper weed after mowing. As a result, fewer aboveground sink and greater herbicide deposition at basal leaves that it can preferentially be translocated to root system. Furthermore, the delay between mowing and resproutingsynchronized maximal below ground translocation rates with herbicide application timing (Renz and DiTomaso, 2004).

Abundant researches have been conducted to evaluate weed response to sequential glyphosate applications at various rates. Bradley and Hagood (2002) reported that single application of glyphosate at 4.5 kg a.i. $ha^{-1}$ provided 63% mugwort biomass reduction but two sequential glyphosate applications did not enhance mugwort biomass reduction. However, mugwort control was increased to 73% with three sequential glyphosate applications. In the present study, two sequential glyphosate applications at 1.83 kg a.i. $ha^{-1}$ were able to reduce viability of stolon, node number, node sprouting, stolon number and stolon length of drought grass by more than 70%. This may be due to rapid absorption and basipetal translocation of glyphosate into the storage organs in sufficient quantities, thus killing the entire plants before the herbicide can be metabolized by the plants (Vencill, 2002). Nevertheless, Bottoms *et al*. (2011) have demonstrated that single applications of glyphosate at 1.12 kg a.i. $ha^{-1}$ managed to decrease multinode stolon production of creeping rivergrass by at least 70% compared to the non-treated control plants, implying that the effectiveness of glyphosate on control of stoloniferous weed is species dependent.

Many studies have been conducted to determine the efficacy of glyphosate under field conditions. Variable control on annual grassy weed, annual broadleaf or perennial weeds with glyphosate have been reported previously. For instance, Koger*et al*. (2007) reported that glyphosate at 0.84 kg a.i. $ha^{-1}$ provide 90 – 99% control on grassy weeds of johnsongrass and barnyardgrass and broadleaf of velvetleaf and prickly sida in Mississippi field experiment. Meanwhile, Ogogo*et al*. (2008) reported that although glyphosate at 1.8 kg a.i. $ha^{-1}$ effectively eliminated *Imperatacylindrica* L. but not all the rhizome were killed. Thus, they suggested the two integrated weed control of glyphosate at 1.0 kg a.i. $ha^{-1}$ plus *Mucunapruriens* var. *Utilis* a cover crop was the best treatment to control this weed species. On the other hand, Barker and Prostak (2014) reported that mowing followed by application of glyphosate at 2.8 kg a.i. $ha^{-1}$ gave strong initial control of grassy weed of tall fescue (*Festucaarundinacea*Schreb.) and increased in efficacy with the time but its effectiveness did not differ either mowed or un-mowed in season long control of vegetation.

**Conclusions:** A field experiment was subsequently conducted to verify promising results obtained from the glasshouse experiments. Sequential applications of slashing and glyphosate at 1.83 kg a.i. $ha^{-1}$ gave satisfactory control of drought grass by reducing of total weed density and total dry weight by 58 and 54% respectively, as compared with untreated plots. These results clearly shows that an integrated weed management by combining chemical, physical and mechanical methods is effective to provide broad spectrum of weed control while extending duration of weed control in the young coconut plantations for at least three months.

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