Journal of Agricultural Science; Vol. 14, No. 6; 2022
ISSN 1916-9752 E-ISSN 1916-9760
Published by Canadian Center of Science and Education

Effects of Foliar and Soil Application of Gibberellic Acid (GA3) at Different Growth Stages on Agronomic Traits and Yield of Rice (*Oryza sativa* L.)

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Received: March 30, 2022 Accepted: April 29, 2022 Online Published: May 15, 2022

doi:10.5539/jas.v14n6p55 URL: https://doi.org/10.5539/jas.v14n6p55

Abstract

Use of Gibberellic acid (GA3) application in rice cultivation for increasing the grain yield is well documented. However, improper and untimely use of GA3 could result in poor response to GA3 application. This study was aimed at to investigate the timing of application during different growth stages and mode of application of GA3 on the growth and yield of MR219, a popular Indica rice variety, released by Malaysian Agricultural Research and Development Institute (MARDI). Two commercial GA3 formulations, namely ProGibb SG containing 40% GA3 and ProGibb silica Granule containing 0.1% GA3 were used for foliar and soil application, respectively. GA3 was applied at late tillering stage and at 10-30% panicle heading stage. Interestingly, GA3 application as foliar spray during the early reproductive stage, ie at 10-30% panicle heading stage enhanced the grain yield significantly, recording over 27% grain yield increase over the untreated control. Moreover, two applications of GA3 at 7 days' interval has consistently given higher grain yield than single application. However, there is no significant difference in flag leaf characteristics, one thousand grain weight and milling qualities among different treatments. Our study has clearly illustrated that foliar application of GA3 at weekly interval at 10-30% panicle heading stage, can increase rice grain yield significantly.

Keywords: *Oryza sativa*, Gibberellic acid, foliar application, soil application, grain yield, flag leaf, milling

1. Introduction

Rice (*Oryza sativa* L.) is one of the most important staple food, providing a major source of calories and nutrients for more than half of the world's population. It is a direct source of calories for more people than any other crop, and it serves as the main staple for some 560 million chronically hungry people (FAO, 2017). It is one of the most important cereal crop of the world, grown under wide range of climatic conditions. It is grown in more than 110 countries across the world with an area of over 162 million hectares and production of 755 million tonnes with the average productivity of 4.66 metric ton per ha (FAO, 2021). More than 90 per cent of the world's rice is grown and consumed in Asia. With ever increasing population and at the current rate of population growth, the demand for rice is projected to increase by 32 percent, sufficient to meet the projected global rice demand by 2030. However, the production is faced with stiff challenges of decreasing availability of arable land, water, labour, chemical inputs and issues caused by climate change. Therefore, enhancing productivity of rice through different novel approaches and interventions including exogenous application of yield enhancers continue to play key role to meet the growing need.

Southeast Asia is one of the important rice producing region in the world, with over 48 million ha or almost 30% of the world rice production. It produced about 220 million tons of rice in 2018. Malaysia's total paddy cropping area is about 0.70 million hectares, which is the lowest in Southeast Asia. Moreover, the average yield per hectare is also one of the lowest in the region, with productivity varies widely among different granary areas due to location-specific factors such as agronomic practices, environmental conditions, cultivation areas, and soil fertility factors (Khazanah Research Institute, 2019). The Malaysian growth strategy plan, known as Vision 2020 was developed by the government aiming at developing and improving the country in all socio-economic aspects including agriculture (Alam et al., 2016; Lee & Baharuddin, 2018). In its growth plan to attain the vision, agriculture remains one of the important sectors of the country's economy, contributing over 12% to the national Gross Domestic Product (GDP) , providing employment for 16% of the national population, besides uplifting the standard of life among rural communities who are directly or indirectly involved in agriculture (Mannan et al., 2017). Malaysia is still a net importer of rice with the self-sufficiency level hovering between 60-70% (Khazanah Research Institute, 2019). The low level of productivity and self-sufficiency necessitated an urgent need to boost paddy production and productivity on national level. The expectation is to provide at least 75% of the countries rice needs from domestic production and to reduce the dependency on imports. While the cultivable paddy area is expected to remain same or shrink, to meet the target, the additional grain yield should come from higher grain yield and productivity. For achieving the target of increased production and productivity of local rice to 75% within the next three or four years, use of modern agronomic technologies, soil profiling and using better varieties with higher fertilizer use efficiency are going to play a critical role.

There are several potential approaches for enhancing the yield and productivity of rice, of which the major intervention tools are plant nutrition management, developing high yielding hybrid rice varieties through plant breeding, adopting modern agronomic practices and also judicious use of beneficial biostimulants based on microbial and non-microbial sources such as seaweed extracts, amino acids, protein hydrolysates, phosphites, humic acids, chitosan, peptides, etc, besides the use of plant hormones and growth regulators (Rouphael & Colla, 2020; Shahrajabian et al., 2021). All these tools form important components in practicing sustainable agriculture. The introduction of plant growth regulators (PGR) has added a new dimension to the possibility of improving the growth and yield of rice. Foliar applications of plant growth regulators, both natural and synthetic, have known to positively influence and improve crop growth and achieving higher yield under varied growing conditions. They are not phytotoxic under normal use rate and are typically applied at low dosages. In most cases, exogenous application of PGRs can influence the plant's hormonal status. PGRs also can significantly improve the performance of crop plants and their effects by increasing the crop yield and quality. It has been well documented that plant growth regulators including gibberellins (GA3), cytokinin, auxins etc are extensively studied plant growth promoting hormones, shown to be involved in variety of plant growth and physiological functions (Frankenberger & Arshad, 1995; Rademacher, 2016). GA3 is applied to cereal crops, ornamentals and orchards, where it plays a role in seed germination (Finch-Savage & Leubner-Metzger 2006; Chen et al., 2008; Urbanova & Leubner-Metzger, 2016), fruit growth enhancement (Li et al., 2011), and stem elongation (Dayan et al., 2012; Wang et al., 2017b; Shan et al., 2021), flowering (Sharma & Singh, 2009; Muñoz-Fambuena et al., 2012), response to abiotic stress (Colebrook et al., 2014), malting of barley (Briggs, 1963), and other physiological effects that occur in its interaction with other phytohormones (Steffens et al., 2006; Hedden & Sponsel, 2015). GA3 is also reported to improve effective portioning and translocation of photosynthates from source to sink in the field crops (Senthil et al., 2003). Studying on hybrid rice seed production, Gavino et al. (2008) reported that application of GA3 resulted in elongation of plant height, promoting panicle and spikelet exsertion, enhancing stigma exertion and longevity thereby increasing seed set rate and grain yield in rice, which is paramount in rice cultivation.

Use of GA3 for hybrid seed production is well researched and well documented. Peng (2014), and Ma and Yuan (2015) report that hybrid rice seed production technology has increased rice yield by 15-20% and has helped ensure China's food security over the past 30 years. Foliar application of GA3 during panicle emergence stage has been widely adopted as an essential technology for improving panicle exsertion of male sterile lines in hybrid rice seed production. Moreover, many investigations reported that application of GA3 at low concentrations after anthesis can impact stigma properties, including vigor of stigma (Zhou et al., 2017) which ultimately influence the yield potential of seeds. Earlier studies have shown enhanced seed yield during hybrid rice seed production due to GA3 application (Parihar et al., 2012; Thu et al., 2008). Yet, some studies also have indicated that application of GA3 after anthesis has adversely affected 1000-grain weight and seed setting rate, which resulted in reduction in grain yield (Dong et al., 2009, 2012). However, studies by Wang et al. (2019) reported that when GA3 was applied after anthesis significantly enhanced the stigma vigour index and correspondingly increased one thousand grain weight, thus improving seed vigor and increasing seed yield in hybrid seed production.

GA3 is among the most widely used PGRs in agriculture. It is typically applied to commercial crops by foliar spraying, but in some instances soil application of hormone also was reported although plant response to soil applied GA3 depend upon both the rate of GA3 biodegradation in the soil and the extend of GA3 adsorption onto soil solids (Anderson et al., 1988; Arteca et al., 1985; Struik et al., 1989; Sukifto et al., 2020). Interestingly, with these very few exceptions, in most of the studies, GA3 was used as foliar application and some as seed treatment. There are also very few studies on the use of GA3 as soil application on agricultural crops, especially paddy.

Dunand (1993) reported that paddy seed treatment with GA3 can dramatically increase emergence, stand, and seedling vigor of deep-seeded semi-dwarf rice planted into a well-prepared drill seeded seedbed which enable uniformity in the germination, increases the effectiveness of fertilizer and pesticide applications and other management practices based on growth stage of the crop. Paddy seed treatment with GA3 also reported to enhance seed germination with better seedling vigour under low growing temperature (Chen et al., 2005) and flooded condition (Watanabe et al., 2018). This field experiment was initiated to understand the influence of gibberellic acid on rice with the following three major objectives;

- To understand the best time of application based on the biological age of crop-late tillering/early booting stage or panicle exertion stage,
- To evaluate the best mode of application-foliar application or soil application, and
- To understand the effect on the frequency/number of applications-single or two applications.

2. Materials and Methods

This study was conducted during the period March to June 2021, in paddy variety MR 219, which is the first commercial indica rice variety in Malaysia developed by MARDI (Malaysia Agriculture Research and Development Institute) in 2001 (MARDI, 2013). It is a high yield variety, capable of producing over 10 metric tons per hectare. The experiment field was located at Parit 15, Simpang Lima, Sungai Besar, Selangor, Malaysia, situated in the Barat Laut Paddy Project, which is considered as one of the most important rice production areas in Malaysia. The geographic coordinates are 3°42′59.0976″N and 101°1′8.382″E latitude. The area was selected as it is plain, flat, and coastal and come under Integrated Agricultural Development Authority (IADA) and considered as rice granary of the state of Kuala Selangor. The weather during the study season was moderately warm during the study period. Total rainfall received during the experimental period was 621.43 mm, distributed over 111 rainy days. The relative humidity ranged from 74 to 77 per cent with a mean of 76 percent. The soil is considered as coastal plain soil having sandy topsoil and clayey subsoil, with mean soil pH of 5.5, while the mean concentration of organic carbon is in excess of 2%. The mean concentration of total nitrogen is about 0.26%, available P > 50 mg kg^{-1} and exchangeable K > 0.1 cmol kg^{-1}. Twenty-eight days old paddy seedlings were planted at 2 seedling per hill with a spacing of 25 × 16.5 cm to accommodate 24 hills per metre square. Good field cultural management and practices were followed during the experiment.

The experiment had 7 treatments and each treatment had 6 replications with the plot size of 40 sq. meter (5 × 8 meter). Buffer rows were maintained between the plots on all the four sides in order to prevent cross movement of nutrients or water or any other overlapping of treatment effect. In this study, we used ProGibb containing 40% GA3 and ProGibb Granule containing 0.1% active ingredient, supplied by Valent Biosciences LLC, USA. Treatment description is given in Table 1. ProGibb 40SG was used for foliar treatment and ProGibb 0.1G (silica based) was used for soil application. Foliar application of GA3 was given once or twice at two different growth stages, as per the treatment description-either at early booting stage or 30 days after transplanting (30 DAT) or at early panicle initiation stage (57 DAT), as outlined in Table 1. Soil application of GA3 was given either once or twice at booting stage, which is 30 days after transplanting. Fertilizer application schedule and other plant protection measures were carried out uniformly for all the treatments. For the sake of accuracy, the timing of application is also described in terms of BBCH scale, as described in BBCH Monograph (2001).

Table 1.Treatment description of the experiment

Tr.	Description	Method	No. of application	Application time
1	Untreated Control			
2	ProGibb 40SG-at 2.4 g ai/ha	Foliar	1	BBCH 43 (30 DAT)
3	ProGibb 0.1G-at 2.4 g ai/ha	Soil	1	BBCH 43 (30 DAT)
4	ProGibb 40SG-at 2.4 g ai/ha	Foliar	1	BBCH 52 (57 DAT)
5	ProGibb 40SG-at 2.4 + 2.4 g ai/ha	Foliar	2	BBCH 43 & 45 (30 & 37 DAT)
6	ProGibb 0.1G-at 2.4 + 2.4 g ai/ha	Soil	2	BBCH 43 & 45 (30 & 37 DAT)
7	ProGibb 40SG-at 2.4 + 2.4 g ai/ha	Foliar	2	BBCH 52 & 53 (57 & 65 DAT)

In recent years, the BBCH scale has been widely used to describe the phenology of many crops, including cereals (Meier et al., 2009). The fertilizer application of nitrogen, phosphorous and potassium was 275, 66 and 215 kg per ha, respectively. Fifty percent of total amount fertilizer was applied as basal fertilizer during the land preparation; the remaining amount of 50% fertilizer was applied in four split applications, at 10, 20, 65 and 90 DAT. Insect pests, diseases and weeds were closely monitored and controlled timely using appropriate and approved chemicals.

Flag leaf width (cm) and flag leaf length (cm) were measured on the main stem of fifteen uniform plants in each replication. Flag leaf area (cm^2) was measured one week before harvest and calculated following the formula adapted by Yoshida et al. (1976) as follows:

$$\text{Flag leaf area } (cm^2) = \text{Leaf length (cm)} \times \text{Maximum width (cm)} \quad (1)$$

The experimental plots were harvested leaving the border rows in order to avoid border effect. The matured crop was harvested from the net plot area and the grain was hand threshed, winnowed and sun dried to bring the moisture content to 14 per cent and the yield was recorded net plot wise and computed to kg ha^{-1}. In addition, number of panicles per plant, sink capacity (total spikelets number per panicle and 1000-grain weight) and source-sink relationship (total spikelets number to flag leaf area ratio and filled grains number per panicle) were also estimated. Assessment on grain weight and milling as per Rice Knowledge Bank (IRRI, 2010) were carried at the Laboratory of Food and Security, Universiti Putra Malaysia, Kuala Lumpur. Analysis of variance were calculated to determine the effect of treatments on the response to variables and treatment means were compared by Tukey test, using Minitab Version 18 test.

3. Results and Discussion

3.1. Effect on Flag Leaf

The uppermost leaf below the panicle is the flag leaf that provides the most important source of photosynthetic energy during reproduction. Flag leaf is metabolically active and has proved that the flag leaf, stem and head are the closest source food to the grain Effect of GA3 application on the flag leaf characteristics is given in Table 2.

Table 2. Effect of different GA3 treatments on flag leaf characteristics in paddy

Treatment	Description	Timing of application	Flag Leaf Length (cm)	Flag Leaf Width (cm)	Flag Leaf Area (cm2)
1	Untreated Control		27.04a	1.43ab	38.72ab
Single application					
2	ProGibb 40SG-Foliar at 2.4 g ai/ha	BBCH 43 (30 DAT)	25.09a	1.41ab	35.61b
3	ProGibb 0.1G-Soil at 2.4 g ai/ha	BBCH 43 (30 DAT)	25.35a	1.46ab	37.04ab
4	ProGibb 40SG-Foliar at 2.4 g ai/ha	BBCH 52 (57 DAT)	26.10a	1.37ab	35.92ab
Two applications					
5	ProGibb 40SG-Foliar at 2.4+2.4 g ai/ha	BBCH 43 & 45 (30 & 37 DAT)	24.85a	1.41b	35.23b
6	ProGibb 0.1G-Soil at 2.4+2.4 g ai/ha	BBCH 43 & 45 (30 & 37 DAT)	27.81a	1.50a	41.92a
7	ProGibb 40SG-Foliar at 2.4+2.4 g ai/ha	BBCH 52 & 53 (57 & 65 DAT)	26.13a	1.41ab	37.01ab
	P Value		p = 0.057	p = 0.006	p = 0.023
	Significance test		ns	*	*

Note. Means in column followed by different letter(s) are significantly different at $p < 0.05$.

No significant difference could be observed between the treatments and length of the flag leaf, however, significant increase in leaf width could be seen in the plants treated with GA3 as soil application, either once or twice, over other treatments. Correspondingly, the total flag leaf area also was impacted due to GA3 application. Largest flag leaf area was recorded in plants treated with two rounds of GA3 as soil application at 30 DAT and 37 DAT, which is significantly higher than flag leaf area in other treatments and followed by GA3 soil application at 30 DAT. Flag leaf area in other GA3 treatments did not differ significantly from that of flag leaves in the untreated control. In rice, the flag leaf is the main organ for photosynthesis, and it provides the main source of photosynthates assimilation required for plant growth and panicle development (Tian et al., 2015). Larger, wider, and longer flag leaves are generally considered ideal for high yield due to the interception of more radiation (Saitoh et al., 2002). Agronomic traits such as the weight of a thousand grains, the weight of the grains per panicle, and other characteristics related to yield are generally positively related to the size of flag leaf (Wang et al., 2020). Therefore, improving the traits of flag leaves is considered desirable for increasing grain yield in rice (Rahman et al., 2014). In another similar study, Tsai and Arteca (1985) reported that soil application of GA3 on certain dicot and monocot plants resulted in an increase in leaf blade area and/or length of sheath in plants like oat, barley, mung bean, squash, pepper, corn, sorghum and other millets. However, it is not always true as in some studies there is no such correlation established. Suratla and Robles (2004) while working on the hybrid rice seed male lines report that length of flag leaf and panicle was not significantly influenced by rate of GA3 application and there is no correlation to the yield. In another latest study, Makinoa et al. (2021) also reported that there is no correlation between the flag leaf size and grain yield. In their study, the shortest flag leaf showed a higher yield and negatively correlated to the grain yield and other agronomic traits. In the present study also we could not establish any direct correlation between GA3 application and flag leaf size and other agronomic parameters like number of spikelets, one thousand grain weight, grain yield, milling recovery etc. (Figure 1).

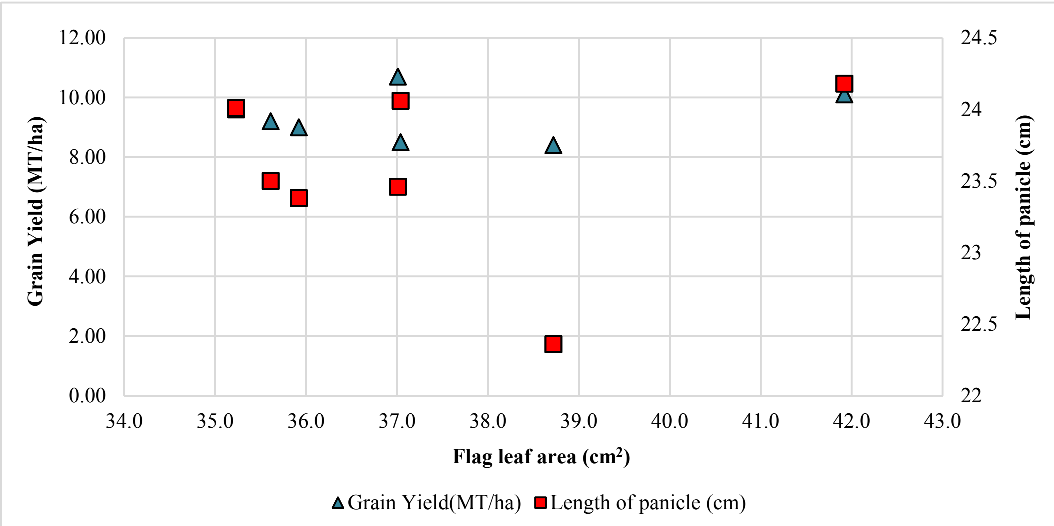


Figure 1. Correlation between size of flag leaf, length of panicle and grain yield in metric ton/hectare (MT/ha)

3.2 Effect on Panicles and Spikelets

Effect of GA3 on the number of panicles, spikelet and spikelet fertility is presented in Table 3. Although GA3 application has increased the panicle production significantly both in foliar and soil applied plots, treatments received GA3 application during late tillering/booting stage (30 DAT and 30 & 37 DAT) numerically had more tillers than the plots received GA3 at panicle heading stage. On the contrary, the number of spikelets was the highest in treatment received GA3 as foliar application, twice, during the panicle heading stage (57 & 65 DAT), which is 29% higher than untreated control. All the plots received GA3 application have recorded significant increase in spikelet count, except treatment received single application of GA3 as soil application during the vegetative growth stage (30 DAT). The highest number of spikelets in a panicle was recorded in treatment received two applications of GA3 as foliar at panicle heading stage which stands at 136.6 spikelets as compared to 117.1 spikelets recorded in the control plot, which is about 16% higher. Pattern of grain filling, however, has shown a different trend; foliar application of GA3 consistently recorded more filled grains per hill as compared to soil application of GA3 with two applications of GA3 outweighed the single application. Plots received GA3 twice as foliar at 30 and 37 DAT recorded the highest grain filling percentage of 83.8%, followed by 82.1% in treatment received single application of GA3 as foliar at 30 DAT. The results clearly reveal that even though the total number of spikelets are more in treatment received foliar application of GA3 during the panicle heading stage (one or two applications), the grain filling percentage was highest when GA3 was applied during late tillering/booting stage.

Table 3. Effect of different GA3 treatment on number of panicles and number of grains

Treatment	Description	Timing of application	Total No. of Panicles/hill	Total Grain/hill	Filled Grain/hill	% Filled grain	Spikelet/panicle
1	Untreated Control	Nil	15.25a	2864.83c	2049.66c	71.55c	117.13b
Single application							
2	ProGibb 40SG-Foliar at 2.4 g ai/ha	BBCH 43 (30 DAT)	18.21b	3221.33b	2645.00b	82.11a	127.07ab
3	ProGibb 0.1G-Soil at 2.4 g ai/ha	BBCH 43 (30 DAT)	17.92b	2448.83c	1771.50c	72.34c	121.87ab
4	ProGibb 40SG-Foliar at 2.4 g ai/ha	BBCH 52 (57 DAT)	18.58b	3269.17b	2437.00b	74.54b	130.07ab
Two applications							
5	ProGibb 40SG-Foliar at 2.4+2.4 g ai/ha	BBCH 43 & 45 (30 & 37 DAT)	18.42b	3342.17b	2801.50b	83.82a	127.07ab
6	ProGibb 0.1G-Soil at 2.4+2.4 g ai/ha	BBCH 43 & 45 (30 & 37 DAT)	18.42b	3428.67b	2636.83b	76.91b	129.33ab
7	ProGibb 40SG-Foliar at 2.4+2.4 g ai/ha	BBCH 52 & 53 (57 & 65 DAT)	18.25b	3701.67a	2974.17a	80.35a	136.53a
	P Value		p = 0.00	p = 0.00	p = 0.00	p = 0.00	p = 0.016
	Significance test		*	*	*	*	*

Note. Means in column followed by different letter(s) are significantly different at $p < 0.05$.

Panicle number in rice is dynamic and adjustable and plant hormones play an important role in regulating the production (Kariali & Mohapatra, 2007). More productive panicles per plant are believed to be closely associated with high seed yield per plant resulting high productivity. Li et al. (2003), and Mai et al. (2021) opine that the tillering ability comprises one of the most important traits of rice plants, which plays a significant role in the determination of rice grain yields, as it is positively related to panicle numbers per unit area. In the present study, we did record higher number of panicles due to GA3 application over the untreated control, especially plots received two applications of GA3 recorded highest number of panicles. Rice inflorescence (panicle) typically consists of a rachis (main axis), ten or more primary rachis branches and spikelets. In all GA3 treatments, the number of secondary branches are more than the primary branches. Number of primary and secondary branches in panicles appear to have clearly influenced by application of GA3, especially with two applications.

Table 4. Effect of different GA3 treatments on the panicle characteristics in paddy

Treatment	Description	Timing of application	Length of panicle (cm)	No. of primary branch/panicle	No. of secondary branch/panicle
1	Untreated Control		22.36b	9.46bc	14.0d
Single application					
2	ProGibb 40SG-Foliar at 2.4 g ai/ha	BBCH 43 (30 DAT)	23.50ab	10.63d	14.06d
3	ProGibb 0.1G-Soil at 2.4 g ai/ha	BBCH 43 (30 DAT)	24.06a	10.26c	16.66c
4	ProGibb 40SG-Foliar at 2.4 g ai/ha	BBCH 52 (57 DAT)	23.38ab	11.86ab	14.33d
Two applications					
5	ProGibb 40SG-Foliar at 2.4+2.4 g ai/ha	BBCH 43 & 45 (30 & 37 DAT)	24.01a	10.73abc	18.06b
6	ProGibb 0.1G-Soil at 2.4+2.4 g ai/ha	BBCH 43 & 45 (30 & 37 DAT)	24.18a	11.81abc	17.53bc
7	ProGibb 40SG-Foliar at 2.4+2.4 g ai/ha	BBCH 52 & 53 (57 & 65 DAT)	23.46ab	12.26a	17.80b
	P Value		p = 0.02	p = 0.000	p = 0.000
	Significance test		*	*	*

Note. Means in column followed by different letter(s) are significantly different at $p < 0.05$.

GA3 application does influence length of panicle as seen in Table 4, however, there is no major difference between treatments received GA3 applications at different time of plant growth and different mode. Interestingly, the highest panicle length was recorded in plots received soil application of GA3, as compared to foliar application. Number of primary and secondary rachis in GA3 treated plants have shown significant increase when compared to the untreated control. Treatments received two applications of GA3 either as foliar or soil, have recorded more number of primary and secondary rachis which is significantly higher than that of untreated control. Generally, larger panicle is associated with high number of grains per panicle resulting into higher productivity; therefore, significant increase in panicle length is desirable for obtaining higher yield (Tiwari et al., 2011). To increase rice grain production, yield-related components such as panicle number per plant, spikelet number per panicle, spikelet fertility, and grain weight are important considerations. The higher grain yield in GA3 treated plots was primarily attributed to the higher number of spikelets per panicle. In this experiment the total number of spikelets ranged from 117 in untreated plot to 136 spikelets in treatment received foliar application of GA3 twice, during panicle heading stage. In general, for a higher yield and productivity, it will be crucial to maintain the balance of agronomic traits; for instance, excessive tillering often gives rise to a decrease in grain production, because tillers can contend with the main culm for resources and negatively affect seed filling rate (Liu et al., 2013). Experimenting on the effect of GA3 on different rice varieties, Yamagishi et al. (1994) found an increase in number of spikelets per panicle in some varieties and they conclude that exogenous GA3 application could determine the number of spikelets in those varieties which are responsive to GA3 application. Mu and Yamagishi (2001) reported that the application of GA3 at the panicle initiation stage did not affect the spikelets per panicle of Nipponbare variety, while Akenohoshi variety increased its spikelets per panicle.

3.3 Effect of GA3 on Grain Yield

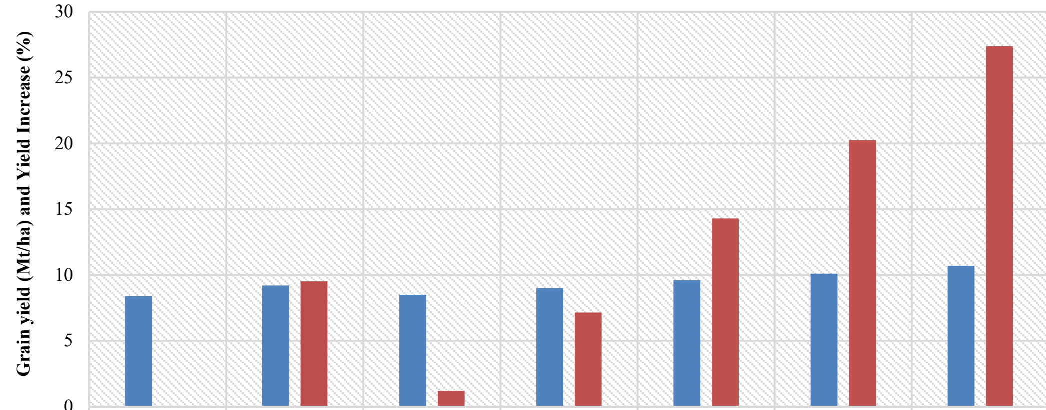
There was perceptible increase in rice grain yield observed due to GA3 application (Table 5 and Figure 2).

Table 5: Effect of different GA3 treatments on grain yield and percentage yield increase

Treatment	Timing of application	1000 grain weight (gm)	Grain Yield (MT/ha)	Yield increase (%)
Untreated Control		25.42 a	8.40 b	-
Single application				
ProGibb 40SG-Foliar	BBCH 43 (30 DAT)	26.64 a	9.20 ab	9.50
ProGibb 0.1G-Soil	BBCH 43 (30 DAT)	26.08 a	8.50b	1.20
ProGibb 40SG-Foliar	BBCH 52 (57 DAT)	26.48 a	9.00 ab	7.10
Two applications				
ProGibb 40SG-Foliar	BBCH 43 & 45 (30 & 37 DAT)	26.60 a	9.60 ab	14.30
ProGibb 0.1G-Soil	BBCH 43 & 45 (30 & 37 DAT)	25.68 a	10.10 ab	20.20
ProGibb 40SG-Foliar	BBCH 52 & 53 (57 & 65 DAT)	25.79 a	10.70 a	27.40
	P Value	p = 0.201	p = 0.011	
	Significance test	ns	*	

Note. Means in column followed by different letter(s) are significantly different at $p < 0.05$.

Among all the treatments, two applications of GA3 either as foliar or soil recorded higher grain yield when compared to single application of GA3. Highest grain yield increase of 27.4% was recorded in treatment received GA3 as foliar, twice, during panicle heading stage and followed by 20.2% yield increase in treatment received two applications of GA3 as soil application. The lowest grain yield increase of 1.2% was recorded in treatment received soil application of GA3 at 30 DAT. The results clearly reveal that two applications of GA3, as foliar application, during 10-30% panicle heading stage is the most ideal stage for getting maximum grain yield increase due to GA3 application and soil application of GA3 is less beneficial. Nevertheless, there is no apparent difference in one thousand grain weight due to GA3 application.



	T1 : UTC	T2 : ProGibb 40SG (30DAT)	T3 : ProGibb Granule (30DAT)	T4 : ProGibb 40SG (57DAT)	T5 : ProGibb 40SG (30DAT &37 DAT)	T6 : ProGibb Granule (30DAT&37 DAT)	T7 : ProGibb 40SG (57 DAT & 65 DAT)
Grain Yield (MT/ha)	8.4	9.2	8.5	9	9.6	10.1	10.7
% Yield Increase	0	9.52	1.19	7.14	14.29	20.24	27.38

Figure 2. Effect of GA3 application on the grain yield (Mt/ha) and percentage yield increase

GA3 application is a key to success of hybrid rice seed production in several countries, especially in China, and there are several studies with GA3 on the timing of application and the application rate to obtain the best seed production (Prasad et al., 1988; Gavino et al., 2008; Li & Yuan, 2000). In these studies, GA3 was used during different growth stages such as tillering stage (Ramesh et al., 2019), booting stage and early panicle initiation stage (Yamagishi et al., 1994; Cheng et al., 2011; Pan et al., 2013; Prakash et al., 2015), late heading stage (Pin et al., 2019), post anthesis stage (Wang et a., 2019; Zheng et al., 2018), etc. and different hybrids and cultivars responded differently. Tiwari et al. (2011) reported significant grain yield increase, besides increase in other agronomic traits, when GA3 was sprayed twice at heading and 50% panicle emergence stage. Similarly, working on hybrid rice seed production, Pin et al. (2019) observed that applying GA3 at 30% panicle heading stage increased seed yield significantly, which is much higher than applying at 0% heading and also 50% heading stage. Results of the present study confirm to the earlier finding of Cheng et al. (2011), and Prakash et al. (2015) that application of GA3 after anthesis/early grain filling stage resulted in highest grain yield. All these above studies clearly revel that the performance of GA3 on rice yield and other agronomic trails differ based on the varieties, stage of application and other agronomic conditions.

Results of the present study indicate that foliar application of GA3 produced higher grain yield when compared to soil application of GA3, which confirms to the earlier observation made by Runkle (2006), who opines that roots can also absorb GA3, but require a higher rate of application to get a comparable response. Since we used the same rate of GA3 both for soil and foliar application in the present experiment, perhaps the yield response to soil application is not as much as foliar application. Not much information is available on studies focusing on the number of GA3 application in paddy cultivation. In the present study, it is clearly demonstrated that two applications consistently performed better than one application, which is very similar to the observation reported by Suralta (2004), who demonstrated that two applications are much superior to single application in terms of grain yield and other agronomic traits.

Effect of GA3 on one thousand grain weight greatly vary in different studies. Pan et al. (2013) reported that GA3 on 1000-grain-weight and percentage of head rice of the earlier-flowered spikelets was greater than that of the later-flowered spikelets. Cheng et al. (2011) found that application of GA3 at the initial heading stage could increase the 1000-grain weight and seed setting rate of rice. However, Wang et al. (2017) reported that the one thousand grain weight was shown to be unaffected by the application of GA3. Yet some other studies have reported that application of GA3 after anthesis has an adverse effect on the 1000-grain weight and seed setting rate, resulting in yield reduction (Dong et al., 2009). This could be due to application timing and concentrations of GA3, which may not be ideal to get a positive response. In the present study, however, there is no significant influence on one thousand grain weight due to GA3 application, although there is numerical increase in all the GA3 treatments.

3.4 Effect of GA3 on Milling Characteristics

There was no apparent difference in milling recovery due to GA3 application in any of the treatments. However, there was a significant increase in head rice percentage in treatment received GA3 twice, either as soil application or foliar application (Table 6). Although all GA3 treatments had shown increase in percentage of head rice when compared to the control, highest value of 86.83% was recorded in treatment received two applications as foliar at 57 and 65 DAT, which is more than 15% increase in head rice as compared to the untreated check.

Table 6. Effect of different GA3 treatments on milling parameters

Treatment	Timing of application	Milling recovery (%)	Head rice (%)	% Increase in head rice	Additional head rice (Kg/ha)
Untreated Control		67.83 a	75.33 a	-	0
Single application					
ProGibb 40SG-Foliar	BBCH 43 (30 DAT)	68.00 a	79.33 ab	5.33	670.6
ProGibb 0.1G-Soil	BBCH 43 (30 DAT)	67.58 a	80.00 ab	6.19	305.1
ProGibb 40SG-Foliar	BBCH 52 (57 DAT)	68.00 a	79.33 ab	5.33	562.8
Two applications					
ProGibb 40SG-Foliar	BBCH 43 & 45 (30 & 37 DAT)	68.08 a	78.33 ab	4.21	827.1
ProGibb 0.1G-Soil	BBCH 43 & 45 (30 & 37 DAT)	69.75 a	83.00 ab	10.20	1556.8
ProGibb 40SG-Foliar	BBCH 52 & 53 (57 & 65 DAT)	66.42 a	86.83 b	15.31	1878.4
	P Value	p = 0.324	p = 0.044		
	Significance test	ns	*		

Note. Means in column followed by different letter(s) are significantly different at $p < 0.05$.

Grain quality in terms of head rice is one of the most important traits in evaluating benefits of PGRs on rice. There were significant effects on grain qualities by spraying exogenous plant growth regulator have been reported. Du et al. (2010) studied that the inferior grains plumpness in rice was enhanced by 5.5% when exogenous GA3 was applied at 5 days before flowering. Higher milling percentage and head rice recovery due to GA3 application has been reported in earlier studies (Prakash et al., 2015; Okasha et al., 2019). In the present study, both foliar and soil application of GA_3 enhanced the head rice percentage, but did not have any significant impact on milling. Paddy variety used in this study, MR219, is known for higher milling and head rice percentage, when grown under ideal growing conditions, with the head rice percentage of 74-76% (Sahari et al., 2018). Head rice percentage of 75% obtained in the untreated control in present study confirms the above observation, but at the same time opens up opportunity to enhance the quantity of head rice to over 80% with GA3 application. Interestingly, improvement in head rice due to GA3 treatment could produce an additional quantity of 305 to 1879 kg of head rice per ha as compared to the untreated control (Table 6).

4. Conclusion

Driven by both population and economic growth in several Asian and African countries, global rice consumption in the coming years will remain strong (Ricepedia, 2020). With further cultivation area expansion is likely to be in a slogger pace, global rice yield and productivity must increase faster, if the world's grain supply needs are to be stabilized at affordable levels for the billions of consumers. Globally, the rice production has to be at least 8–10 million tons more each year, which means an increase of 1.2-1.5% per annum over the coming decade, equivalent to an average yield increase of 0.6 t/ha during the present decade. Intervention with sustainable productivity enhancement tools with natural biostimulants can be one of the key tools in this direction. Plant growth regulators such as GA3 significantly influence agronomical traits in rice and can potentially result in significant increase in grain yield and head rice. The present study demonstrates that foliar application of GA3 is superior to soil application, when applied during the panicle exertion stage for enhancing grain yield.

Acknowledgements

The authors wish to thank Valent Biosciences LLC, USA for providing experimental samples of ProGibb 40SG and ProGibb silica Granule for the study. Thanks are also due to the technicians at Laboratory of Food and Security, Universiti Putra Malaysia for assistance in milling assessment.

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