Asian Journal of Crop Science

ISSN 1994-7879

OPEN ACCESS

Asian Journal of Crop Science

ISSN 1994-7879
DOI: 10.3923/ajcs.2023.16.23

Research Article
Effect of Gibberellic Acid on Malaysia Cytoplasmic Male Sterile Line During Hybrid Rice Seed Multiplication

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Abstract
Background and Objective: Poor cross-pollination or out-crossing between parental lines is the major constraint in hybrid rice seed production. One of the reasons is due to poor panicle exertion of the cytoplasmic male sterile (CMS) line. The aim of this study was to improve the out-crossing rate and grain yield during CMS seed multiplication. **Materials and Methods:** The study was conducted at the Centre of Excellent for Rice, MARDI Seberang Perai, Penang, Malaysia using a randomized complete block design with 3 replications. Four concentrations of gibberellic acid (GA_3) were applied to both 0047A CMS and 0047B maintainer lines when 10% of the panicles emerged out from the sheath. **Results:** The result showed that the application of GA_3 had significantly affected the plant height, panicle exsertion rate, seed setting, grain yield and harvest index of 0047A CMS. The internode of 0047A CMS also showed some similarities and variations between the untreated and treated plants (the CMS was applied with and 30 mg L^{-1} GA_3). **Conclusion:** The application of GA_3 at the concentration of 15 and 30 mg L^{-1} is recommended to improve the panicle exertion of 0047A CMS during CMS seed multiplication.

Key words: Exogenous hormones, out-crossing rate, panicle elongation, hybrid rice

Citation: Hashim, S., P. Ding and M.N. Fadzli, 2023. Effect of gibberellic acid on Malaysia cytoplasmic male sterile line during hybrid rice seed multiplication. Asian J. Crop Sci., 15: 16-23.

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Plant hormones are naturally synthesized by the plants and it is an organic compound synthesized in one part of the plant and translocated to another part, where in very low concentrations it causes a physiological response[1]. Besides naturally synthesized hormones, there is synthetically produced can be found in the market. This synthetic compound is known as a plant growth regulator (PGR). This PGR can be applied exogenously to plants. Previous studies have demonstrated that plants can respond to exogenous PGR[2]. An exogenous application of PGR affects the endogenous hormonal pattern of the plant, either by supplementation of sub-optimal levels or by interaction with their synthesis, translocation or inactivation of existing hormone levels[3].

The application of exogenous hormone is essential in the rice industry, particularly in hybrid rice seed production. This is because poor panicle exertion or natural deficiency of 'panicle enclosure' of a female parent or cytoplasmic male sterile (CMS) is one of the major constraints in hybrid rice seed production[4,5]. Thirty to forty percent of the panicles partially or fully remained in the flag leaf sheath, thereby making these florets unavailable for out-crossing or cross-pollination between the CMS and male parent (maintainer or restorer lines), and this situation consequently affects the seed setting and finally the grain yield[7]. Yin *et al.*[6] have revealed that the occurrence of male sterility in the CMS line caused a deficiency of indole-3-acetic acid in the panicle, which consequently caused a decrease of gibberellin 1 (GA_1) in the uppermost internode. The reduced GA_1 resulted in a shortened uppermost internode and therefore, the panicle failed to protrude out from the flag leaf sheath.

In China, foliar application of exogenous gibberellic acid (GA_3) was proven as a significant approach to improve panicle exertion and led to successful seed production in hybrid rice[7]. The application of GA_3 is also implemented widely in other hybrid rice producer countries such as Bangladesh[8] and Indonesia[2]. Besides improving panicle exertion through cell elongation, GA_3 was also identified to increase the duration of floret opening, improve the stigma exertion rate, lengthen the duration of stigma receptivity, adjust the plant height of both parents and increase the tillering ability of secondary and tertiary tillers so that they bear panicles[2,8,9].

High concentrations of GA_3 (150-300 g ha^{-1}) have been used in order to get high seed yield by hybrid rice seed growers in China[9]. However, outside China, the high cost of GA_3 limits seed growers to use only 45-50 g ha^{-1}. In Malaysia, 50 g ha^{-1} of GA_3 which is equivalent to 100 mg L^{-1} has been applied in local hybrid rice seed production. However, the concentration of GA_3 was used 20 years ago on imported hybrid rice lines. Since Malaysia has successfully developed a semi local hybrid rice parental line with poor panicle exertion, as this study was conducted in order to determine the appropriate concentration of GA_3 to be applied in local CMS hybrid seed production and its effect on plant growth and yield performance. Inter-cellular of the plant stem was also observed after harvest in order to determine the changes in the internodes of the CMS applied with GA_3 solution.

MATERIALS AND METHODS

Study area: The study was conducted at a plant house of Centre of Excellent for Rice, MARDI Seberang Perai, Penang, Malaysia (5.54°N and 100.47°E) during main-season, 2016 and off-season, 2016.

Planting materials and nursery management: A locally developed hybrid namely 0047A CMS/0047B maintainer was obtained from Gene Bank, MARDI Seberang Perai, Penang, Malaysia and were used in this study. The seed of 0047B maintainer was sown two days after the seed of 0047A CMS was sown in order to ensure flowering synchronization between parental lines. Eighteen days seedlings of both CMS and maintainer lines were manually transplanted to the pot with a size of 1x1x0.5 m. The row proportion or row ratio refers to the number of rows of the maintainer line to that of the CMS line in a seed production plot. In this study, the row ratio used was 2:5 with a spacing of 10x10 and 15x15 cm between the CMS and maintainer lines, respectively. A single seedling of CMS and two seedlings of maintainer was transplanted per point. The study area was isolated using more than 1.5 m height of nylon cloth as an artificial barrier during the flowering period to ensure the purity of the seed. Roguing activities were done during flowering and maturity to ensure the purity of the seed by removing the off-types plant. The fertilizer rate used was 120 kg N: 70 kg P_2O_5: 80 kg K_2O. Flag leaf clipping of CMS and maintainer lines was done at the booting stage to ensure easy dispersal of pollen and higher out-crossing between parental lines. Two third of the flag leaf blades were cut using a brush cutter (SUM328SE, Tanaka, Japan). Supplementary pollination was done by shaking the maintainer lines with bamboo sticks during flowering period. This operation was done 3-4 times in between 10 a.m. to 1 p.m. within 7 days.

Preparation of gibberellic acid: Preparation of different concentrations of GA_3 was done at Water and Soil Science Laboratory, MARDI Seberang Perai, Penang, Malaysia by referring to the method of Virmani[10] with slight modification. Four concentrations of GA_3 were tested in this study namely 0, 15, 30 and 45 mg L^{-1} GA_3 and the GA_3 solution was sprayed on the upper and middle parts of the plant stem for both CMS and maintainer lines. The area planted with CMS plants for each pot was 0.2 m^2. Ninety percent gibberellins A_3 (G7645, Sigma-Aldrich, Germany) powder in the amount of 0.17, 0.34 and 0.51 mg were weighed using an analytical balance (Sartorius, Germany) in order to obtain 15, 30 and 45 mg L^{-1} GA_3. The respective GA_3 powder was then dissolved in 50 mL of 90% ethanol before it was mixed with water because the powder does not dissolve in water. Five milliliter liquid dish detergent was added to the GA_3 solution. The detergent makes GA_3 solution stick to the plant for better coverage. Virmani[10] recommended that 500 liters of water per hectare need to be added to the GA_3 solution. Hence, in this case, distilled water was added according to the respective planting area.

Application of gibberellic acid: This study was conducted using a randomized complete block design with 3 replications. Gibberellic acid solution at the concentration of 0, 15, 30 and 45 mg L^{-1} was sprayed to both CMS and maintainer lines when 10% of the panicles emerged out from the flag leaf sheath and it was sprayed once a day for two consecutive days. The respective solution was sprayed from the upper to middle parts of the plant stem. Zero mg L^{-1} was considered as a control treatment. The spraying activities were done between 8 to 10 a.m. using a hand pump sprayer.

Data collection: A series of agronomic traits of the CMS were recorded throughout this study by selecting four hills randomly from each pot. The plant height of maintainer lines was also recorded in order to compare with the CMS lines. Plant height was recorded after the flowering period by measuring the plants from the soil surface until the tip of the highest leaf or panicle (whichever is longer) using a measuring tape. Other parameters such as number of productive tillers, duration of opening floret, angle of opened floret, stigma exertion rate (SER) and panicle exertion rate (PER) were also recorded. The SER and PER were determined and calculated according to Virmani[10].

Determination of selected yield components, grain yield and harvest index of cytoplasmic male sterile lines: At maturity, the hill which comprises of the stem, leaves and panicles of the plant were cut at about 2 cm above from the soil surface using a sickle. The samples were then brought to the laboratory and each panicle per hill was removed from the whole plant using a scissor. The panicle was then placed in a brown paper bag with a label and it was then oven-dried (UF30, Memmert, Germany) at 60°C until constant weight. Ten panicles were then randomly selected and data on yield components such as number of spikelet per panicle, 1000-grain weight and out-crossing rate were recorded.

Grain yield and harvest index: The remaining balance of the CMS plants per pot was harvested and the grain was removed from the whole plant parts and placed in a brown paper bag. The separated grain and the remaining plants or straw biomass were then oven-dried in an oven (UF30, Memmert, Germany) at 70°C for 72 hrs. The grain yield of CMS was recorded by weighing all the filled grain and it was adjusted to 14% moisture content. Moisture content of the grain was recorded using a moisture meter (GMK-303RS, G-WON, Korea). The grain yield recorded was then converted to per hectare basis. The weight of straw biomass was also recorded and converted to per hectare basis. The formula of harvest index (HI) was as follows:

$$\text{Harvest index} = \frac{\text{Grain yield (t ha}^{-1}\text{)}}{\text{Total straw biomass (t ha}^{-1}\text{)}} \times 100$$

Observation on the changes in plant internode of Malaysia cytoplasmic male sterile: The second internodes of the 0047A CMS plants were collected from the plants treated with 0 and 30 mg L^{-1} GA_3 during milking stage and immediately fixed in 70% formaldehyde-acetic acid-alcohol solution (FAA) for 2-3 days according to Wu *et al.*[11] with slight modification. It was then placed overnight in a vacuum oven. Samples were then washed with distilled water followed by post-fixed in 1% osmium tetroxide for 24 hrs. After the post-fixation, samples were rinsed at least 10 sec with distilled water to remove any free osmium and dehydrated through degraded series of alcohol (30, 40, 50, 60, 70, 80, 90 and 100%) at 2 min per change. Approximately 1 mm hand-cut transverse sections were critical point dried in a Baltec CPD 039 (Germany), mounted on aluminium stub and coated with gold in a sputter coater (JFC-1600, JEOL, Japan). The samples were viewed under a scanning electron microscope (JSM-IT100, JEOL, Japan) at 15 kV.

Statistical analysis: All the calculated data were tabulated and SAS version 9.3 (SAS Institute 2011, Gary, NC) was used to analyze the data using Analysis of Variance (ANOVA). Mean differences were compared by Duncan's Multiple Range Test at 5% level of significance[12].

RESULTS

Effects of gibberellic acid on agronomic traits of Malaysia cytoplasmic male sterile lines: Application of GA_3 had significantly affected the plant height of both parental lines (0047A CMS and 0047B maintainer) and panicle exertion rate of 0047A CMS as shown in Table 1. The plant height of 0047A CMS increased by 10.5-10.7, 13.0-13.2 and 18.9-20.5% than the control after GA_3 at the concentrations of 15, 30 and 45 mg L^{-1} was applied to the CMS plants during the initiation of heading stage (10% of the panicles emerged from the sheath), respectively. The plant height of 0047B maintainer was also increased as higher concentrations of GA_3 were applied to the plant. The plant height of the maintainer increased by 3.0-4.0, 5.3-5.7 and 9.7-10.5% as compared to control after GA_3 at the concentrations of 15, 30 and 45 mg L^{-1} was applied to the maintainer, respectively.

Panicle exertion rate of 0047A CMS was significantly improved as GA_3 was applied to the plants (Table 1). The PER of 0047A CMS increased by 7.5-8.9, 8.8-12.4 and 13.9-18.5% as compared to the control plants after the plants were applied with GA_3 at the concentrations of 15, 30 and 45 mg L^{-1}, respectively.

Effects of gibberellic acid on yield performance of Malaysia cytoplasmic male sterile lines: Analysis of variance as shown in Table 2 indicated that application of GA_3 significantly affected the seed set, grain yield and harvest index of CMS 0047A at maturity. A significant increase of seed set by 29.0-47.8% was found in 0047A CMS treated with 15 mg L^{-1} GA_3 as compared to the control plants. Application of GA_3 solution at the concentration of 30 mg L^{-1} caused a significant increase in seed set of 0047A CMS by 43.15-69.17% in the plants treated with 15 mg L^{-1} GA_3 solutions. However, as the GA_3 at the rate of 45 mg L^{-1} was applied to the plant, a significant reduction in the seed setting was recorded.

A similar finding was also observed in the grain yield of the 0047A CMS. It was recorded that the application of GA_3 at the concentration of 15 mg L^{-1} significantly improved the grain yield of the 0047A CMS by 80-133.33% as compared to control (Table 2). A significant increase in the seed set of 0047A CMS by 42.86-44.44% was also observed in the 0047A CMS applied with 30 mg L^{-1} GA_3 as compared to the 0047A CMS applied with 15 mg L^{-1} GA_3. An application of GA_3 up to 45 mg L^{-1} significantly reduced the seed set of 0047A CMS by 38.46-40% as compared to the 0047A CMS that was applied with 15 mg L^{-1} GA_3. Results also revealed that

Table 1: Effect of Gibberellic acid concentrations (GA_3) on agronomic traits of 0047A cytoplasmic male sterile and plant height of its maintainer, 0047B during seed multiplication

Factor	Plant height (cm±SD) 0047A	Plant height (cm±SD) 0047B	Number of tiller per sq meter (Mean±SD)	Duration of floret opening (min±SD)	Angle of floret opening (°±SD)	Panicle exsertion rate (%±SD)
GA_3 concentration (mg L^{-1})						
0	80.3±2.8^{cz}	89.4±1.6^{b}	240.0±31.3^{ab}	185.7±21.3^{a}	35.9±1.2^{a}	76.7±1.6^{b}
15	88.8±3.2^{b}	92.5±2.1^{a}	224.0±28.6^{ab}	183.8±18.4^{a}	38.9±1.1^{a}	83.0±2.3^{a}
30	90.8±3.1^{b}	94.3±1.5^{a}	266.8±32.5^{a}	187.0±25.6^{a}	38.6±1.4^{a}	84.8±0.4^{a}
45	96.1±2.7^{a}	98.4±2.2^{a}	216.6±33.5^{b}	184.7±18.4^{a}	38.4±0.1^{a}	89.1±0.1^{a}

[z]Means within the column followed by the same letters are not significantly different by DMRT at 5% and SD: Standard deviation

Table 2: Effect of Gibberellic acid concentrations (GA_3) on grain yield, yield components and harvest index of 0047A cytoplasmic male sterile during seed multiplication

Factor	Number of spikelet/panicle (Mean±SD)	1000-grain weight (g±SD)	Seed set (%±SD)	Grain yield (t ha^{-1}±SD)	Harvest index (Mean±SD)
GA_3 concentration (mg L^{-1})					
0	127.0±10.4^{az}	15.1±0.3^{a}	9.60±0.3^{c}	0.8±0.2^{c}	0.3±0.03^{b}
15	110.7±14.2^{a}	15.6±1.3^{a}	13.3±1.3^{b}	1.6±0.2^{b}	0.5±0.04^{a}
30	115.7±12.7^{a}	16.1±2.1^{a}	20.6±0.3^{a}	2.3±0.3^{a}	0.5±0.01^{a}
45	101.3±18.3^{a}	16.2±2.3^{a}	18.2±0.1^{b}	1.4±0.2^{b}	0.5±0.02^{a}

[z]Means within the column followed by the same letters are not significantly different by DMRT at 5% and SD: Standard deviation

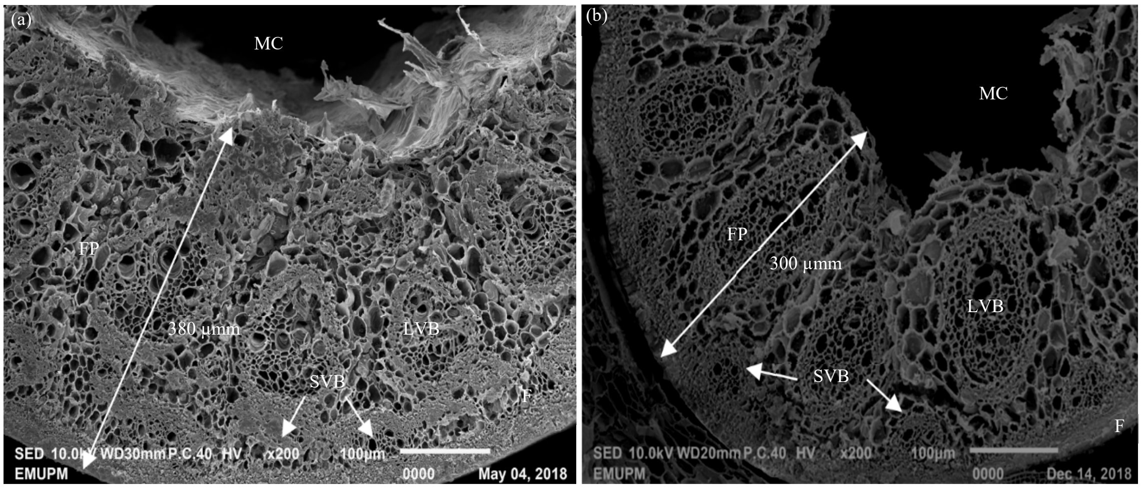

Fig. 1(a-b): Scanning electron micrographs of second internode of CMS 0047A applied with (a) 0 mg L^{-1} GA3 (x200) and (b) 30 mg L^{-1} GA3 (x200). The cross sections revealed that 90% of the internodal wall of both treated and untreated CMS internodes consists of fundamental parenchyma tissue (FP) while the other 10% is a cortical fibre (F). A hollow cylinder or known as medullary cavity (MC) was seen developed in the central part of both CMS internodes. A number of large vascular bundles (LVBs) and small vascular bundles (SVBs) were seen in the fundamental parenchyma tissue and cortical fibre, respectively

there was a significant increase in the HI of the 0047A CMS when the plants were treated with GA_3 at the concentrations of 15, 30 and 45 mg L^{-1} where, the HI increased by 63.6-70.4, 54.4-81.6 and 57.6-77.8% as compared to the control, respectively.

Changes in internodes of Malaysia cytoplasmic male sterile due to application of gibberellic acid: The cross section of the internode of 0047A CMS showed some similarities and variations between the untreated and treated plants (the CMS was applied with and 30 mg L^{-1} GA_3) as shown in Fig. 1(a-b), respectively.

When the internodes were viewed using a scanning electron microscope, it was observed that both CMS internodes have hollow cylinders developed in the central part of the internode and it is called lacuna or medullary cavity. The medullary cavity develops in the central part of the internode after the internode elongates. The internode tissue, except the medullary cavity, is known as the internodal wall and it consists of fundamental parenchyma tissues, vascular bundles and cortical fiber.

A number of small vascular bundles were seen in the cortical fiber while large vascular bundles existed in the fundamental parenchyma tissue of both CMS internodes. The large vascular bundles were arranged systematically in the fundamental parenchyma tissue of both CMS internodes.

However, a few differences were observed in treated and untreated CMS internode. The untreated CMS internode (Fig. 1a) was 26.67% thicker than the treated CMS internode (Fig. 1b). The fundamental parenchyma tissues of the treated CMS internode were seen bigger in size and the tissues were not as compact as the control CMS internode. It can be seen clearly that the shape of the large vascular bundles of the treated CMS was elliptical. Observation also shows that more small vascular bundles were seen in the treated CMS internode than in the untreated CMS.

In the large vascular bundles of both treated and untreated CMS internodes, several small vessels are differentiated outside the phloem Fig. 2(a-b). The vascular bundles of the treated CMS internode were seen as elliptical and they were wider than the untreated CMS as indicated by the arrows in Fig. 2(a-b), respectively. More number of lumens was also observed in the vascular bundles of the treated CMS and this situation contributes to the treated vascular bundles appearing more compact than the untreated CMS internode. The cell layers encircling the vascular bundles known as mestome sheath also differed between the untreated and treated CMS internode. The mestome sheath of the untreated CMS internode was thicker than the treated CMS internode as shown in Fig. 2(a-b), respectively. Figure 2(a-b) also showed that the mestome sheath of treated CMS looked collapsed and unclear.

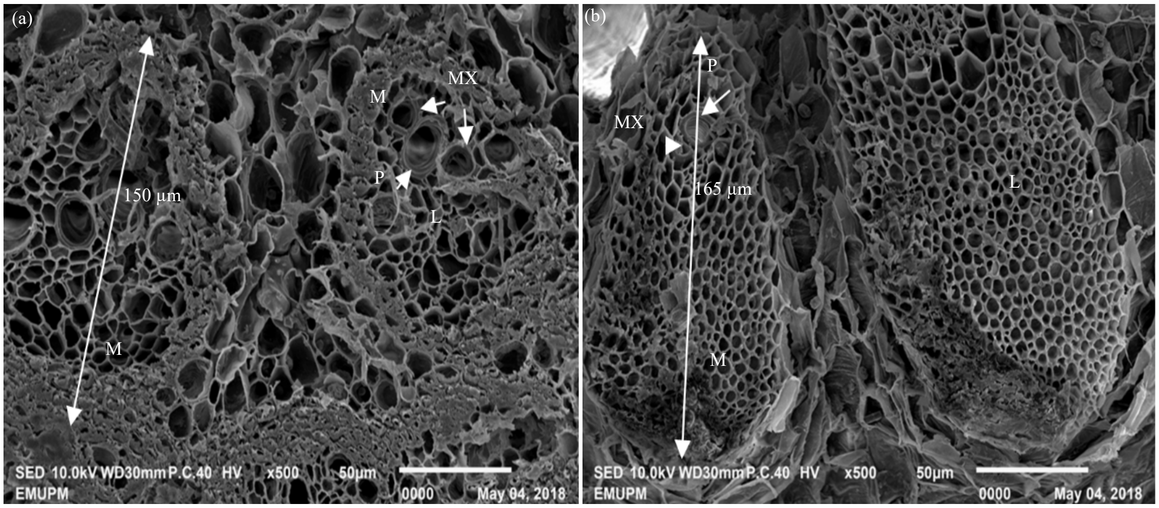

Fig. 2(a-b): Cross section of the large vascular bundles (LVBs) of CMS 0047A treated with (a) 0 mg L^{-1} GA_3 and (b) 30 mg L^{-1} GA_3 viewed under scanning electron microscope (x500). The orientation of the metaxylem vessels (MX) and phloem (P) of both CMS internodes looked similar and the phloem was surrounded with a number of metaxylem vessels. A layer which surrounded the cells of vascular bundle is known as mestome sheath (M) and it was thicker in untreated CMS internode than the treated CMS. A compact look of treated CMS could be seen due to its more number of lumens (L) as compared to the untreated CMS

DISCUSSION

Plant height is one of the important agronomic characteristics that contribute to the success of the out-crossing rate between the parental lines. The present studies indicated that the plant height of both 0047A CMS and 0047B maintainer lines was significantly higher than the control after the plants were applied with GA_3 solution. This result implies that the application of GA_3 increases the plant height by activating and elongating the stem of the plants. The result showed that an application of GA_3 at a concentration of as low as 15 mg L^{-1} significantly increased the plant height of both parental lines. This result suggested that endogenous gibberellin in the plant itself may be insufficient and the application of an exogenous hormone might be relevant to promote stem elongation and consequently increasing the plant height.

Based on visual observation, the stems of maintainer lines sprayed with 45 mg L^{-1} GA_3 were seen broken due to wind and supplementary pollination sticks used during flowering (unpublished data). This observation is aligned with the results shown in Table 1 where the plant height of 0047A CMS applied with 45 mg L^{-1} GA_3 was significantly higher if compared to 15 and 30 mg L^{-1} GA_3. This result indicated that excessive concentration of GA_3 may lead to uncontrolled stem elongation and due to these reasons, the taller plant is easily broken down, particularly during windy conditions. Therefore, it is necessary to find out an appropriate concentration/s of GA_3 because excessive application of GA_3 to the plants may associate with the mechanical strength of the stem or lodging resistance. It is also expected that once the stem had broken, the transportation of photoassimilates and water in the plants via the translocation process might be disturbed and this situation may affect the yield performance of the CMS.

Panicle exertion rate is an important parameter when applying GA_3 in hybrid rice because it indicates the efficacy of the exogenous gibberellins in stimulating and promoting plant growth. The present studies indicated that the application of GA_3 at the concentrations of 15, 30 and 45 mg L^{-1} significantly improved the panicle exertion of 0047A CMS as compared to control CMS. This result was similar to the plant height thus indicating that an elongated stem by the GA_3 is not the only factor affecting the plant height but as the stem elongated, it also encourages the hidden florets in the leaf sheath to protrude from the leaf sheath. Eventually, the possibility of these florets being out-crossed with their respective maintainer increases and consequently affecting its seed set and grain yield as well.

Seed setting or percentage of filled grain is the other parameter that indicates the efficacy of GA_3 in hybrid rice seed production. It is associated with the successfulness of out-crossing between parental lines during the flowering period[10]. Application of GA_3 to 0047A CMS showed different effects on its yield performance. The results indicated that plants treated with GA_3 showed significant changes in seed set, grain yield and harvest index as compared to the control plants. This result revealed that GA_3 plays a role in assisting the plant to get better yield performance. However, there was a reduction in seed set and grain yield of 0047A CMS after the CMS was applied with 45 mg L^{-1} GA_3. The seed set and grain yield of the respective treatment decreased by 10.84-12.44% if compared to the CMS applied with 30 mg L^{-1} GA_3. This result suggested plant height may play a certain role in causing broken panicles (data not published). As the stem elongated, the plant height also increased and lead to the weaker mechanical strength of the stem. By referring to Fig. 1, the fundamental parenchyma tissue of CMS treated with GA_3 looked less dense and bigger in lumen size as compared to the control CMS. Hence, a windy environment and bamboo sticks used for supplementary pollination during the flowering period may lead to fragile panicles and consequently contribute to the reduction of yield.

Rice is a gramineous plant and its stem is made up of a series of nodes and hollow internodes[13]. The internode in the plant part is associated with stem elongation caused by gibberellins[14]. Results from present studies demonstrated that the application of GA_3 had significantly improved some of the plant growth characters and yield performance of the CMS line. These findings were seen associated with the changes in the internode of the CMS caused by exogenous GA_3.

The inter-cellular studies revealed that there were a few differences found in the internode of GA_3 treated and control CMS (Fig. 1 and 2). It is obviously observed that the internode of the treated CMS showed different changes than the control CMS even though the same position of internode was observed (2nd internode from the top of the CMS). The internode of treated CMS was composed of many small vascular bundles (SVBs) as compared to control CMS. These SVBs were observed abundantly near the cortical fiber and inner part of the internode. It is understood that gibberellins elongated the stem by dividing more cells. As the cells divided including the vascular bundles, it is expected that more assimilates or photosynthetic products will be transported from the stem to the panicle. This finding was aligned with the statement of Fukuyama and Takayama[15] who mentioned that the phloem of vascular bundles in the stem plays a significant role in transporting photosynthetic products from source to sink. The divided cell or elongated stem by the gibberellins is not only contributing to the protrusion of the panicle from the leaf sheath, but it also increased the out-crossing rate and encourages more assimilates to be transported to the panicles as found in this study.

The fundamental parenchyma tissues of the treated CMS internode were seen thinner as compared to the internode of controls CMS as shown in Fig. 2. The tissues of the treated CMS internode were also seen as bigger in size and less compact as compared to the control CMS. The result suggested that as the stem elongated, the thickness and mechanical strength of the stem may be affected and this situation may lead to lodging incidence. It was proven that the application of GA_3 at the rate of 45 mg L^{-1} had caused lodging incidence. This lodging incidence was even obvious when the CMS was applied with 45 mg L^{-1} GA_3. There was a significant yield reduction recorded when the CMS was applied with 45 mg L^{-1} GA_3 as compared to 30 mg L^{-1} GA_3 CMS due to broken panicles.

CONCLUSION

It is recommended to use 15 and 30 mg L^{-1} GA_3 to improve panicle exsertion of 0047A CMS during CMS seed multiplication. This is because by using these rates, the panicles of the respective CMS exserted significantly as compared to the control treatment (0 mg L^{-1} GA_3). As panicle exsertion improved, the out-crossing rate also increased and consequently affecting the grain yield. Application of GA_3 at the concentration of 45 mg L^{-1} is not recommended because it is associated with prone to lodging incidence. This finding suggested that the mechanical strength of the stem of treated CMS might be weaker due to thinner fundamental parenchyma tissue of CMS treated with GA_3 as compared to the untreated CMS.

SIGNIFICANCE STATEMENT

This study reveals the effect of exogenous GA_3 on plant growth, yield performance and changes in the intercellular of CMS during CMS seed multiplication. Application of GA_3 up to 30 mg L^{-1} was seen to improve the seed setting and consequently the grain yield of Malaysia CMS. The findings may be used by seed producers, farmers and researchers who are involved in hybrid rice industries.

ACKNOWLEDGMENT

The authors would like to acknowledge Putra Grant (GP-IPS) given by Universiti Putra Malaysia (UPM/700-2/1/GP-IPS/2016/9487300) for funding this project.

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