Effect of Plant Growth Regulators, Basal Media Strength and Carbon Sources on Hylocereus Costaricensis (Red Dragon Fruit) Seed Germination

Nor Hasima Mahmod[1]& Abubakar Abdullahi Lema [2] & Siti Fazlina Kamarudin[3]& Norsyaya Shari[4]& Tajul Afif Abdullah[5]& Abdulrahman Mahmoud Dogara[6]

[1,2,3,4, &5]Department of Plant Science and Biotechnology, Faculty of Bioresources and Food Industry (FBIM), Universiti Sultan Zainal Abidin (UniSZA), Terengganu, Malaysia
[2]Department of Biological, Sciences, College of Natural and Applied Sciences, Al-Qalam University, Katsina, Nigeria
[6]Department of Biology, Faculty of Education, Tishk International University-Erbil, Kurdistan Region, Iraq
Correspondence: Nor Hasima Mahmod, Universiti Sultan Zainal Abidin (UniSZA), Terengganu, Malaysia.
Email: norhasima@unisza.edu.my

Doi:10.23918/eajse.v7i2p149

Abstract: Hylocereus costaricensis, commonly known as red dragon fruit or 'pitaya,' is famous for its purple-red flesh. It has achieved international recognition as an ornamental plant and secondly as a commercial fruit crop. Containing high level of vitamin C and antioxidants like betalans, hydroxycinnamates and flavonoids that includes edible fibre with numerous health benefits including weight loss, improve digestion, lower LDL cholesterol, and boost immune system. However, there is no large-scale production of dragon fruit because the seed storage viability is not reliable; the present study aims to employ tissue culture techniques supplied with different plant growth regulators (PGRs), the strength of growth media, and carbon sources. Seed germination was determined by growing the seeds in media enriched with benzyl aminopurine (BAP) 1 mg/L, 2 mg/L BAP, and 15 mg/L gibberellins (GA). Then, the seed growth was measured under different strengths of Murashige and Skoog (MS) basal media, which are full strength, 1/2, 1/4 and 1/8 strength, that promoted the seed germination the most. The percentage of seed germination was determined, and the effects of seed germination was measured by the period of seed to germinate, shoot length, and root length of the plants. In this study, all the dragon fruit seeds were successfully germinated. The seed shows the higher germination is MS + 2 mg/L BAP within 2 days, but the elongation of the shoot is not the fastest. The seed germination is effective in 1/8 media strength on day 2; the seeds are already germinated. However, the elongation for roots and shoots is the best in 1/2 media strength. The effect of carbon sources influences the fastest day to germinate, demonstrated in sucrose 30 g/L but not for root and shoot length. The 30 g/L glucose enhances the root length the highest, which is 2.48 cm. The average shoot length shows in TCS0 and TCS3 was similar, which was 1.38 cm. The effect of carbon sources that boost the shoot length the most is in TCS3 (2.04 cm), fructose 15 g/L. Those factors should be considered to enhance the best condition for dragon fruit planting and achieve the large-scale production,

Keywords: Dragon Fruit, Carbon Sources, Plant Growth Regulators, Media Strength, Germination

Received: September 17, 2021
Accepted: December 25, 2021
Mahmod, N.H., & Lema, A.A., & Kamarudin, S.F., & Shari, N., & Abdullah, T.A., & Dogara, A.M. (2021). Effect of Plant Growth Regulators, Basal Media Strength and Carbon Sources on Hylocereus Costaricensis (Red Dragon Fruit) Seed Germination. *Eurasian Journal of Science and Engineering*, 7(2), 149-162.

1. Introduction

Dragon fruit is a cactus vine species that is a member of the family Cactaceae (Patwary et al., 2013). Due of its unique appearance, its plant is attractive (Liaotrakoon, 2013). Nutritious and therapeutic characteristics of dragon fruits are gaining popularity (Sonawane, 2017) this fruit, it is regarded as an important economic fruit species worldwide (Rifat et al., 2019). Dragon fruit is not regarded as an indigenous crop in Malaysia but as one of the most popular fruits (Khandaker et al., 2020). Dragon fruit is found in three varieties: Hylocereus undatus (red peel with white flesh), Hylocereus polyrhizus (red peel with red flesh), and Hylocereus megalanthus (red peel with red flesh) (yellow peel with white flesh) but red peel with white flesh and red peel with red flesh is widely considered depending on the region or county (Yusof et al., 2020). In Malaysia, only two types of dragon fruit are suitable for planting: red and white flesh dragon fruit. Dragon fruit species and variants may be distinguished by their skin color and pulp (Pagliaccia et al., 2015). Due to customer demand, red-fleshed pitaya has always been more popular with growers than white-fleshed pitaya (Abdulrahman et al., 2018).

Pitaya is a notable tropical fruit that may be processed into various products, including jams, drinks, and candied fruits (Lee-Hoon, 2016). Additionally, several investigations suggest that the red flesh dragon fruit has a significant concentration of antioxidants and anti-proliferation characteristics, weight loss, improve digestion, lower LDL cholesterol levels in the blood, and enhance the immune system. Hydroxycinnamates contribute to cancer prevention, whereas flavonoids work on brain cells and blood arteries, lowering the risk of heart disease. Additionally, it protects against germs and fungi and aids in the body's overall functioning (Lema et al., 2022). Furthermore, compared to its competitors, H. costaricensis has better nutritional levels that help lower cholesterol, manage diabetes, regulate blood pressure, and support oral health. They are also capable of neutralizing heavy metals (Sheng et al., 2016). Dragon fruit potential yield in Malaysia is expected to reach approximately 10 to 12 metric tonnes (mt) per hectare (ha) each year. There are currently roughly 680 hectares of planted land, generating 6,407 mt or 6 million US dollars in 2017. About 36% of the Malaysian dragon fruit has a market of US$ 2 million for the last 5 years, mostly in Singapore, Taiwan and Hong Kong (Mohd et al., n.d.). In different countries, numbers of commercial producers gradually expand because of the attractive its price (Hossain et al., 2021).

Since seed viability of stored dragon fruit is not reliable, however, micropropagation has also been used. Micropropagation, also known as in-vitro culture, is a method for selectively producing disease-free, virus-free, herbicide-resistant, and high-yielding plants. In comparison to other conventional plant growth methods, cultivating plants in vitro functions as a means of conserving commercial, rare, and endangered plants rapidly (Dahanayake et al., 2018). Additionally, Plant tissue culture and micropropagation are methods for growing plants on a large scale from tiny portions of the source plant. Furthermore, people employ plant tissue culture since it has no adverse effect on the environment and may help rescue the plant from extinction (Thinesh et al., 2015). Because of its unusual ovule form, the seed has a low germination rate and a prolonged germination time. Furthermore, there is no large-scale production of dragon fruit planting supplies (Dahanayake et al., 2018). In response to this challenge, this research aims to enhance H. costaricensis seed germination in the presence of several plant growth regulators. The goal is to grow in various basal medium strengths and use a variety of carbon resources to boost seed viability and acquire a large number of high-quality dragon fruit plants.

2. Materials and Methods

2.1 Seeds Preparation

Fresh, ripe dragon fruit was obtained from a neighboring market in Besut, and the seeds were harvested, washed, and soaked in 70% ethanol for two minutes. After soaking the seeds in a solution of 1% Clorox and Tween 20, they were blotted dry on sterile paper and rinsed three times with distilled water (Halliru et al., 2021).

2.2 PGRs Media Preparation

The MS medium Sheng et al. (2016) was prepared and supplemented with PGR. The macro solution, micro solution, Fe EDTA, and vitamins, sucrose, PGR and GelriteTM were added in the order listed in the Table 1. Before autoclaving for 15 minutes, the pH of the medium was adjusted to 5.8. Then, each media was poured into a test tube in a quantity of 20 mL. Five replicates of each treatment had been grown. Each test tube contained a single dragon fruit seed grown, yielding a total of 20 duplicates for all PGR treatments. Cultures were kept on a rack in a growing environment with a 16-hour photoperiod and fluorescent light at a temperature of 25 ± 2oC (Jendy et al., 2019).

Table 1: Preparation of MS media with different PGRs

No	Stock/ingredient	Stock concentrations	TP0 (Control)	TP1 (MS+ 1mg/L BAP)	TP2 (MS+ 2mg/L BAP)	TP3 (MS+ 15mg/L GA)
1	Macro solution	10x	50 ml	50 ml	50 ml	50 ml
2	Micro solution	100x	5 ml	5 ml	5 ml	5 ml
3	Fe EDTA	100x	5 ml	5 ml	5 ml	5 ml
4	Vitamins	100x	5 ml	5 ml	5 ml	5 ml
5	PGRs	100x	-	1 ml	2ml	15ml
6	Sucrose	-	15 g	15 g	15 g	15 g
7	Gelrite TM (3g/L)	-	1.5 g	1.5 g	1.5 g	1.5 g

2.3 Preparation of Media Using Different of Media Strengths

The MS medium (Murashige & Skoog, 1962) was prepared and supplemented with PGR. The macro solution, micro solution, Fe EDTA, and vitamin, sucrose, PGR, and GelriteTM were added in the order listed in the Table 2. Before autoclaving for 15 minutes, the pH of the medium was adjusted to 5.8. Then, each media was poured into a test tube in a volume of 20 mL. The medium was supplied with various concentrations of basal media according to the treatments listed in Table 2. The items were then disinfected by autoclaving 15 minutes. Cultures were kept on a rack in a growth environment with a 16-hour photoperiod and fluorescent light at 25 ± 2oC.

Table 2: Preparation of MS media with different strength

No	Stock/ingredient	Stock	TMS0 (Control)	TMS1 (Half strength MS)	TMS2 (Quarter strength MS)	TMS3 (Eighth strength MS)
1	Macro solution	10x	50 mL	25 mL	12.5 mL	6.25 mL
2	Micro solution	100x	5 mL	2.5 mL	1.25 mL	0.625 mL
3	Fe EDTA	100x	5 mL	2.5 mL	1.25 mL	0.625 mL
4	Vitamins	100x	5 mL	2.5 mL	1.25 mL	0.625 mL
5	Sucrose (30 g/L)	-	15 g	15 g	15 g	15 g
6	GelriteTM (3g/L)	-	1.5 g	1.5 g	1.5 g	1.5 g

2.4 Preparation of Media Using Different Carbon Sources

The MS medium (Murashige & Skoog, 1962) with different carbon sources was prepared Table 3. Before autoclaving for 15 minutes, the pH of the medium was adjusted to 5.8. Each of the media was poured 20 ml per test tube. For each concentration, a seed was cultured on the MS medium in a test tube. Each of the treatments includes five replicates. Cultures were maintained in a growth room with a 16-hour photoperiod under fluorescent light at 25 ± 2ºC (Gao et al., 2021; Vijay, Shukla, & Saxena, 2016).

Table 3: Preparation of MS media with different carbon sources

No	Stock/ ingredient	TCS0 (Control)	TCS1 (15g/L sucrose)	TCS2 (30g/L Fructose)	TCS3 (15g/L Fructose)	TCS4 (30g/L Glucose)	TCS5 (15g/L Glucose)
1	Macro solution	50 mL	50 mL	50 mL	50 mL	50 mL	50 mL
2	Micro solution	5 mL	5 ml	5 mL	5 mL	5 mL	5 mL
3.	Fe EDTA	5 mL	5 mL	5 mL	5 mL	5 mL	5 mL
4	Vitamins	5 mL	5 mL	5 mL	5 mL	5 mL	5 mL
5	Sucrose	15 g	7.5 g	-	-	-	-
6	Fructose	-	-	15 g	7.5 g	-	-
7	Glucose	-	-	-	-	15 g	7.5 g
8	Gelrite™	1.5 g	1.5 g	1.5 g	1.5 g	1.5 g	1.5 g

2.5 Culture Conditions

All the reagents and equipment used in the tissue culture procedure, such as forceps, test tubes, beakers, micropipette, and also distilled water, were in aseptic conditions to prevent any contamination. The culture apparatus was autoclaved for 15 minutes and sprayed with 70% ethanol before used in the laminar airflow. Before using the laminar airflow, UV light was switched on for 15 minutes and wiped with 70% ethanol. All of the aseptic cultures were maintained in a culture room.

2.6 Statistical Analysis

The data collection was recorded based on the germinated seed. As a result, only mature shoots and roots were measured. Seed germination data was collected every day, while shoot and root length data

were collected. The percentage of germination observed at 0-7 days was used to compute the percentage of germination. ANOVA (Analysis of variance) was used to determine whether there were any statistically significant differences between the means of each group at p value ≤ 0.05 (Abdulrahman et al., 2019).

3. Results and Discussion

3.1 Seed Germination of Hylocereus costaricensis

All seeds inoculated successfully germinated regardless of the treatments, giving a percentage germination of 100%. The growth seeds, in general, are shown in Figure 1.

3.1.1 Effect of Different Plant Growth Regulators on a Day to Germinate

In this study, all the seed germinated in all the treatments. The application of plant growth regulators has significantly affected the seed germination of H. costaricensis Table 4. The seeds germination in TP2, which was in MS + 2 mg/L BAP, demonstrated the fastest seed germination with an average time of 2 days in this investigation (Figure 2). On the other hand, BAP showed the slowest seed germination in TP1, which was MS + 1 mg/L, with an average period of 6.2. Furthermore, the seed germinated slower in TP0 and TP3 than in TP2, with average days of 3.4 and 3, respectively.

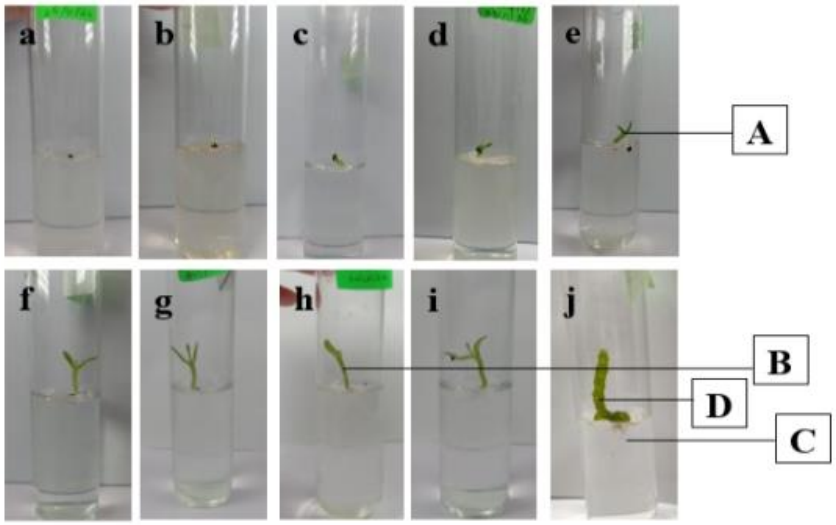

Figure 1: The observation of seed germination with PGRs and Basal Media in different day to germinate

Key: (a) Day 0, (b) Day 4, (c) Day 8, (d) Day 12, (e) Day 16, (f) Day 20, (g) Day 24, (h) Day 28, (i) Day 32, (j) Day 42. (A) Shoot, (B) Stem, (C) Root and (D) areole.

There is multiplication coefficient (5.41) other variety of dragon fruit, Halley's Comet, grown on medium MS enriched with 2.0 mg/L BAP. Sheng et al. (11) also reported similar findings. Their study looked at the proportion of seeds that germinated in a semi-solid MS medium supplemented with 1 ppm BAP and three dosages of Indole-3-butyric acid (IBA) (0.0-0.50.8 ppm). They claimed that the treatment with the most remarkable germination rate was a mixture of 1 ppm BAP and 0 ppm IBA (93.33 %). The combination of 3.6 ppm 2-4D and 1.8 ppm BAP resulted in high callus induction rate

of (75%) for H. costaricensis. This research showed that the addition of MS to 2 mg/L BAP increased seed germination.

Table 4: Day to germinate in plant growth regulators

Treatments	R1	R2	R3	R4	R5	Average
TP0	3	3	4	3	4	3.40 ± 0.55*
TP1	6	6	6	6	7	6.20 ± 0.45*
TP2	1	1	3	4	1	2.00 ± 1.41 *
TP3	2	3	3	3	4	3.00 ± 0.71*

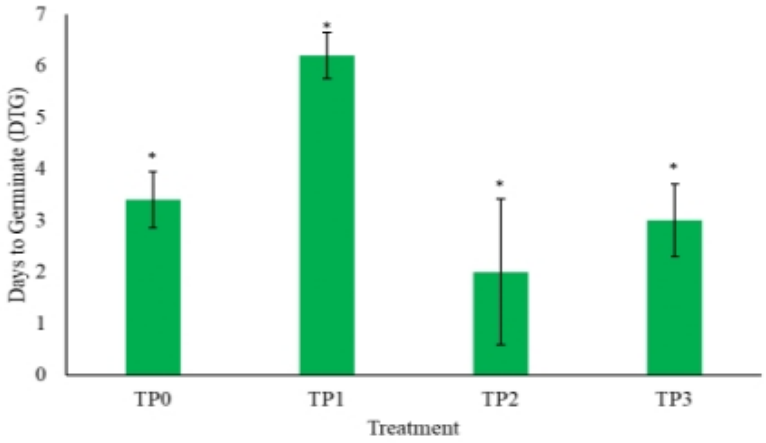

Figure 2: Day to germinate in plant growth regulators

3.2 Effect of PGRs on Root Length

The red dragon fruit treated with different plant growth hormone have higher root length except in TP3 Table 5 and figure 3. According to Figure 3, TP1 has the lowest root length 0.1 cm. The application of gibberellic acid (GA) in TP3 did not affect root length. According to the findings, MS medium without hormones aided root development more than MS medium with hormones. Recent research found that the MS medium supplemented with 0.1 mg/L BAP and 0.1 mg/L 1-Naphthalene Acetic Acid (NAA) combinations produced the greatest mean number of roots in the vertical culture position (Dahanayake & Ranawake, 2011). According to Bozkurt et al. (2020) MS medium supplemented with 1 mg/l IBA is the optimum medium for rooting. Furthermore, it has been established that MS medium without PGRs can also be used for rooting. In this investigation, it was discovered that TP0, which contains no PGR, is likewise the greatest therapy for promoting root length.

Table 5: Root length measured as effect of growth in different plant growth regulators

Treatments	R1	R2	R3	R4	R5	Average
TP0	1.1	2.7	0.2	1.9	1	1.38 ±0.95*
TP1	0.1	0.1	0.1	0.1	0.1	0.10 ± 0.00*
TP2	1.4	1	0.8	2	1.5	1.34 ± 0.47*
TP3	0.4	0.9	1	0.5	0.8	0.72 ± 0.23

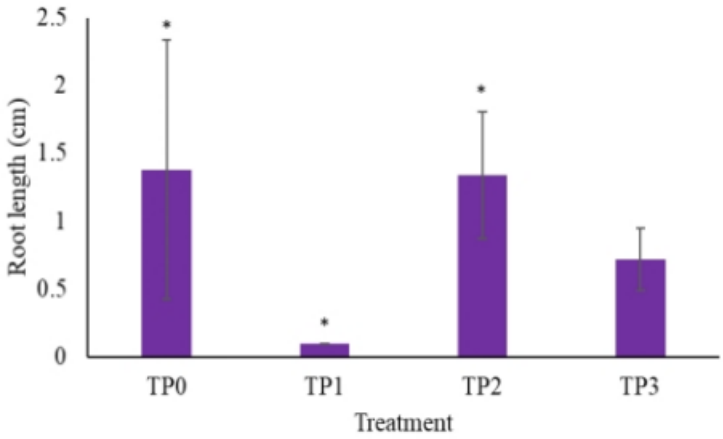

Figure 3: Root length measured as effect of growth in different plant growth regulators

3.3 Effect Different Plant PGRs on Shoot Length

The shoot length was measured on day 32 after inoculation. PGRs positively affected shoot growth, which increased shoot height Table 6. Figure 4 depicts the effect of various plant growth hormones on shoot length. TP2 had the highest shoot length 0.94 cm, compared to TP1 and TP3, which were 0.54 cm and 0.68 cm, respectively. On the other hand, the shoot has the longest length in TP0, which is 1.76 cm. This research showed that even in the absence of hormones, the shoot could stretch to its maximum length. Even at high BAP concentrations, vitrification could not be seen on axillary shoots (Monostori, Tanács, & Mile, 2010). However, this research found that a greater concentration of BAP may enhance shoot elongation more than a lower concentration of BAP. Furthermore, BAP induced morphological alterations in cactaceae shoot development in vitro (Torres-Silva et al., 2018). According to (Bozkurt et al., 2020), the medium containing 4.0 mg/L BAP had the most incredible shoot average per explants.

Table 6: Shoot length in different plant growth regulators

Treatments	R1	R2	R3	R4	R5	Average
TP0	1.5	2.5	1.8	1.4	1.6	1.76 ± 0.44*
TP1	0.5	0.5	0.7	0.5	0.5	0.54 ± 0.09*
TP2	1.2	0.9	0.7	0.7	1.2	0.94 ± 0.25*
TP3	0.5	0.7	0.6	1.1	0.5	0.68 ±0.25*

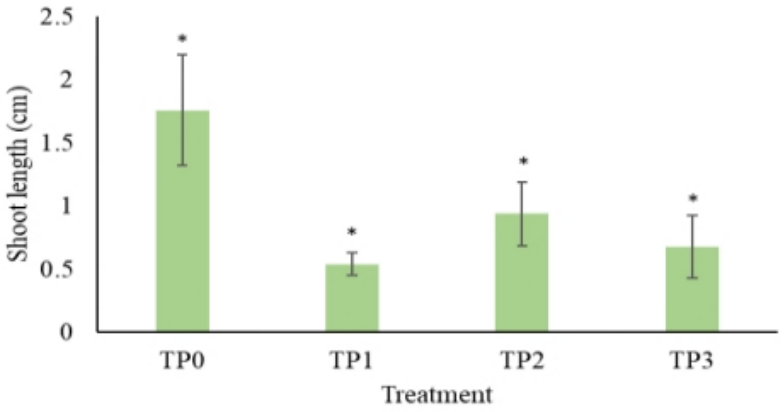

Figure 4: Shoot length in different plant growth regulators

3.4 Effect of Basal Media on a Day to Germinate of Hylocereus costaricensis

Basal media shows high effects on the seed germination Table 7. Based on the figure 5, the seeds of red dragon fruit in different media strength show that the rapid germination in the quarter strength (1/4) is in 2 days, the roots already show up. While in eighth strength (1/8), treatment shows the slowest seeds germination of dragon fruit which is 5 days needed for the seed to germinate. The induction and elongation of roots usually are better in half and full strength (Shekhawat et al., 2016). However, this has confirmed that quarter strength can induce seed germination faster.

Table 7: Day to germinate in different basal media strength

Treatments	R1	R2	R3	R4	R5	Average
TMS0	3	3	4	3	4	3.4 ± 0.55*
TMS1	2	3	2	2	3	2.4 ±0.55*
TMS2	1	2	1	1	3	1.6 ± 0.89*
TMS3	4	5	3	4	4	4.0 ±0.71*

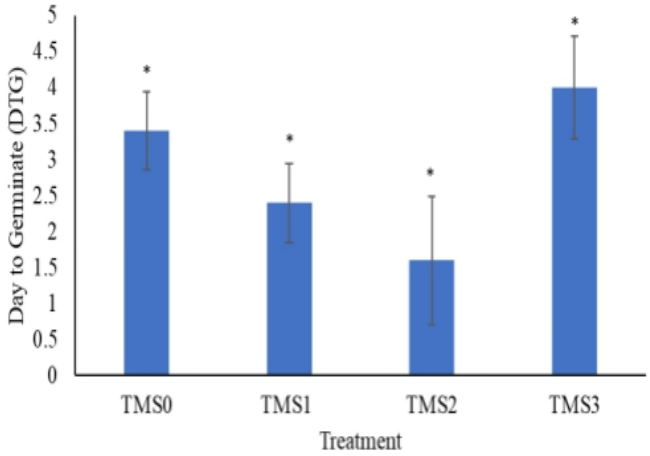

Figure 5: Day to germinate in different basal media strength

3.5 Effect of with Different Basal Media on Root Length

The findings can be elucidated based on Figure 6 and Table 8. It can be safely assumed that the root length is not altered by TMS0, TMS1, TMS2, and TMS3. The results demonstrate that there is no significant difference in media strengths treatment. According to Figure 6, there is no apparent change in root length across treatments. TMS1 increased root length by 0.18 cm greater than TMS2 (2.0), TMS3 (1.9), and TMS0 (1.38). TMS0 had the lowest root length of 1.38 cm, while TMS3 had the longest root length of 1.9 cm. Based on this finding, it was revealed that the influence of medium strength significantly impacted the roots. Half strength was shown to be the best strength for root length. (Rezali et al., 2017) Said in prior research for root formation that as medium strength reduced, the quantity and length of roots rose. However, the findings of this research reveal that when media strength decreases (half strength, quarter strength, and eight strength), so does root length. However, it cannot be used at full strength, and even at maximum strength, it cannot enhance root growth.

Table 8: Root length in MS medium supplemented with different basal media strength

Treatments	R1	R2	R3	R4	R5	Average
TMS0	1.1	2.7	0.2	1.9	1	1.38 ± 0.95
TMS1	2	3	1.8	1.5	2.6	2.18 ± 0.61
TMS2	2.1	1.4	1.6	1.8	3.1	2.0 ± 0.67
TMS3	2.5	1.5	2	1.7	1.8	1.9 ± 0.38

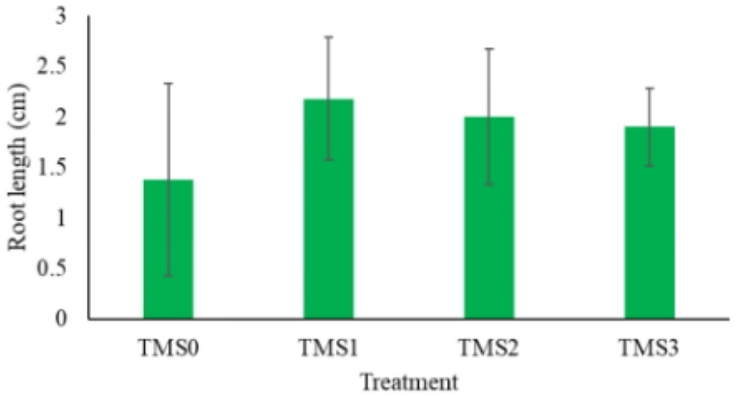

Figure 6: Root length in MS medium supplemented with different basal media strength

3.6 Effect of Different Basal Media Strengths on Shoot Length

The result obtained in Figure 7 and Table 9 respectively shows the application of media strength on shoot length of red dragon fruit. The shoot length was observed and measured in the different strength of media. Figure 7 depicts the elongation of the shoot length till day 32. There is no apparent difference in the shoot lengths of H. costaricensis. TMS1's shoot grew rapidly than TMS0, TMS2, and TMS3. TMS1 has a shoot length of 2.1 cm, followed by TMS2 (1.8 cm), TMS0 (1.76 cm), and TMS3 (1.36 cm). Previous research found that increasing the strength of the media reduces the height and number of shoots in solid media (Rezali et al., 2017). According to certain studies, full-strength influences shoot length development, followed by half strength. However, this research found that decreasing the strength of the medium slowed shoot development. Nonetheless, half-strength media is preferable to

full-strength media. Full strength, without any treatment, may also slow down the growth of shoot length (Kari et al., 2010). Figure 8 and Table 10 shows that there is no significant difference in seed germination. Sucrose is commonly utilized in plant tissue culture due to its growth-promoting properties. The medium supplemented with 30 g/L sucrose yielded the highest leaf number (Muslihatin et al., 2012). Sucrose is a key carbohydrate in most plants. It acts as a signaling molecule in regulating seedling development and germination (Xu et al., 2010). Even though the carbon sources are the same, the concentration of carbon sources affects the day of germination of the seeds. This study determined that the use of 15 g/L sucrose shows the best concentration and effect among other treatments.

Table 9: Shoot length in MS medium supplemented with different basal media strength

Treatments	R1	R2	R3	R4	R5	Average
TMS0	1.5	2.5	1.8	1.4	1.6	1.76 ± 0.44
TMS1	2.5	3.2	1.4	0.9	2.5	2.10 ± 0.93
TMS2	1.9	0.8	2	2.1	2.2	1.80 ± 0.57
TMS3	1	1.5	1.4	1.6	1.3	1.36 ± 0.23

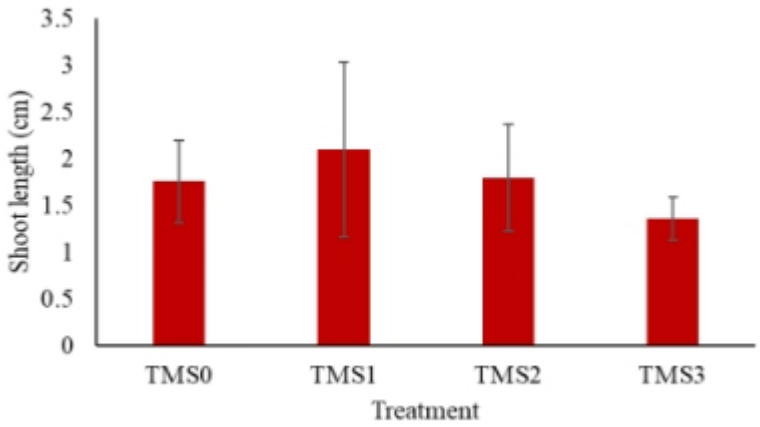

Figure 7: Shoot length in MS medium supplemented with different media strength

3.7 Effect of Different Carbon Sources on Root Length

Result obtained in Figure 8 and Table 10 shows the effect of carbon sources on root length of H. costaricensis. According to the graph above, the root length in TCS4 is the longest at 2.46 cm. TCS4 is treated with 30g/L glucose. TCS0 and TCS3 both show the same average shoot length of 1.38 cm. TCS2 (1.4 cm) has a 0.4 cm higher root length than TCS1 (1.1 cm), and it has the shortest root length among all treatments. Many studies on sucrose for development processes have been conducted. This study, on the other hand, demonstrated that glucose could hasten germination while inhibiting root development. The use of sucrose and fructose in various concentrations has no effect on excessive root growth. Excessive heat-induced fructose emits a toxic chemical called 5-hydroxymethyl-2-furaldehyde, which exacerbated hyper hydricity and lowered leaf water potential. However, 15 g/L and 30 g/L glucose promote root growth, with 30 g/L glucose being the best for root development. During the root induction phase, adding glucose to 49.0 M indole-3-butyric acid (IBA) resulted in an 85 percent rooting success rate. Furthermore, compared to other therapies, it was revealed that glucose

in the root induction medium is necessary for a more significant number of root initials to be recruited (Yasodha et al., 2008).

Table 10: Root length in MS medium supplemented with different carbon sources

Treatments	R1	R2	R3	R4	R5	Average
TCS0	1.1	2.7	0.2	1.9	1.0	1.38 ± 0.95
TCS1	0.7	1.5	0.5	0.9	1.9	1.1 ± 0.58
TCS2	1.5	1.5	0.4	2.4	1.2	1.4 ± 0.72
TCS3	1.4	1.5	1.7	0.8	1.5	1.38 ± 0.34
TCS4	3	3.1	4.7	0.5	1.0	2.46 ± 1.71
TCS5	2.3	0.5	2	2.3	2.9	2.0 ± 0.90

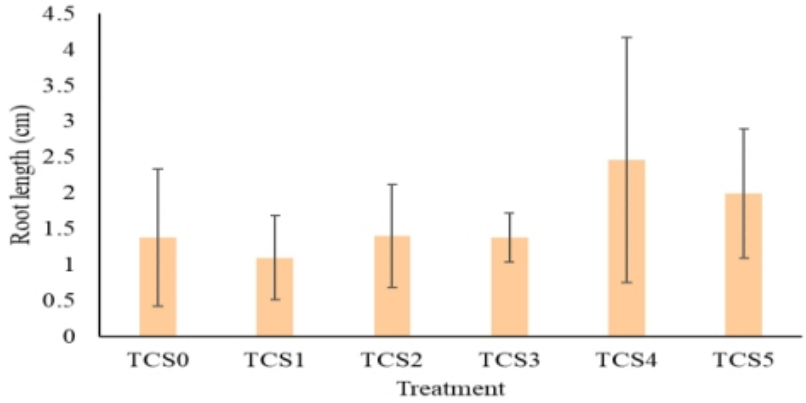

Figure 8: Root length in MS medium supplemented with different carbon sources

3.8 Effect of with Carbon Sources on Shoot Length

Result in Figure 9 and Table 11 shows the effect of carbon sources on shoot elongation. The length of shoots differed depending on the different carbon sources that had been used. The effect of carbon sources that increased shoot length TCS3 (2.04 cm) was found to be the highest, which is in 15 g/L fructose, whereas TCS5 (15 g/L glucose) has the shortest shoot length (1.28 cm). Even though, there was no significant difference in shoot length between treatments. Sucrose is commonly thought to be the best carbon source, but 15 g/L fructose and 30 g/L glucose outperformed the others in this study.

Table 12: Shoot length in MS medium supplemented with different carbon sources

Treatments	R1	R2	R3	R4	R5	Average
TCS0	1.5	2.5	1.8	1.4	1.6	1.76 ± 0.44
TCS1	1.5	2	2.5	0.9	0.6	1.5 ± 0.78
TCS2	2	1.2	0.9	1.8	1	1.38 ± 0.49
TCS3	2.1	2	2.2	1.7	2.2	2.04 ± 0.21
TCS4	1.7	2.5	2.6	1	1.9	1.94 ± 0.65
TCS5	0.7	1	1.2	1.3	2.2	1.28 ± 0.56

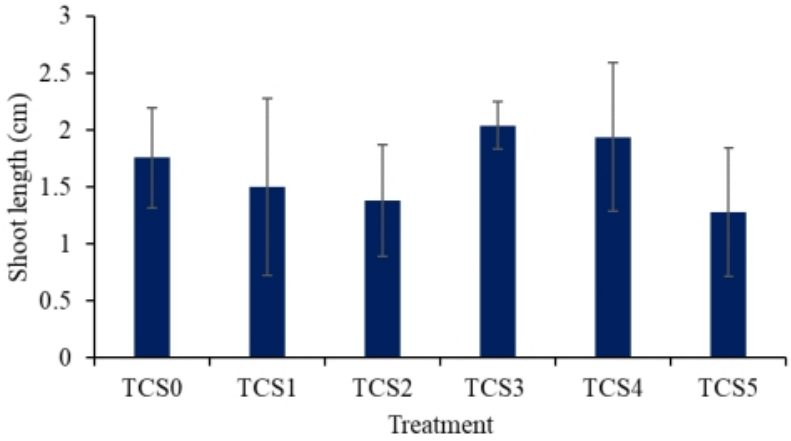

Figure 9: Shoot length in MS medium supplemented with different carbon sources

In general, glucose and fructose are thought to aid in the growth of specific tissues. This is demonstrated in this study because shoots grow faster in 15 g/L fructose and 30 g/L glucose than sucrose. Previous research on shoot response observed that as sugar concentration increased, root and shoot length decreased. Thus, the concentration of sugar used has influenced growth responses. However, this effect is only visible when the concentrations of fructose and glucose are different and not when the concentrations of sucrose and glucose are different. Yasodha et al. (2008) reported the increasing shoot multiplication using glucose as a carbon source was effective Apart from root growth, glucose has been discovered to operate as a signaling molecule for gene expression, cell proliferation, inflorescence creation, leaf expansion and senescence, and growth hormone-like activities. The findings of this study demonstrated that glucose could also aid in shoot division because the difference between the shoots in fructose and glucose did not show a contrasting length.

4. Conclusion

Dragon fruit seeds have successfully been grown with a 100% germination rate regardless of the treatment. The seeds grew healthily in all treatments and produced roots and shoots. All seeds provide positive feedback in all treatments on the growth of root and shoot. MS media supplemented with 2 mg/L BAP was the most effective in assessing numerous plant growth regulators for speeding germination and boosting root and shoot development. The MS medium enriched with 1 mg/L BAP, on the other hand, demonstrated the slowest root growth. The most rapid germination of the seeds was noticed in quarter media strength in terms of media strength, although root and shoots grew in media of half-strength significantly. The findings of the seed germination were incoherent from carbon sources. It was found that 15 g/L sucrose recorded the fastest germination of the seed. But glucose was excellent for root development in 30 g/L, while fructose concentrations were better for shooting elongation in 15 g/L. Finally, the effects of plant growth regulators, growth media strengths, and carbon sources were identified. The results give a suitable methodology for in vitro germination of seeds, in particular for cactus. The optimal in vitro cultivation procedure has been successfully devised for the germination of the Hylocereus costaricensis plant. The influence of plant development controls, the medium strengths, and carbon sources was detected to germinate the seeds of H. constaricensis.

References

Abdulrahman, M. D., Ali, A. M., Fatihah, H., Khandaker, M. M., & Mat, N. (2018). Traditional medicinal knowledge of Malays in Terengganu, Peninsular Malaysia. *Malayan Nature Journal, 70*(3), 349-364.

Abdulrahman, M. D., Hasan Nudin, N., Khandaker, M. M., Ali, A. M., & Mat, N. (2019). In vitro biological investigations on Syzygium polyanthum cultivars. *International Journal of Agriculture and Biology, 22*(6), 1399-1406.

Bozkurt, T., İnan, S., & Dündar, İ. (2020). Micropropagation of different pitaya varieties. *International Journal of Agricultural and Natural Sciences, 13*(1), 39-46.

Dahanayake, N., & Ranawake, A. (2011). Regeneration of Dragon fruit (Hylecereus undatus) plantlets from leaf and stem explants. *Tropical Agricultural Research and Extension, 14*(4), 85-89.

Halliru, B. S., Abdulrahman, M. D., Hamad, S. W., & Aliero, B. L. (2021). Identification of Weeds Resistant to Paraeforce (R) and Aminoforce (R) Herbicides. *Euphrates Journal of Agriculture Science, 13*(4), 20-26.

Gao, Y., Wang, Q.-M., An, Q., Cui, J., Zhou, Y., Qi, X., Li, L. (2021). A novel micropropagation of Lycium ruthenicum and epigenetic fidelity assessment of three types of micropropagated plants in vitro and ex vitro. *PloS one, [*Internet]. 2021;16(2 February):1–20. Available from: http://dx.doi.org/10.1371/journal.pone.0247666.

Hossain, F. M., Numan, S. M., & Akhtar, S. (2021). Cultivation, Nutritional Value, and Health Benefits of Dragon Fruit (Hylocereus spp.): A Review. *International Journal of Horticultural Science and Technology, 8*(3), 239-249.

Jendy, J. A., Ha, H. C., Zainuddin, R., Zawawi, D. D., & Ing, N. S. (2019). Micro-propagation of Aquatic Plant Brazilian Micro Sword (Lileaopsis brasiliensis). *Journal of Agrobiotechnology, 10*(1S), 29-34.

Kari, R., Lukman, A. L., & Zainuddin, R. (2010). Basal Media for in Vitro Germination of Red-Purple Dragon Fruit Hylocereus polyrhizus. *Journal of Agrobiotechnology, 1*, 87-93.

Khandaker, M. M., Syafiq, M., Abdulrahman, M. D., Mohd, K. S., Yusoff, N., Sajili, M. H., & Badaluddin, N. A. (2020). Research Article Influence of Paclobutrazol on Growth, Yield and Quality of Eggplant (Solanum melongena). *Asian Journal of Plant Science.* 19 (4): 361-371, 2020.

Lee-Hoon, H. (2016). Nutritional composition, physical properties, and sensory evaluation of cookies prepared from wheat flour and pitaya (Hylocereus undatus) peel flour blends. *Cogent Food & Agriculture, 2*(1). 231-245.

Lema, A. A., Mahmod, N. H., Khandaker, M. M., & Abdulrahman, M. D. (2022). Roselle anthocyanin stability profile and its potential role in post-harvest deterioration: A review. *Plant Science Today, 9*(1), 119–131-119–131.

Liaotrakoon, W. (2013). *Characterization of dragon fruit (Hylocereus spp.) components with valorization potential.* Ghent University,

Mohd, M. H. (n.d.). Status and challenges of dragon fruit production in Malaysia status and challenges of dragon fruit production in Malaysia. Status and Challenges of Dragon Fruit Production in Malaysia | FFTC Agricultural Policy Platform (FFTC-AP).

Monostori, T., Tanács, L., & Mile, L. (2010). *Studies on in vitro propagation methods in cactus species of the genera Melocactus, Cereus and Lobivia.* Paper presented at the XXVIII International Horticultural Congress on Science and Horticulture for People (IHC2010): International Symposium. 937, 255-261

Murashige, T., & Skoog, F. (1962). A revised medium for rapid growth and bio assays with tobacco tissue cultures. *Physiologia Plantarum, 15*(3), 473-497.

Muslihatin, W., & Ratnadewi, D. (2012). Effect of carbohydrate source on growth and performance of in vitro sago palm (Metroxylon sagu Rottb.) plantlets. *Hayati Journal of Biosciences, 19*(2), 88-92.

Pagliaccia, D., Vidalakis, G., Douhan, G. W., Lobo, R., & Tanizaki, G. (2015). Genetic characterization of pitahaya accessions based on amplified fragment length polymorphism analysis. *HortScience, 50*(3), 332-336.

Patwary, M. A., Rahman, M., Barua, H., Sarkar, S., & Alam, M. S. (2013). Study on the growth and development of two dragon fruit (Hylocereus undatus) genotypes. *The Agriculturists, 11*(2), 52-57.

Rahnama, T., & Abbaszade, R. (2018). Effects of cold plasma on color changes, anthocyanins content and microbial quality of barberry juice. *Food Science and Technology, 15*(82), 373-385.

Rezali, N. I., Sidik, N. J., Saleh, A., Osman, N. I., & Adam, N. A. M. (2017). The effects of different strength of MS media in solid and liquid media on in vitro growth of Typhonium flagelliforme. *Asian Pacific Journal of Tropical Biomedicine, 7*(2), 151-156.

Rifat, T., Khan, K., & Islam, M. S. (2019). Genetic diversity in dragon fruit (Hylocereus sp) germplasms revealed by rapd marker. *J. Anim. Plant Sci, 29*(3), 809-818.

Shekhawat, M. S., Manokari, M., & Ravindran, C. (2016). Micropropagation, micromorphological studies, and in vitro flowering in Rungia pectinata L. *Scientifica, 2016*. https://doi.org/10.1155/2016/5813851.

Sheng, W. K. W., Sundarasekar, J., Sathasivam, K., & Subramaniam, S. (2016). Effects of plant growth regulators on seed germination and callus induction of Hylocereus costaricensis. *Pakistan Journal of Botany, 48*(3), 977-982.

Sonawane, M. S. (2017). Nutritive and medicinal value of dragon fruit. *Asian Journal Of Horticulture, 12*(2), 267-271.

Thinesh, A., & Seran, T. (2015). In vitro callogenesis from bud and stem explants of dragon fruit (Hylocereus undatus). *Asian J Pharma Sci Tech, 5*(4), 253-256.

Torres-Silva, G., Resende, S., Lima-Brito, A., Bezerra, H., De Santana, J., & Schnadelbach, A. (2018). In vitro shoot production, morphological alterations and genetic instability of Melocactus glaucescens (Cactaceae), an endangered species endemic to eastern Brazil. *South African Journal of Botany, 115*, 100-107.

Vijay, R., Shukla, J., & Saxena, R. (2016). Micropropagation of Plumbago zeylanica L.: An important medicinal plant of India. *Int J Pure App Biosci, 4*(6), 59-167.

Xu, F., Tan, X., & Wang, Z. (2010). Effects of sucrose on germination and seedling development of Brassica napus. *International journal of Biology, 2*(1), 150.

Yasodha, R., Kamala, S., Kumar, S. A., Kumar, P. D., & Kalaiarasi, K. (2008). Effect of glucose on in vitro rooting of mature plants of Bambusa nutans. *Scientia Horticulturae, 116*(1), 113-116.

Yusof, N., Adzahan, N. M., & Muhammad, K. (2020). Optimization of Spray Drying Parameters for White Dragon Fruit (Hylocereus undatus) Juice Powder using Response Surface Methodology (RSM). *Malaysian Journal of Applied Sciences, 5*(2), 45-56.