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EFFECT OF ORGANIC SUBSTRATES ON GINGER GROWTH, YIELD AND [6]-GINGEROL CONTENT CULTIVATED USING SOILLESS CULTURE SYSTEM

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ABSTRACT

Ginger (*Zingiber officinale* Rosc.) belongs to a tropical and sub-tropical *Zingiberaceae* family, which originated from Southeast Asia. Ginger is one of the most widely used herbs contains several interesting bioactive constituents including [6]-gingerol that has potent antioxidant activity and health promoting properties. Substrates plays an important role for plants to growth in the soilless culture system. Given the nature of the substrate may play a crucial role in determining water and nutrient availability for the plant and hence may affect the metabolic pathways involved in the synthesis of specific biochemical compounds, this study was conducted to determine the effects of organic soilless substrates such as coir dust and burnt paddy husks on ginger growth, yield and [6]-gingerol content using soilless culture system. The treatments were arranged in a randomized complete block design (RCBD) with five levels of treatment with three replicates. The treatments include, T1 = 100% coir dust; T2 = 100% burnt paddy husks; T3 = 70% coir dust and 30% burnt paddy husks; T4 = 30% coir dust and 70% burnt paddy husks; and T5 = 50% coir dust and 50% burnt paddy husks. Results showed that plant in T1 gave rise to highest rhizomes yield (5480 ± 325 gm) compare to other treatments. Media containing high amount of coir dust (70 – 100%) showed good growth and increased the rhizome yield up to 36% compared to those containing high amount of burnt paddy husks. There was no significant difference between all treatments in term of [6]-gingerol content in the fresh and dry ginger rhizomes. The studies suggested that the secondary metabolites like [6]-gingerol content and accumulation were not affected by the substrates. It can be concluded that 100% coir dust are the best substrates for growing ginger in soilless culture system.

Keywords: *ginger, substrates, [6]-gingerol, soilless culture system.*

INTRODUCTION

Soilless culture system is the most intensive production method in Malaysia today's agriculture industry, which can result in higher yields even in limited and adverse growing conditions. Yields of chillies, rock melons and tomatoes cultivated in soilless system increased 3 – 5 times compared to those using conventional method (De Rijck and Schrevens 1998). In soilless production system, many types of growing media or substrates such as rockwool, perlite, vermiculite and peat have been used to grow many kinds of crops (Komada et al. 1997). Media such as rockwool, perlite and vermiculite are expensive because they have to be imported. Hence, alternative substrates that are cheaper and locally available such as coconut husks and burnt paddy husks should be used as alternative media (Ortega et al. 1996). Substrates plays an important role for plants to growth in the soilless culture system.

Ginger is one of the most widely used herbs that contains several interesting bioactive constituents and possesses health promoting properties. [6]-gingerol, a major pungent ingredient of ginger, also has potent antioxidant activity (He et al., 1998). Several studies have assessed and modified component of soilless culture production system including irrigation techniques and nutrient elements to increase the plant yields and quality (Hargreaves et al., 2008, 2009). Limited studies have yet investigated the role of substrates may have on the plant productivity and final product such as fruit quality and composition. Given the nature of the substrate may play a crucial role in determining water and nutrient availability for the play and hence may affect the metabolic pathways involved in the synthesis of specific biochemical compounds, this study was conducted to determine the effects of organic soilless substrates such as coir dust and burnt paddy husks on ginger growth, yield and [6]-gingerol content using soilless culture system.

MATERIALS AND METHODS

Bentong ginger was selected and used in this study. Each of the rhizomes was cut into smaller pieces of about 4 cm long and 40 g in weight. Each of the seed rhizomes contained 2 – 3 point buds. The seed rhizomes were treated with previcur-N prior to planting. A side-netted rain shelter of 30 m long x 10 m wide x 4.5 m high located in MARDI Station, Kluang, Johor was used in the study. The treatments were arranged in a randomized complete block design (RCBD) with five levels of treatment with three replicates and 30 plants per treatment. The coir dust and burnt paddy husks were weighed in accordance to the quantity required for each treatment. There were five coir dusts and burnt paddy husks mixtures used as treatments in this study. These treatments were as follows: T1 = 100% coir dust; T2 = 100% burnt paddy husks; T3 = 70% coir dust and 30% burnt paddy husks; T4 = 30% coir dust and 70% burnt paddy husks; and T5 = 50% coir dust and 50% burnt paddy husks. Each mixture was thoroughly mixed in a 10-litre pail before filled into 60 cm x 60 cm black polyethylene bags. The seed rhizomes were sown into the substrate according to the treatments. Each polyethylene bag was placed randomly on four irrigation lines under the side-netted rain shelter and individually irrigated with nutrient solution via a dripper on the surface of the medium. The irrigation system, which was built in the side-netted rain shelter, consisted of a 1,500-litre tank, 1.5 Hp water pump, water liter, pressure meter and four lateral lines (28 m each) which looped to each other. Each of the lateral lines was equipped with 100 drippers that were placed into 100 polyethylene bags, side by side. The distance between each line was 1.5 m and the distance between each dripper point in the lateral line was 0.3 m. The nutrient solutions were supplied through 0.3 m micro tubes and arrow drippers.

The fertilizer was formulated by MARDI based on the needs of the plant rhizomes (Yaseer Suhaimi et al. 2009). All the fertiliser components were water soluble. The fertiliser stocks were prepared according to Yaseer Suhaimi et al. (2011).

The irrigation solutions were prepared in a 1,500-litre tank. Stock A and stock B were added into the tank at 1:1 ratio until the needed electricity conductivity (EC) was achieved. The EC of the fertigation solution was between 1.8 uS and 2.3 uS. The irrigation scheduling was automatically implemented by a digital timer, three times per day in the first 3 months (0800 h, 1200 h and 1600 h), six times per day in the 4^{th} – 7^{th} months (0700 h, 0800 h, 1000 h, 1200 h, 1400 h and 1600 h), and once per day in the last 2 months (1000 h).

The duration of irrigation was 3 min and an identical amount of fertilizer solution was applied to all polyethylene bags. The daily irrigation volumes per plant were 675 ml in the first 3 months, 1,350 ml in the 4^{th} – 7^{th} months, and 75 ml in the last 2 months. Routine horticultural practices for pest, disease and weed control were followed. Insecticide (*Malathion*) and fungicide (*Benlate*) were applied once every 2 weeks.

The growth of the ginger plants was measured monthly by measuring the height and weight of leaves/shoot and rhizomes. The ginger plants were randomly selected and the rhizomes were harvested after 3 – 9 months of sowing to determine the yield and growth of rhizomes. The weight was measured immediately after harvest to prevent desiccation and water loss from the rhizomes.

Fresh ginger rhizomes were graded, washed thoroughly with tap water, peeled and cut into cross-sections of 2 ± 1 mm thickness. Cut ginger samples were dried in an air dryer at 50 ± 2 $^{\circ}$C to achieve 10-12 % moisture content for 3 hours then the samples were taken for the analyses of [6]-gingerol content for dried ginger. 2 g of dry ginger were put in a 250 ml conical flask contain 150 ml of methanol then were shaking about 7 hours on the orbital shaker at 250 rpm. The methanol extract was filtered using 0.20 µm Nylon membrane filter (Whatman, England). 10g of cut fresh ginger were blended with 50 ml methanol (HPLC grade) by electrical blender for 1min and centrifuged at 5,000 rpm for 5 min. The supernatant was subsequently filtered through a 0.20 µm Nylon membrane filter (Whatman, England). A 20 µl ginger extract from both dry and fresh ginger extract were then subjected to HPLC for the [6]-gingerol analysis. The analysis method of 6-gingerol content was done according to Sharizan et al. (2014) with modification.

Data obtained were subjected to statistical analysis using analysis of variance (ANOVA) procedures to test the significant effect of all the variables investigated using SAS version 9.1. Means were separated using Duncan Multiple Range Test (DMRT) as the test of significance at $p \leq 0.05$.

RESULTS AND DISCUSSION

There were significant differences in plant height between treatments (*Table 2*). The tallest plants were produced by ginger cultivated in 100% coir dust with an average height of 123 ± 23 cm and the lowest were those cultivated in mixture of 30% coir dust and 70% burnt paddy husks (average height 105 ± 8 cm). Treatment containing 100% coir dust produced the tallest plants compared to burnt paddy husks and mixtures of both substrates. This could be due to the higher porosity of coir dust compared to the other treatments. This higher porosity property drained out the excess fertiliser solution between the irrigation schedules more quickly. Raviv et al. (2001) found that the number of rose flowers was 19 per cent higher in coir dusk than in others substrates. The obtained results were agreed with Neamati et al. (2010) who observed the highest stem and root length tomato cultivated in the coco peat media. The mixtures between coir dust and burnt paddy husks could have increased the water holding capacity and subsequently decreased the dissolved oxygen availability in the growing medium. Plant height grown in mixture of 30% coir dust and 70% burnt paddy husks was significantly affected. The higher content of burnt paddy husks in the medium added more moisture content that lowered dissolved oxygen in the media, which consequently reduced height of the ginger plant compared to 100% coir dust. Similar studies also showed that high water holding capacity reduces the growth and yield of cucumber (Peyvast et al. 2010). The shoots were cut and trimmed 2 weeks before harvesting. This allowed the rhizomes to harden in the media (Paul et al. 2004). There were significant differences in shoot fresh weight between treatments. The highest shoot fresh weight was recorded from plants cultivated in 100% coir dust with an average weight of 1,340 ± 235 g (*Table 2*), while the lowest weight was obtained from plants cultivated in mixture of 30% coir dust and 70% burnt paddy husks. The shoot fresh weights were higher with higher content of coir dust in the growing media. However, there was no significant difference between 100% burnt paddy husks, mixture of 70% coir dust and 30% burnt paddy husks, and mixture of 50% coir dust and 50% burnt paddy husks at $p \leq 0.05$. A study by Fukuda and Anami (2002) also revealed an increase in melon biomass when grown in coir dust.

Table 2. Plant growth and rhizomes yield after 9 months of cultivation periods

Treatment	Plant height (cm)	Shoot fresh weight (g)	Average rhizome yield per plant (g)	Rhizome to shoot ratio
100% CD	123 ± 23a	1,340 ± 235a	5,480 ± 325a	4.09a
100% BPH	114 ± 15b	1,210 ± 223b	3,480 ± 150d	2.87d
70% CD + 30% BPH	112 ± 12c	1,130 ± 127b	4,580 ± 170b	4.05b
30% CD + 70% BPH	105 ± 8d	1,090 ± 115c	2,570 ± 135e	2.33e
50% CD + 50% BPH	115 ± 16b	1,120 ± 120b	4,400 ± 180c	4.03c

Mean values in the same column followed by the same letter are not significantly different at $p < 0.05$
CD = Coir dust; BPH = Burnt paddy husks

For commercial purposes, ginger rhizomes are harvested 7 – 9 months after sowing (Wilson and Ovid 1993). In this study, the rhizomes were harvested after 3 – 9 months and the fresh weight of the rhizomes were measured. The interior fresh and epidermis were lighter in colour than the mother seed piece. There was also fibre development in the interior fresh. The rhizomes also produced a pungent odour with a distinctive ginger flavour. They were marketable as fresh young ginger between 3 and 6 months of cultivation and mature ginger between 8 and 9 months. There were significant differences in rhizome yield between treatments after 9 months of cultivation. The highest average fresh rhizome yield was obtained from plants cultivated in 100% coir dust, followed by mixtures of 70% coir dust and 30% burnt paddy husks, 50% coir dust and 50% burnt paddy husks, 100% burnt paddy husks, and 30% coir dust and 70% burnt paddy husks. These results showed that ginger cultivated in higher amount of coir dust media increased the rhizome yield up to 36% compared to those grown in media containing higher amount of burnt paddy husks. High oxygen availability in the coir dust media supported the underground rhizomes requirement for high oxygen for growth. Tomatoes grown very well in coir dusk compared to others substrates, even under temperature stress (Islam et al., 2002). It has been shown that a number of potted plants, nursery, leafy and fruity vegetables performed better in coir dusk (de Kreij and van Leeuwen, 2001). For crops grown in containers, it is important to consider the tendency of most root systems to grow gravitropically to form a dense layer at the bottom of the containers (Raviv et al. 2001). Coir dust has a strong capillarity that provides more uniform moisture conditions for roots. These conditions are able to increase aeration in the base mix and reduce drying of the surface by lifting the moisture higher up in the polyethylene bags. This increases the volume of the mixture that is suitable for root development and improve access to moisture and fertilizer. This redistribution of moisture is perhaps one of the reasons for plants grown in pure coir dust to have higher rhizome yield. Aeration in the growing medium is positively related to AFP and negatively to water content (Raviv and Lieth 2008).

The coir dust is less acidic with a pH suitable to facilitate ginger to grow and consequently allows the plant roots to absorb nutrient efficiently.
In the early cultivation period between 1 and 3 months, the growth of rhizomes between treatments was similar. The exponential growth of the rhizomes began in the fifth month and the rhizomes in 100% coir dust treatment showed the highest growth compared to other treatments (*Figure 1*). Media with higher content of burnt paddy husks gave lower rhizome yield throughout the cultivation period with a mixture of 30% coir dust and 70% burnt paddy husks exhibited the lowest rhizome yield.
These results were similar with the study conducted by Kratky (1998), who found that ginger rhizome yield increased significantly when grown using soilless system under rain shelter. Previous study done by Hayden et al. (2004) found that the growth of rhizomes is dependent on the type of medium. The growing medium acts as heat insulator and provides heat that enhances the growth of rhizomes.

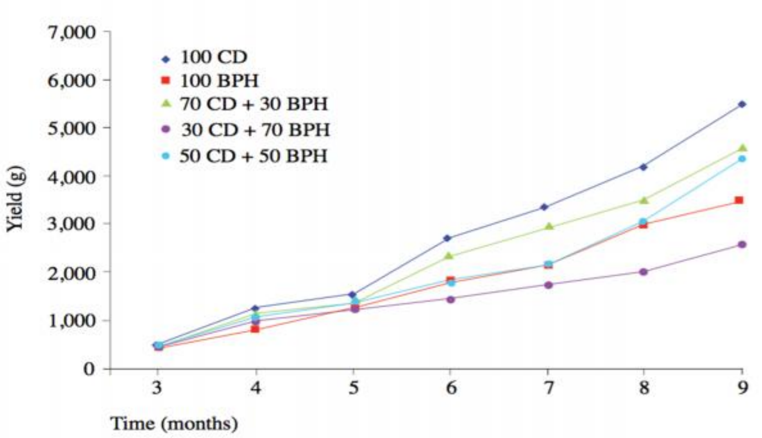

Figure 1. Growth of ginger rhizomes between third and ninth months of cultivation

Overall biomass of ginger plants can be divided into two parts: above ground biomass consisting of leaves and stem (shoots), and underground biomass consisting of rhizomes and roots. In this study, there were significant differences between treatments in rhizomes to shoot ratio. The ratio of underground biomass to aboveground biomass was highest in plants cultivated in 100% coir dust with a ratio of 4.09 (*Table 3*). There was higher underground biomass compared to aboveground biomass in plant grown in the 100% coir dust. The higher ratio of underground biomass to aboveground biomass reflects that the roots were well able to supply the top of the plant with water, nutrient, stored carbohydrates and certain growth regulators (Harris 1992). The rhizome to shoot ratio in plants cultivated in high coir dust media was 4 to 1, while that in plants cultivated in high burnt paddy husks was 2 to 1. Long and quicker root development of tomatoes and strawberry were observed in the coir dusk growth substrate (Lopez- Medina et al., 2004).

Table 3. The dry weight of shoot and rhizomes biomass after 9 months of cultivation periods

Treatment	Shoot dry weight (g)	Rhizome dry weight (g)
100% CD	804	3288
100% BPH	726	2088
70% CD + 30% BPH	678	2748
30% CD + 70% BPH	654	1542
50% CD + 50% BPH	672	2640

[6]-gingerol was the major ginger oleoresin. The amounts of [6]-gingerol content from fresh and dried ginger are shown in Table 1. β-hydroxyl keto functional group is main molecular structure of gingerol which was thermally labile. [6]-gingerols tends to degrade into shogaols and aliphatic aldehydes due to heat or thermal process during occurrence of drying process (Bhattarai et al., 2001). As a result, the dried ginger had a smaller amount of [6]-gingerol compared to the fresh product. The HPLC chromatograms of [6]-gingerol standard are shown in Figure 2. The retention time of [6]-gingerol standard as shown in the chromatograms at 11.5 minutes.

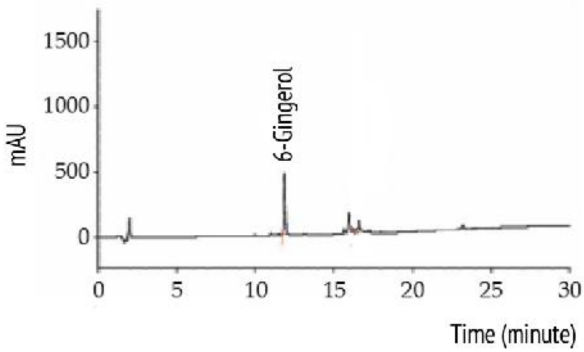

Figure 2: The retention time of [6]-gingerol standard injected into HPLC instrument

There were no significant different between all treatments in term of [6]-gingerol content in the fresh and dry ginger rhizomes. However, plant cultivated in the media contained coir dust showed slightly higher [6]-gingerol content in the fresh weight of ginger (Table 4). Similar pattern also was observed in [6]-gingerol content in the dry weight of ginger (Table 5). The accumulation of [6]-gingerol in both fresh and dry ginger samples were higher from month three to month six and started to decrease at month seven towards the end of the cultivation period. The [6]-gingerol content was higher in the month six and lowest at months nine of cultivation periods. Both fresh and dry ginger samples showed the similar pattern. Studies by Sharizan et al. (2014) revealed that accumulation of [6]-gingerol compound were higher in 6-month ginger rhizomes and lower in 9 and 12-month ginger rhizomes. No differences in [6]-gingerol were observed according to the type growth media. Investigation by Rodriguez et al. (2006) on the effect of different substrates for hydroponic production of 'Galia' muskmelons (Cucumis melo L.) revealed that fruit quality and composition were not affected by substrate. Others studies also had showed that rising concentration of electric conductivity (EC) of nutrient solution and nutrient solution composition can caused increase in total soluble solids, organic acids and plant secondary metabolites for tomatoes (Petersen et al., 1998), sweet pepper and cucumber (Trajkova et al., 2006). Overall, the nature of the soilless media substrate did not affect overall rhizomes quality and chemical compounds. Appropriate root zone aeration is effective for higher fruit yield and bioactivity of the fruit in perlite culture, but excessive aeration inhibited root respiration, nutrients, bioactivity, and water uptake, and it resulted in the reduction of plant growth and fruit yield (Jong et al., 2014)

Table 4. Accumulation of [6]-Gingerol in the fresh weight of rhizomes from three to nine months if cultivation periods (mg/g fresh weight)

Treatment/ months / mg/g fresh weight	**3**	**4**	**5**	**6**	**7**	**8**	**9**
100% CD	1.879	2.045	2.389	2.546	1.767	1.542	1.183
100% BPH	1.521	1.756	2.023	2.298	1.437	1.247	1.094
70% CD + 30% BPH	1.732	1.906	2.274	2.456	1.612	1.434	1.013
30% CD + 70% BPH	1.597	1.845	2.154	2.318	1.556	1.228	0.754
50% CD + 50% BPH	1.621	1.889	2.171	2.308	1.678	1.390	0.806

Table 5. Accumulation of [6]-Gingerol in the dry weight of rhizomes from three to nine months if cultivation periods (mg/g fresh weight)

Treatment/ months / mg/g dry weight	**3**	**4**	**5**	**6**	**7**	**8**	**9**
100% CD	1.474	1.045	0.643	0.434	0.367	0.236	0.122
100% BPH	1.125	0.906	0.512	0.302	0.290	0.104	0.093
70% CD + 30% BPH	1.305	1.006	0.734	0.404	0.313	0.209	0.114
30% CD + 70% BPH	1.138	0.943	0.705	0.398	0.285	0.201	0.105
50% CD + 50% BPH	1.224	0.912	0.684	0.351	0.255	0.112	0.100

CONCLUSIONS

The yield and quality of the products reflects to the different cultivation guidelines that applied to each substrate. The mixture of coir dust and burnt paddy husks significantly affected plant height, shoot biomass, rhizome yield, and rhizome to shoot ratio. Media containing high amount of coir dust (70 – 100%) showed good growth and increased the rhizome yield up to 36% compared to those containing high amount of burnt paddy husks. The [6]-gingerol content were higher in 100% coir dust throughout the cultivation periods. However, there were not significant difference in the [6]-gingerol content between each treatment. The studies suggested that the secondary metabolites like [6]-gingerol content and accumulation did not effected by the substrates or substrates mixtures alone. It can be concluded that 100% coir dust or any combinations with high amount of coir dust are the best substrates for growing ginger in soilless culture system.

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