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Short Communication

First Reported Observation on Aerial Tuber Formation in *Dioscorea hispida* Dennst.

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ABSTRACT

During an experiment to study the effects of irrigation on growth and development of *Dioscorea hispida* Dennst., we observed aerial tuberization after six to nine months of *D. hispida* growth. In this experiment, there were five irrigation schedules applied on *D. hispida*: daily watering (T2), watering at one day interval (T1/control), two day intervals (T3), five day intervals (T4) and six day intervals (T5). Each plant was given 440 mL of water in every application. Treatment T2 showed the highest percentage of tuberization (80%), while for T5 no aerial tuber formation was observed. To our knowledge, aerial tuber formation of *D. hispida* has never been reported until now. Aerial tubers have the potential to be used as an alternative and clean planting material for propagation and if they can be induced to germinate, it would be possible to use them for the tissue culture and commercialization of *D. hispida*.

Keywords: *Dioscorea hispida*, aerial tuber, irrigation, propagation

ABSTRAK

Dalam satu eksperimen untuk mengkaji kesan pengairan terhadap pertumbuhan dan perkembangan tumbuhan *Dioscorea hispida* Dennst., kami telah mendapati pembentukan ubi *aerial* selepas tujuh hingga sembilan bulan pertumbuhan *D. hispida*. Dalam eksperimen ini, terdapat lima jadual aplikasi pengairan bagi tumbuhan *D. hispida* seperti berikut: siraman bagi setiap hari (T2), selang sehari (T1/kawalan), selang dua hari (T3), selang lima hari (T4) dan selang enam hari (T5). Setiap tumbuhan telah diberikan 440 mL air untuk setiap aplikasi siraman.

Rawatan T2 menunjukkan peratusan pembentukan ubi *aerial* yang paling tinggi (80%). Bagi kekerapan pengairan yang paling kurang iaitu T5, tiada pembentukan ubi *aerial*. Menariknya, pembentukan ubi *aerial* bagi tumbuhan *D. hispida* tidak pernah dilaporkan sebelum ini, oleh itu kita percaya bahawa ini adalah kali pertama ia didokumentasi. Ubi *aerial* boleh digunakan sebagai benih alternatif dan bersih untuk pembiakan tanaman, dan sekiranya ubi *aerial* tersebut boleh diaruh untuk bercambah, maka ianya mungkin berguna untuk tujuan kultur tisu dan pemerdagangan *D. hispida*.

Kata kunci: *Dioscorea hispida*, ubi *aerial*, pengairan, pembiakan

INTRODUCTION

Yam is one of the oldest groups of angiosperms and monocotyledonous plants which were found about 3,500 years ago. Some species of yams are herbaceous climbers that twined and their vegetative propagation is from rhizomes and tubers which are also the storage organs (Craufurd *et al.,* 2001; Kulkarni *et al.,* 2007; Behera *et al.,* 2008; Marie-Vianney *et al.,* 2008). In certain yam species like *Dioscorea bulbifera* L., tubers originated from the leaf axils and are known as bulbils (Tchabi, 2008). On the other hand, *D. hispida* has other tubers which are known as underground tubers and these tubers have economic values because they are consumed as food (Craufurd *et al.,* 2001). In Malaysia, *D. hispida* tubers are known to be consumed as food by the Malays in the Terengganu areas whenever food is scarce (Nashriyah *et al.,* 2011; Nashriyah *et al.,* 2012).

The propagation of *Dioscorea hispida* is quite similar to other yams, which was traditionally propagated vegetatively from whole tubers (seed), large (setts) and small (minisetts) tuber pieces (Craufurd *et al.,* 2001). On the other hand, underground tubers of *D. hispida* had been proven to be more palatable compared to aerial tubers. Therefore aerial tubers can be used as replacement seedlings for *D. hispida* cultivation. Aerial tubers can be used as clean planting materials as they are usually devoid of infestation by soil-borne pests such as nematodes. In yams, the use of nematode-free planting material is a practical and economical way to prevent damage to the crop, with the aerial tubers *D. bulbifera* and some forms of *Dioscorea alata* L., being proposed as examples to be used for propagation (Tchabi, 2008).

MATERIALS AND METHODS

Materials

Fifty *Dioscorea hispida* underground tubers were collected in September 2012 at Ladang Karas, Merchang, Terengganu, Peninsular Malaysia (location N 05^{o} 02.125' E 103^{o} 14.767'). Each tuber was weighed and planted in a polyethylene bag at the Nursery of Faculty of Agriculture, Biotechnology and Food Sciences, Universiti Sultan Zainal Abidin, Gong Badak Campus, Kuala Terengganu, Peninsular Malaysia; a day after sampling. Sixty percent of these tubers sprouted within two

months (November 2011) after collection; and 30 out of these 50 tubers were chosen for the experiment. They were assigned randomly for each treatment.

Experimental Design

A Complete Randomized Design (CRD) with six plants for every treatment was arranged at a distance of 0.3 m from each plant and 0.7 m between rows. The treatments were 1) watered every day each week (T2); 2) watered at two day intervals each week (T3); 3) watered at five day intervals each week (T4); and 4) watered at six day intervals each week (T5). The control frequency had been estimated as watered at one day interval each week (T1/control) according to *Dioscorea rotundata* Poir. by Charles-Dominique *et al.* (2008).

To each *D. hispida* plant, NPK green (15 N: 15 P_2O_5: 15 K_2O) chemical fertilizer was applied at 20 g per plant around the plant once after five months of plant growth (March 2012) until harvested (July 2012) to supply nutrients. Weeds in the plot were kept cleared by hand weeding until harvested. The amount of water per day was determined and set at 440 mL per plant; this was calculated from the rainfall data of Kuala Terengganu that was obtained from the Malaysian Meteorological Department (Table 1). The average three-year rainfall data (2008 until 2010) was used to calculate the amount of water required per day with equation (1) and then, equation (2) was applied to get the exact amount of water according to the area of a polyethylene bag (DID, 2009). Weather forecast can be used as an indicator to schedule irrigation due to the lack of crop evapotranspiration data (Wang *et al.,* 2006).

$$\mathrm{DDIR} = \frac{\mathrm{RAW}}{\mathrm{fx}} \tag{1}$$

$$\text{Volume (kL)} = \text{Depth (mm)} \times \text{Area (m}^2\text{)} \div 1000 \tag{2}$$

where,

DDIR = design daily irrigation requirement (mm)
RAW = readily available water (mm)
fx = irrigation interval (day)

Number of Aerial Tubers

Dioscorea hispida has not been reported to have aerial tuber formations by Wilson (1988). The aerial tuber formed was counted manually at harvesting (July 2012).

Statistical Analysis

Data collected was evaluated by descriptive statistics and one-way analysis of variance (ANOVA) with comparison between treatments carried out by Least Significant Difference (LSD) using SPSS (version 17.0, IBM).

Table 1. Annual rainfall data in Kuala Terengganu between the year 2008 until 2010 (mm).

Month	Year 2008 (mm)	Year 2009 (mm)	Year 2010 (mm)	Average (mm)
Jan	157.2	122.2	203.4	160.9
Feb	130.8	93.0	18.2	80.7
March	47.0	416.4	148.6	204.0
April	47.8	185.2	160.0	131.0
May	187.8	318.6	94.0	200.1
June	93.6	128.6	42.4	88.2
July	142.4	99.6	81.8	107.9
Aug	128.2	155.2	80.0	121.1
Sept	123.0	161.0	109.6	131.2
Oct	109.0	404.4	327.8	280.4
Nov	1580.4	1170.2	393.2	1047.9
Dec	862.4	339.2	704.2	635.3
Annual	3609.6	3593.6	2363.2	3188.8

Source: Malaysian Meteorological Department, 2008-2010.

RESULTS AND DISCUSSION

Different Locations of Aerial Tuber Formation

In this study, aerial tubers were observed at the petiole axis and these tubers were smaller compared to underground tubers, similar to other *Dioscorea* species which had aerial tuber (Craufurd *et al.,* 2001).

There were three positions of aerial tuber formation in *D. hispida*. Firstly, at the basal level that touched the ground as shown in Figure 1. The roots of this aerial tuber penetrated into the soil and provided better establishment of the tuber to become progenies of *D. hispida* plant.

Fig. 1. The first type of aerial tuber formation, where the aerial tuber touches the ground at eight months growth (June 2012) of the *Dioscorea hispida* plant.

The second type of aerial tuber formed was shown in Figure 2, where the aerial tuber formation was at the middle level of the plant. The root growth was quite intense at the beginning of tuberization but eventually the root tips became necrotic and finally died (Fig. 2a); and about one month later, the roots disappeared and the tuber increased in size (Fig. 2b).

(a) **(b)**

Fig. 2. Second type of aerial tuberization on the middle part of the plant. a) after eight months of growth (June 2012) of the *D. hispida* plant; b) after nine months of growth (July 2012) of the *Dioscorea hispida* plant. Notes that photos (a) & (b) are from the same plant.

For the third type of aerial tuber, the sequence of the formation of this type of aerial tuber was depicted in Figure 3. Figure 3a shows the beginning of its formation, where whitish roots were seen at the petiole pulvinus and this occurred at the middle period of plant growth, which was about six months after the tubers were planted. Then at the middle stage of tuberization, the tuber started to expand and was green in color and this occurred one month after the roots appeared (Fig. 3b). At the end of the observation period of plant growth (after nine months), the aerial tuber started to change color from green to brownish (Fig. 3c).

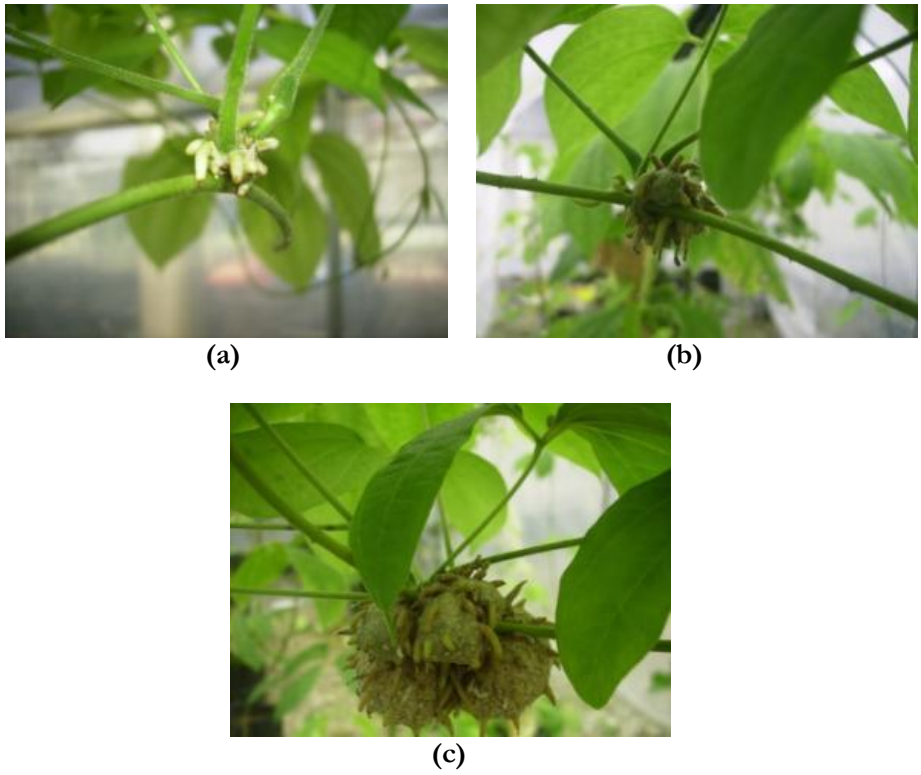

Fig. 3. Third type of aerial tuber formation on top part of the plant. Age of plants after planting a) after six months (April 2012); b) after seven months (May 2012); c) after nine months (July 2012).

Effects of Different Irrigation Treatments on the Number of Aerial Tuber Formation

There was a significant difference of the irrigation treatment on the number of aerial tubers formed at $P = 0.049$ ($P \leq 0.05$). The highest number of aerial tubers formed was shown in plants subjected to T2 treatment where 80% out of the six plants (total number of sprouted *D. hispida* plants in that treatment) yielded aerial tubers. Plants that were subjected to T5 treatment (*i.e.,* watered at six day intervals) showed no aerial tuber formation. Table 2 also shows that T2 treatment had a significant difference when compared to other treatments.

Table 2. Descriptive statistics on the number of aerial tuber formation.

Treatment	N	Number of Tubers Formed*	Standard Error
T1/control (one day interval)	6	1.0 ^{b}	0.7
T2 (daily watering)	6	4.8 ^{a}	2.6
T3 (two day intervals)	6	0.5 ^{b}	0.3
T4 (five day intervals)	6	0.2 ^{b}	0.2
T5 (six day intervals)	6	0.0 ^{b}	0.0

Note: *Significant difference between treatments was indicated by different letters in the column.

Wilson (1988) had classified *Dioscorea hispida* as a non-aerial tuber producing yam. The formation of aerial tuber has never been reported before in *D. hispida*. Therefore, an interesting first observation was made in this study, of *D. hispida* aerial tuber formation under the different irrigation schemes implemented. Aerial tuberization was described by Craufurd *et al.* (2001) in *Dioscorea rotundata* Poir., whereby the first macroscopically visible sign of tuber initiation is the bursting of the suberous layer of this amorphous mass. The mass of cells soon differentiates into a growing point, usually whitish in color, which began to elongate with a recognizable head (proximal) and tail (distal) ends.

The unique distribution on the vine where the *Dioscorea hispida* aerial tubers were formed, observed in this experiment, might explain the distribution of this plant in the wild. This plant probably disperses their progenies far away from the mother plant to avoid competition. This phenomenon of aerial tuberization, which was detached from the ground level, proved the assumption that distribution of its progeny was *via* aerial tuberization. This may have happened when the mother plant with the underground tubers died, the aerial tubers above the ground might continue the regeneration of the species, and became the progeny of the mother plant.

Observation in the natural habitat showed that *Dioscorea hispida* tubers were not eaten by wild animals, especially mammals such as wild boar in the wild. This ethnobotanical opinion was supported by reports on its bitter taste and the

presence of toxic compounds which are known as dioscorine (Nagata *et al.,* 1999; Murthy *et al.,* 2011; Muhamad Azhar *et al.,* 2012). The alkaloid can trigger fatal paralysis of the nervous system when even a fragment of the tuber weighing 100 g is ingested (Bhandari and Kawabata, 2005).

When these plants were irrigated daily with a water volume of 440 mL per plant, the percentage of aerial tubers formed was at its highest. Even though Wilson (1988) had classified *D. hispida* as a non-aerial tuber producing yam, our observations here clearly contradicted his report. We are currently investigating the application of hormones to induce aerial roots and tubers for further multiplication of *D. hispida*. If aerial tubers can be successfully produced, they can be used as an alternative source of clean planting materials in tissue culture while the underground tubers can be used to generate income for the small farmers.

CONCLUSION

As a conclusion, we can induce and produce aerial tubers in *Dioscorea hispida* at the petiole axils *via* daily irrigation. The best irrigation frequency was the daily irrigation (T2), with a water volume of 440 mL per plant, which gave the highest percentage of aerial tuber formation.

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