ISSN 2012-9912; ©2012 Open University of Sri Lanka

FEASIBILITY STUDY OF GROWING LUNUWILA (*BACOPA MONNIERI* L.) UNDER HYDROPONICS

G.D. Krishantha, H.K.L.K. Gunasekera[1*], K.S.S.Sugathadasa[2] and K.W.D.S. Prasangika

Department of Agricultural and Plantation Engineering,

The Open University of Sri Lanka, Nawala, Nugegoda

INTRODUCTION

Lunuwila (*Bacopa monnieri* L.) or Brahmi is a dwarf herbaceous medicinal herb belonging to family Scrophulariaceae (Kapoor,1990). The whole plant of Lunuwila has been used in Ayurvedic medicine as a tonic for nerves and mental disorders. It has even been used to treat rheumatism, blood diseases, congestive heart failures, urinary track infections, hepatitis, high blood pressure and as a leafy vegetable due to its high nutritional value (Das, 2006). The annual demand of ayurvedic drug co-operation for Lunuwila has been estimated to be 1700 Kg (Anonymous, 2004). However, there is no commercial cultivation of Lunuwila recorded in Sri Lanka. Hence, entire requirement is fulfilled from the natural population and thus leading to a rapid depletion of the plants from its natural habitat. Furthermore, International Union for Conservation of Natural Resources has listed Lunuwila as an endangered species (Joshi *et al.*, 2010). Therefore, to meet the growing demand for safe and quality plant material, commercial cultivation in hydroponic could be adopted. It has been reported that many herbaceous medicinal plants can be successfully grown under hydroponics (Canterl *et al.*, 2005). Hydroponics is a science of growing plants without soil, giving nutrients to the plant through water under protected environmental conditions (Resh, 2001). Therefore, in this study it was investigated the feasibility of growing Lunuwila hydroponically by using a simple hydroponic system applicable to Sri Lanka.

METHODOLOGY

The experiment was conducted in a plant house (10 m x 12 m) at the Botany division of the Bandaranayake Memorial Research Institute, Nawinna, Maharagama, during the period from June, 2011 to February, 2012. The experiment was conducted in two steps, i.e. as a field experiment in a plant house and a laboratory experiment. Lunuwila plants were grown under commercially available Albert's solution in Sri Lanka (CIC Fertilizer Pvt. Ltd.). The experiment was conducted in a Complete Randomized Design (CRD). Three replicates or regifrm (styrofoam) boxes were allocated for each treatment. Seventy two Lunuwila cuttings (10 cm length with 10 leaves) were randomly selected to grow under hydroponically and eighteen cuttings were selected to grow under natural marshy condition as control. The five treatments imposed were as follows.

Treatment -1 (T1)	100% concentration of Albert's solution
Treatment -2 (T2)	75% concentration of Albert's solution
Treatment -3 (T3)	50% concentration of Albert's solution
Treatment -4 (T4)	25% concentration of Albert's solution
Treatment -5 (T5)- (control)	Natural marshy soil medium

100% concentration of Albert's solution (T1) was prepared according to the Department of Agriculture (DOA) recommended concentration and other treatments T2, T3 and T4 were prepared by diluting it further with water. The Electrical conductivity (1.5 -2.5ds/m) and pH values (5.8 -6.5) were measured twice a month (Anonymous,2002). Quantitative and qualitative evaluation of harvested plants was done in the laboratory.

1*All correspondence should be addressed to Dr. H.K.L.K. Gunasekera , Department of Agricultural and Plantation Engineering, The Open University of Sri Lanka (*Email: hkgun@ou.ac.lk*)
2 Scientist, Department of Botany, Bandaranayake Memorial Research Institute, Nawinna, Maharagama

The total dry weight, shoot: root ratio and leaf: stem ratio of hydroponically grown plants (HGP) and soil grown plants (SGP) were assessed. Thin Layer Chromatography (TLC) test was done according to the Wanger et al., (1984) for all treatments to check whether the chemical constituents of Lunuwila, mostly Saponins are affected by the hydroponics culture. Data were tabulated and analyzed by using Analysis of Variance (ANOVA) procedure of Statistical Analysis System (SAS). The Least Significant Difference (LSD) test was used to compare differences among the treatment means at p= 0.05.

RESULTS AND DISCUSSION

Quantitative assessment

Results of the quantitative analysis are shown in Table 1. Hydroponically grown plants (T1 – T4) showed a higher dry matter accumulation than that of soil grown plants. Resh (2001) also reported that many crops showed at least more than two times higher yield under hydroponics than under soil culture. Among the different treatments tested, plants grown in 50% concentration of Albert's solution showed the highest dry matter accumulation while the lowest was shown in soil grown plants/control (T5). Furthermore, shoot: root ratio in HGP was higher than that of SGP (control). As the shoot: root ratio represents the proportion of total biomass allocated to shoots, higher values obtained in HGP. The leaf:stem ratio of HGP and SGP was not significantly different ($p>0.05$). This result could not be considered as a disadvantage of Lunuwila as the whole plant is used for medicinal purpose.

Table 1 Assessment growth parameters of hydroponically grown plants (HGP) and soil grown plants (SGP)

Treatments	Fresh weight (g)	Dry weight (g)	Shoot : Root ratio	Leaf : Stem ratio
(a). HGP				
T1- (100%)	49.86b	10.49^{b}	2.54^{bc}	0.67^{a}
T2- (75%)	47.94^{b}	9.86^{b}	2.71^{b}	0.57^{a}
T3- (50%)	71.41^{a}	14.24^{a}	3.85^{a}	0.56^{a}
T4- (25%)	47.03^{b}	10.67^{b}	2.90^{b}	0.61^{a}
(b) SGP				
T5- (Control)	12.77^{c}	3.86^{c}	1.91^{c}	0.66^{a}
LSD	16.708	2.321	0.650	0.183
CV	20.050	12.989	12.793	16.377

Note: Within each column, means followed by the same letter are not significantly different at p=0.05. Measurements are the means of three replicates.

Qualitative assessment

Prompitayarat et al (2007) reported that the Triterpinoid saponins as a major component in Lunuwila and there were several types of saponins. Some of them are Bacoside A_3, Bacoside B, Bacopasaponin C, Bacopaside I and II and Jojubojenin. Thin Layer Chromatography (TLC) test was done according to the Wanger et al (1984) for all treatments to check whether chemical constituents of Lunuwila, mostly saponins are changed or not under hydroponics. Three solvent systems (i.e. chloroform 95: methanol 5, chloroform 70: methanol 30: water 4 and chloroform 64: methanol 50: water 10) were used to detect active saponins present in the plant. About 10µl of a solution of the substance tested (70% ethanol extracts of Lunuwila) was chromatographed by using three solvent samples separately to detect the mixture of active saponins in Lunuwila..

Thin layer chromatography using solvent sample I (chloroform 95: methanol 5)

Spraying the chromatogram with (anisaldehyde, sulphuric acid, glacial acetic acid and methanol) regent revealed several colour bands (Figure 1). Saponins, the major compound in

Lunuwila could be seen in purple coloured spots when observed under visible light (Anees, 2010). Spraying the chromatogram with (anisaldehyde, sulphuric acid, glacial acetic acid and methanol) regent revealed several colour bands (Figure 1). Spraying the chromatogram with each regent revealed purple coloured spots with Rf values ranging from 0.07 to 0.63 (Table 2). The results also revealed that T3 was more or less similar to the control treatment (T5). The Rf values of T1, T2, T3 and T5 were also similar. However, there were bands responsible for the Rf 0.07, 0.10, 0.14 and 0.63 only in the chromatogram of T4 and bands belonging to the Rf values of 0.21 and 0.31 were absent in the T4 (Table 2).

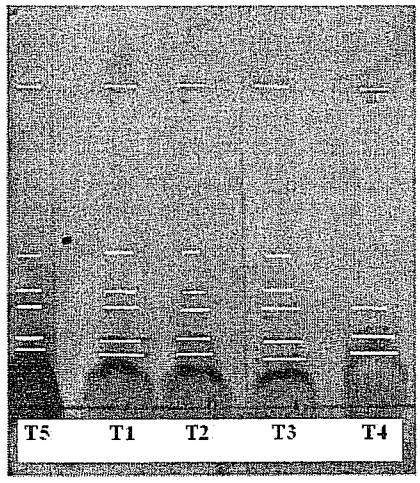

Figure 1:Thin layer chromatograph of five ethanol extract of Lunuwila, T5-(control)-column 1, T1-column 2, T2-column 3, T3-column 4 and T4-column 5, developed with solvent system I and visualized under visible light, after the spraying of anysaldehyde/ H_2SO_4 reagent.

Table 2 R_f values (retention factor) of chromatograms developed with solvent system I

Chromatogram Treatments	R_f values						
T1	0.07	0.10	0.14	0.21	0.31	0.63	0.63
T2	0.07	0.10	0.14	0.21	0.31	0.63	0.63
T3	0.07	0.10	0.14	0.21	0.31	0.63	0.63
T4	0.07	0.10	0.14	-	-	0.63	0.63
T5 (control)	0.07	0.10	0.14	0.21	0.31	0.63	0.63

Thin layer chromatography using solvent sample II (chloroform70: methanol30: water4)

According to the chromatogram, T1, T2 and T3 were identical and were also similar to TLC pattern obtained with control (T5). Purple coloured spots with R_f values of 0.19 to 0.53 and the other spots of chromatograms with R_f values were 0.25, 0.32 and 0.42 were observed. However, the TLC pattern of T4 was different to that of others (Table 3).

Table 3 R_f values (retention factor) of chromatograms developed with solvent system II

Chromatogram Treatments	R_f values				
T1	0.19	0.25	0.32	0.42	0.53
T2	0.19	0.25	0.32	0.42	0.53
T3	0.19	0.25	0.32	0.42	0.53
T4	0.19	0.25	-	-	-
T5 (control)	0.19	0.25	0.32	0.42	0.53

Thin layer chromatography using solvent sample III (chloroform 64: methanol 50: water10)

Spraying the chromatogram with each regent revealed purple coloured spots with Rf values ranging from 0.16 to 0.90. Purple coloured bands could be observed with Rf 0.05, 0.16, 0.23,

0.36, 0.58 and 0.90. These bands are for different kinds of saponins in Lunuwila. In chromatogram of plants grown under the treatment T1, the first band belonged to Rf 0.05 was absent and the other bands were similar to the TLC pattern of SGP (control –T5). In the chromatogram of T2, the lowest Rf value was 0.16 and highest was 0.89. On the other hand chromatograms of the treatments T1 and T4 were identical (Table 4).

Table 4 R_f values (retention factor) of chromatograms developed with solvent system III

Chromatogram Treatments	R_f values						
T1	-	0.16	0.23	0.36	0.58	-	0.90
T2	-	0.16	0.23	0.36	0.58	0.89	-
T3	0.05	0.16	0.23	0.36	0.58	0.89	0.90
T4	-	0.16	0.23	0.36	0.58	0.89	-
T5 (control)	0.05	0.16	0.23	0.36	0.58	-	0.90

CONCLUSION

Growth and yield parameters of Lunuwila were significantly higher ($p<0.05$) in the hydroponically grown plants (HGPs) than that of plants grown under natural marshy soil medium (SGPs). Thin layer chromatography (TLC) was carried out to investigate the effects of hydroponics on chemical constituents of Lunuwila and the results did not show a significant difference between HGPs and SGPs . Therefore, the findings of this study clearly revealed that the hydroponic technique is suitable for cultivation of Lunuwila. Furthermore, cost benefit analysis showed that the commercially available Albert's solution (50% concentration) is more economical than the other concentrations of nutrient solution that were tested.

REFERENCES

Anees, A.S. (2010). Pharmaceutical Analysis. 2nd ed. Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Jaima Hamdard, Hamdard Nagar, New Delhi, India. pp 198-213.

Anonymous (2004). Compendium of Medicinal Plants, A Sri Lankan Study.Vol iv, Ayurvedic Drug Co-operation, Sri Lanka. pp 39-41.

Anonymous (2002).Hydroponics. Extension and training centre, Department of Agriculture, Sri Lanka.

Das, S.N. (2006). Medicinal Plants For Health and Welth, Agrotech publishing Academy, Udaypur, pp. 57-59.

Joshi A.G., Ashutosh R. P. and Asha M. (2010). Environmental and experimental biology, High Frequency of Shoot Regeneration on Leaf Explants of *Bacopa monnieri,* Department of Botany, Faculty of Science, The Maharaja Sayajirao University of Baroda, India, pp 81-84.

Kapoor, L.D. (1990). Ayurvedic Medicinal Plants, CRC press Washington D.C, USA, pp 61.

Prompittyarat, W., Jetiyanon, K. and Putalun, W. (2007). Determination of Saponin Glycosidesin Bacopa monnieri by Reversed phase High Performance Liquid Chromatography, Thai Pharmaceutical and Health Science Journal, 2 (1): 26 - 35.

Resh, H.M. (2001). Hydroponics food production. A definitive guide book of soil-less food growing method. 6th ed. Woodbridge Press Publishing Company. California.

Wanger, H., Bladf, S and Zgaiki, E.M. (1984), Plant Drug Analysis, Berlin, Germany. pp. 226-299