

UNIVERSITI PUTRA MALAYSIA

GERMINATION OF *Phyllanthus urinaria* AS INFLUENCED BY MEDIA, ENVIRONMENT AND BRANCH POSITION

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GERMINATION OF Phyllanthus urinaria AS INFLUENCED BY MEDIA,

ENVIRONMENT AND BRANCH POSITION

By

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Sincerely,

WONG YING YONG

CERTIFICATION

This study report entitled "GERMINATION OF *Phyllanthus urinaria* AS INFLUENCED BY MEDIA, ENVIRONMENT AND BRANCH POSITION" is prepared by Wong Ying Yong and submitted to the Faculty of Agriculture in fulfillment of the requirement of PRT 4999 (Final Year Project) for the award of the degree of Bachelor of Horticultural Science.

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LIST OF ABBREVATIONS

ANOVA	Analysis of Variance
DAA	Day After Anthesis
DF	Degree of Freedom
GP	Germination Percentage
gr UPM	Germination Rate
LSD	Least Significant Difference
MDG	Mean Daily Germination
NKEA	National Key Economic Area
NS	Not Significant
Р	Phyllanthus
RCBD	Randomized Complete Block Design
RM	Ringgit Malaysia
VI	Vigor Index

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ABSTRACT

Phyllanthus urinaria or 'Pokok Dukung Anak' is a common weed which can be found in cultivated land as well as abandoned area. This plant also serves as traditional herbs in many parts of the world to treat malaria, asthma, hepatitis and other diseases. The main propagation method of P. urinaria is through seeds, however it has been reported that the germination is low. Therefore, in this study, the seeds of P. amarus were grown to maturity and the naturally dehisced seed were harvested. In the first experiment, the seeds were treated with 5 different growing media in combination with 3 different environments in order to obtain high germination in P. urinaria. In the second experiment, the seeds were grown under the structured house and the seeds were harvested on 13 day after anthesis from main stem and first branch for germination improvement. The treatments were considered as two factorials. The seeds were subjected to germination test and monitored for 28 days. The experimental design used was be RCBD. The data was collected every day. The best result obtained from experiment one treatment M2E2 - 50% peat with 50% perlite under structured house. The germination percentage was as high as 85% with mean daily germination which was about 1.7 seeds/ day. The vigor index was 727.70. For experiment two, the best result was obtained in treatment M2B1 - 50% peat with 50% perlite peat where the seeds were harvested from first branch. The germination percentage was 81.7% with mean daily germination which was about 1.7 seeds/ day. The treatment has the highest vigor index (571.67) among the treatments. For better germination in P. urinaria, the recommended media to be used was 50% peat with 50% perlite under structured house. Also, seeds harvested from first branch will give better germination.

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ABSTRAK

Phyllanthus urinaria atau 'Pokok Dukung Anak' ialah satu rumpai yang popular dan boleh dijumpai di tempat terbiar. Pokok ini juga digunakan sebagai herbal tradisional di merata tempat di dunia untuk mengubati penyakit malaria, asma, hepatitis and penyakit yang lain. Cara penanaman P. urinaria adalah melalui biji benih, namun germinasinya adalah sangat rendah. Oleh itu, dalam kajian ini, P. urinaria di tanam sampai matang dan biji benih yang meletup secara semula jadi di tuai. Dalam eksperimen yang pertama, biji benih ditanam di 5 media dengan 3 combinasi persekitaran yan<mark>g berlainan yang bertujuan mendapatkan germinasi yang</mark> tinggi di P. urinaria. Dalam eksperimen yang kedua, beji benih ditanam di bawah rumah struktur dan pada hari ke-13 bunga kembang, biji benih akan dituai dari batang utama dan batang yang pertama untuk memperbaikkan lagi germinasi. Rawatan tersebut akan dilihat sebagai dua faktorial. Biji benih akan ditujukan untuk ujian germinasi dan diperhatikan selama 28 hari. Reka bentuk eksperimen yang digunakan adalah RCBD dan data direkodkan setiap hari. Keputusan yang paling baik adalah didapati daripada rawatan M2E2 – 50% peat dengan 50% perlite dibawah rumah struktur. Peratus percambahan adalah sebanyak 85% dengan purata percambahan setiap hari adalah 1.7 biji benih/ hari. Indeks vigor yang direkodkan adalah 727.70. Untuk eksperimen kedua, keputusan yang terbaik adalah didapati di rawatan M2B1 – 50% peat dengan 50% perlite dimana biji benih tersebut adalah didapati di batang yang pertama. Peratus percambahan yang direkodkan adalah sebanyak 81.67% dengan purata germinasi setiap hari adalah 1.7 biji benih/ hari. Indeks vigor juga adalah yang terbanyak (571.67). Untuk mendapati germinasi yang tinggi dalam P. urinaria, media yang disyorkan adalah

50% peat dengan 50% perlite di bawah rumah struktur. Di samping itu, biji benih yang dituai daripada batang yang pertama juga akan beri germinasi yang tinggi.



CHAPTER 1

INTRODUCTION

Phyllanthus urinaria is a common weed which can be found in cultivated land as well as abandoned areas. The genus Phyllanthus comprises about 800 annual, perennial, shrub, climber, and floating aquatic species. They are commonly found in the tropic and the subtropic regions. Some of the species are termed as 'Pokok Dukung Anak' in Malaysia and Indonesia (Zaki, 2007; Tharakan, 2012). Phyllanthus was initially categorized under the family Euphorbiaceae. However, with more systematic and scientific researches being carried out on this crop, it has now been categorized into the family *Phyllanthaceae*, due to the presence of a biochemical compound which is phyllanthin. Phyllanthin (extracted from the leaves) is a special characteristic for species within the family Phyllanthaceae as phyllanthin does not exist in other families. Due to its biochemical compounds, the local people in India usually use *Phyllanthus species* to treat malaria, asthma, hepatitis and other diseases (Calixto et al., 1998). Intense research has been conducted to study the phytochemical compounds in *P. urinaria* and the compounds extracted have been found to have hepaprotective, antiviral, anticancer, antioxidant, anti - inflammatory, antimalarial, antiplasmodial, diuretic and antibacterial properties (Patel et al., 2011). Due to its extensive medicinal uses, the plant has also become popular in Malaysia and the market price for dried Phyllanthus plant is RM70/kg in Malaysia. According to the World Bank, the global market value of the herbal plants is estimated to rise up to 5 trillion in year 2050. In view of the economic values possessed by P. urinaria, the Malaysian government has listed P. urinaria as one of the top five herbs on focus in the National Key Economic Area (NKEA) in order to generate the interest of the growers to cultivate this herb in large scale and generate more income to boost the economy.

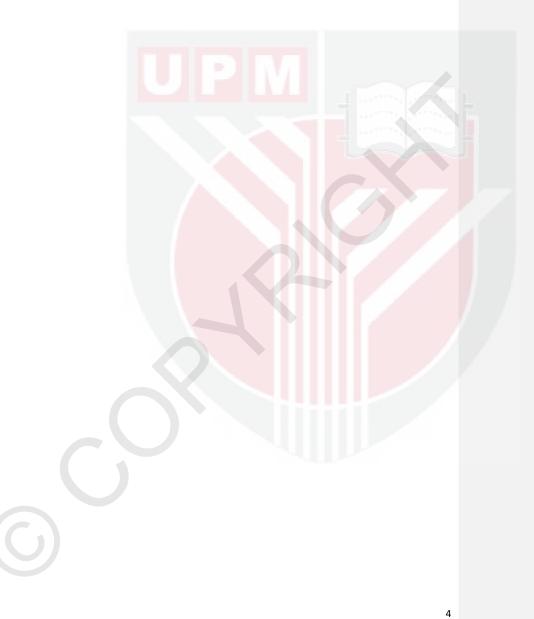
In order to cultivate *P. urinaria*, the main propagation method of the herbs is through seeds. High quality seeds which give high germination percentage are a must for the growers expecting high yield. However, the seeds produced in *Phyllanthus* are reported to be very low in germination, less than 50% in 10 days after sowing (Unander *et al.*, 1995; Rajeswara – Rao, 2012). Also, it had been reported that low germination in *Phyllanthus* could be due to the dormancy in the seeds (Unander *et al.*, 1995). Dormancy in the seeds was mainly due to the surrounding environment such as high temperature, absence of light and so on. Thus, these factors are possibly restricting the interest of farmers to cultivate *Phyllanthus* on large scale. Hence, it is necessary to find a method to either break dormancy or to select a suitable environment for germination of *Phyllanthus* seeds.

Various factors have been shown to influence germination in seeds. Although, Unander *et al.*, (1995) and Rajeswara – Rao (2012) have mentioned that *Phyllanthus* seeds have dormancy, but others have shown that it may not be dormant as the percentage germination changes based on the condition where germination is carried out. One of the options that can be used to improve the germination percentage is by using different planting media and environment during the germination. These two factors have been successfully employed to improve the seed germination rate in *Jatropha curcas* (Gairola *et al.*, 2011). Different growing media can be varied in terms of its pH, water holding capacity, aeration and so on in order to favor the germination of seeds. Different types of media not only effects germination of seeds but also directly influence the rooting system of the plants (Awang *et al.*, 2009). Apart from this, the environment, especially temperature and light during germination processes is also important in enhancing the rate of seed germination. Different plant species may require different optimal temperature and light intensity in order to ensure maximum germination. According to Relf (2009), it is very important to use suitable growing media and maintenance of optimum temperature to achieve high percentage of germination. This is because, when the seeds are sown in minimum temperature, the germination process will occur very slowly as the respiration rate has been lowered. Protein denaturation could happen if the seeds are sowed in maximum temperature (Miller, 2007; Relf, 2009). Hence, sowing the seeds in optimum temperature is very important to avoid slow germination and protein denaturation and to improve germination percentage. Elliott and French (1959) also stated that for germination to occur, the process depended on light and temperature.

Other than media and environment which could influence the germination percentage, it has also been found that seeds harvested from different position of the mother plants could enhance the germination as well. Khatun and Bhuiyan (2011) had claimed that the seeds harvested from the primary branch were better in germination and vigor index than the seeds harvested from secondary and tertiary branches. This could be the seeds near to the nutrient source will get more nutrients than the one located in the far. Selecting seeds from different position of the mother plant of *P. urinaria* and then germinating the seeds under favorable temperature and media might help to enhance the germination of the seeds.

Therefore, this study on *P. urinaria* was undertaken in order to propose a suitable environment for uniform germination of seeds. The specific objectives are as follows:

- i. To determine the germination of *P. urinaria* seeds in different media and surrounding temperature.
- ii. To improve the germination of *P. urinaria* seeds by selecting seeds from different branches.



REFERENCES

- Adusei Fosu, K., Elegba, W., Annor, C., Klu, G., & Danso, K. (2012). In vitro regeneration and morphogenesis in *Phyllanthus niruri* L., an anti-plasmodial herb. *African Journal of Biotechnology*, 11(80): 14542-14552.
- Alan, R., Zulkadir, A., & Padem, H. (1993). The influence of growing media on growth, yield and quality of tomato grown under greenhouse conditions. *II* Symposium on Protected Cultivation of Solanacea in Mild Winter Climates, 366: 429-436.
- Angelis, G., Papadantonakis, N., Spano, T., & Petrakis, C. (2001). Effect of substrate and genetic variation on fruit quality in greenhouse tomatoes: Preliminary results. *Acta Horticulturae*, 548: 497-502.
- Anjanawe, S., Kanpure, R., Kachouli, B., & Mandloi, D. (2013) Effect of plant growth regulators and growth media on seed germination and growth vigour of papaya. *Annals of Plant and Soil Research*, 15(1): 31 – 34.
- Aref, I.M. (2002). The effect of light intensity on seed germination and seedling growth of Cassia fistula (Linn.), Enterolobium saman (Jacq.) Prain ex King. and Delonix regia (Boj) Raf. Alexandria Journal of Agricultural Research. 47(2): 73-80.
- Awang, Y., Shaharom, A. S., Mohamad, R. B., & Selamat, A. (2009). Chemical and physical characteristics of cocopeat-based media mixtures and their effects on the growth and development of *Celosia cristata*. *American Journal of Agricultural and Biological Sciences*, 4(1): 63.
- Bahuguna, V. K., Sood, O. P., and Rawat, M. M. S. (1987). Preliminary studies on the viability, germination and longevity of *Terminalia myriocarpa* seed. *Journal* of *Tropical Forestry*, 3(4): 314-323.
- Baskin, J. M., & Baskin, C. C. (2004). A classification system for seed dormancy. Seed Science Research, 14(1): 1-16.
- Baskin, C. C., & Baskin, J. M. (2014). Seeds: Ecology, biogeography, and evolution of dormancy and germination (2nd edition ed.) Elsevier.
- Bewley, J. D., and Black, M. (1978). Physiology and biochemistry of seeds in relation to germination. Germany: Springer Verlag Berlin Heidelberg.
- Bhardwaj, R. (2014). Effect of growing media on seed germination and seedling growth of papaya cv. red lady. *African Journal of Plant Science*, 8(4): 178-184.
- Bigelow, C. A., Bowman, D., & Cassel, K. (2000). Sand based rootzone modification with inorganic soil amendments and sphagnum peat moss. USGA Green Section Record, 38(4): 7-13.

- Böhme, M., Schevchenko, J., Pinker, I., & Herfort, S. (2005). Cucumber grown in sheepwool slabs treated with biostimulator compared to other organic and mineral substrates. *International Symposium on Growing Media*, 779: 299-306.
- Burton, P. J. (1982). The effect of temperature and light on *Metrosideros* polymorpha seed germination. Pac. Sci. 36(2): 229-240.
- Calixto, J. B., Santos, A. R., & Yunes, R. A. (1998). A review of the plants of the genus *Phyllanthus*: Their chemistry, pharmacology, and therapeutic potential. *Medicinal Research Reviews*, 18(4): 225-258.
- Chapter 3: Tree planting techniques. Farmers' tree planting manual. Retrieved 7th September 2014 from http://www.treeseedfa.org/doc/Manual_English/Chapter 31TreePlantingTechniques.pdf
- Contreras, S. (2008). (2) Seed dormancy. Retrieved from http://seedbiology.osu.edu/HCS631_files/8A%20Seed%20Dormancy,%20after %20MM.pdf
- Cros, V., Martínez-Sánchez, J. J., & Franco, J. A. (2007). Good yields of common purslane with a high fatty acid content can be obtained in a peat-based floating system. *Horttechnology*, 17(1): 14-20.
- Czabator, F. J. (1962). Germination value: An index combining speed and completeness of pine seed germination. *Forest Science*, 8(4): 386-396.
- Elliott, R. F., & French, C. S. (1959). Germination of light sensitive seed in crossed gradiants of temperature and light. *Plant physiology*, 34(4): 454 456.
- Ekpong, B., & Sukprakarn, S. (2008). Seed physiological maturity in dill (*Anethum graveolens* L.). *Kasetsart J*, 42: 1-6.
- Evenari, M. (1952). The germination of lettuce seeds. I. Light, temperature and coumarin as germination factors. *Palestine Journal of Botany*, 5:138-160.
- Fandi, M., Al Muhtaseb, J. A., & Hussein, M. A. (2008). Yield and fruit quality of tomato as affected by the substrate in an open soilless culture. *Jordan J.Agric.Sci*, 4(1): 65-72.
- Fariman, Z. K., Azizi, M., & Noori, S. (2011). Seed germination and dormancy breaking techniques for *Echinacea purpurea* L. J. Biol. Environ. Sci, 5(13): 7-10.
- Filtration and microfiltration catalogue. Retrieved 22nd August 2014 from http://www.chmlab.com/pdf/CHMLABcatalogue.pdf
- Finch Savage, W. E., & Leubner Metzger, G. (2006). Seed dormancy and the control of germination. *New Phytologist*, 171(3): 501-523.

- Gairola, K., Nautiyal, A., & Dwivedi, A. (2011). Effect of temperatures and germination media on seed germination of *Jatropha curcas* Linn. *Adv Biores*, 2: 66-71.
- Gao, H., Zhang, T., Lu, G., Zhang, G., Wu, X., Li, J., & Gong, B. (2006). Effects of different compound substrates on growth, yield and fruit quality of cucumber. *International Symposium on Vegetable Safety and Human Health*, 856: 173-180.
- Gehlot, M., & Kasera, P. K. (2012). Improvement of seed germination behaviour in *Phyllanthus amarus* by acid and mechanical scarification pretreatments. *An International Journal of Ecology*, 19: 1-5.
- Green, M., Lima, W. A. A., Figueiredo, A. F., Atroch, A. L., Lopes, R., Cunha, R. N. V., and Teixeira, P. C. (2013). Heat-treatment and germination of oil palm seeds. *Journal of Seed Science*, 35(3): 296 301.
- Grunert, O., Perneel, M., & Vandaele, S. (2008). Peat-based organic growbags as a solution to the mineral wool waste problem. *Mires and Peat*, 3: 1-5.
- Hanna, H. Y. (2009). Influence of cultivar, growing media, and cluster pruning on greenhouse tomato yield and fruit quality. *Horttechnology*, 19(2): 395-399.
- Hanudin, E., Wismarini, H., Hertiani, T., & Sunarminto, B. H. (2012). Effect of shading, nitrogen and magnesium fertilizer on phyllanthin and total flavonoid yield of *Phyllanthus niruri* in Indonesia soil. *Journal of Medicinal Plants Research*, 6(30): 4586-4592.
- Hartmann, K. T., and Kester, D. E. (1975). Plant propagation, principles and practices (3rd edition). Prentice Hall. Inc Englewood Cliffs, New Jersey.
- Hassani, S., Saboora, A., Radjabian, T., & Husseini, H. F. (2009). Effects of temperature, GA3 and cytokinins on breaking seed dormancy of *Ferula assa – foetidal. Iran. J. Sci. Technol*, 33: 75-85.
- Hipparagi, S., Reddy, D., Gowda, R., & Vishwanath, K. (2007). Studies on physiological maturity in *Phyllanthus amarus* schum and thonn. Seed Research – New Delhi, 35(2): 202.
- Ikeda, H., Tan, X., Ao, Y., & Oda, M. (2001). Effects of soilless medium on the growth and fruit yield of tomatoes supplied with urea and/or nitrate. Acta Horticulturae, 548: 157 -164.
- Khatun, A., & Bhuiyan, M. (2011). Evaluation of seed quality of chickpea (*Cicer arietinum* L.) collected from different branches. *Bangladesh Journal of Scientific and Industrial Research*, 46(4): 507-512.
- Kobryn, J. (2002). The effect of substrate type on the yield and quality of tomato fruits (*Lycopersicon esculentum* mill.) in glasshouse cultivation. *Folia Horticulturae*, 659: 417 424.

- Koller, D. (1972). Environmental control of seed germination. In Kozlowski, T. T. (Ed), Seed biology. Vol II. New York: Academic Press.
- Kuepper, G., & Everett, K. (2004). Potting mixes for certified organic production. (pp. 1–20). Arkansas, US: National Centre for Appropriate Technology.
- Lalithamba, A. (2012). Identification manual for some species of the genus *Phyllanthus* L. of phyllanthaceae with special reference to the Indian subcontinent. In Kuttan, R and Harikumar, K. B. (Ed.), *Phyllanthus species : Scientific evaluation and medicinal applications*. USA: Taylor and Francis Group.
- Larcher, W. (2003). Physiological plant ecology: Ecophysiology and stress physiology of functional groups. Germany: Springer Verlag Berlin Heidelberg.
- Lee B. S., Cho J. Y., Park S. K., Ahn J. S., & Chung S. J. (1999). Effects of substrates on the growth and fruit quality of hydroponically grown tomatoes (*Lycopersicon esculentum* Mill.) cv. Katinka. *Acta Horticulturae*, 483: 147–153.
- Machado, C., Oliveira, P., & Mentz, L. (2003). Scanning electron microscopy of the seed coat in some herbaceous *Phyllanthus* L. (EUPHORBIACEAE). Acta Microscopica, 16(1): 31 – 41.
- Mani, V., & Thulasimani, S. (2012). Antiaging effects of Phyllanthus species. In Kuttan, R., and Harikumar, K. B. (Ed.), *Phyllanthus* species scientific evaluation and medicinal applications (pp. 47-64). United State of America: Taylor and Francis Group, LLC.
- Martins, D. C., Vilela, F. K. J., Guimarães, R. M., Gomes, L. A. A., & Silva, P. A. d. (2012). Physiological maturity of eggplant seeds. *Revista Brasileira De Sementes*, 34(4): 534-540.
- Miller B. Mcdonald. Physiology of seed germination. (2007). Retrieved from http://seedbiology.osu.edu/HCS631_files/4A%20Seed%20germination.pdf
- Narbona, E., Arista, M., & Ortiz, P. L. (2007). Seed germination ecology of the perennial *Euphorbia boetica*, an endemic spurge of the southern Iberian peninsula. *Annales Botanici Fennici*, 44(4): 276-282.
- Olle, M., Ngouajio, M., & Siomos, A. (2012). Vegetable quality and productivity as influenced by growing medium. *Agriculture*, 99(4): 399-408.
- Olubukanla, T. O. (1980). Germination and growth of *Celosia cristata* L., under various light and temperature regimes. *Amer. J. Bot.* 67(6): 854–858.
- Pang, J. H. S., Huang, S. T., Yang, R. C., & Wu, H. T. (2012). The *in vivo* and *in vitro* proapoptotic and antiangiogenic effects of *Phyllanthus* species. In Kuttan, R., and Harikumar, K. B. (Ed.), *Phyllanthus* species scientific evaluation and

medicinal applications (pp. 47-64). United State of America: Taylor and Francis Group, LLC.

- Patel, J. R., Tripathi, P., Sharma, V., Chauhan, N. S., & Dixit, V. K. (2011). *Phyllanthus amarus*: Ethnomedicinal uses, phytochemistry and pharmacology. *Journal of Ethnopharmacology*, 138(2): 286-313.
- Rahimi, Z., Aboutalebi, A., and Abdolrasoul, Z. (2013). Comparison of different medium for production of sweet pepper transplant. *International Research Journal of Applied and Basic Sciences*, 4(2): 307 - 310.
- Rajeswara Rao, B. R. (2012). Cultivation, economics, and marketing of *Phyllanthus* species. In Kuttan, R., and Harikumar, K. B. (Ed.), *Phyllanthus* species scientific evaluation and medicinal applications (pp. 47-64). United State of America: Taylor and Francis Group, LLC.
- Raviv, M., Wallach, R., Silber, A., & Bar-Tal, A. (2002). Substrates and their analysis. (pp 25-101). Athens, Greece: Embryo Publication.
- Relf, D. (2009). Plant propagation from seed. Retrieved from https://pubs.ext.vt.edu/426/426-001/426-001_pdf.pdf
- Reynolds, T., & Thompson, P. (1973). Effects of kinetin, gibberellins and (±) abscisic acid on the germination of lettuce (*Lactuca sativa*). *Physiologia Plantarum*, 28(3): 516-522.
- San Bautista, A., Rueda, R., Pascual, B., Maroto, J., & Lopez Galarza, S. (2005). Influence of different substrates and nutrient solutions on the yields and the incidence of abiotic disorders of broccoli. *Acta Horticulturae*, 697: 275-280.
- Sangkkara, U. R. (1993). Influence of position of panicle on seed and seedling characteristics of rice. *Pertanika Journal of Tropical Agricultural Science*, 16(1): 11 - 15.
- Santhan, P. R. (2010). Variations in the agro climate of *Phyllanthus amarus* in different zones of peninsular India. *National Seminar on Technological Intervention for Sustaining Production of Commercially Valuable Medicinal Crops in India*, TNAU Coimbatore, India.
- Sawan, O., Eissa, A., & Abou-Hadid, A. (1997). The effect of different growing media on cucumber seedling production, fruit yield and quality under greenhouse conditions. *International Symposium Greenhouse Management for Better Yield & Quality in Mild Winter Climates* 491: 369-376.
- Shakila, A., & Rajeswari, R. (2006). Effect of GA3, certain chemicals and media on germination, growth and yield of *Phyllanthus niruri*. *Plant Archives*, 6(1): 121-125.

- Simon, E. W. (1984). Early events in germination. In D. R. Murray (Ed.), Seed physiology. Volume 2. Germination and reserve mobilization. (pp. 77 - 110). Orlando, Florida: Academic Press Australia.
- Swaminathan, C., & Revathy, R. (2013). Improving seed germination in Sapindus emarginatus vahl. Pinnacle Agricultural Research & Management, 1(1): 101 – 103.
- Tharakan, S. T. (2012). Taxonomy of the genus *Phyllanthus* and identification manual. In Kuttan, R., and Harikumar, K. B. (Ed.), *Phyllanthus species: Scientific evaluation and medicinal applications*. USA: Taylor and Francis Group.
- Tzortzakis, N. G., & Economakis, C. D. (2005). Shredded maize stems as an alternative substrate medium: Effect on growth, flowering and yield of tomato in soilless culture. *Journal of Vegetable Science*, 11(2): 57 – 70.
- Unander, D. W., Bryan, H. H., Lance, C. J., & Mcmillan, R. T. (1995). Factors affecting germination and stand establishment of *Phyllanthus amarus*. *Economic Botany*, 49(1): 49-55.
- Wulff, R., and Arias, I. (1972). A bimodal temperature response and effect of light intensity in the photocontrol of germination of seeds in *Jussiaea suffruticosa*. *Planta (Berl.)*, 107: 369 – 373.
- Wolka, K., & Habte, Y. (2014). Effect of seed storage period and condition on viability of *Jatropha curcas* L. seed. Research Journal of Forestry, 8(2): 56-63.
- Zaki, W. W., & Musa, Y. (2007). Effect of planting density on growth and biomass yields of two dukung anak species, *Phyllanthus debilis* and *Phyllanthus urinaria*, grown on alluvial soil. *Journal of Tropical Agriculture and Food Science*, 35(1): 1.