RICERCA INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY RESEARCH AND INNOVATION
Peer Reviewed
ISSN: 2583- 083X

Impacts of Flooding Stress in Fruit Crops and Its Adaptation Strategies

Deena Dayalan, M., *Ramesh Kumar, A., Senthilkumar,S., Srivignesh, S and Manivannan, S.
Department of Horticulture, School of Life Sciences, Central University of Tamil Nadu, Thiruvarur- 610005, TN, India.

Abstract

The climate change has forced the environment pattern to alter globally. Climate change is one of the major causes of abiotic stresses. It has become a major threat to the crop production, limiting productivity and quality of the fruit crops at various stages of the plant life cycle viz., the vegetative and reproductive growth. The major abiotic stresses affecting fruit crop production are drought (moisture deficit stress), flooding (excess moisture stress), high temperature (heat stress), low temperature (cold stress), salt stress (salinity, sodicity stress), acid soils, dust pollution etc.Flooding is one among the stresses affecting the plants growth and development that includes physiological, biochemical and anatomical changes in the plants. Flooding stress affects many fruit crops when there is a prolonged rainfall with poor drainage in the soil. Flooding will have negative effects on the soil and plant environment which includes pest and diseases occurrence, pathogen involvement, and impairs the metabolic process in plants, in severe condition leads to death of the plants. By the knowledgeon the flood stress effects on fruit crops,it's easy to categorize the flood tolerant crops and helps in developing the cultivation aspects for managing the stress condition and mechanisms undergoing in the plants under stress conditions can also be revealed. This review focuses on the impacts of flooding stress in fruit crops, responses of plants to stress, adaptation strategies developed by plants to combat the flood condition.

Key words: Adaptation mechanisms, Flooding stress, Fruit crops, Plant responses.

Introduction

Flooding or waterlogging condition in the soil may lead several adverse actions in the fruit crops those are sensitive to wet conditions. The plants very often produce common symptoms in response to the wet conditions includes chlorosis, wilting of leaves and abscission, epinasty in leaves, hypertrophy, wet conditions develop some injury and diseases in root like *Phytophthora* root rot, *Pythium* and at prolonged condition plants may die. Plants growth and productivity are affected based on their sensitivity to the flood conditions.

Flooding stress

Flooding is defined as the situation of excess water due to high or unusual rainfall events and poor drainage capacity leading to the flooding stress in plants. The waterlogged soil results in poor aeration in soil which is deleterious for plants normal growth and functioning. The waterlogged condition also would develop the plants for susceptibility to various diseases and reduces resistance mechanism, reduced gaseous exchange, leaching of nutrients may occur and all these factors are detrimental for plants survival. The adaptation strategies are to be developed in order to reduce the yield or economic loss aided by flooding stress.

Plants response to waterlogged condition

The plants under stress condition reduced the aerobic respiration process due to the absence of oxygen. Setter *et al*. (1987) reported that the plants under excess water in soil switched for anaerobic respiration which lead to carbohydrate starvation and produced toxic products like ethanol, lactate, malate, and alanine. The plants also reduced the stomatal conductance and reduced chlorophyll content (Neog *et al.*, 2002) resulting in the reduced photosynthesis since the CO_2 diffusion is interfered. The excess water in the soil affects the roots metabolism in water and nutrient uptake thus resulting in wilting of the plants and in severe condition leads to death of the plants.

Influence of flooding stress on respiration

The waterlogged soil results the soil with deficit of oxygen which affecting the root respiration. The first effect of anaerobic soil conditions is a decrease in aerobic respiration in roots due to the anoxic conditions developed by flooding (Armstrong, 1979,Bailey-Serres &Voesenek, 2008). Since the oxygen supply is severely limited leads to the reduced root activity and lacking energy availability. The hypoxia or anoxia condition switches to anaerobic

respiration for energy synthesis as survival of stress which results in the increased accumulations of toxic substances *viz*.,ethanol, acetaldehyde, lactic acid and the ATP is reduced. Therefore it is necessary for plants to increase the anaerobic respiration to increase the amounts of ATP molecules for survival under stress conditions. The prolonged condition of waterlogging in plants results in increased ROS production acts as a secondary messenger under stress conditions and also causingoxidative damage and membrane damages interfering the metabolism of the plants **(**Sharma *et al*., 2012**;**Hasanuzzaman *et al*., 2017**).**

Influence of flooding stress on Photosynthesis

The process photosynthesis in plants requires adequate CO_2 for further growth and development. Theplants in flood conditions have reduced CO_2 assimilation due to the closure of stomata or decreased stomatal conductance and chlorophyll content in the leaves thus resulting in the decrease in photosynthesis in plants. This reduced photosynthetic rate can be inhibited by developing some adaptive strategies in plants that regulates the normal metabolism and functioning in response to the stress conditions.

Influence of flooding stress on endogenous levels of plant growth regulators

Endogenous hormones are essential for a plants appropriate growth and development. In response to the flood condition the endogenous levels of phyto-hormones are disturbed. The stomatal conductance indicates the increased level of ABA which mediates in the closure of stomata and enhances the gibberellin biosynthesis. Anoxia condition in the soil induced ethylene production which may cause premature leaf senescence and chlorosis, as well as adventitious root formation(Jackson and Campell, 1976).The cytokinin level also get reduced in waterlogged condition.This change in the endogenous levels may be one of the stress responsive traits in plants for tolerance under waterlogged or flooded condition.

Influence of flooding stress on Water and nutrient uptake

The efficacy of roots for water and nutrient uptake is harmed by excessive moisture-induced damage to the cell membrane of root tissues. The plant growth is associated with the sufficient nutrient availability in the soil. The submerged soil with absence of oxygen results in decreased ion homeostasis associated with nutrient imbalance under waterlogged condition.Loss of vital nutrients through leaching from the rhizosphere and intermediate metabolites from the roots may cause nutritional stress. The micro (B,Fe,Mn,Zn) and macro nutrient (N,P,K) concentrations are tend to be decreased in the oxygen deficient soil in many fruit crops.

Influence of flooding stress on fruit crops

Many researchers contributed to this study of effects of flooding stress on fruit crop growth. The fruit trees in response to flooded condition over a period of time showing the symptoms like reduce in net CO_2 absorption through stomatal closure, transpiration and under long periods of flooding plants symptoms are withering, and a stop of root and shoot growth reduced the nutritional absorption and in many cases, tree mortality(Schaffer *et al*., 1992).

Mango

According to Schaffer *et al*. (1994) mango is moderately tolerant to flooded conditions. In mango, the flood condition negatively effects the shoot growth due to the decreased CO_2 assimilation which required for growth (Whiley and Schaffer, 1997) and reduced root growth (Larson *et al*., 1991). Adventitious roots (Schaffer *et al*., 1994; Whiley and Schaffer, 1997) are formed in mango when plants under flooded condition, these roots are believed to aid in the absorption and transfer of O_2 to the waterlogged roots in the soil (Kozlowski, 1997). Flooded mango trees responses for survival of long period under waterlogged condition by developing hypertrophic stem lenticels with increased intercellular spaces (Larson *et al*., 1991) which helps in enhancing the diffusion of oxygen in the roots (Kozlowski, 1984).

Banana

In Banana**,** the root metabolism gets affected under waterlogged condition in which the air circulation in roots are absent for normal respiration process to be accompanied and hence the plants shows yellowing symptoms and severe stage results in death of the plant(Ravi and MayilVaganan, 2016).

Avocado

According to Schaffer *et al*.(1992),the avocado trees after flooded, the net assimilation of CO_2 falls, at severe affected condition causing *Phytophthora* root rot. In addition, there is a negative effect of inundation on stomatal conductance and photosynthesis. Schaffer and Whiley (1994) revealed that there was a reduced transpiration in Avocado trees as a result of waterlogging partly due to a reduction in hydraulic conductivity.

Apple

The apple trees exposed to flood stress during the spring season had stunted root growth and reduced the leaves growth. Such leaves dried up quickly in hot days (Heinicke, 1932) and thus the reduced shoot growth.

Papaya

Papaya is most sensitive to flooding stress which requires adequate drainage for better growth and development. Khondakerand Ozawa (2007) studied the effects of papaya growth in relation to deficient oxygen in soil and reported that the oxygen deficit soil in papaya had reduced the plant growth and root development, photosynthesis rate also decreased and dry matter content too got reduced.

Custard apple

In Custardapple, all the commercial species are sensitive to waterlogging (Morton, 1987; Popenoe, 1920) and had resulted in reduction of growth, flowering and fruit set has been reduced except *Annona glabra*a tolerant species of Annonaceae can be utilized for rootstocks in flooded conditions which resulted in high CO_2 assimilation rates.

Adaptation strategies under waterlogged condition

The degree of tolerance to various stress stimuli is generally determined by the particular plantspecies, genotype and developmental stage.Avoidance, tolerance (with standing the stress) and acclimation are the different stress resistance mechanisms in any fruit crop. Several flood tolerant crop species having anatomical and morphological characteristics seem to be linked to their ability of withstanding the long period of flooding stress.

Aerenchyma formation(Anatomical response)

Improved aeration is one of the solutions for dealing with submergence. The plants develop some adaptive strategies to combat the flooding stress which includes the aerenchyma development which is the formation of gas filled spaces in the roots, thus increasing the internal gas movement (Armstrong *et al*., 1991) in the plants for their survival during flood stressed condition. Aerenchyma are formed by different processes includes cell death, cell lysis, modification of intercellular spaces etc.,thus facilitating the oxygen transport in the plants (Seago *et al*., 2005).

Formation of adventitious roots (Morphological response)

Anatomical and morphological structures such as hypertrophied lenticels and adventitious roots grow in many plants, including fruit crops which develop tolerance mechanism under waterlogged conditions. The adventitious root formation is a response of flooding stress which replaces the activity of roots in water and nutrient uptake (Kozlowski and Pallardy, 1984). Fruits crops are also observed with hypertrophic lenticels characterized by increased intercellular spaces and cell lysis which is a tolerance mechanism developed by plants under flood conditions. Thus the adventitious roots and hypertrophy increases the air exchange, water and nutrient uptake and also the excretory site for the toxic substances formed during anaerobic respiration. The adventitious roots are formed by the auxin and ethylene in the plants for survival of plants in the flood condition (Vidoz *et al*., 2010).

Ethylene role in mitigation of flooding stress

Plants under stresses possess various physiological and biochemical mechanisms at cellular level that induces stress tolerance and in improving crop growth and productivity (Upreti and Sharma, 2016). Ethylene is a gaseous hormone that aids in the development of adaptive mechanisms in plants in flooded situations. The flooding response of plants includes rapid accumulation of ethylene, indicates a signaling of stress conditions (Sasidharan and Voesenek, 2015). The oxygen deficit in the roots under flooded conditions drives ACC production and converts to ethylene which gets oxidized when transported to the upward direction in the shoots via xylem (Jackson, 2002), thus also encouraging in the aerenchyma formation (Moore *et al*. 1998 and Colmer 2003).

Rootstock for flooding stress tolerance

Rootstocks play crucial role and mostly determined for the flood tolerance of fruit crops. *Pyrus betulaefolia*, an extremely tolerant species of flooding, in citrus trifoliate orange is tolerant, and *Annona glabra* (Nunez-Elisea *et al*., 1997) can be used as rootstocks for *Annona* species under flood condition which showed increased levels of CO_2 assimilation.

Melatonin role in flooding stress tolerance

The study on application of melatonin for flooding tolerance in *Malus baccata* (Linn.) (Zheng *et al*, 2017) has been reported that Melatonin greatly increased the endurance of apple seedlings to waterlogging stress. Melatonin maintains aerobic respiration, retains photosynthesis, and minimises risks of oxidative stress in plants under waterlogging stress. As reported by Xianbin Gu *et al*. (2020), the exogenous application of melatonin in *Prunus persica* had significant effects on the plants growth, chlorophyll content and maintained the better root activity. Melatonin application also resulted in the increased antioxidant enzymes thus scavenging the ROS and also suppression of anaerobic respiration and enhancing the aerobic process through aerenchyma in roots.

Conclusion

The climate change is one of the factors that is likely to increase several stresses in plants includes uneven rainfall patterns and some poor drainage factors are responsible for detrimental effects in plant growth and productivity. Therefore the efforts to be made on further impacts studies of flooding in fruit crops and the adaptation strategies to be improved for the survival and sustaining the yields.

References

1. Armstrong, W. (1979). Aeration in higher plants. Advances in Botanical Research. 7: 225–332.
2. Armstrong, W., Beckett, P.M., Justin, S.H.F.W. and Lythe, S. (1991).Modelling and others aspects of root aeration by diffusion. In: *Plant Life under Oxygen Deprivation*, pp. 267-282 (Jackson, M.B., Davies, D.D. and Lambers, H., Eds.), SPB Academic Publ., Hague, Netherlands.
3. Bailey–Serres, J. and Voesenek L.A.C.J. (2008). Flooding stress: acclimations and genetic diversity.Annual Review of Plant Biology.59: 313-339.
4. Colmer T.D. (2003). Long-distance transport of gases in plants: a perspective on internal aeration and radial oxygen loss from roots. Plant Cell Environ. 26:17-36
5. Gu, X., Xue, L., Lu, L. (2020). Melatonin enhances the waterlogging tolerance of *Prunus persica* by modulating antioxidant metabolism and anaerobic respiration. J. Plant Growth Regul. https://doi.org/10.1007/s00344-020-10263-5
6. Hasanuzzaman, M., Al Mahmud, J., Nahar, K., Anee, T. I., Inafuku, M., Oku, H.and Fujita, M. (2017). Responses, adaptation and ROS metabolism in plants exposed to waterlogging stress. Reactive Oxygen Species and Antioxidant Systems in Plants: Role and Regulation under Abiotic Stress.*257–281*. doi:10.1007/978-981-10-5254-5-10.
7. Heinicke, A. J. (1932). The effect of submerging the roots of apple trees at different times ofthe year. Proc. Am. Soc. Hort. Sci.,29:205-207.
8. Iyyakutty Ravi and M. Mayil Vaganan.(2016). Abiotic stress physiology of horticultural crops, Abiotic Stress Tolerance in Banana. 12: 207-22

9. Jackson, M.B. (2002). Long-distance signalling from roots to shoots assessed: The flooding story. J. Exp. Bot., 53: 175-181.

10. Jackson M.B and Campell, D. J. (1976). Water logging and petiole epinasty in tomato. The role of ethylene and low oxygen.New Phytol. 76: 21-29.

11. Khondaker, N. A. and Ozawa, K. (2007). Papaya plant growth as affected by soil air oxygen deficiency. ActaHorticulturae.740: 225-232.

12. Kozlowski, T.T. (1984). Responses of woody plants to flooding. In: Kozlowski T.T, ed. Flooding and plant growth. New York Academic Press, 129-163.

13. Kozlowski, T. T. and S. G. Pallardy.(1984). Effect of flooding on water, carbohydrate, andmineral relations. p. 165-193. In: T. T. Kozlowski (ed.), Flooding and Plant Growth.Academic Press, Inc., New York.

14. Kozlowski, T.T. (1997). Responses of woody plants to flooding and salinity.Tree Physiology.Online Monograph No.1.www.heronpublishing.com/tp/monograph/kozlowski.pdp.

15. Larson, K.D., Davies, F.S. and Schaffer, B. (1991).Floodwater temperature and stem lenticel hypertrophy in *Mangifera indica* (Anacardiaceae). American Journal of Botany.78: 1397-1403.

16. Moore, R., Clark, W.D and Voodpich, D.S. (1998). Botany.WBC/ McGraw-Hill, NewYork.

17. Morton, J.F. (1987). Fruits of warm climates. Greensboro, North Carolina: Media, Inc.

18. Nunez-Elisea, R., Schaffer, B., Crane,J.HandColls, A.M. (1997). Effects of flooding on growth and leaf gas exchange of Annona species. Proceedings Congresso Internacional de Anonaceas, University of Chapingo, Texcoco, Mexico, 12–14 November, 1997.124–132.

19. Neog, B., Gogoi, N., Baruah, K.K and Gogoi, N. (2002). Morphological changes associated with waterlogging in rice (*Oryza sativa*). Indian J.Agril Sci., 72(7): 404-407.

20. Popenoe, W. (1920). Manual of tropical and subtropical fruits. London: Hafner Press, Mcmillan Publishers.

21. Sasidharan, R. and Voesenek, L. A. C. J. (2015). Ethylene-Mediated Acclimations to Flooding Stress. Plant Physiology, 169(1): 3-12.

22. Schaffer, B., Andersen, P.C and Ploetz, R.C. (1992). Responses of fruit trees to flooding. Horticultural Reviews.13: 257-313.

23. Schaffer, B., Whiley,A.W and Crane, J.H. (1994). Mango. In: Schaffer B, Andersen PC, eds. Handbook of environmental physiology of fruit crops, Volume 2. Subtropical and tropical crops. Boca Raton, Florida: CRC Press, 165-197.

24. Schaffer, B. (1995). The environment, the urban jungle and politics versus fruit production in south Florida, with special reference to avocado.Proceeding of the Australian Avocado Growers' Federation Conference, Fremantle Australia.127-134.

25. Schaffer, B. (1998). Flooding Responses and Water-use Efficiency of Subtropical and tropical Fruit Trees in an Environmentally-sensitive Wetland. Annals of Botany, 81(4), 475- 481.

26. Seago, J.L., Marsh, L.C., Stevens, K.J., Soukup, A., Vortubová, O. and Enstone, D.E. (2005).A re–examination of the root cortex in wetland flowering plants with respect to aerenchyma. Annals of Botany.96: 565–579.

27. Setter, T.L and Belford, B. (1990). Water logging: how it reduces plant growth and how plants overcome its effects. Western Aust. J. Agric. 31: 51-57.

28. Sharma, P., Jha, A. B., Dubey, R. S. and Pessarakli, M. (2012).Reactive oxygen species, oxidative damage, and anti-oxidative defence mechanism in plants under stressful conditions. J. Bot. 217-37.

29. Upreti, K. K. and Sharma, M. (2016). Role of Plant Growth Regulators in Abiotic Stress Tolerance. Abiotic Stress Physiology of Horticultural Crops, 19–46.

30. Vidoz, M.L., Loreti, E., Mensuali, A., Alpi, A and Perata, P. (2010). Hormonal interplay during adventitious root formation flooded tomato plants. Plant J., 63:551-562

31. Whiley, A.W. and Schaffer, B. (1997). Stress physiology. In: Litz RE, ed. The mango:botany,production and uses. London: CAB International Press, 147-173.

32. Zheng, X., Zhou, J., Tan, D.-X., Wang, N., Wang, L., Shan, D., and Kong, J. (2017). Melatonin Improves Waterlogging Tolerance of *Malus baccata* (Linn.)Borkh.Seedlings by Maintaining Aerobic Respiration, Photosynthesis and ROS Migration.Frontiers in Plant Science, 08.

Importance of Plant Growth Promoting Rhizobacteria (PGPR) in Sustainable Vegetables Production

V. Dhanushkodi, Noorjehan A.K.A. Hanif and N. Tamilselvan
ICAR-Krishi Vigyan Kendra, Sirugamani,
Tiruchirappalli -639 115, Tamil Nadu

Abstract

Vegetable cultivation is an important economic activity of small and margincal farmer's for their livelihood security. But, the yield of vegetables is highly depends on more pesticide application as they frequently experience yield losses due to pest and disease. Recently, use of plant growth promoting rhizobacteria (PGPR) is measured as a major asset for efficient pest management. PGPR CSR Bio is a unique microbial consortia developed for improving yield and income of farmers. In this context, demonstrations were conducted on performance of CSR Bio on major vegetable cultivation (okra, onion and brinjal). Where, CSR Bio was used for seed treatment (3%), soil application (5kg/ha) and foliar spray (3%). The results shows that, application of CSR Bio recorded more yield in okra (21.1 t/ha), onion (15.5 t/ha) and brinjal (18.3 t/ha). Where as in farmers practice, 17.4t/ha, 13.5t/ha and 17.2 t/ha of yield in okra, onion and brinjal was recorded respectively. Favourable net return of Rs. 192024/-, Rs. 232970/- and Rs. 344792/- per hectare and Cost benefit ratio of 2.74, 3.13 and 4.0 in okra, onion and brinjal respectively was obtained through CSR Bio application. Sucking pest and disease incidence was reduced (50-60%) under CSR Bio application. The results of the survey indicated that CSR Bio was useful in getting more income and reduce pesticide application. But, timely non availability of PGPR and lack of awareness were identified as major reasons for non adoption of PGPR application. Hence, it is essential to enhance the availability of PGPR in all vegetable growing areas, create awareness regarding the adoption of PGPR.

Keywords: PGPR, Pest incidence, Vegetables, Yield

Introduction

Plant Growth Promoting Rhizobacteria (PGPR) is a group of beneficial bacteria that actively colonize in plant roots. The application of PGPR to control diseases is an biological approach to increase yield and income without affecting the soil and environment. In general, PGPR application induced the biocontrol activity to release of nutrients, niche exclusion, induced systemic resistance and antifungal metabolites production. Interaction of beneficial rhizobacteria with the plant roots can result in plant resistance against some pathogenic bacteria, fungi, and viruses. PGPR improve plant growth directly or indirectly through supply of nutrients and inhibitory effect on pest and disease. Commonly available PGPR such as *Pseudomonas, Trichoderma, Bacillus, Azospirillum are* capable of facilitating the growth and yield of vegetables.

Vegetables are an integral part of human dietary systems for healthy life. Because, they supply several important nutrients, vitamins, antioxidants for daily need of human health. Annual vegetable crops such as okra, onion and brinjal are cultivated in tropical and subtropical regions. Okra is considered a high-value vegetable crop owing to its high levels of vitamins, minerals, carbohydrates, and fats (Habib, 2016). Onion is a very common crop grown all over the India and is consumed by every family either as raw in the form of salad or as cooked along other spices and vegetables, sometimes flowering shoot 'scape' is also used as vegetable. Onion contains nutrients, carbohydrate, proteins and vitamins etc., Besides it also contains useful medicinal properties. Correspondingly brinjal is also a popular vegetable, highly cosmopolitan and is considered as poor man's crop, thereby grown in almost all parts of India, all round the year except higher altitudes (Jayalakshmi and Praneetha, 2018). Brinjal has high nutritive value and it contains high amount of carbohydrates (6.4%), protein (1.3%), fat (0.3%), calcium (0.02%), phosphorus (0.02%), iron (0.0013%) and other mineral matters. The production and demand for okra, onion and brinjal are relatively high. These crops are grown by farmers during *Kharif* as well as *Rabi* seasons.

Vegetable growers frequently complain yield losses due to insect pests, the yield of these vegetable per hectare is highly depends on use more pesticide. To overcome the yield loss caused by pest and disease incidence, farmers penetrating in adoption of disease-resistant varieties, crop rotation and other disease control measures, but all these

methods have not been successful and effective. The indiscriminate use of agrochemicals including pesticides in vegetable production adversely affects the soil fertility and consequently it accumulate inside the plant parts, thus making it unhealthy for human consumption (Rizvi *et al*., 2017). Now-a-days, the plants inoculation with PGPR is a major asset for organic agriculture. Recently PGPR is receiving more attention as a way to reduce the chemicals without facing yield loss (Pacome *et al*., 2013). CSR Bio is a unique microbial consortia developed by Central Soil Salinity Research Institute, Lucknow for improving yield and income of farmers. It contains *Bacillus pumilus*, *Bacillus thuringensis* and *Trichoderma harzianum*. CSR Bio act as a nutrient mobilizer, soil vitalizer, biocontrol agents for soil born diseases and growth enhancer for salt affected soil. In this context, KVK Tiruchirappalli demonstrated the importance of PGPR in combination on the major vegetable crops such as okra, onion and brinjal grown in Tiruchirappalli region of Tamil Nadu to create the awareness among the farming community to reduce the overload of chemicals on soil and environment.

Materials and Methods

Krishi Vigyan Kendra, Tiruchirappali District, Tamil Nadu conducted demonstration in 15 ha of land area on the performance of PGPR on okra, onion and brinjal in Trichy district during 2016-17, 2017-18 and 2020-21 respectively under KVK operational area. Through field survey, farmers meeting and field diagnostic visit during the cropping period, income of the farmers affected by pest and disease was conceived for this demonstrations. During the demonstrations, improved and recommended technologies under okra, onion and brinjal cultivation were adopted as intervention during the course of front line demonstration. Under the technology demonstration seeds were treated with CSR Bio 3%, further soil application (5kg/ha) and foliar spray at flowering and 15 days later (3%) was done. Trial has been taken up in 15 ha (each 5 ha in 5 locations) of targeted area. Visit of the farmers and extension functionaries were organized at trial plots to disseminate the message at large scale. Yield and economic data were collected from farmers practice (control) and demo plot (CSR Bio applied) and cost of cultivation, net income and cost benefit ratio were computed and analysed.

An interview schedule was developed during 2021 to collect the data on farmers' adoption and constraints faced in PGPR adoption with 75 farmers in the same village. The survey included several open-ended questions to elicit farmers perceptions regarding the system and the broader aspects of income changes in their field. All categories of farmers *i.e.* small (upto 2 ha), medium (>2- 4 ha) and large (>4 ha) were selected and 25 vegetable growers were selected from each group through random technique. The responses were scored on 4 points scales fitting to the statements as very much (4), much (3) not so much (2) and not at all (1) important.

Table 1. Effect of PGPR CSR bio application on the performance of vegetables.

S.No.	Crop	Pests observed	Incidence (%)		Yield (t/ha)		Net income (Rs.)		BC ratio	
			FP	Demo	FP	Demo	FP	Demo	FP	Demo
1.	Okra	Fruit Borer	23	11	17.4	21.1	161038	192024	2.38	2.74
2.	Small Onion	Basal rot	32.5	9.4	13.5	15.5	185150	232970	2.74	3.13
3.	Brinjal	Fruit Borer	28	15	17.2	18.3	310650	344792	3.6	4.0

FP - Farmers Practice

Results

Effect of PGPR on the yield of vegetables

The yield of vegetables was recorded in CSR bio applied plots and compared with farmers practice Table 1. Results indicated that application of CSR bio produced more yield in okra (21.1 t/ha), onion (15.5t/ha) and brinjal (18.3

t/ha), with an increased yield level of 21.2%, 14.8 % and 6.3% respectively. Where as in farmers practices, 17.4t/ha, 13.5 t/ha and 17.2 t/ha of fruit yield were recorded in okra, onion and brinjal respectively. The increased yield of vegetables by the application of PGPR could be due to the application of PGPR-CSR bio involved in enzymes, proteins, antibiotics, etc.. Similar results was obtained by Yagmur *et al*. (2021) and Damodaran *et al*. (2013).

Effect of PGPR on Economics in vegetable production

The economic indicators *viz.*, net income and benefit cost ratio is presented in Table 1. Favouable net return of Rs. 192024/- and cost benefit ratio of 2.74 in okra, net return Rs. 232970/- and cost benefit ratio of 3.13 in onion and net return Rs. 344792/- with cost benefit ratio of 4.0 in brinjal was obtained through CSR bio application. They clearly revealed that the additional net return of Rs. 30986/-, RS.47820/- and Rs.34142/- from the trials were subsistence higher than control i.e. farmers practices in okra, onion and brinjal respectively. Higher economic status might be due to the increasing yield of vegetables and of lesser pest incidence in the PGPR CSR bio applied plots.

Effect of PGPR on pest and disease incidence

Effect of PGPR CSR Bio in pest management is shown in Table 1. Fruit borer is major pest affecting the yield in most of the vegetables. Fruit borer affect fruit quality and yield loss reached more than 30 -45%. Application of CSR Bio reduced 50-60% of fruit borer in okra and brinjal, which could be due to the indirectly benefit, antibiotic production, parasitism of CSR Bio on deleterious microorganisms or root pathogens that inhibit plant growth (Bhattacharyya and Jha, 2012). With respect to basal rot in onion, it was observed minimum damage (9.4%) in CSR Bio applied plot than in farmers practice (32.5 %). Where CSR Bio consists *Bacillus* pumilus, *Bacillus thuringiensis and Trichoderma harzianum,* which act as a beneficial microorganism that are widely used as a bio pesticide and also improve the resistance for pest and disease incidence (Gangwar, 2017). Gholamreza Salehi Jouzani *et al.* (2017) and Jamshidnia, *et al*. (2018) expressed the same view in application of PGPR in Tomato. Hence, to optimize the vegetable production with reduced input, PGPR in vegetable cultivation is recommended (Mekonnen and Kibret, 2021). Pravin Vijan *et al*. (2016) expressed that application of PGPR like *Bacillus*, *Pseudomonas*, *Serratia*, *Arthrobacter*, and *Stenotrophomonas* are involved in systnesis of volatile compound like enzymes, proteins, antibiotics, which help to prevent pest and disease incidence.

Extend of adoption and contraints in PGPR application

The reason for adoption and contraints in use of CSR Bio was diversified and diffères from individual to individual (Table 2). Most of the farmers expressed that application of PGPR useful in getting more income, easy for application, safe for soil and environment through reduction in chemical application. But, timely non availability of input and lack of awareness were identified as major reasons for non adoption of PGPR. The survey indicated that most of the constraints were related to government actions that need to be solved to make use of PGPR effectively. All farmers favoured its adoption in vegetable cultivation. So it was found that all the sampled farmers were favoured in principle, but the farmers were seeking government assistance for adoption.

Table 2: Farmers feedback on adoption and contraints in PGPR application

S.No.	Particulars	M.F M.S	S.F M.S	B.F M.S	Average M.S.
I	**Reason for adoption of PGPR CSR bio**				
1.	Easy for application	3.2	3.7	3.6	3.5
2.	Reduction in pest and disease incidence	2.8	3.2	3.5	3.2
3.	Helpful for yield enhancement	3.4	3.1	3.5	3.3
4.	To obtain more income	3.2	3.7	3.8	3.6
5.	Low cost of input	3.1	3.4	3.2	3.2
6.	Beneficial for soil and environment	3.3	3.5	3.7	3.5
7.	Helpful to reduce pesticide usage	3.1	3.5	3.8	3.5

II	Constraints in adoption of PGPR CSR bio				
	Lack of awareness	3.4	2.8	3.1	3.1
	Timely non availability of inputs	3.5	3.1	3.8	3.5
	Lack of guidance from extensional personnel	2.5	2.9	3.6	3.0
	In adequate training to famers	2.7	2.9	3.4	3.0
	Inadequate demonstration of new technology	2.9	3.5	2.3	2.9
	Deficiency in technical know-how	2.9	2.4	2.8	2.7
	Insufficient follow up service	2.1	2.5	2.9	2.5

M.F.-Marginal farmers , S.F. -Small farmers, B.F- Big farmers, M.S.-Mean score

Conclusion

The findings of the study concluded that application of PGPR favourably influence the yield and reduce the pest damage in vegetable. Therefore, PGPR is highly recommended to reduce the heavy usage of pesticides and to improve the yield and quality of vegetables. Application of PGPR is a cost effective and outstanding approach to attain sustainable vegetable production and also for environmental sustainability. It was evident from the study that farmers were aware of the use of PGPR to save environment, soil and living beings. But, timely non availability of input and lack of awareness were identified as major reasons for non adoption of PGPR. Hence, it is essential to enhance the availability of suitable PGPR in all vegetable growing areas, create awareness to improve knowledge, skill and attitude regarding the adoption of PGPR in vegetable production by extension organizations through organizing various training, demonstration, exposure visit and awareness programmes *etc*.

References

1. Bhattacharyya, P. and Jha, D. (2012). Plant growth-promoting rhizobacteria (PGPR): emergence in agriculture. World Journal of Microbiology and Biotechnology. 28: 1327-1350.
2. Damodaran, T, Rai, R. B,, Jha, S. K,, Dhama, K,, Mishra, V.K., Sharma, D. K., Singh, A. K. and Himanshu Dixit. (2013). Impact of CSR bio - an eco-friendly bio-growth enhancer on increasing the profitability of horticultural crops to small and marginal land holders. International Journal of Current Research. 5(9): 2682-2685.
3. Gangwar, R.K. (2017). Role of biological control agents in integrated pest management approaches. Acta Sci Agri.1:9–11.
4. Gholamreza Salehi Jouzani , Elena Valijanian and Reza Sharafi. (2017). Bacillus thuringiensis: a successful insecticide with new environmental features and tidings. Appl Microbiol Biotechnol.101(7): 2691-2711.
5. Habib, S.H., Hossain Kausar., and Halimi Mohd Saud. (2016). Plant Growth-Promoting Rhizobacteria Enhance Salinity Stress Tolerance in Okra through ROS-Scavenging Enzymes. BioMed Research International. 1-10.
6. Jamshidnia, A., Abdoli, S, Farrokhi, S and Sadeghi R. (2018). Efficiency of spinosad, *Bacillus thuringiensis* and *Trichogramma brassicae* against the tomato leaf miner in greenhouse. Biocontrol. 63(5): 619–627.
7. Jayalakshmi, K. and Praneetha, S. (2018). Evaluation of brinjal (Solanum melongena L.) local types for yield and its quality characters. International Journal of Chemical Studies. 6(3): 292-297
8. Mekonnen, H. and Mulugeta Kibret. (2021). The roles of plant growth promoting rhizobacteria in sustainable vegetable production in Ethiopia. Chem. Biol. Technol. Agric. 8:1-11
9. Pacome, A.N., Eméric, K., Yedeou, O. D., Adolphe, A., Marcellin, A., Rachidatou,S., Emma W. G., Simeon O. K., Lamine, B.M. (2013). Effect of Different Plant Growth Promoting Rhizobacteria on Maize Seed Germination and Seedling Development. American Journal of Plant Sciences. 4: 1013-1021.
10. Pravin Vejan , Rosazlin Abdullah, Tumirah Khadiran, Salmah Ismail and Amru Nasrulhaq Boyce. (2016). Role of Plant Growth Promoting Rhizobacteria in Agricultural Sustainability—A Review. 21: 573-190.
11. Rizvi, A., Almas, Z., Khan, M.S, Saif, S., Ahmed, M. and Shahid, M. (2017). Growth Improvement and Management of Vegetable Diseases by Plant Growth-Promoting Rhizobacteria. In: Growth Improvement and Management of Vegetable (A. Zaidi, M.S. Khan, eds.). Springer International Publishing AG. 99-123.

12. Yagmur, B. and Gunes, A. (2021). Evaluation of the Effects of Plant Growth Promoting Rhizobacteria (PGPR) on Yield and Quality Parameters of Tomato Plants in Organic Agriculture by Principal Component Analysis (PCA). Gesu*nde Pflanzen*. 73: 219-228.

Prospects in Organic Agriculture and Agro Forestry

Noorjehan A.K.A.Hanif[1], V.Dhanushkodi[2] and N. Tamilselvan[3]

Tamil Nadu Agricultural University
[3]ICAR KVK Sirugamani– 639 115, Tiruchirappalli district, Tamil Nadu

Abstract

Intensive agriculture has led to soil fertility issues due to improper crop rotation and excessive application of synthetic fertilizers paving way for organic agriculture in a systematic method. In India Organic Agriculture is practiced in lesser, however, the number of organic growers are the highest here. At present, Organic agriculture is refers to any farming system that uses organic methods based on the Principles of Organic Agriculture, irrespective of its certification. Driving forces of organic agriculture are Consumer or market-driven, Service-driven and Farmer-driven. Different type of organic farming refers to Natural, Zero-chemical, Bio dynamic, Biological, Humus, Compost, Sewage and Integrated farming. In India, the factors that affect the adoption of organic farming are economic, social, marketing, cultivation and government policy. For sustainable agriculture and increased food production, agroforestry provides the best solution through integration of biodiverse processes to increase yields, minimizing harmful effects. Major agro forestry practices are alley cropping, silvopasture, riparian buffers, windbreaks and forest farming. India's position in global trade would enhance enormously with the help of capacity building and creation of awareness of the producers. Henceforth organic farming can be promoted successfully in India through government schemes and imparting exclusive training in organic farming and financial support for extensive adoption.

Key words: Organic agriculture, agro forestry, factors of adoption and constraints in organic farming

Introduction

Intensive agriculture has led to severe soil fertility issues including macro and micro nutrient deficiencies due to improper crop rotation. Excessive application of synthetic fertilizers and pesticides has paved way for organic agriculture in a systematic method. The excessive fertilizers especially nitrate runoff in catchment area contaminates water bodies thereby causing water pollution at the reservoirs. Pulverized soil due to deep ploughing when met with heavy rains leads to soil erosion. Use of heavy machinery leaves the soil compacted in addition to increasing fuel requirements for cultivation. Indiscriminate use of pesticides and growth hormones has left the agricultural produce unworthy. Unethical animal husbandry practices like cruelty is being imposed on draught animals at the time of housing, feeding, and slaughtering. Monoculture practice leads to irrevocable loss of biodiversity. Hybrid varieties and Genetically Modified Organisms have reached a stage where the native species are under elimination threat (Pointer, 2021). Hence a dire need is felt for a basic shift from intensive farming to sustainable natural organic farming to feed population of future in a healthy way.

Organic farming policy was introduced in India during 2005. In India, about two per cent of the net cultivated area *viz*. 78 million hectares are covered under organic farming under National Programme for Organic Production (NPOP), Mission Organic Value Chain Development for North Eastern Regions (MOVCD)and under Paramparagat KrishiVikasYojna (PKVY).With regard to number of organic farmers, India ranks first in the world. India possess 1.9 million farmers of which 1.3% of 146 million are agricultural landholders. Further, the natural growers, especially tribal cultivators, who do not use synthetics are not covered in the number count. Sikkim is the first Indian fully organicstate. Many states have announced their organic policy to catch the momentum in growing natural food. Three stateswhich accounts for the largest area under organic cultivation with 4.9%, 2.0%, and 1.6% are Madhya Pradesh, Rajasthan, and Maharashtra respectively (Pointer, 2021).

Concepts and Definitions – Organic Agriculture

"Organic agriculture is a holistic production management system which promotes and enhances agro-ecosystem health, including biodiversity, biological cycles, and soil biological activity. It emphasizes the use of management practices in preference to the use of off-farm inputs, taking into account that regional conditions require locally

adapted systems. This is accomplished by using, where possible, agronomic, biological, and mechanical methods, as opposed to using synthetic materials, to fulfil any specific function within the system." (FAO/WHO Codex Alimentarius Commission, 1999).

As per IFOAM General Assembly, 2008, Organic Agriculture is a production system that sustains the health of soils, ecosystems, and people. It relies on ecological processes, biodiversity and cycles adapted to local conditions, rather than the use of inputs with adverse effects. Organic Agriculture combines tradition, innovation, and science to benefit the shared environment and promote fair relationships and good quality of life for all involved.

At present, Organic agriculture is refers to any farming system that uses organic methods based on the Principles of Organic Agriculture, irrespective of its certification (IFOAM, 2015).

Organic agriculture systems and products are not always certified and are referred to as "non-certified organic agriculture or products". This excludes agriculture systems that do not use synthetic inputs by default (e.g. systems that lack soil building practices and degrade land). Three different driving forces that can be identified for organic agriculture: (FAO/WHO Codex Alimentarius Commission, 1999)

- *In Consumer or market-driven organic agriculture,* products are clearly identified through certification and labelling and consumers has a strong influence over organic production, processing, handling and marketing.
- *Service-driven organic agriculture*demands for subsidies for organic agriculture such as in the European Union (EU) to generate environmental goods and services, like reducing groundwater pollution and for creating a more biologically diverse landscape.
- *Farmer-driven organic agriculture* envisages that conventional agriculture is unsustainable to improve family health, farm economies and/or self-reliance of farmers. Organic agriculture is adopted as a method to improve household food security or to achieve a reduction of input costs and certification. Small farmers have developed direct channels to deliver non-certified organic produce to consumers in case o developing countries.

Organic agriculture grows and develops based on the Principles of Health, Ecology, Fairness and Care. These principles express that organic agriculture can make a difference and improve overall agriculture in a global context.

1. *Principle of Health:* Organic agriculture should sustain and enhance the health of soil, plant, animal, human and planet as one and indivisible.
2. *Principle of Ecology:* Organic agriculture should be based on living ecological systems and cycles, work with them, emulate them and help sustain them.
3. *Principle of Fairness:* Organic agriculture should build on relationships that ensure fairness with regard to the common environment and life opportunities.
4. *Principle of Care :*Organic agriculture should be managed in a precautionary and responsible manner to protect the health and well-being of current and future generations and the environment (IFOAM General Assembly, 2008).

The meaning comprehensive definition of organic farming revolves around promotion and maintenance of biological activity and biological cycles to achieve an ecologically stable agricultural production. The supply of nutrients and control of pests are the natural corollaries of biologically stable system. Hence, understanding the concept of organic farming within the limited provision of nutrients by controlling pest restricts its broad view of organic farming (Joshi, 2016).

Many new terms are popularly used to represent different type of organic farming. Some of them are (Joshi, 2016):

1. **Natural Farming:** It signifies the method of farming, wherein a crop is cultivated within the natural forces at any given place *i.e.* with least human interference involving least operations of weeding, manure/fertilizer application and control of pest; everything is left to nature. This may be a useful system to develop soil microflora and soil fertility in long run. But it is certainly not an encouraging method of farming to achieve higher agricultural production.

2. **Zero-chemical farming:** It is a method offarming, where the use of synthetic chemical is avoided in to that it is synchronized with the harmony of local eco system with supply of natural nutrient and pest control.
3. **Bio dynamic farming:** It emphasizes on use of forces with living nature which include practices of herbal preparations against pests, methods to enhance decomposition in manures and composts, use of cosmic rays to enhance soil fertility (Gold, 2007).
4. **Biological farming:** It is a system of crop production, in which the producer attempts to improve the biological balance between the flora and fauna, of that a stabilized situation of agro ecosystem situation helps in natural control of pests or in supply of nutrients. (Gold, 2007)
5. **Humus farming:** It is a type of farming, wherein the crop is expected to grow by utilizing the humus formed out of constant decomposition of biomass. (typical for tropical climate)
6. **Compost farming:** In this system of farming, adequate amount of compost prepared out of agricultural wastes, cow dung, poultry manure or out of vermicultureis applied to meet the nutrient requirement of crops.
7. **Zero budgetfarming**: This type of farming encourages the resource poor farmers to meet their input requirement from minimum cattle rearing and usage of cow dung and cow urine obtained from the farm reared cow. These resources are converted to value added products thereby leading to reduction in cost of cultivation.
8. **Integrated farming**: It is a type of arming, where animal based enterprises are integrated with crop husbandry activities in such a way that each one is supportive and dependent on other. Integrated farming System (IFS) justifies most objectives of organic farming.

Discussion

Presently conventional agriculture is unsustainable and inadequate to face climate change, environmental pollution, food security and dependence on fossil energy along with decline of natural resources and biodiversity. These problems are related to monocultureand consequently simplified agroecosystem. There is need to improve individual agronomic techniques to increase the use-efficiency of external inputs (e.g. synthetic inputs, fossil fuels), without modifying the structure and functions of the whole system,. Current organic farming systems adopting the so-called input substitution approach remain intensive and highly specialized and require system diversification and redesign of the agroecosystem to increase the spatial and temporal diversification of all its components and promote positive ecological relationships between them. Agroforestry is an best agricultural approach on the diversification of the agroecosystem production components (woody perennials, such as trees or shrubs, plus crops and/or livestock) and its interactive agroecological relationships. Based on the above facts, adoption of agroforestry practices pave way for increasing the sustainability of organic farming (Rosati *et al*., 2021).

In India, farmers have cultivated crops with green manures, green leaf, farmyard manures, sheep penning and thus have grown crops under natural way for time immemorial Sustainable Development Goal targets to end hunger, achieve food security and to improve nutrition and promote sustainable agriculture (Pointer, 2021).

Threats to Organic Farming in India

- ✓ *Awareness* among farmers about the latest organic farming practice and its potential benefit is a hurdle in development of organic farming in India
- ✓ *Feasibility of Marketing* are to be ascertained over conventional produce **b**efore the beginning of organic crop cultivation.
- ✓ *Nominal prices* for the produce are not realized for the organic products is a threat to organic growers.
- ✓ *Limited organic nutrient supply in organic farming due to lesser biomass availability* is another challenge.
- ✓ *Adequate Infrastructure facility* and support are yet to be formulated by State governments apart from NPOP guidelines.
- ✓ Inadequate certifying agencies is still demanding in Indian States.
- ✓ *Costs of critical inputs are higher* thanthose of industrially-produced agrochemicals used in the conventional farming system.

Remedial measures to Promote Organic Farming

- *Substantial Financial support* is necessary to promote organic farming in India through government subsidies and to meet out the marketing price demnd o organic growers.
- *Development of Markets* for organic produces through various farmer and consumer associations / groups.
- *Creating vast awareness* on the benefits of organic farming against conventional farming through campaigns and mass media among the farmers and consumers.
- *Identification of suitable crops* for organic farming is most significant for its promotion. For example, soybean cultivation in Madhya Pradesh

Review of Research Findings on Organic Farming

Noorjehan (2004) reported that cent percent of paddy farmers adopted practices *viz.*, application of farmyard manure, green manures, green leaf manures, weed compost, *Azospirillum, Phophobacteria*, neem oil 3% and neem seed kernel extract 5% while cent percent sugarcane growers adopted application of farmyard manures, green manures, crop residue composts, sewage waste and pressmud. While least adoption was found for latest bio control agent like *Trichogramma chilonis, Platygaster oryzae and other bio pesticides.*

Iman *et al.* (2010) revealed thatbuying coffee husk for growing pineapple expensive, problem of weeds, absence of niche market and premium price for organic fruits in the local and regional markets, more time needed for weeding than herbicides, cannot substitute cereals or other crop for family consumption, companies do not often come to buy organic, pineapple thorns pricking while weeding and pests and diseases problems are the major constraints for organic agriculture among pineapple growers of Uganda.

Bera *et al.* (2013) indicated better performance of IRF organic package as compared to all other organic packages of practice in terms of crop productivity, cost of production and soil quality development in Organic Tea cultivation.

Hill (2016) concluded from the case study research in West Bengal that Participatory Guarantee System provides an ideal system formarginal and small farmers for eay organic certification of their produce enabling them for sale in local and urban markets boosting their incomes and that organic farming led to health life.

According toKhosla (2006) following are the Organic Standards to be followed

1. Prohibition of Synthetic chemical fertilizers
2. Organic fertilizers only to be used
3. Prohibition of Synthetic chemical pesticides and herbicides
4. Botanical pesticides are allowed
5. Proper cleaning of Farming equipment that is used for conventional farming is a must
6. Clear labelling of bags used for harvest of produce
7. Prohibition of all GMOs
8. Proper measures to check erosion is to be adopted by farmers
9. Burning of green material and crop residues should be minimized
10. Humane way of treating livestock must be followed Conversion period allowed for full organic is three years
11. The organic farmer must attend in the PGS meetings of their local group and take an organic pledge
12. Organic farmers must complete a peer-appraisal of another farm; and also have a successful peer review of his own farm

Azam and Shaheen (2019) found five major factors that affect the adoption of organic farming (economic, social, marketing, cultivation, government policy) in India and that marketing and government policy factors were most crucial in influencing all types of farmers irrespective of their educational level. The farmers with more farming experience were more concerned about social factors and the farmers using lease farms were found to be concerned about the economic viability of organic farming.

Azam and Shaheen (2019) suggested that without government support, the adoption of organic agriculture seems to be a highly challenging task in a situation, where majority of the farmers fall under the small and marginal category.

Agroforestry

Agro forestry is a concept of growing trees with agriculture encouraging positive interactions to improve farm resilience, leading to enhanced productivity, biodiversity and other mutual benefits.

Agroforestry is a farming where in integrated land use combines both the elements of agriculture and forestry in a sustainable production system *i.e.* agroforestry can be described as 'growing trees on farms' integrating both ecologically and economically of the woody elements *viz.*, hedgerows, windbreaks, buffer zones, trees in pasture, and small woodlands. Agroforestry systems in wider adaptation include standard trees, orchard trees and/or coppice systems are grown in rows between crops or pasture in an alley-cropping design. Interactions between the trees and crops and/or livestock elements in an agroforestry system lead to higher productivity compared to conventional systems. Further improved soil management, microclimate modification, shelter, weed control, natural fencing, carbon sequestration and nutrient recycling are envisaged.Through Agroforestry systems farmers' livelihood are supported through food, fuel, timber, fodder and forage, fibre, gums and resins. Also provides materials for thatching, hedging, gardening and services through recreation, craft products and medicinal products.

Five agro forestry practices in general practiced are (Matthew and Sarah, 2016)

1. ***Alley cropping*** is growing field crops between rows of treesgrown for timber or fruits and nuts providing longer-term revenue. The alley crops which provides short-term income includes a variety of grains, vegetables, or forages cut for hay. The research findings concluded that the system produces 40% more product per given area than if the two crops were grown separately.
2. ***Silvopasture*** is a system which incorporates livestock along with mixture of trees and pasture in such a way that the spacing of the trees is so planned for allowing enough sunlight for the forages below and the livestock are kept away from damaging the treesin the initial period. There is a shade effect of trees for livestock protecting from heat in summer and from wind strokes in cold winter.
3. ***Riparian buffers*** is planting of two or three "zones" of vegetation based on the proximity to the waterway, slope, and producer needs to evade from erosion, nutrient leaching, or habitat loss. Usually there are vegetation that vary in composition.
4. ***Windbreaks*** or shelterbelts is a agroforestry practice to prevent wind erosion, provide habitat for wildlife, increases water availability to nearby crops through reduced evapotranspiration and minimizes snow scorching effects.
5. ***Forest farming*** is agroforestry practice which includes mushrooms, medicinal herbs (eg. ginseng and goldenseal) and woody ornamental plants cultivation.

For sustainable agriculture and increased food production, agroforestry provides the best solution through integration of biodiverse processes to increase yields, minimizing harmful effects (Matthew and Sarah, 2016).

Conclusion

Organic agriculture is gaining momentum as an alternative method to the modern system. The ill effects of the conventional farming system are felt in India in terms of the unsustainability of agricultural production, environmental degradation, health and sanitation problems, etc. able to convert 2-10 per cent of their cultivated areas into organic farming in in many countries. The demand for organic products is growing fast (at the rate of 20 per cent per annum in the major developed countries). The difficulties like laying down of the National Standards for Organic Production (NSOP) and the approval of four accreditation agencies whose expertise is limited to a few crops makes slower progress in adopting organic farming.

Organic agriculture in India can be envisaged based on

1. Through governments (Central, state and lower level bodies) substantial financial support is very much essential to promote organic farming. Similar to the very liberal subsidies provided by the governments in other countries, Indian organic farmers need to be providedwith required government subsidies.
2. Market facility for the organic products is significant to promote domestic sales and balance demand and supply for organic products in the country with proper links.
3. Reduce the costs of certification and simplify the process by linking with Farmer producer organizations to get accredited for inspection and certification in accordance with the NSOP.
4. Increase the awareness of the farmers and consumers on the benefits of organic farming against the conventional system through campaign and identify crops for organic cultivation. Eg. Soyabean in Madhya Pradesh

Educating and empowering the farmers with adequate technical support and creating awareness, organic farmers could soon be reinforcing their rightful place in the global agriculture trade. In a world bruised by the COVID pandemic, the demand for safe and healthy food is showing an upward trend and hence could be an opportune moment to be captured for a win-win situation for our farmers, consumers, and the environment. [Pointer, 2021]Henceforth organic farming can be promoted successfully in India through government schemes and imparting exclusive training in organic farming and financial support for extensive adoption.

References

1. Azam, M.S. and Shaheen, M. (2019), "Decisional factors driving farmers to adopt organic farming in India: a cross-sectional study", *International Journal of Social Economics*, Vol. 46 No. 4, pp. 562-580. https://doi.org/10.1108/IJSE-05-2018-0282
2. Barik, G.C., and Mazumdar A.K. (2013). Need for a comprehensive approach to ensure sustainable and cost-effective organic tea cultivation - An experience from Model Farm Maud T.E. (Assam), under FAO-CFC-TBI Project in National Seminar On "Organic Tea', Organized by: Tea Research Association, Toklai Experimental Station, Assam, & Tea Board of India, January 8, 2013.
3. FAO/WHO Codex Alimentarius Commission, 1999http://www.fao.org/organicag/oa-faq/oa-faq1/en/
4. Gold, Mary,V. (2007). Sustainable Agriculture: definitions and terms, USDA publications for Alternative farming systems Information Centre, pp.1-13.
5. Hill, Joe K. W. (2016). Organic agriculture in India and Participatory guarantee systems (pgs): A case study from West Bengal. Jharkhand Journal of Development and Management Studies, XISS, Ranchi, Vol. 14, No.2, June 2016, pp. 7037-7055
6. IFOAM (2015). *Position paper. The full diversity of organic agriculture: What we call organic*. Bonn: IFOAM . Organics International
7. IFOAM, General Assembly (2008) Definition of organic agriculture. https://www.ifoam.bio/why-organic/organic-landmarks/definition-organic
8. Iman Raj Chongtham, Andreas de Neergaard and Didier Pillot. (2010). Assessment of the strategies of organic fruit production and fruit drying in Uganda Journal of Agriculture and Rural Development in the Tropics and Subtropics, Vol. 111 No. 1 (2010) 23-34
9. Joshi, Mukund. (2016). New vistas of Organic Farming 2nd revised and enlarged edition. Scientific Publishers. India.
10. Khosla, R. (2006). *A participatory organic guarantee system for India.* Food and Agriculture Organization. Retrieved from http://tianguisorganicos.org.mx/wp-content/uploads/SPG/biblioteca/SPGindia.pdf p. 21
11. Matthew Heron Wilson and Sarah Taylor Lovell. (2016). Agroforestry—The Next Step in Sustainable and Resilient Agriculture. Sustainability 2016, 8, 574; (www.mdpi.com/journal/sustainability)
12. Noorjehan, A. K. A. Hanif. (2004). Organic farming in Tamil Nadu: A multidimensional analysis. Ph.D thesis topical research work, Tamil Nadu Agricultural University, Coimbatore, pp 47.

13. Pointer. (2021). State of organic farming in India April 2021, https://www.10pointer.com/current-affairs/state-of-organic-farming-in-india
14. Rosati, Adolfo, Robert Borek and Stefano Canal. 2021. Agroforestry and organic agriculture. Agroforestry System June 2021 95 (4) pp: 805–821
15. Agroforest Syst (2021) 95:805–821
16. https://doi.org/10.1007/s10457-020-00559-
17. Agroforest Syst (2021) 95:805–821
18. https://doi.org/10.1007/s10457-020-00559-
19. Agroforestry and organic agriculture

Production Technology of Rambutan

V. Krishnamoorthy*, V.P. Shanthi, and K. Indhumathi
Department of Fruit Crops, Horticultural College and Research Institute for Women,
Tamil Nadu Agricultural University, Trichy-620 027, Tamil Nadu.

Abstract

Rambutan (Nephelium lappaceum var. lappaceum) is a unique fruit tree from the Sapindaceae family. It is a delicious juicy fruit with a good flavor and aroma. The edible part is jelly like aril being attracted by children. It is widely cultivated in South East Asian countries and exported to other countries. It can be cultivated in the areas having temperature of 22–30°C. It best suitable for Sandy loam to clay loam soil. In India, Arka Coorg Peetabhand Arka Coorg Arun varieties available cultivation. The commercially cultivated varieties are Lebakbulus, Binjai, Seematjan, and Rapiah from Indonesia (Anonymous, 1992); R134, GulaBatu, Muar Gading, Khaw Tow Bak, Lee Long, and Daun Hijau from Malaysia; Deli Cheng and Jit Lee from Singapore; Seematjan, Seenjonja and Mahalika from the Philippines (Anonymous, 2003) and Rongrien, Seechohmpoo and Bangyeekhan from Thailand. The vegetatively propagated materials may be planted at 10X10m spacing.The full grown 12 years old tree may supplied with 2.4kg, 0.30kg, and 1.5kg of NPK per tree per year. There are three types of flowers in Rambutan - Staminate (m), hermaphrodite functional female (hff) and hermaphrodite functional male (hfm). In the hff type, bifid stigma split open during anthesis facilitates pollination but the stamens remained closely appressed to the sepals and the anthers never dehisced. In the hfm type, cross pollination is essential for fruit set, the stamens are erect at the time ofanthesis and the anthers dehisce viable pollen but the stigma of the rudimentary pistil will not open. The panicle was found to be comprised of 94.41 and 5.99 per cent of hermaphrodite functional female and hermaphrodite functional male respectively.Hermaphrodite cultivars are economical. The duration for fruit maturity from flowering is up to five months.

Key words: Aril, Hermaphrodite, *Nephelium lappaceum*, Rambutan, Spinterns,

Introduction

Rambutan (*Nephelium lappaceum* var. *lappaceum*) is a unique fruit tree from the Sapindaceae family. It is indigenous to the Malay Archipelago, which includes Indonesia, Malaysia and southern Thailand. Rambutan is grown in Malaysia, Thailand,Philippines, Northern Australia, Sri Lanka, India, Madagascar, Costa Rica, Congo and some South American countries. Thailand, Malaysia and Indonesia are the world's largest producers of rambutan (Coronel,1983). The area under cultivation is expanding in Indonesia, Malaysia, Australia, Philippines and Hawaii (Morton, 1987 and Zee, 1983). In India rambutan is cultivated in Kerala, Karnataka and Tamil Nadu. There is good potential for expansion this crop in Kerala, Tamil Nadu and Karnataka (Ravisankar, 2007).

The word *rambutan*, derived from the Malay word *rambut*. It is largely grown throughout Southeast Asia. It is an evergreen tree grows upto 12–20 m. The leaves are pinnate alternate, length is 10–30 cm, pinnate, with 3-11 leaflets, each leaflet length is 5–15 cm and breadth is3-10 cm, with an entire margin. The flower is very small 2.5–5 mm, apetalous and emerge in erect terminal panicles 15–30 cm wide. Rambutan trees are male, female, or hermaphroditic. The male tree bears only male flowers which are apetalous with 5-7 stamens andaccommodates a rudimentary ovary.A single panicle contains 300 to 500 flowers. The hermaphrodite treesbearsbisexual flowers with bilocular ovary and bifid stigma and six nonfunctional stamens. A panicle bears 500 -800 flowers. The hermaphrodite trees also bear 0.5 to 0.8 % male flowers, while female tree bears only female flowers. The fruit skin carries numerous long, thick yellow or pink and red colored hairy growths (Spinterns). The fruit is a round to oval drupe with a diameter of 3–8 cm and a length of 3–8 cm, borne in a loose pendant cluster of 10-20. The fruit skin is leathery, red or yellowcolour. The outer surface has soft fleshyhairs called spintern. The edible portion is white or rose scented is ariladhering to seed, coourless and sweet taste. The seed is oblong and flattened, glossy brown and 2–3 cm long. (Tripathi *et al*., 2014).

Food Uses

It is a delicious fruit with a good flavor and aroma. It is a good source of sugars, vitamin C and other vitamins and minerals. It can be used as a fresh dessert fruit or can be processed into different items like dried fruit pulp, jams, jelly, fruit concentrates etc (Wills *et al*., 1986).

Rambutans are most commonly consumed after removing the spintern skin. It does not cling to the flesh. The seeds are consumed after roasting by traditional people but the raw seeds are said to be poisonous. The fruit contains moisture of about 82.3%, carbohydrate 16%, calcium (10.6mg), phosphorous (12.9mg) and ascorbic acid (30mg) and the total soluble solids range from 14-20 ^{0}Bx (Linn, 1992).

Other Uses

Seed fat

The seed kernel contains37-43% of a solid, white or yellow fat resembling cacao butter. The oil is heated to become yellow to get agreeable flavor.Palmitic (2.0%), stearic (13.8%), arachidic (34.7%), oleic (45.3%), and ericosenoic (4.2%) acids, as well as fully saturated glycerides, are present in the oil (1.4 percent). The oil is used in making soaps and candles (Tee, 1982).

Medicinal Uses

The mature fruit is astringent, stomachic; acts as a vermifuge, febrifuge, and is taken to relieve diarrhoea and dysentery. The leaves are used for relieving. The bark is astringent, decoction prepared used as a remedy for thrush. A decoction of the roots is taken as a febrifuge (Suganthi and Marry Josephine, 2016)

Toxicity

Traces of alkaloids present in the seed, saponin and tannin are present in testa. The seeds are poisonous, bitter and narcotic. The outer fruit skin contains toxic saponin and tannin.

Climate

Rambutan is commercially grown in warm tropical climate. It can be cultivated upto the elevation of 700 metre above sealevel. It requires optimum temperature of 22–30°C for better growth. It is very sensitive to low temperature and growth of plant is severely affected when the temperature goes below 10^{0}C.Defoliation occurs as a result of the reduced temperature, which impacts panicle emergence and fruiting. The temperature above 40^{o}C affects the growth and development of plants. Fruit development is hampered by low humidity. Rambutan requires rainfall of 200-500cm distributed throughout the year.The initiation of floral bud takes place after a short dry period.High rainfall during maturity period causes fruit cracking (Anonymous, 2003).

Soil

Sandy loam to clay loam soils with high organic matter with good drainage are most suitable for rambutan cultivation. Soil pH of 4.5 to 6.5 is best for its growth. Water logged conditions unsuitable growing rambutan.

Varieties

There are more than 200 cultivars available throughout tropical Asia. Most of the commercial cultivars are hermaphroditic. The commercially cultivatedvarietiesare Lebakbulus, Binjai, Seematjan, and Rapiah from Indonesia (Anonymous, 1992); R134, GulaBatu, Muar Gading, Khaw Tow Bak, Lee Long, and Daun Hijau from Malaysia; Deli Cheng and Jit Lee from Singapore; Seematjan, Seenjonja and Mahalika from the Philippines (Anonymous, 2003) and Rongrien, Seechohmpoo and Bangyeekhan from Thailand. In India, the Indian Institute of Horticulture Research's (IIHR) Horticultural Experiment Station, Chettalli, released two varieties and identified two promising lines (Tripathi, 2014).

Arka Coorg Arun :Itis an early maturing variety. The trees are medium in size and semi-spreading in nature. Fruits are dark red in colour. Average fruit weight is between 40 to 45 g. Fruits aril can be easily removed from the seed without piece of testa. The aril is white in colour, thick, firm, dry and sweet (TSS- 20 ^{0}Bx) with recovery of about 42 per cent (Tripathi, 2014).

Arka Coorg Peetabh: The tree canopy is semi-spreading,midseasonandregular bearer. Yellow colour fruits with

25- 30g weight. Aril colour is white, juicy, sweet with total soluble solids-21^{0}Brix and gives 41 percent aril recovery (Tripathi, 2014).

CHES-26: The trees are medium sized and semi-spreading in nature. This is an early maturing variety. Fruits are dark red colour with the average fruit weight of 35 to40g. The aril is attached to the testa and seed firmly. White colouraril,juicy, sweet with the total soluble solids of 19^{0}Bx gives 41 percent aril recovery(Tripathi, 2014).

CHES-14: The trees are semi-spreading type and is a regular bearer. This is a late variety. A six yearoldtree yields around 50-100kgper tree. Fruits are yellow in colour. Average fruit weight is 45 to 50 g. Fruits aril can be easily removed from the seed without attachment to the testa. The aril is white in colour and sweet (TSS -19.5 ^{0}Bx) with recovery of about 44 percent.

Lebakbooloos: Tree canopy is wide, fruits are darkred fruits with less spinterns. Thespinterns are 1.5 cm long and flesh is grayishwhite, tough, subacid , frequently difficult to separate from the seed and often flesh come with portion of testa. Suitable for long distance transport.

Seematjan: Tree canopy is open crown and long, flexible branches. Fruits are dark red with spines upto 2 cm long. It is susceptible to various pest.. It is cultivated in Java, India and Philippines.It yields 16 kg fruits per tree. There are twotypes in this variety.i.) Seematjanbesar: Has small fruit with thin rind, less spines; very sweet, coarse texture flesh, fibrous testa tightly adheres; ii) Seematjanketjil (Koombang): the fruit flesh is soft, less sweet and seed coat does not tightly adhere.

Binjai: The fruits are orange coloured, round in shape, the flesh recovery is 40 percent, the spintern length is 14mm and spintern density is 6.5 per square centimetre.

Rapiah: The fruits are red in colour, round shaped, the flesh recovery is 41 percent, the spintern length is 16.6mm and spintern density is 7.0 per square centimetre.

Jitlee:The fruits are orange in colour, round shaped, the flesh recovery is 36 percent, the spintern length is 15mm and spintern density is 6.2 per square centimetre.

Maharlika: The tree has broad canopy and fruits are medium large sized, globose fruits weighs 29-45g. The red spines are widely spaced, pliable, fine and 1.0 to 1.2 cm long. The fruit skin is thin and remains yellow for a many days later changed to red. The aril is about 0.5 cm thick, pearl white, firm, medium juicy, subacid to sweet and has very good quality. The pulp is loosely attached to the seed with some part of the seedcoat adhering. The fruit contains 50 per cent as edible portion.

Seenjonja: The trunk is shortand the canopy is lax. Small sized fruits of 3.8-4.2 cm length, 2.5-3.5 cm breadth and 18-23 g in weight and ovoid shape. The skin is thin, dark red with fine spinterns about 0.8-1.0 cm long. The aril is colourless, sweet, juicy, thin,and adheres tightly with the seed. Quality is good. The edible portion is about 40% of the fruit by weight. The seed shape is oblong, 2.6 cm length and 1.3 cm width.

Rongrein:Medium statured tree, with a dome shape canopy, oval shapeleaves, shortthin rounded apex and short petiole. The fruits are large size around 40-50g weight, length 50-55 mm; width 38-40 mm, the shape is ovoid to globose. Thefruitskinis thin with long coarse spinterns which changes colour from green to dark red at ripening and the tip remaining green. The aril is pearl white, thick with a good flavor and easily separates from the seed. The seedsarelaterally compressed and shape is oblong to elliptic. The total soluble solids ranged from18-21^{0}Bx.

Sri Chompoo: The tree canopy is dense and develops a large size canopy. Leaves are elliptic and large. The globose

fruits are large, 50 mm long and 39 mm wide and weighs 28-35g. The fruit skin is thin and pink or red. The flesh has a good flavour. The total soluble solids range from18-20^{0}Bx.

Aguilar-1: This variety developed by the Aklan State University, Banga, Aklan. The tree is strong with a spreading canopy. It has the yield potential of 225 kgfruitsper tree in a year. The globose to ovoid fruit is large, 4.7 mmlong, 10mm wide, and weighs 32.3 g. The fruits with thin leathery skin, weighs 14.18 g and ripened fruit colour changes to reddish orange. The aril is pearly white, smooth and firm, thick, juicy, sweet and aril is superior quality 45 per cent recovery. It has 21.9 ^{0}Bxtotal soluble solids. Theseed is small and flat.

Roja: This variety was owned by Mr. Mario Tenorio of Calauan, Laguna. The tree has strong branches and with the yield potential of 70-80 kgs. The fruitsareoblong, 50.40 mm length, 45.60 mm diameter, and 45.85g weight. The fruit has long spinterns and colour change to red when ripe. The flesh is white,smooth, sweet and juicy. It has a total soluble solid (TSS) of 20.88^{0}Bx. The fruit has an edible portion of 55.9 %.

Amarillo: This variety was owned by Dr.PoncianoBatugal of UPLB, College, Laguna. The tree has strong semi-upright branches andhigh yield potential of 50-60 kgs at 10 years old. The fruit shape is oblong, weighs 34.12 kgs, 52.65 mm length and 36.89 mm diameter. The fruit colour is yellow, thin leathery with lengthy spinterns. The aril is white,juicy and sweet. It has a total soluble solid content (TSS) of 22.2^{0}Bx. The fruit has an edible portion of about 60.67%.

Goyena: The tree originated from a seedling selection by Mr. Jaime M. Goyena, Sr. of Lamot II, Caluan, Laguna. The treesare 10 m tall at 15 years, strong with spreading growth habit and very prolific. It is regular bearing variety. The fruit shape is ovoidand weighs 36.9 g. The ripened fruits will have deep red skin, tough and leathery, 3.81 mm thick and weighs 17.0g. The thick, pearl white flesh is smooth, firm, sweet, moderately juicy and easily separates from the seed. It has 20.94 Bx of total soluble solids (TSS).

Propagation

Seed

Rambutan is propagated through seed, budding, grafting and layering. Seed propagation is easy. However, male trees will account for more than half of the plants. The seedlings will be utilised as root stocks. Rambutan seeds, extracted from the fruit, washed thoroughly and are planted horizontally with the flattened side downward in order that the seedling will grow straight and have a normal, strong tap root system. Seeds were sown in the poly bags and it takes around 25-28 days for germination. The seeds are recalcitrant, lose their viability very soon and should be sown immediately after removing from the fruit. The germination of 2-dayold seeds is 87% to 95% as compared to 50-60% in one week old seed.Sun drying for 8hour skills seeds within a week. The fresh seeds can be stored on moist sawdust or sphagnum moss or charcoal for 3-4 weeks. The seedlings become ready for grafting after 9-10 months (Lam and Kosiyachinda,1987).

Vegetative propagation

Rambutan requires vegetative propagation since seedlings take a long time to fruit and female to male trees ratios are 4 or 5 to 7. It can be vegetatively propagated through approach grafting (Inarching), air layering and budding.

Grafting

The grafts are produced by approach grafting method.The seedlings of 8 to 12 month old are used as rootstocks. Grafting during November and December months is more successful.

Air layering

The erect upright branches (having 1.0 –1.5 cm stem diameter and 30-60 cm long) of well-developed trees have to be selected for air layering. IBA at 5,000 ppm should applied and coir pith can be used as rooting media. About 25% success rate has been recorded.

Chip budding and patch budding

Patch-budding is chosen over other budding methods because it has a higher success rate. In the months of May and July, budding is done on well-grown rootstocks that are 8 to 12 months old. The success rate is reported to be as high as 83.6% in other countries. T –budding was also found successful. The experiment results of CHES, Chettalli reported that higher humidity and moderate temperature are essential for higher success.At temperature less than 20 ^{0}C success rate was low. The selection of bud wood is very important for the success. The bud wood collected from selected high yielding trees. The scion shoots should be obtained 3-4 months after harvesting. A one-year-old budwood of 1.5 to 2 cm diameter is found more suitable. Bud wood from procured shoot provide better success.

Spacing and Planting

The seedling trees are vigourous and spreading type and require wide space for growth. In the Philippines, it is advised that trees be planted at least 10 metres apart in each direction, with 12 metres being excessive in rich soil. If it is planted closely the branches become overcrowded in few years and fruit production will be severely affected. Grafted or budded plants used for planting means it grow small in size and it can be planted at 8x8 or 8x6 m spacing. The close planting 6x6 m^2 need regular pruning to control the tree size. For orchards with mechanization, wider row spacing should be given. The pits of 1x1x1m size are taken 2 to 3 weeks prior to planting and filled with a mixture containing three parts of top soil and one part of organic manures. Planting is done preferably during the onset of monsoon so that the plants are well established at the end of the monsoon.

Training and Pruning

Rambutan trees grows vertically and have a tendency to produce erect upright branches. Early training and pruning to form an open center tree is recommended. After harvesting, fruited twigs are pruned back to stimulate new growthof up to four new side shoots, of which 22 per cent of the shoots will bear fruits in the following season. Water suckers and dead branches should be removed on a regular basis (Tindall, 1994).

Nutrition

The quantity of NPK recommended is 200:25:130g per tree per year of age. After 12 years 2.4 kg, 0.30 kg, and 1.5kg of NPK can be applied. Fertilizers should be applied in four equal applications at three-month intervals for the first four years. The fruit bearing may supplied with onefourth (25% RDF) of the yearly fertilizer should be applied four weeks after fruit sets, half the quantity (50% RDF) should be applied immediately after harvest and the remaining onefourth (25% RDF) at nine weeks after harvest. In the hilly region 400g of dolomite per tree per year of age, upto ten years and remain constant thereafter has to be applied during slow growing months.

Irrigation

Irrigation should be given during summer season. Strong winds can easily injure rambutan trees during flowering and fruiting, so they should be shielded by other tall trees. It should not be irrigated for one month before flowering in full-grown plants to encourage flowering. The plants are then irrigated for 10-15 days to encourage floral bud initiation.

Inter-cultural operations

Mulching is essential during establishment and dry periods and not require prior to flowering. The glyphosate herbicide should not be used near irrigation channels or tree basin as it could cause severe yellowing and abscission of the lower leaves. The pineapple or vegetables recommended as intercrop in the rambutanorchards.

Pollination and fruit set

There are three types of flowers in Rambutan – Staminate (m), hermaphrodite functional female (hff) and hermaphrodite functional male (hfm). In the hff type, bifid stigma split open during anthesis facilitates pollination but the stamens remained closely appressed to the sepals and the anthers never dehisced. In the hfm type, cross

pollination is essential for fruit set, the stamens are erect at the time of anthesis and the anthers dehisce viable pollen but the stigma of the rudimentary pistil will not open. The panicle was found to be comprised of 94.41 and 5.99 per cent of hermaphrodite functional female and hermaphrodite functional male respectively.

Hermaphrodite cultivars generate only functionally female blooms and necessitate pollination by male trees. While planting, the male trees should also be planted. Male trees produce around 3000 flowers per panicle, flowers are greenish white each with 5-7 anthers and a non-functional ovary. Male flowers have nectaries and 5-7 stamens. Hermaphrodite flower panicle produce 500 greenishyellow flowers. Each flower has six anthers with bilobed stigma, and each one ovule two locules. The flowers are receptive for about one day before pollination, and they remain receptive until pollination. The peak flowering period is March- April. Flowers are white in colour. The panicle length is 22.9 cm. One shoot have 10 panicles each panicle with 466 flowers. The anthesis takes place between 5.00 am and 6:00 a.m (Lim, 1984).

Among the pollinating insects, *Apiscerana* was found to be the predominant followed by *A. florea, Trigona*spp., ants and wasp. The pollinating insects visits flowers between 7:00 a.m to 10:30 a.m. When fruit setting has occurred, water and fertilizer should be applied regularly and adequately for better yield. Increased pollinator populations, such as honey bees and flies, also aid in fruit set.During flowering, two colonies of bees (*Apiscerena indica*) should be kept per acre for better pollination.

The fruit set per panicle was found to be 3.41 per cent. The number of days taken from flowering to initial fruit set was 63 days. Fruit length and the diameter were equal during a period of 30 days, a rapid increment in fruit growth was observed which, continued up to 100 days. Following then, fruit growth slowed to 100 days and remained steady for the next 120 days. The Rambutan fruits takes about 110-120 days from flowering to harvest (Yan Diczbalis, 2002).

Harvesting and Yield

Grafted trees start bearing of fruits from four yearsafter planting. It may take up to five months for the fruits to develop into ripe fruits after fruit set. In Southeast Asian countries, rambutan It produces fruits two times in a year, the first crop (Main) during June and second crop during December. In India produce one crop per year. The fruits usually ripe month of July to October. Harvesting is done by cutting fruit bunch stalk and the fruits are individually cut off and packed for sale. The fruits are plucked without falling on ground. If fall on ground fruits easily bruised and have a short shelf life. The ten year old tree may produce 60-70kg fruits per tree. It may vary from year to year as it exhibits alternate bearing.

After the harvest, the fruits must be sold as soon as possible since they begin to shrivel and perish.The low temperature storage of fruits maintains fruits texture; the sucrose contents also get increased and good appearance. Storage temperatures less than 7 ^{0}C cause chilling injuries to the peel and hairs.Fruits can be packed in polythene bags and stored at a temperature of 13-15 ^{0}C to extend their shelf life.

Conclusion

The fruit is attractive, sweet, tasty and juicy it is most preferred by children. The aril is best salad-based dishes. In India it is cultivated only in homestead in Kerala. It has good market potential and the favorable climate available region of India can grow as most remunerative crop.

References

1. Anonymous, (1992). Plant Resources of South-East Asia No. 2. In: Edible Fruits and Nuts. Bogor, Indonesia.
2. Anonymous, (2003). DIS Techno guide Series. In: Rambutan Production. No. 02-03 Series. Philippines.
3. Anonymous. (2003). National Seed Industry Council. In: Approved Crop Varieties- Catalogue, Philippines.
4. Coronel, R.E. (1983). Promising Fruits of the Philippines. College of Agriculture, University of the Philippines at Los

Baños. P 508.

5. Lam, P.F. and Kosiyachinda, R. (1987). Rambutan Production. Asian KOFAF, Jakarta, Indonesia.
6. Lim A.L. (1984). The reproductive biology of Rambutan (*Nephelium lappaceum* L.) (Sapindaceae). Gardens Bulletin, Singapur 37(2) 181-192.
7. Lim, TK and Diczbalis, Y. (1998). Rambutan. Agnote, Australia
8. Linn ,T.K.(1992). Rambutan Industry in the Northern Territory. Acta Hortic. 321.62-70.
9. Morton, J. (1987). Rambutan. In Fruits of Warm Climates. Miami, Florida. pp 26.
10. Ravisankar, H., Sakthivel, T., Samual D. K. and Shivaramu. (2007). Rambutan-An underutilized fruit crops for Tropical humid climate. CHES, Chettalli, IIHR, Bangalore Tech Bull.32. PP-8.
11. Suganthi, A. and Marry Josephine, R. (2016). *Nephelium Lappaceum* (L.): An overview. International Journal of Pharmaceutical Science and Research 1(5): 36-39.
12. Tee, E. S. (1982). Nutrient Composition of Malaysian Food. A preliminary table. Division of Human Nutrition, Institute of Medical, Kuala Lumpur, Malaysia.
13. Tindall, H D (1994). Rambutan Cultivation. FAO Plant Production and protection Paper 121.P163.
14. Tripathi, P.C., ,G.Karunakaran, T., Sakthivel, V. Sankar and Senthil kumar, R. (2014). Rambutan Cultivation in India. Technical Bulletin, Central Horticultural Experiment Station, Indian Institute of Horticultural Research, Chettalli – 571 248, Kodagu, Karnataka
15. Wills, R.B. H, Lim. S. K and Greenfield, H. (1986). Composition of Australian Foods. Tropical and subtropical Fruits Food Technology in Australia 38(3):118-123.
16. Yan Diczbalis. (2002). Rambutan, Improving Yield and Quality. A report for the Rural Industries Research and Development Corporation. Barton.
17. Zee, F.T. (1983). Rambutan and Pilinuts. Potential crops for Hawaii. New Crops. I. Janick and J.E. Simen (Eds) John Wiley and sons .Inc. New York 461-466.

In vitro regeneration in pigeonpea: Effect of explants and culture media

Manjinder Singh and Ajinder Kaur*

School of Agricultural Biotechnology, Punjab Agricultural University, Ludhiana, Punjab

Abstract

Pigeonpea is a key pulse crop cultivated in tropical and sub-tropical countries with India as the largest pigeonpea growing country in the world. Several biotic, abiotic stresses are key constraints in the crop production. Conventional breeding methods to develop biotic, abiotic stress resistant pigeonpea varieties are not much successful owing to inadequate resistant sources in compatible germplasm along with non-crossability with wild species. Genetic transformation is a promising approach for introducing gene(s) of interest in commercial varieties. In vitro shoot differentiation is a main requirement for carrying out stable genetic transformation in any crop. This manuscript reviews the progress towards achieving plant regeneration in pigeonpea under in vitro conditions.

Keywords *Cajanus cajan*, *In vitro* plantlet development, Organogenesis pathway, Somatic embryogenesis pathway

Introduction

Pigeonpea (*Cajanus cajan* L.) or red gram of family Fabaceae is one of the key pulse crops cultivated in the semi-arid tropics. It is cultivated throughout in sub-tropical and tropical countries, particularly Asia, Africa, America and Australia. Globally, pigeonpea cultivation is distributed across more than 22 different countries with five major growers, including India (4.65 mha), Myanmar (0.65 mha), Tanzania, Malawai (0.22 mha) and Kenya (0.14 mha) (FAO, 2014). Out of the total world pigeonpea production, India accounts for >80 % production (Popelka *et al.*, 2004). Pigeonpea is a source of protein (21%) for humans, used as fodder for animals, for making roofs, windbreaks, hedges and also finds use as fuel wood and green manure. Global production of pigeonpea is limited because of biotic stresses, e.g. damage by insects, pathogens, and abiotic stresses, e.g. water logging, drought, cold sensitivity etc. There is non-availability of suitable genetic resources for genetic improvement of pigeonpea as primary gene pool of the crop lacks genetic variation for desired traits. The breeders opt for wild relatives from secondary, tertiary and quaternary gene pools, however lack of proper genetic information on desirable traits, e.g. insect resistance, and strong sexual barriers between cultivated and wild species hamper the utilization of wild species in conventional breeding programs (Sharma and Mathur, 2016). Plant genetic transformation is a good option for introducing gene(s) of interest in a high-yielding commercial background. Efficient regeneration and recovery of stable transformants are two basic requirements for genetic transformation in any crop. Therefore, an attempt is made to review the work done so far on use of different explants, culture media for obtaining *in vitro* shoot and root differentiation in pigeonpea by different researchers.

Modes of *in vitro* regeneration in pigeonpea

Regeneration is the process of formation of complete plant from initial explants cultured under *in vitro* conditions. There are two different modes for regeneration, *viz*. organogenesis and somatic embryogenesis. Organogenesis mode involves the development of adventitious organs, either from cultured explants or undifferentiated cell mass (callus). Usually, shoots are induced first via organogenesis and then shoots are transferred to rooting medium for root induction. The organogenesis is classified into two types, direct and indirect organogenesis. The direct organogenesis mode does not involve callus formation, rather shoots or roots are directly induced from pre-existing cells in explants. There are a large number of reports on direct development of shoots from leaf parts (Eapen et al., 1998; Dayal *et al*., 2003; Villiers *et al*., 2008), cotyledonary nodal explants (Singh *et al.*, 2003; Thangella and Fakrudin, 2016), embryonic axis (Krishna *et al*., 2011; Parekh *et al.*, 2014) and shoot apices (Naidu *et al*., 1995; Geetha *et al.*, 1998) in pigeonpea (Table 1). On the other hand, indirect organogenesis involves intervening callus formation from which shoots and roots are formed, and has been reported in various studies (Mohan and Krishnamurthy, 1998; Tyagi *et al*., 2001).

Somatic embryogenesis mode involves *in vitro* induction of somatic embryos from diploid somatic tissues, which are then cultured to develop into plantlets. Patel *et al*. (1994) reported somatic embryogenesis in pigeonpea for the first time. Somatic embryogenesis is also categorized into direct and indirect types of somatic embryogenesis. The former does not involve callus phase, while latter involves callus phase. From genotype adaptability perspective, organogenesis in pigeonpea is widely used due to higher regeneration capacity and plant production per explant, but from genetic transformation viewpoint, somatic embryogenesis is most preferred mode due to prolific multiplication of embryogenic calli (Hansen and Wright, 1999).

Effect of culture media and plant growth regulators on *in vitro* regeneration of pigeonpea

Culture medium is the basic requirement for any tissue culture as it provides macro and micro nutrients for growth and development of complete plantlet from a single cell under *in vitro* conditions. Before starting with tissue culture, it is important to prepare a medium that meet requirements for culturing a specific tissue. Kumar *et al*. (1983) tested various types of media for effective plantlet regeneration in pigeonpea, namely White's medium (White, 1939), Wood's medium (Wood and Braun, 1961), MS medium of Murashige and Skoog (1962), LS medium given by Linsmaier and Skoog (1965), Blaydes medium (Blaydes, 1966), B_5 medium devised by Gamborg *et al*. (1968), Schenk and Hildebrandt (1972) medium and EC_6 medium (Maheswaran and Williams, 1984); amongst these, Blaydes medium was observed to be most effectual for callus induction and shoot bud differentiation. Most commonly used media in pigeonpea regeneration include MS, B_5 and EC_6. These can be used either singly or in combination with each other (Table 1). Mohan and Krishnamurthy (2002) used combination of MS, B_5 and EC_6 for plantlet regeneration from cotyledonary explants of pigeonpea via somatic embryogenesis. Parekh *et al*. (2014) documented pigeonpea plantlet regeneration from apical meristem cultured on MS medium.

For successful accomplishment of any tissue culture experiment, inclusion of plant growth hormones in culture medium is of utmost importance. There are four different classes of growth regulators, namely cytokinins, auxins, gibberellins and ABA (abscisic acid), out of which auxins and cytokinins are considered to be main growth regulators without which a tissue culture experiment cannot be initiated. Gibberellins, ethylene, ABA and other hormones are usually not added to culture medium, but their role can never be ignored as these interact with added auxins, cytokinins, and promote cell growth and development in a hidden manner. Different growth hormones each with varying concentration have intense influence on callus induction as well as on the entire process of regeneration (George, 1993). Both auxins and cytokinins have been known to be used in combination with each other in a specific ratio. These may act synergistically or antagonistically. High ratio of auxin to cytokinin in the culture medium promotes root induction, while a low ratio of the same results in shoot differentiation (Skoog and Miller, 1957). Auxins, like Indole-3-acetic acid or IAA, Indole-3-butyric acid or IBA, Naphthalene acetic acid or NAA and 2,4-Dichlorophenoxyacetic acid or 2,4-D have been used in various concentrations and combinations with cytokinins, e.g. kinetin, 6-Benzylaminopurine or BAP, Thidiazuron or TDZ (George and Eapen, 1994; Shiva Prakash *et al*., 1994; Eapen *et al*., 1998; Geetha *et al*., 1998; Anbazhagan and Ganapathi, 1999; Dayal *et al*., 2003) to promote cell division, shoot regeneration and enhance proliferation of axillary buds. Kinetin, BAP and zeatin are commonly used cytokinins; among these, BAP is most preferred because of high shoot bud regeneration both in organogenesis mode and somatic embryogenesis mode. The use of BAP (5 mg/l, 10 mg/l) promoted shoot regeneration at high frequency from cotyledonary node and whole seed explants, respectively (Shiva Prakash *et al*., 1994; Naidu *et al*., 1995). At lower BAP concentration (2 mg/l), similar number of shoots was still obtained, but by using it in combination with IAA, shoot regeneration frequency increased by 50% (Naidu et al., 1995). Eapen and George (1993) for the first time reported use of BAP along with IAA. Yadav and Chand (2001) reported 100% shoot formation in pigeonpea through use of BAP alone and/or in combination with IAA. Singh et al. (2003) demonstrated that TDZ at low and intermediary concentrations induces clusters of leafy structures and multiple shoots from cotyledonary nodal regions, but at high concentration, it completely switches the pathway to somatic embryo development. The frequency of shoot bud formation on 2.5 mg/l BAP has been reported to be higher than on medium containing both IAA and NAA, in organogenesis pathway. TDZ having both activities of an auxin and a cytokinin is widely used for somatic embryo regeneration (Thomas and Katterman, 1986; Saxena *et al*., 1992). Optimum concentration of TDZ is necessary for somatic embryo regeneration; otherwise it may lead to organogenesis at extreme low and high

concentrations (Sreenivasu *et al.*, 1998; Singh *et al.*, 2003). A total of 38 embryos were produced on medium augmented with 1 to 2 mg/l thidiazuron (Mohan and Krishnamurthy, 2002). Likewise, TDZ produced 30.8 globular embryos at 0.6 mg/l concentration (Krishna *et al.*, 2011), whereas BAP yielded only 13.9 globular embryos at 1.1 mg/l concentration. Naidu et al. (1995) used BAP + kinetin for shoot differentiation and development. Parekh et al. (2014) reported 31.73% frequency of shoot multiplication on medium augmented with BAP (0.5 mg/l) + TDZ (0.2 mg/l) with average shoot length of 5.2 cm. Multiple shoot formation was seen from cotyledonary nodal segments on MS medium + BAP (5 mg/l) + kinetin (2 mg/l) after 2 weeks of culturing, the shoots were inoculated on MS medium containing adenine sulphate for elongation, and rooted *ex vitro* using ROOTEX powder (Dev and Kaur, 2017). However, Dayal et al. (2003)) achieved 95% shoot regeneration and Villiers et al. (2008) reported 46% shoot induction using BAP and kinetin at same concentration (5 µM). A comprehensive list on *in vitro* shoot and plantlet regeneration in pigeonpea is presented in Table 1.

Conclusion

Various types of explants and culture media used by different researchers to achieve *in vitro* regeneration in pigeonpea have been presented; one can use these for reference during development of *in vitro* shoot and root regeneration protocol in a new pigeonpea variety. The standardized protocol can then be utilized for carrying out genetic modification of pigeonpea to obtain transgenic plants resistant or tolerant to different types of stresses.

Table 1: *In vitro* shoot/plantlet regeneration in pigeonpea via organogenesis (O) and somatic embryogenesis (SE) pathways

Genotype	Explant (Mode of regeneration)	Shoot induction medium	Rooting medium	Remarks	References
T-21	Seed, hypocotyls (O)	-	-	Gamma radiation effect on cell multiplication, regeneration in explants was studied.	Rao and Narayanaswamy (1975)
T-21	Anther (SE)	MS + B_5 + Kin (2 mg/l) + IAA (4 mg/l)	-	Regeneration frequency of 1.8% was obtained.	Bajaj *et al*. (1980)
Prabhat	Cotyledon (O)	MS + BAP	-	Regeneration of plantlets was achieved.	Mehta and Ram (1980)
ICP 6917, ICP 7035, BDN-1, T-21, ICP 2836, ICP 7186	Leaf, cotyledon (O)	Blaydes medium + 2,4-D (2 mg/l) + Kin (0.5 mg/l) for callus formation, Blaydes medium + BAP (0.5 mg/l) + NAA (0.1 mg/l) for shoot bud regeneration	Blaydes medium + NAA (0.6 mg/l) + Kin (0.01 mg/l), Blaydes medium + IAA (1 mg/l) + Kin (0.01 mg/l)	Leaves were most responsive to callus induction. ICP 7035 showed highest number of shoot induction (17.4) from cotyledons. Complete plantlet regeneration occurred.	Kumar *et al.* (1983)
ICP 6917, ICP 6974, ICP 7119, ICP 7263	Apical meristem, cotyledon, epicotyl (O)	MS + B_5 + BAP	-	Multiple shoots from cultured explants were obtained.	Kumar *et al*. (1984)
AL 15, Hyderabad C	Apical meristem, hypocotyl, cotyledon (O)	MS + Kin/BAP (0.5-3 mg/l)	MS	Shoot buds (2-3) with varying shoot length (2.5 - 4 cm) from cotyledons and hypocotyls were obtained. Complete plants were obtained when BAP was omitted from the medium.	Cheema and Bawa (1991)
Cajanus cajan L.	Cotyledon, mature embryo axis (O)	MS + B_5 + BAP (0.5 mg/l)	-	Direct shoot proliferation was reported.	Sarangi and Gleba (1991)

ICPL 161	Leaf (O)	MS + BA (10 µM) + IAA-l-aspartic acid (0.5 µM)	½MS + NAA (1 µM)	Maximum regeneration frequency of 15.7% (14 shoots/explants) was obtained. A total of 25 plants were transplanted to soil.	Eapen and George (1993)
ICPL 161	Cotyledon, epicotyl, mature cotyledonary segment, leaf, root, immature embryo axis, seed (O)	MS + BA (1 mg/l) + IAA (0.1 mg/l)	½MS + B_5 + NAA (0.2 mg/l)	Leaf explants gave best response with eight shoots/explant on an average and complete plantlets were obtained.	George and Eapen (1994)
	Cotyledon, immature embryo axis (SE)	MS + 2,4-D/ Picloram/NAA (5 mg/l)	-	Cotyledons gave best response on picloram medium, while embryo axes responded best on 2,4-D medium for somatic embryo production, however no plants could not be obtained.	
BP 86-34, CC (2376, 11295), Gaut-(88-29, 89-8), SPMA-4	Cotyledonary node (O)	MS + B_5 + BAP (2 mg/l) supplemented topically with IAA	MS + B_5 + IBA (0.5 mg/l)	CC 11295 was most responsive with maximum 43 shoots/explant, while lowest number of shoots (21) was observed in CC 2376. About 85% plants survived upon transfer to soil.	Shiva Prakash *et al*. (1994)
Bandapalera, Gaut-82-90, NP (WR) 15, T-15-15	Mature cotyledonary segment (SE)	MS + B_5 + Adenine sulphate + BAP + Kin	-	Regeneration was achieved through somatic embryogenesis.	Patel *et al.* (1994)
Visakha-1, ICPL87, N-290-21, PT-22, T-15-15, Gaut-82-90, BDN-2, T-21	Cotyledon, seed, embryo axis, epicotyl (O)	MS + B_5 + BAP (22.2 µM) + Kin (2.32 µM) for shoot bud induction, MS + B_5 + Kin (0.46 µM) + NAA (0.53 µM) + GA_3 (0.29 µM) for shoot elongation	½MS + B_5 + IBA (4.9 µM)	Only PT-22 and ICPL 87 showed shoot regeneration (25 - 80%). Complete plants were obtained with 70 - 80% survival rate upon transfer to soil.	Naidu *et al*. (1995)

ICPL87	Leaf, root, epicotyl, cotyledon (SE)	MS + B_5 + BAP (1 mg/l) + NAA (1 mg/l)	-	Cotyledons gave better result with a regeneration frequency of 3%. The somatic embryos induced on cotyledons developed into complete plants.	Nalini *et al*. (1996)
ICPL161, ICPL 88039, UPAS 120	Primary leaf (O)	MS + B_5 + TDZ (1 - 2 mg/l) + IAA (0.1 mg/l) for shoot regeneration, MS + B_5 + BA (1 mg/l) + IAA (0.1 mg/l) + GA_3 (1 mg/l) for shoot elongation	MS + NAA (1 mg/l)	An average of 47, 44 and 43 shoots developed/explant in ICPL (161, 88039) and UPAS 120, respectively.	Eapen *et al*. (1998)
Vamban	Cotyledonary node, apical meristem (O)	MS + BAP (13.31 µM)	½ MS + full strength organic addenda + IBA (2.46 µM)	Maximum shoot number (49/ explant) was achieved using cotyledonary node explants. Plantlets were grown in soil that produced viable seeds.	Franklin *et al*. (1998)
Hyderabad C	Epicotyl, hypocotyl, leaf, cotyledon, cotyledonary node (O)	MS + B_5 + BAP (2 mg/l) + Kin for shoot bud differentiation, MS + B_5 + NAA (0.1 mg/l) + BAP (1 mg/l) + GA_3 (1 to 5 mg/l) for shoot elongation	MS + B_5 + IBA (0.2 mg/l)	Regeneration frequency was 93.2, 71.4, 87.5, 80.2 and 65.4% from cotyledonary node explant, cotyledon, epicotyl part, hypocotyl and leaf explants, respectively. The plants exhibited 95% survival in soil.	Geetha *et al*. (1998)

BDN-1, BDN-2, ICP-7182, ICPL- 87, -87119, TV-1, Gaut 82-90, T-15-15	Cotyledon (O)	MS + B_5 + BAP (22.2 µM) + Kin (2.3 µM) + Adenine sulphate (271 µM) for shoot bud induction, MS + B_5 + BAP (2.22 µM) + NAA (0.54 µM) and half strength MS + GA_3 (2.89 µM) for shoot elongation	MS + B_5 + IAA (4.92 µM)	Maximum number of shoots obtained was 33 and 23 in T-15-15 and Gaut 82-90, respectively, while in other genotypes no shoots were obtained. The elongated shoots exhibited 87% rooting.	Mohan and Krishnamurthy (1998)
Vamban 1	Apical meristem (O)	MS + B_5 + BAP (13.31 µM)	½MS + B_5 + IBA (0.49-24.6 µM)	Multiple number of shoots regenerated from apical and axillary meristems.	Pudukkottai (1998)
Pusa 606, 609, 852, 855, 856, H-86-25	Leaf, cotyledonary node (SE)	MS + B_5 + TDZ (10 µM)	½MS	SE (73.6%) was achieved using leaf explants in Pusa 606, while 46.2% SE was achieved from cotyledonary nodes in Pusa 852. Seventy percent somatic embryos got converted into plantlets and survival rate of plants in soil was 80%.	Sreenivasu *et al*. (1998)
Vamban1	Immature leaf (SE)	MS + B_5 + 2,4-D (6.78 µM)	-	Maximum regeneration frequency obtained was 36%.	Anbazhagan and Ganapathi (1999)
ICPL 88039	Cotyledonary node (O)	MS + B_5 + BAP (5 mg/l) for induction of shoots, MS + B_5 + BAP (1 mg/l) for shoot elongation	-	Mean number of 32 shoots/ explant were obtained.	Ramchandar (1999)
Bahar	Decapitated embryonic axis (O)	MS + BAP (1 mg/l) + IAA (0.5 mg/l)	MS + IBA (0.3 mg/l)	Maximum regeneration frequency of 56.66% and 6.8 shoots/explant were obtained. Complete plants were obtained from decapitated embryo axes with 80% survival rate.	Rathore and Chand (1999)

VBN1, VBN2, SA1, CO5	Mature embryo axis (O)	MS + B_5 + BAP (8.86 µM) + NAA (1.07 µM)	½ MS + IBA (2.41µM)	Maximum number of shoots obtained was 9.5 in case of VBN2, in other genotypes shoot number ranged from 5 to 6.5.	Franklin *et al*. (2000)
Tanzania-7, ICPL 93086, F_1 hybrid	Shoot, leaf, root (O)	KM medium + BAP + NAA + Kin	KM medium + IAA + 2 iP + GA_3 + BAP	All explants exhibited 82 - 88% shoot regeneration in different genotypes.	Tyagi *et al*. (2001)
Cajanus cajan L.	Cotyledonary leaf (O)	MS + B_5 + BAP (8.9 µM) + NAA (5.37 µM)	-	Plants were regenerated via organogenesis.	Kumari *et al*. (2001)
Bahar, UPAS120	Decapitated embryonic axis (O)	MS + B_5 + BAP (5 mg/l) + IAA (0.5 mg/l)	MS + B_5 + IAA (0.5 mg/l)	Plantlets were regenerated from decapitated embryonic axes.	Yadav and Chand (2003)
T-15-15, GAUT-82- 90, GAUT-82-99	Cotyledon, mature cotyledonary segment (SE)	EC_6 + BAP (2.22 - 22.2 µM) + TDZ (0.45 - 13.62 µM) for somatic embryo induction, MS + B_5 + GA_3 (2.89 - 14.43 µM)for elongation	½ MS + ABA (0.38 µM)	Cotyledons exhibited maximum regeneration frequency of 92% in GAUT-82-99.	Mohan and Krishnam urthy (2002)
CC11295	Cotyledon (SE)	MS + TDZ (9.08 µM)	-	High frequency of SE (75.6%) was achieved. However, these somatic embryos showed poor plant conversion rate.	Chandra *et al*. (2003)
ICP 8863, ICPH 8, Pusa 33, UPAS 120	Cotyledonary node (O)	MS + B_5 + TDZ (0.05 - 1 µM)	MS + IBA (2.5 µM)	Maximum number of shoots obtained was 6.2 in ICP 8863, 4.7 each in ICPH 8, UPAS and 13 in Pusa 33. The shoots showed prolific root formation and complete plantlets were obtained with 95% survival rate.	Singh *et al*. (2003)

ICP 151, ICP 8863, ICPH 8, Pusa 33, Pusa 853, RWL 19, UPAS 120	Cotyledonary node, leaf (SE)	MS + B_5 + TDZ (10 µM)	MS + IBA (2.5 µM)	Maximum number of embryos (20.8) was obtained in Pusa 853 using cotyledonary node explants, while only a few embryos (8.2) were obtained using leaf explants in UPAS 120. About 15% somatic embryos formed roots and developed into complete plants.	
ICPL93115	Cotyledon (O)	MS +BA (5 mg/l) for regeneration of shoots, MS + BA (1 mg/l) for elongation of shoots	MS + IAA (1 mg/l) + Kin (0.1 mg/l)	Maximum shoot number obtained was 16.5/cotyledon. Plants obtained were hardened on hormone-free basal MS medium.	Yadav and Padmaja (2003)
ICPL 93115	Seedling leaf (O)	MS + B_5 + BA (5 mg/l) for induction of shoots, MS + B_5 + BA (1 mg/l) for elongation	MS + B_5 + IAA (1 mg/l) + Kin (0.1 mg/l)	Regeneration from immature leaf explants was reported.	Slater *et al*. (2003)
ICPL-91011, -88009, -84031, -87, -2376, -8705, -332, -85063, -87119	Leaf (O)	MS + B_5 + BA (5 µM) + Kin (5 µM) for shoot induction, MS + B_5 + BA (5 µM) + Kin (5 µM) + GA_3 (0.58 µM) for shoot elongation	MS + B_5 + IAA (11.42 µM)	Multiple adventitious shoot induction was observed on majority of leaves excised from seedlings (4 to5 day-old) raised *in vitro*. Complete plants were obtained on IAA medium.	Dayal *et al*. (2003)

Bahar	Leaf (O)	MS + BAP (0.25 mg/l) + Kin (0.25 mg/l) + IAA (0.05 mg/l) + Adenine sulphate (40 mg/l) for induction of shoot buds, MS + BAP (0.01 mg/l) + IAA (0.01 mg/l) + 2 Chloro ethyl trimethyl ammonium chloride (10 mg/l) for shoot elongation	MS + IAA (0.25 mg/l)	Complete plantlets were formed with 90% survival rate.	Jain and Chaturvedi (2004)
ICP 26, ICP 28	Leaf petiole (O)	MS + B_5 + BAP (2 - 5 mg/l + NAA (0.1 mg/l)	MS + B_5 + IBA (0.3 mg/l)	Maximum regeneration frequency of 70% and 60% was obtained from distal cut-ends of petioles in ICP 26 and ICP 28, respectively.	Srinivasan *et al*. (2004)
ICEAP (00557, 00020, 00040, 00053, 00554), ICPL 86012, ICPL 88039 00040,00554 and 0053), ICPL (88039, 86012and 87091)	Leaf (O)	MS + BA (5 µM) for shoot induction, MS + BA (5 µM) + GA_3 (0.58 µM) for shoot elongation	MS + B_5 + IAA (11.4 µM) + Sucrose (1%)	Regeneration frequency of 77%, 48% and 46% was obtained in ICEAP 00053, ICPL 86012 and ICPL 88039, respectively. All tested varieties showed healthy plantlet formation.	Villiers *et al*. (2008)
LGG-29	Embryo (O)	MS +BAP (1 mg/l) + NAA (0.1 mg/l) for shoot bud differentiation, MS + GA_3 (0.4 mg/l) for shoot	MS + IBA (1 mg/l)	Rregeneration (62.5%) with maximum number of shoots as 7 was obtained. The plants were grown in sand, clay and vermiculate in a ratio of 1:1:1.	Guru Prasad *et al*. (2011)

		elongation			
ICPL 87-118, ICPL 151	Mature cotyledon (SE)	MS + B_5 + 2,4-D or TDZ (2 mg/l)	MS + BA (0.1 mg/l)	No. of somatic embryos obtained/explant was 29.40 for ICPL 87-118 and 21.20 for ICPL 151. Complete plantlets were formed with 32% survival rate.	Aboshama (2011)
Pusa 9, IPA-(34, 61, 98-3, 204, 242, 337, 341, 2013, 3088), T-7	Leaf (O)	MS + B_5 + BAP (3 mg/l), MS + B_5 + NAA (0.05 mg/l)	MS + B_5 + NAA (0.1 mg/l)	Among 11 genotypes, IPA-2013 was found to be best for multiple shoot bud and complete plantlet formation.	Kashyap *et al*. (2011)
JKR105	Embryonic axis (O)	MS + B_5 + BAP (2.5 mg/l)	Half strength MS + B_5 + IBA (0.5 mg/l) + Sucrose (2%)	Highest no. of shoot bud differentiation was 17.5 using 2.5 mg/l BAP.	Krishna *et al*. (2011)
JKR105	Cotyledon (SE)	MS + TDZ (0.6 mg/l), ½ MS + BAP (0.1 mg/l)	-	Maximum no. of somatic embryos/explant (30.8) was obtained on TDZ + sucrose. However, less no. of plants was produced in comparison to organogenesis pathway irrespective of high number of somatic embryos.	
Cajanus cajan L.	Epicotyl, cotyledon	MS + B_5 + IAA (1 mg/l) + Kin (0.9 mg/l) for callus formation	-	Callus formation (95%) was observed on epicotyl as well as cotyledon explants.	Prabhakaran *et al.* (2011)
AL 201	Cotyledonary nodal segment (O)	MS + B_5 + BAP (3 mg/l) + Kin (1 mg/l) + 0.2% (w/v) Charcoal	½ MS + 0.2% (w/v) Charcoal	Average no. of 3.19 shoots, and highest shoot elongation obtained was 11.27 cm. Complete plantlets were obtained in 45 days.	Kaur *et al.* (2012)

LRG-41	Cotyledon (O)	MS + B_5 + BAP (2 mg/l) + NAA (0.1 mg/l) + GA_3 (0.4 mg/l)	MS + B_5 + IBA (1 mg/l)	Regenerated plants survived in soil with up to 95% frequency.	Raghavendra *et al*. (2012)
LRG-41	Shoot tip (O)	MS + BAP (2 mg/l) + NAA (0.1 mg/l)	MS + IBA (1 mg/l)	Shoots with a frequency of 61.9% were obtained. Plant survival rate in soil rite was 91%.	Raghavendra and Sudhakar (2014)
ICPL 87119 (Asha)	Mature zygotic embryo (SE)	MS + B_5 + TDZ (2 mg/l) for shoot bud formation, MS + B_5 + TDZ (1 mg/l) for shoot elongation	MS + IBA (3 mg/l)	Plant regeneration was achieved.	Shekhar *et al*. (2012)
	Mature zygotic embryo (O)	MS + TDZ (4 mg/l)	MS + IBA (3 mg/l)	A regeneration frequency of 73% was obtained.	
ICPL 87119, BSMR 736	Embryo disc with half cotyledon, cotyledonary node (O)	MS + BAP (2 mg/l)	MS + IBA (0.5 mg/l)	Multiple shoot induction was more in BSMR 736 from cotyledonary node explants.	Manohar (2014)
GT-102	Apical meristem (O)	MS + BAP (0.5 mg/l) + TDZ (0.2 mg/l)	½ MS + IBA (0.3 mg/l)	Shoot regeneration frequency of 31.73%, no. of shoots/explant (7.67) and shoot length (5.2 cm) were obtained. Plantlets were obtained on IBA medium that exhibited 80% survival upon transfer to soil.	Parekh *et al*. (2014)
Phule Rajeshwari, Vipula	Apical meristem, leaf (O)	MS + BAP (0.5 mg/l)	MS + IBA (2 mg/l)	Phule Rajeshwari responded better than Vipula with apical meristem explant; no shoot bud differentiation was recorded from leaf explant. Complete plants obtained were transferred to soil with 80% survival frequency.	Pawar *et al*. (2014)

ICPL 81179	Embryonal axis (SE)	MS + Specified phytohormones + Growth factors + Phospholipid precursors	-	Somatic embryos were obtained by micro droplet cell culture system. Complete plantlets were derived and transferred to soil.	Kumar *et al*. (2015)
NTL-30	Cotyledonary node, embryonic axis, scutellum (O)	MS + Zeatin (1.35 µM) + Kin (0.93 µM) + Silver nitrate (2.94 µM) for shoot formation, MS + GA_3 (0.5 µM) for shoot elongation	MS + IAA (0.57 µM)	Embryonic axes showed highest shoot induction frequency (96%).	Raut *et al*. (2015)
GT 101	Cotyledonary node (O)	MS + BAP (3 mg/l) + NAA (0.5 mg/l) for shoot induction, MS + BAP (0.5 mg/l) + GA_3 (0.5 mg/l) for shoot elongation	MS + IBA (0.5 mg/l)	Maximum shoot regeneration frequency of 73.33% with mean no. of shoots/explant as 13.7, and highest shoot elongation of 4.2 cm were obtained. The developed plants produced viable seeds.	Jasani *et al*. (2016)
ICPL 87	Leaf, cotyledonary node, embryo, embryonal axis, epicotyls (O)	MS + Kin (2 mg/l) + BAP (5 mg/l) for shoot induction, MS + Adenine suphate (30 mg/l) for shoot elongation	ROOTEX powder	Maximum shoot induction (67.11%) occurred from cotyledonary node explants. ROOTEX powder induced 85-95% rooting within 20 days of transplanting shoots in sand.	Dev and Kaur (2017)
BDN-2, GT-101	Leaf, hypocotyl, root (O)	MS + BA (5 mg/l) + IAA (0.5 mg/l) + Adenine (2 mg/l) for shoot induction, MS + NAA (1 mg/l) + Adenine (4 mg/l) for shoot elongation	MS + IAA (1.0 mg/l)	GT-101 was more responsive with 54.6% shooting frequency. The hypocotyls produced maximum shoot number (7.3) per callus with maximum shoot length of 6.9 cm. BDN-2 was more responsive to rooting.	Abhijeeta and Rajesh (2018)

ICP 8863	Leaf petiole, cotyledonary node (O)	MS + BAP (2 mg/l) for shoot bud induction, MS + NAA (0.1 mg/l) + BAP (1 mg/l) + GA_3 (2 mg/l) for shoot elongation	MS + IBA (0.5 mg/l)	Shoot bud induction (92%) was achieved, and well developed shoots showed 95% rooting. The plants were acclimatized and finally shifted to green house.	Nalluri and Karri (2019)

References

1. Abhijeeta, K.N. and Rajesh, B.M. (2018). *In vitro* plant regeneration in pigeonpea [*Cajanus cajan* (L.) Millsp.] using various explants. Legume Research. 41: 226-229.
2. Aboshama, H.M.S. (2011). Somatic embryogenesis proliferation, maturation and germination in *Cajanus cajan*. World Journal of Agricultural Sciences. 7: 86-95.
3. Anbazhagan, V.R. and Ganapathi, A. (1999). Somatic embryogenesis in cell suspension cultures of pigeonpea (*Cajanus cajan* L. Millsp.). Plant Cell, Tissue and Organ Culture. 56: 179-184.
4. Bajaj, Y.P.S., Singh, H. and Gosal, S.S. (1980). Haploid embryogenesis in anther cultures of pigeon pea (*Cajanus cajan*). Theoretical and Applied Genetics. 58: 157-159.
5. Blaydes, D.F. (1966). Interaction of kinetin and various inhibitors in the growth of soybean tissue. Physiologia Plantarum. 19: 748-753.
6. Chandra, A., Gupta, V., Burma, P. and Pental, D. (2003). Patterns of morphogenesis from cotyledon explants of pigeonpea. In Vitro Cellular and Developoemtal Biology – Plant. 39: 514-519.
7. Cheema, H.K. and Bawa, J. (1991). Clonal multiplication via multiple shoots in some legumes (*Vigna unguiculata* and *Cajanus Cajan*). Acta Horticulturae. 289: 93-94.
8. Dayal, S., Lavanya, M., Devi, P. and Sharma, K.K. (2003). An efficient protocol for shoot regeneration and genetic transformation of pigeon pea [*Cajanus cajan* (L.) Millsp.] by using leaf explants. Plant Cell Reports 21: 1072-1079.
9. Dev, A., and Kaur, A. (2017). Successful propagation of pigeonpea (*Cajanus cajan* L.) through *in vitro* shoot regeneration and *ex vitro* rooting. International Journal of Biotech Trends and Technology. 7: 27-32.
10. Eapen, S., Tivarekar, S. and George, L. (1998). Thidiazuron-induced shoot regeneration in pigeon pea (*Cajanus cajan* L.). Plant Cell, Tissue and Organ Culture. 53: 217-220.
11. Eapen, S. and George, L. (1993). Plant regeneration from leaf discs of peanut and pigeonpea: Influence of benzyladenine, indoleacetic acid and indoleacetic acid-amino acid conjugates. Plant Cell Tissue and Organ Culture. 35: 223-227.
12. FAO (2014). Agriculture data (Food and Agriculture Organization, Rome, Italy), http://faostat.fao.org/.
13. Franklin, G., Jeyachandran, R. and Ignacimuthu, S. (2000). Factors affecting regeneration of pigeon pea (*Cajanus cajan* L. Millsp.) from mature embryonal axes. Plant Growth Regulation. 30: 31-36.
14. Franklin, G., Jeyachandran, R., Melchias, G. and Ignacimuthu, S. (1998). Multiple shoot induction and regeneration of pigeon pea [*Cajanus cajan* (L.) Millsp.] cv. Vamban from apical and axillary meristem. Current Science. 74: 936-937.
15. Gamborg, O.L., Miller, R.A. and Ojima, K. (1968). Nutrient requirements of suspension cultures of soybean root cells. Experimental Cell Research. 50. doi: 10.1016/0014-4827(68) 90403-5.
16. Geetha, N., Venkatachalam, P., Prakash, V. and Sita, G.L. (1998). High frequency induction of multiple shoots and plant regeneration from seedling explants of pigeon pea (*Cajanus cajan* L.). Current Science. 75: 1036-1041.
17. George, E.F. (1993). Plant propagation by tissue culture, part 1: the technology (Exegetics Limited, Westbury, UK).
18. George, L. and Eapen, S. (1994). Organogenesis and embryogenesis from diverse explants in pigeon pea (*Cajanus cajan L.*). Plant Cell Reports. 13: 417-420.
19. Guru Prasad, M., Prasad, N.V.K.V.T. and Sudhakar, P. (2011). *In vitro* proliferation of shoot regeneration from embryo of *Cajanus cajan* L. (var. LGG-29). Journal of Developmental Biology and Tissue Engineering. 3: 62-65.
20. Hansen, G,and Wright, M.S. (1999). Recent advances in the transformation of plants. Trends in Plant Science. 4: 226-231.
21. Jain, M. and Chaturvedi, H.C. (2004). *In vitro* cloning of *Cajanus cajan* var. Bahar through prolific shoot bud differentiation in leaf segments and production of fertile plants. Indian Journal of Biotechnology. 3: 258-262.
22. Jasani, H.V., Nimita, G., Umretiya Kapuria, M.N., Dharajiya, D.T., Khatrani, T.J., Pagi, N.K., Pachchigar, K.P. and Parmar, L.D. (2016). *In vitro* regeneration of pigeonpea [*Cajanus cajan* (L.) Millsp.] genotype GT 101 using cotyledonary node. Indian Journal of Science and Technology. 9: 1-05.

23. Kashyap, V., Sarangi, B.K., Yadav, M.K. and Yadav, D. (2011). Assessment of *in vitro* multiple shoot bud induction from leaf explants among eleven Indian cultivars of pigeonpea (*Cajanus cajan* L. Millsp.). Biotechnology. 10: 534-539.

24. Kaur, A., Devi, R. and Dev, A. (2012). Efficient *in vitro* regeneration in pigeonpea from cotyledonary nodal explants. Journal of Cell and Tissue Research. 12: 3075-3080.

25. Krishna, G., Reddy, P.S., Ramteke, P.W., Rambabu, P., Tawar, K.B. and Bhattacharya, P. (2011) *Agrobacterium*-mediated genetic transformation of pigeon pea [*Cajanus cajan* (L.) Millsp.] for resistance to legume pod borer *Helicoverpa armigera*. Journal of Crop Science and Biotechnology. 14: 197-204.