EFFECT OF GROWING MEDIA AND BENZYL AMINO PURINE ON MACROPROPAGATION OF BANANA CV. 'MALBHOG'

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EFFECT OF GROWING MEDIA AND BENZYL AMINO PURINE ON MACROPROPAGATION OF BANANA CV. 'MALBHOG'

BY

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CERTIFICATE

This is to certify that the thesis entitled **EFFECT OF GROWING MEDIA AND BENZYL AMINO PURINE ON MACROPROPAGATION OF BANANA CV. 'MALBHOG'** *submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of* ***MASTER OF SCIENCE (MS) in HORTICULTURE****, embodies the result of a piece of bona fide research work carried out by* ***MD. MATIUL ALAM,*** *Registration No. 04-**01324** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.*

I further certify that such help or source of information, as has been availed of during the course of this investigation has been duly acknowledged and style of this thesis have been approved and recommended for submission.

.....
Dated: JUNE, 2020
Place: Dhaka, Bangladesh

Dr. Md. Nazrul Islam
Professor
Supervisor

Dedicated To

My Respected Parents , Beloved Spouse

And Affectionate Children

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ABSTRACT

An Experiment was conducted at Horticulture Centre, Department of Agricultural Extension (DAE) , Burirhat , Rangpur during the period of February 2020 to May 2020 to investigate the macro propagation of banana cv.'Malbhog' as influenced by different growing media and Benzyl Amino Purine in different concentrations . The experiment was laid out in Randomized Complete Block Design (RCBD) with three replications. Treatments were different media viz, Mo: Control (Soil), M_1: Rice husk, M_2: Saw dust, M_3: Cocodust and four levels of BAP, viz, So: Control (0ml/L), S_1: 20mg/L, S_2: 30mg/L, and S_3: 40mg/L concentrations. Results indicated that days of first sucker emergence and highest values of vegetative growth i.e; number of suckers, suckers height, number of leaves, sucker collar diameter (cm), number of roots and roots length attributed the highest ramifications for the treatment of M_3 (Cocodust) and S_3(40 mg/L) of BAP. Combined applications M_3S_3 produced the best result for first time sucker emergence, number of sucker, sucker height and other vegetative growth and development. So, it can be concluded that cocodust media with 40 mg/L concentrations of BAP treatment combination is suitable for better macro propagation of banana cv.'Malbhog'.

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LIST OF ABBREVIATED TERMS

ABBREVIATIONS	ELABORATIONS
AOV	: Analysis of Variance
AEZ	: Agro- Ecological Zone
BARI	: Bangladesh Agricultural Research Institute
BAP	: Benzyl Amino Purine
BBS	: Bangladesh Bureau of Statistics
BAMIS	: Bangladesh Agro Meteorological Information Service
BMD	: Bangladesh Meteorological Department
CV%	: Percent of Co-efficient of Variance
DAE	: Department of Agricultural Extension
NS	: Non Significant
SRDI	: Soil Resources Development Institute

CHAPTER I

INTRODUCTION

1.1 Background Information

Banana (*Musa spp*.) belongs to the Musaceae family. Banana is originated from South East Asia, a region considered as the primary center of diversification of the crop and where the earliest domestication occurred (Simmonds, 1962). The name banana comes from and Arabic word meaning 'finger' (Banana Link, 2016).In Bangladesh bananas were cultivated in 120,709 ha of land and total production was 8,33,309 MT in 2018-2019 (BBS, 2019). FAOSTAT estimated that in 2018 a total of 155.2 million tons of banana were produced in the world. In Asia the highest half of the total some 62.6 million metric tons of banana were produced.

In Asia and pacific regions, banana has great socio economic significance. Bangladesh ranks 34th largest banana producing country. India is the top most (30.80 million tons) producer of banana in the world followed by China (13.32 million tons) (FAOSTAT, 2019). Banana is one of the most important commercial tropical fruits traded. Eve was said to have used banana leaves to cover the modesty in the garden of paradise as revealed from antiquity. Banana is thus called 'Apple of paradise'. It is also known as 'Adam Fig' (International Tropical Fruits Network, 2016). Banana is a valuable source of potassium, vitamin A, $B_{6,}$ and C. They are perennial and are a source of steady income all the year round. Bananas can be eaten fresh when ripe or after cooking or processing.

The total per capita consumption of banana in Bangladesh is about 4.7 kg. This is very much lower than that consumed by Europe specially Belgium (26.7 kg), Sweden (16.7 kg), and Germany (14.5kg). While USA consumed 13.1 kg and UK 10.5 kg (Siti Hawa, 1998). Bangladesh exports Champa kola (English name apple banana, Scientific name: *Musa sapientum*) throughout the year (Hortex Foundation, 2013).

1.2 Statement of the problem

Banana (*Musa spp*.) are seed sterile and parthenocarpic in nature and are normally propagated by suckers. In this method the rate of multiplication is highly limited (Ascenso, 1967; Baker 1959; Hamilton, 1965). Banana production sometimes become seriously affected by different fungal and viral diseases such as panama and bunchy top diseases.

As a result banana productivity decreases and the yield becomes very poor and static as well. Moreover, it is very difficult to carry the bulk volume of suckers from one place to another (Rahman *et. al*; 2002). Another major constraint to the expansion of banana and plantain cultivation is the scarcity of healthy planting material (Schill *et al*., 1997, Nkendah and Akyeampong , 2003).

Planting materials produced through tissue culture becomes costlier and the small and the marginal farmers cannot afford the higher cost. Under the above circumstances macro propagation of banana becomes popular (Njau *et al*. 2011). Macropropagation techniques involve methods that employ whole suckers or relatively large pieces of corm tissue to produce planting material in a propagator (Tenkouaam *et.al* ; 2006).

The quality of nursery potting medium is important to the successful growing of plants in containers (Bunt, 1988). It is easy to think of soil as a good medium, but most soils when used alone are very poor growing medium. Soil has been indicated as the easiest way through which seedlings become infected by diseases such as root knot nematode and seedling root rots (Egunjobi and Ekundare, 1981). Use of suitable growing media or substrates is essential for production of quality horticultural crops. It directly affects the development and later maintenance of the extensive functional rooting system. A good growing medium would provide sufficient anchorage or support to the plant, serves as reservoir for nutrients and water, allow oxygen diffusion to the roots and permits gaseous exchange between the roots atmosphere outside the root substrate (Abad *et al*. 2002; Bunt, 1988).

Organic substrates provide adequate nutrients to the seedlings , better root substrate relation than conventional soil mixed less predispose the seedlings to soil borne pests and diseases(Adams *et al.* 2003; anenbei *et al*; 2002). Rice husk is obtained as waste product of rice. It is light in weight, has uniform quality, resistant to decayed depletion of available nitrogen by microorganisms. Rice husk has the advantages of being easily incorporated into media for improved drainage and aeration (Baiyeri, 2005). Sawdust is the most commonly and widely used wood residues in in agriculture for potting mixers (Albery, 1975).

Cocopeat is an agricultural by-product obtained after the extraction of fibre from the coconut husk (Abad *et al.* 2002). Cocopeat is considered as a good growing media component with acceptable $P^{H,}$ electrical conductivity and other chemical attributes (Abad *et al.* 2002). Benzyl Amino purine (BAP) is a synthetic cytokine (Salisbury and Ross, 1985), which is used to induce multiple shoots production (vuylsteke, 1989). *In vivo* multiplication of suckers can be increased through application of BAP which induces sprouting of axillary buds and adventitious shoots (Singh, *et al*., 2011; Langford *et al*., 2012)

1.3 Objectives

However, considering the above circumstances, the present study was undertaken with the following objectives. The overall objective of this experiment was to measure and develop the production of quality planting materials of banana cv. Malbhog through Macro propagation in different growing media with application of BAP. Specifically two objectives are mentioned beneath.

i. To justify the better growing media for macro propagation of banana cv. Malbhog

ii. To determine the effect of Benzyl Amino purine (BAP) Concentrations on Macro propagation of banana cv. Malbhog.

CHAPTER II

REVIEW OF LITERATURE

Banana is one of the most important fruit as well as vegetable crops widely grown under field condition all over the world. This crop received much attention of researchers, extension agents, policymakers in every country of the globe. Various investigation have been carried out for its successful cultivation. The relevant literature on banana, growing media and propagation of banana related things have been reviewed here to the present study.

Thiemele *et al.* (2015) conducted an experiment in Africa, plantain is one of the most important starchy food and cash crops. Nonetheless, one of the major constraints for its production was the unavailability of healthy planting materials at planting time. This constraint could be lifted using the cloning of planting materials via the in vitro micropropagation or in vivo macropropagation techniques. Shelled corms from four cultivars, known as PITA 3, FHIA 21, ORISHELE and CORNE 1, were used. Three treatments differing in three hormonal concentrations, especially 20.0, 30.0 and 40.0 mg L^{-1} were tested. The control one was hormone free. Tested treatments were laid out in a split plot design. The decorticated banana corms were sprayed twofold at 2 weeks interval with BAP solution when placed in sterilized soil in high humidity plastic tunnel. It emerged from results, regarding BAP concentration effect, that BAP treatment with 40 mg L^{-1} significantly reduced the emergence time of shoots at 20 days as against 25.1, 28.3 and 28.5 for the 2 tested other treatments as well as control, respectively. Likewise, the concentrations 40.0 mg L^{-1} both recorded the largest number of sprouted buds per corm and number of shoots per corm. With respect to banana cultivar effect, PITA 3 showed the largest number of shoots per corm. Basing on such findings, it is concluded that MSD technique combined with BAP at 40.0 mg L^{-1} is a suitable technique for improving of the in vivo macropropagation of plantain. This concentration increased at least 50 % of sucker production compared to control.

Al-Amin *et. al*. (2009) conducted an experiment at the Biotechnology Laboratory, Biotechnology Division, Bangladesh Agricultural Research Institute (BARI), Gazipur during the period from September 2004 to June 2005 to investigate the effect of different concentrations of BAP and NAA on virus free plant regeneration, shoot multiplication and different concentrations of IBA and IAA on *in vitro* root formation of banana cv. BARI Banana-I. The culture meristem first turned brown in colour in 4-5 days which grew into a green globular hard coat mass after 30-35 days. From this ball like structure, adventitious plantlets were developed. Among the different concentrations, 7.5 mg/l BAP + 0.5 mg/l NAA showed highest shoot proliferation of 0.75, 2.75 and 6.25 shoots per explant at 10, 20 and 30 DAI, respectively. The longest shoot (1.03, 2.45 and 3.38 cm) at 10, 20 and 30 DAI, respectively, was produced by the treatment combination of 7.5 mg/l BAP + 0.5 mg/l NAA. The maximum number of leaves (2.50, 3.25 and 7.00 leaves/explant at 10, 20 and 30 DAI) were produced on the medium supplemented with the same treatment and it also produced the longest leaves, 0.85, 2.70 and 4.23 cm at 10, 20 and 30 DAI, respectively. For root initiation half strength MS medium supplemented with different levels of IBA (0, 0.5, 1 .0 and 1.50 mg/l) and IAA (0, 0.5 and 1.0 mg/l) was used. Root numbers varied with different concentrations of IBA and IAA. The highest number of roots were produced by 0.5 mg/l IAA + 0.5 mg/l IBA. The highest length (2.93, 4.63 and 5.88 cm) was recorded at 10, 20 and 30 DAI in the same treatment which was statistically significant. Meristem derived plantlets were transferred to poly bags containing 1:1 (ground soil : cowdung) mixture after 7 days hardening in room temperature (28-30°C) and established plantlet was ready for planting.

Ramirez *et al.* (2017) conducted an experiment in Research Department , Isabela State University , Echague, Isabela, Phillipins because the main production constrain of banana is the availability of reliable and safe planting material. Planting materials obtained through conventional methods do not meet the increasing demand for planting and they are of poor quality. Tissue culture approach can solve these problems but it is yet to benefit majority of small scale farmers because of the high costs and sophisticated skills associated with the technology. Therefore, to increase banana production in small scale systems, there is a need for affordable and simple technique for seedling production.

Macro propagation is one such technique and can improve banana production if adopted. In view of these characteristics, the effect of the humidifier and types of growth enhancer were evaluated using factorial in completely randomized design. The main plots consisted of types of humidifier with 2 levels: A1=with misting system, A2=without and for sub plots different growth enhancer was applied at 3 levels: B0=Control, B1=Benzyl Amino Purine at 1.5mg/L and B2= Naphthalene acetic acid at 0.93g/L. Result showed that humidifier led to a significant increase ($p<0.01$) in all the growth parameter tested during the 1st and 2nd cycle. On the other hand, growth enhancer affects significantly the number of days to emergence ($p<0.01$) First Generation Shoots (FGS), Second Generation Shoots (SGS), number of shoots emerged ($p<0.01$) (FGS, SGS), shoot collar diameter emergence ($p<0.01$) (FGS), ($p<0.05$) (SGS) and total leaf area ($p<0.05$) (FGS), ($p<0.01$) (SGS). However, no significant effect was observed on the interaction of the two factors for FGS but interaction of factors during SGS affect significantly the number of days to emerge ($P \leq 0.05$), number of shoots emerged ($P \leq 0.01$) and total leaf area ($P \leq 0.05$). Interaction of factors did not significantly affect the shoot collar diameter.

Lohidas, J. and Sujin, D. (2015) carried out an experiment in Department of Botany and Research Centre, Scott Christian College (Autonomous), Nagercoil - 629 003, Kanyakumari District (Tamilnadu), India. This study was micro propagation of banana cv. Matti. The sword suckers of three months old were used as explants.The explants sterilised thoroughly and MS media supplemented with the cytokinins like BAP, kinetin, zeatin and adenine sulphate in different concentration (0.5 to 5.5 mg/l) were used for the multiple shoot generation and shoot growth. Among the tested cytokinins BAP in medium concentration performed well and higher concentration of all tested cytokinins showed declining effect. Generated shoot initials were transferred to the rooting media after three sub cultures. For rooting MS media was supplemented with different levels of IBA. Rooting was highly promoted in the medium concentration of IBA. After 115^{th} day of inoculation the plantlets were transferred to green house for hardening and subsequently planted in the field after 30 days of acclimatization.

Rahman *et al*. (2013) performed an experiment to investigate the best plant growth regulators for shoot proliferation and multiplication; and to determine the optimum concentrations of phytohormones for shoot tip culture of banana cv. Agnishwar by observing the effect of different culture conditions during induction . A micro-propagation protocol for banana (*Musa* sp.) cv. Agnishwar was established by using shoot tip culture. Shoot tips obtained by *removing* leaf sheaths from sucker were cultured aseptically in MS (Murashige and Skoog) medium supplemented with different concentrations of cytokines viz. 6-benzylaminopurine (BAP), kinetin (kin), N_6 - (2-isopentyl) adenine (2iP) for multiplication of shoot and auxins viz. Indole-3- butyric acid (IBA), α-naphthalene acetic acid (NAA) for induction of root. Maximum multiplication (95%) was obtained in MS medium containing 4.0 mg/1 BAP. The highest average number of shoots for each explant (5.9) was found in MS medium fortified with 4.0 mg/l BAP while maximum elongation of shoot (4.9cm) was observed in MS medium having 5.0 mg/l BAP. IBA at a concentration of 1.0 mg/l was found most suitable for rooting of shoot. The rooted shoots were acclimatized and successfully transferred to plastic pots. After hardening, they were transferred to the main field and the survival rate was around90%.This protocol might be used for the massive *in vitro* production of the plantlets of banana cv. Agnishwar.

Kelta *et al.* (2018) worked out a research on explants (Suckers) of two banana cultivars on Murashige and Skoog (MS) medium supplemented with different concentrations of BAP and Kinetin sole and in combination for shoot initiation and multiplication. The concentration of BAP alone tested was (0.5mg/l and 1.0mg/l) and kinetin (0.5mg/l and1.0mg/l) whereas, the combination of 0.5mg/l BAP+0.5mg/l kin and 1.0mg/l BAP+0.5mg/l kin were used for shoot initiation. For multiplication, concentration of BAP(2.0,2.5 and 2.5mg/l) alone and in combination BAP+ Kin (1.5mg/l +1.5mg/l,2.0mg/l +2.0mg/l and 2.5mg/l +2.5mg/l) were used. The rapid shoot initiation obtained from MS medium supplemented with the combination of 1.0mg/l BAP with 0.5mg/l kin (8 and 10 days) in both Poyo and Giant Cavendish, respectively. The highest multiple shoot (6.0 and 4.5/explants), in Poyo and Giant Cavendish were observed on the MS medium fortified with 2.5mg/ l BAP+2.5mg/l kin and 2.0mg/l BAP +2.0mg/l Kin respectively. For root induction 1.5mg/l IBA and 1.5mg/l IAA each tested separately on

MS medium. IBA showed best performance with 5.12 and 4.69 root/ plantlet after four weeks of inoculation in Poyo and Giant Cavendish respectively. After 12 weeks in vitro plantlets were transferred to green house for acclimatization where 82% and 88% survival rate was recorded in Poyo and Giant Cavendish respectively. The two cultivars studied exhibited variation in shoot initiation, shoot multiplication and rooting. Among the two cultivars tested Poyo found to be more responsive for in-vitro techniques. It had highest rate of shoot initiation and multiplication.

Sajith *et al.* (2014) observed that Stimulation of lateral bud development and plantlet production is generally accomplished through decapitation methods in banana. Attempts were made in the present study to enhance the efficacy of decortication in elite cv. Bangladesh Malbhog using additives like bio-fertilizers and plant growth hormones. This trial was carried out with suckers weighing 1.0-1.5 kg and sawdust as substrate.

All treatments tested, showed good response in terms of plantlet production and enhanced bud proliferation, growth and better root profiles compared to Micro propagation assures rapid production of healthy, vigorous, and disease-free planting material. However, due to the large capital investments required for tissue culture facility, the plantlets produced are fairly expensive and beyond the reach of resource poor farmers. Thus, tissue culture as a method of generating planting material is not an option for small-scale farmers; hence, there is a need for cheap and simple technique that increase the sucker multiplication at farm level and warranting minimum technical skill. Macro propagation is one such cost effective technique where repression of apical meristem will stimulate the regeneration of lateral meristem (Uma *et al*., 13). Increased suckering rate can be achieved through complete/ partial decapitation on a field grown plant or detached corm technique (Baiyeri and Aba, 2) and sawdust is the best substrate over others like rice hull, sand etc. with higher water holding capacity (Baiyeri and Aba, 1). In the present study, attempts have been made to enhance the rate of plantlet production through macropropagation by the addition of bio-fertilizers (AMF, *Trichoderma viride*, *Bacillus subtilis*, *Pseudomonas fluorescens* and *Azospirillum*) and phytohormones (BAP and IBA) to the explant/ substrate.

Njau *et al*. (2012) found that Banana (*Musa spp*.) is one of the most important food crops that contribute to the food security of the majority small holders in Kenya. However, due

to diseases, banana production has not reached its full potential. Fusarium wilt caused by Fusarium oxysporum f. sp. cubense (Foc); black and yellow sigatoka leaf spot caused by Mycosphaerella fijiensis and Mycosphaerella musicola respectively, weevils (Cosmopolites sordidus) and plant parasitic nematodes have been the major diseases affecting banana seedlings. In addition, banana Xanthomonas wilt (BXW) caused by the bacterium *Xanthomonas vasicola* pv. musacearum is a disease that rapidly destroys plantations.

Most of these diseases are spread by use of infected planting material. Macro propagation is a cost effective seedling production technology which can be implemented with little capital and skill to provide planting material. The study was carried out to determine the potential of the technology to produce healthy banana seedlings. From the study, the macro propagation technique has potential for banana seedlings multiplication only if the mother corms were healthy.

Patel, M.K. and Rath, S.S. (2018) stated that Banana in India is mostly a crop of marginal farmers with little affordability to tissue culture plants which are 4-8 times higher than the sucker cost. Hence, a simple and farmer friendly method has been developed to bridge the gap in supply of adopt this especially to enhance the planting material production of traditional cultivars. Healthy planting material with an affordable cost through macro-propagation. This method generates plantlets from sword suckers and initial explants so farmers can Banana and plantains are propagated vegetative through sword suckers and other types of planting materials like bits, butts and peepers. But the most common limiting factor for enhanced productivity is the non-availability of clean and disease free planting material. To address the problem of poor suckering nature of the crop, tissue culture technology is used for the mass production of the planting material.

Thungon *et al.* (2017) conducted an experiment to standardize the suitable growing media for macroprogation of Malbhog banana. Three growing media viz. sawdust (M_1), Paddy husks (M_2) and cocopeat (M_3) were selected for the study. The experiment was laid out in factorial randomized block design with three replications under polyhouse condition. It was observed that the emergence of first primary sucker

was significantly influenced by the growing media. Cocopeat required shortest period (18.57) while paddy husk required longer period for emergence of primary suckers (3.88), secondary suckers (8.26) and tertiary suckers (23.84) per corm and similarly, high numbers were also observed when cocopeat was treated with BAP (0.04%). Out of three media used, the highest weight (316.92 g) was recorded in cocopeat which was at par with 309.0 g in sawdust.

Cocopeat recorded the shortest time for primary sucker emergence (18.57), decortications of primary suckers (49.95 days), separation of tertiary suckers (82.55 days), higher number of leaves (5.80), and roots (25.16) and bigger sucker (316.92 g). This was the first report of successful macroprogation of Malbhog banana using cocopeat growing medium.

Jimson *et al*. (2016) found that the main production constrain of banana is the availability of reliable and safe planting material. Macropropagation can improve banana production if adopted. In view of these characteristics, the effect of the humidifier and types of growth enhancer were evaluated using factorial in completely randomized design. Factor A consisted of types of humidifier: A1=with misting system, A2=without and different growth enhancer was applied for Factor B: B0=Control, B1=Benzyl Amino Purine at 1.5mg/L and B2= Naphthalene acetic acid at 0.93g/L. Result showed that humidifier led to a significant increase ($p<0.01$) in all the growth parameter tested during the 1st and 2nd cycle. On the other hand, growth enhancer affect significantly the number of days to emergence ($p<0.01$) First Generation Shoots (FGS), Second Generation Shoots (SGS), number of shoots emerged ($p<0.01$)(FGS, SGS), shoot collar diameter emergence ($p<0.01$) (FGS), ($p<0.05$)(SGS) and total leaf area ($p<0.05$)(FGS), ($p<0.01$)(SGS). However, no significant effect was observed on the interaction of the two factors for FGS but interaction of factors during SGS affect significantly the number of days to emerge ($P \leq 0.05$), number of shoots emerged ($P \leq 0.01$) and total leaf area ($P \leq 0.05$). Interaction of factors did not significantly affect the shoot collar diameter.

Keeriol *et al*. (2018) conducted an experiment to study the multiplication rates of banana shoot tips derived from different suckers under in vitro conditions during successive sub-culture of FHIA-23, KM5 and Basari and assessment of rapid amplified polymorphic DNA

(RAPD) techniques of banana cultivars through molecular marker. Shoot tips of banana varieties were cultured on MS medium supplemented with different concentrations of Benzaline alkaline phosphate (BAP). MS ½ with Indole butaric acid (IBA) 1 mg^{L-1} + 5.0 g, 10 g, 20 g, 30 g and 40 g sugar $L^{-1}$for root length, number of root and root weight were used for root induction.

The results revealed that the best shoot initiation was observed in Basari using MS medium containing BAP 4 mg L^{-1}. Maximum shoot length was observed in KM5 under the concentration of BAP 3.0 mg L^{-1}. Maximum numbers of shoots and shoot weight were observed in KM5 followed by FHIA-23 and Basari variety. Maximum root length was observed in KM5 followed by FHIA-23 under the concentration of sugar 30 g L^{-1} + IBA 1.0 mg L^{-1}. Maximum numbers of roots were observed in KM5, followed by FHIA-23 and Basari variety under concentration of 30 g L^{-1} + IBA 1.0 mg L^{-1}. It was concluded that the KM5 variety was superior, followed by FHIA-23 and Basari. The concentration of 3.0 to 4.0 mg L^{-1} of BAP were found better for shooting and 1.0 mg L^{-1} of IBA + 30 and 40 g L^{-1}sugars for root induction of banana micro propagation.

Awang *et al*. (2009) stated that Cocopeat is considered as a good growing media component with acceptable pH, electrical conductivity and other chemical attributes but it has been recognized to have high water holding capacity which causes poor air-water relationship, leading to low aeration within the medium, thus affecting the oxygen diffusion to the roots. Incorporation of coarser materials into cocopeat could improve the aeration status of the media. Approach: Selected chemical and physical characteristics of five types of growing media comprising of (v/v) 100% cocopeat, 70% cocopeat: 30% burnt rice hull, 70% cocopeat: 30% perlite, 70% cocopeat: 30% kenaf core fiber and 40% cocopeat: 60% kenaf core fiber were determined and their suitability as growing media was tested using Celosia cristata. Data on pH, Electrical Conductivity (EC) and various aspects of air-water relationships of the media, as well on growth and flowering of test plant and leaf nutrient contents were collected. Results: Initial pH for 100% cocopeat and 70% cocopeat: 30% kenaf core fiber was higher than the other media but the values were eventually similar by the end of the study. The bulk density and EC of media containing burnt rice hull was markedly higher than the other media (0.12 g cm3 and 0.48 mS cm−1, respectively). Media

comprising of 70% cocopeat: 30% burnt rice hull and 70% cocopeat: 30% perlite contained higher air content. The former held the highest volume of available water. Incorporation of burnt ricehull and perlite into cocopeat increased water absorption ability of the media which reached saturation earlier than the other media.

Addition of burnt rice hull (30%), perlite (30%) and kenaf core fiber (30%) to cocopeat elevated the Air-Filled Porosity (AFP) of the media. The growth and flowering of Celosia cristata were the greatest when grown in a mixture of 70% cocopeat: 30% burnt rice hull and perhaps linked with a good balance in the aeration and moisture relationship of the media.

Njau *et al.* (2010*),* conducted an experiment on banana and stated that banana (*Musa spp*.) is one of the most important food and cash crops in parts of Kenya. The crop provides food security, nutrition and income for many smallholder farmers. Bananas can be eaten fresh, cooked or processed into numerous value added products, depending on the variety. Despite the importance of the crop, it faces major production challenges including scarcity of high quality seedlings, insect pests and diseases. Naturally produced suckers are more likely to carry pests and diseases leading to reduced productivity and shortened lifetime of new plantations. Demand for disease free high quality planting materials has been on the increase. To address this demand macropropagation has been introduced as an alternative seedling production technology. The technology requires little capital and skill to implement, and can therefore be promoted to small scale seedling entrepreneurs and farmers. However, some aspects of the technology require further research to ensure quality of seedlings. This study is being carried out to establish the effectiveness of macropropagation technology to produce disease free banana seedlings.

Muhammad *et al*. (2004) studied In-vitro multiplication of banana (*Musa spp*.) cv. Basrai. Shoot tips were cultured on Murashige & Skoog basal medium supplemented with 5.0 mg/l BAP. Observations were recorded at an interval of four weeks for five subculturings. Evaluations were done at each subculture by counting the number of new shoots produced. Shoot tips coming from different rhizomes behaved differently under in vitro conditions. Some being highly productive while others produced less number of shoots. On the average, 124 plants were produced from each shoot tip after five subculturing.

Kindimba (2013) found that in vivo macropropagation either alone or in combination with benzyl amino purine (BAP) is an alternative simple technique for banana multiplication but has not been applied to recalcitrant plantain such as cv. 'Itoke Sege'. This study was conducted to determine the effect of BAP concentration on in vivo multiplication and genetic stability among in vivo derived regenerants of plantain cv. 'Itoke sege'. An experiment was laid out in RCBD with four treatments each replicated three times. The treatments consisted of four BAP concentrations (1.5, 3.0, 6.0 mg/l and untreated control). Data were collected on number of days to first shoot emergence, number of shoots per corm, shoot size and morphological and genetic stability of in vivo derived shoots. Morphological stability was assessed using banana morphotaxonomic descriptors while genetic instability was assessed based on analysis of 2C nuclear DNA content of in vivo derived suckers. Results showed that BAP concentration at 1.5 mg/l significantly ($P \leq 0.05$) enhanced first the shoot emergence at 15.78 days followed by BAP at 3.0, 6.0 mg/l and untreated control with 25.18, 28.39 and 36.43 days, respectively. Moreover, BAP concentration at 1.5 mg/l significantly ($P < 0.05$) produced the largest number of suckers of 17.11 suckers per corm followed by untreated control and BAP concentration at 3.0 and 6.0 mg/l and with 15.23, 13.08 and 12.96 suckers per corm, respectively. Similarly, BAP at 1.5 mg /l and untreated control significantly ($P \leq 0.05$) showed the lowest frequencies of off-types with 10.89 and 10.23 % compared to BAP at 3.0 and 6.0 mg/l with 12.08 and 12.86 % of off-types, respectively. However, ploidy analysis revealed that the off-type and normal banana suckers had significantly ($P \leq 0.05$) equal 2C nuclear content and ploidy level. The findings of this research provide evidence for the use of in vivo macropropagation coupled with BAP at 1.5 mg L-1 as an alternative technology for rapid production of planting materials of recalcitrant plantain varieties.

Dagnew *et al.* (2012) carried out an experiment at the Tissue Culture Laboratory of Melkassa Agricultural Research Centre, Ethiopian Institute of Agricultural Research

(EIAR) to investigate the effects of different types and concentrations of cytokinins and auxins on shoot initiation and multiplication, and in vitro shoot rooting of three banana varieties using shoot-tip explants. Shoot initiation was greater on Murashige and Skoog (MS) basal medium supplemented with 3 mg/l N6-benzylaminopurine (BAP) for Dwarf

and Giant Cavendish while 2 mg/l for Poyo varieties. Among the different concentrations of plant growth regulators (PGR) tested, MS medium supplemented with combinations of BAP and indole-3-acetic acid (IAA) at 3+0.4, 4+0.4 and 3+0.2 mg/l for Dwarf, Giant and Poyo, respectively, were best combinations for high rates of shoot proliferation and elongation.

Further multiplication of shoots required up to 5 times sub culturing of 1 month each on the same media combination. In this study, about 3-fold multiplication rate was achieved during every subculture. Better rooting was obtained when the shoots were cultured on MS medium with 2.12 mg/l α-naphthalene acetic acid (NAA) for Dwarf and Giant while 1.74 mg/l indol-3-butyric acid (IBA) for Poyo. In vitro rooted plantlets were transferred to the lathouse for acclimatization and hardening. The best growth was recorded for plantlets transplanted on potting media containing a 3:1 ratio (v/v) of sugarcane filter cake and sand. The hardened plants were transferred and well established to the field.

From the above review of literature it is revealed that use of different growing media, rooting and shooting hormones in different concentrations have significant effect on days for sucker emergence, sucker height, number of suckers, leaves, roots and root length of suckers in macro propagation of banana.

CHAPTER III

MATERIALS AND METHODS

The investigation was carried out at the experimental field of the Horticulture Centre, of the Department of Agricultural Extension (DAE), Burirhat, Rangpur, during the period from February 2020 to May 2020 to study the "Effect of growing media and Benzyl amino purine (BAP) on macro propagation of banana cultivar: Malbhog". A brief description about the locations of the experimental site, characteristics of soil, climate, materials, layout and design of the experiment, land and pit bed preparation, separation of suckers from corms, intercultural operations, bagging, hardening, data recording procedure, economic and statistical analysis etc. which are presented as follows:

3.1. Experimental site

The research work was conducted at the nursery field of the Horticulture Centre, the Department of Agricultural Extension (DAE), Burirhat, Rangpur during the period from February 2020 to May 2020.

3.2 Geographical Location

The experimental area is situated between 25.48° and 25.57° North latitudes and in between 89.05° and 89.21° East longitudes and at an altitude of 34 meter above the sea level. The experimental field belongs to the Agro-ecological zone of The "Active Teesta Floodplain", AEZ-2 (bamis). This is a region of irregular patterns of grey stratified sands and silts. They are moderately acidic throughout and parent alluvium is medium in weather able k minerals. Organic matter contents and soil fertility level are low to medium.

3.3 Climate

Area has classified as warm and temperate climate. The summers are much rainier than the winters in Rangpur. The average temperature in Rangpur is 24.9º C. The annual rainfall is 2192 mm . Area is characterized by scanty rainfall associated with moderately low temperature during the Rabi season (October-March) and high rainfall, high temperature during rest of the year. Meteorological information regarding temperature, relative humidity, rainfall and sunshine hours prevailed at the experimental site during the study period was recorded and presented in Appendix I.

3.4 Characteristics of soil

Soil of the experimental site belongs to the general alluvial soil type, Shallow Red Brown. Top soils were clay loam in texture, olive-gray with common fine to medium distinct dark brown mottles. Soil pH ranged from 4.8- 6.5 and had organic matters are 1.55-1.82%. Experimental area was flat having available irrigation and drainage system and above flood level. The soil related data and information were collected from Soil Resource and Development Institute (SRDI), Dhaka.

3.5 Planting materials

The banana crop cultivar Malbhog was used as a test crop.The planting materials were collected from a farmer named Shahjahan Islam Shamim, village: Chawksolagari, Upazilla: Pirganj, Distrit:Rangpur. About 06 months old suckers were used for the experiment.

3.6 Treatment of the Experiment:

The Experiment consisted of two factors as follows:

Factor A: Different Media

a. Mo = Soil (control)
b. M_1 = Rice husk
c. M_2 = Saw dust
d. M_3 = Coco dust

Factor B : BAP application

a. So = 0 mg/L (No BAP application)
b. S_1 = 20mg/L
c. S_2 = 30 mg/L
d. S_3 = 40mg/L

There were altogether 16 treatment combinations thus planned as following:

$MoSo$, MoS_1, MoS_2 , MoS_3, M_1So, M_1S_1, M_1S_2, M_1S_3 , M_2So, M_2S_1, M_2S_2, M_2S_3 , M_3So, M_3S_1, M_3S_2, M_3S_3

3.7 Design and layout of the experiment

The experiment was laid out Randomized Complete Block Design (RCBD) with three replications. The experimental area was divided into three equal blocks. Each block was divided into 16 equal blocks. Every replication had sixteen lots where 16 treatments were allotted at random. The total number of lot was 48. The size of each plot was 1m×1m. The distance between two blocks was 0.50 m. A layout of the experiment has been shown in figure 1.

3.8 Collection of planting materials

Selected germplasm of banana corms were collected from a farmer plot of Pirganj upazilla, Rangpur district on 12th February 2020. A careful selection of sword suckers was done from healthy mother plants grown by that farmer. Sword suckers were pared to remove roots and packed into plastic bags for transport to Horticulture centre, Burirhat, Rangpur.

3.8.1 Preparation of planting materials

The remnants of the pseudostem and roots were removed and external layer of the corm was scrapped using a sharp knife to ensure freeness from all nematodes and external root borne pathogens. The apical meristem was removed to a depth of 2 cm leaving a cavity of 2cm diameter in the rhizome. The rest of the corm was given 4 cross cuts and incised upto 0.25-0.50 cm depending on the sucker size. Cross cuts were made on the exposed axillary buds and apical meristem to encourage sprouting of axillary buds and to kill apical dominance respectively. The corms were washed with fungicides Tilt-250 EC at 2ml/L solution for 30 minutes and air dried 3-4 hours before planting. The decapitated corms were planted individually in different media peat. Corms were buried 6cm deep in the substrate to protect from intensity of direct sunlight and then respective treatments were imposed .The apical meristem when scooped out to a depth of 2 cm near the crown region and the corms were given 4-6 transverse incisions to a depth of 2 mm, in this cavity 4 ml of each concentrated BAP was poured into left by the removal of the apical meristem. The same treatment was imposed after first time sucker collection to start second time sucker regeneration.

Plate 1. Collection of 5-6 months sucker

Plate 2. Decapitation of sucker

Plate 3. Decapitated sucker

Plate 4. Cavity made on rhizome for pouring BAP

Plate 5. Cavity made on different rhizomes

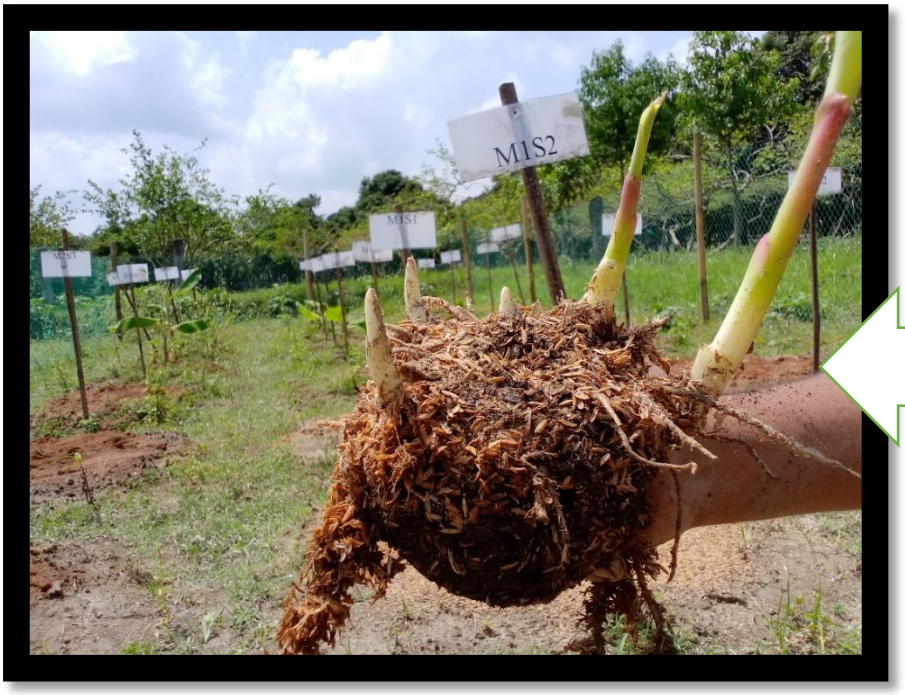


Plate 6. Sprouted bud

Plate 7. Sprouted small sucker, ready for collection

Plate 8. Separation of sucker from rhizome

Plate 9.
Collected small sucker, ready for bagging

Plate 10. Bagging of produced sucker

Plate 11. Bagged small sucker carrying for hardening in shade

3.9 Land and Plot preparation

The experiment plot was prepared by several ploughing and cross ploughing followed by laddering and harrowing with tractor and power tiller to bring about good tilth. Weeds and other stables were removed carefully from the experimental plots and plots were prepared as per layout. Saw dust, cocodust and paddy husk were taken as substrate/ growing media for the experiment. Rice husk and saw dust were collected from rice mill and saw mill in Burirhat and cocodust was collected from Paglapir, Rangpur. Each plot was 30 cm depth. Control media (Mo) Soil was treated with H_2O_2. Other media except soil like Rice husk, Saw dust and Coco dust were sterilized by boiling in water at 100º C for 30 minutes.

3.10 Preparation of BAP

Benzyl Amino Purine (BAP) in different concentrations of 0, 20, 30 and 40 mg/L were prepared by following the procedure mentioned below. 20 mg powder of BAP was poured into 1(one) liter of distilled water. This way 20 mg/L solution was made. In the same way 30mg/L and 40 mg/L concentrated BAP solutions were also made. Control corms cavity that is 0mg/L water, No BAP with water was poured.

3.11 Intercultural Operations

Intercultural operations such as weeding and irrigation etc. were done when necessary for keeping viability and regeneration capacity of corms. No insecticide was used at all. Proper shading was given to protect the young buds and seedlings from scorching sunshine during the day time.

3.11.1 Weeding

The first weeding was done after 20 days of transplanting to keep the corms free from weeds. Weeding was also done in several times when it was needed.

3.11.2 Irrigation

Primarily watering was done once weekly by water cane in each bed. In mature stage, when propagules grew around 15-30 cm height then flood irrigation was done to the field when it was necessary for the crop.

3.11.3 Separation and collection of small produced suckers

After primary decortication, the emerging small suckers were allowed to grow for about 25-30 days after emergence. And when they attained 3-5 leaves with height of about 15-20 cm. Small newly produced suckers were collected after 60 days of treatment for the first time and after 90 days second time collection was done. They were carefully separated with a sterilized sharp knife and transplanted into the polybags having 5-6 pierced holes (20cm × 20cm size) containing the mixture of soil and decomposed cowdung at the ratio of 1:1 for hardening in nursery shade .

3.12 Data recording

Observations were recorded for the following parameters

I. Number of days to first sucker emergence
II. Number of sucker emerged per corm
III. Sucker height (cm)
IV. Number of leaves per sucker
V. Sucker collar diameter (cm)
VI. Number of roots per sucker
VII. Length of root

3.13 Data collection procedure

3.13.1 Number of days to first sucker emergence

Days to first shoot emergence was collected by counting the days from when corms were planted to the day when the tallest shoot appeared above the media (soil, rice husk, saw dust, coco dust).

3.13.2 Number of suckers emerged per corm

It was recorded by counting number of shoots at the time of collection of sucker by separating them from corm in each cycle.

3.13.3 Sucker height (cm)

Shoot height was measured using vernier caliper from the root collar to the tip of cigar leaf.

3.13.4 Number of leaves per sucker

It was recorded by counting number of leaves contain in each sucker when collected from sucker.

3.13.5 Sucker collar diameter (cm)

Collar girth was recorded by measuring 3.0 cm apart from root collar region using vernier caliper.

3.13.6 Number of roots per sucker

It was recorded by counting number of shoots at the time of collection of sucker by separating them from corm in each cycle.

3.13.7 Length of roots

Root length was measured using vernier caliper from the root source region to end of the root.

3.14 Statistical analysis

The recorded data on different parameters of height, length, and other contributing characters were statistically analyzed using statistix 10 software to find out the significance of variation due to applied treatments. The mean for all the calculated and the analysis of variance for each of the characters under study was done by F (variance) ratio test for Randomized Complete Block Design (RCBD) and mean separation was done by Tukey HSD test at 5% level of probability.

CHAPTER IV

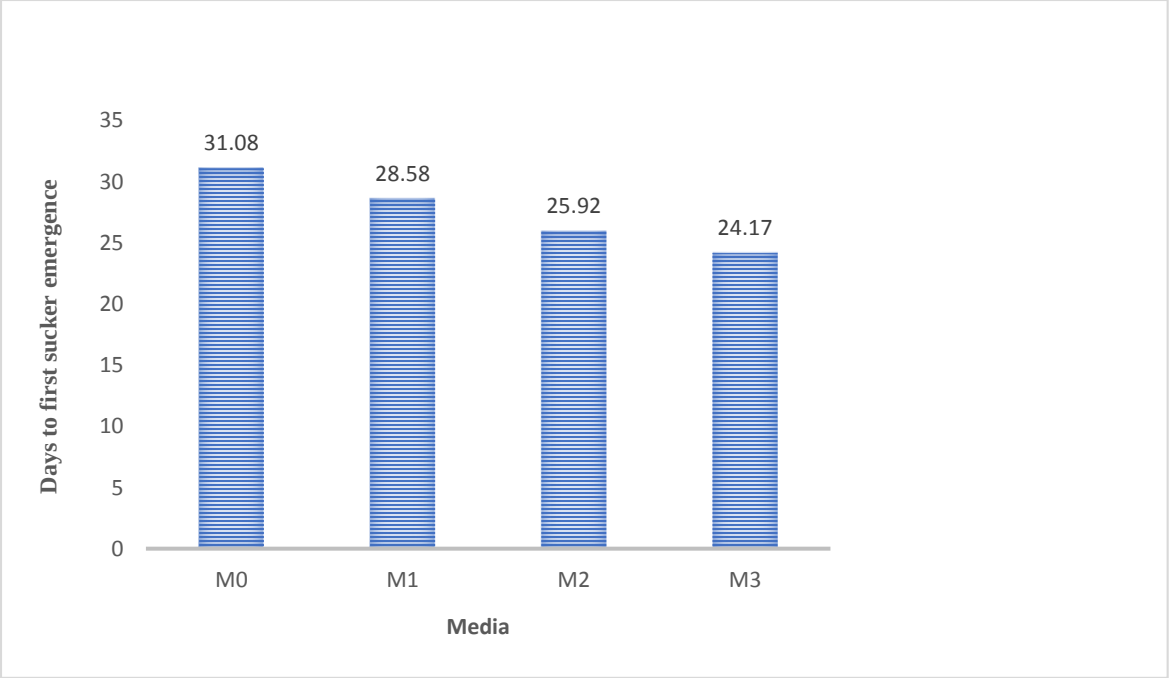
RESULTS AND DISCUSSION

The experiment was conducted to study the macro propagation of banana (*Musa spp*.) cultivar, Malbhog as influenced by Media and Benzyl Amino Purine (BAP) concentrations. Data on first sucker emergence, number of suckers, sucker height, number of leaves, sucker collar diameter, number of roots and roots length were recorded. The analysis of variance (ANOVA) of the data on different parameters are presented in Appendix section. The results have been presented with the help of graphs, tables and possible interpretations given under the following headings.

4.1 Days of first sucker emergence:

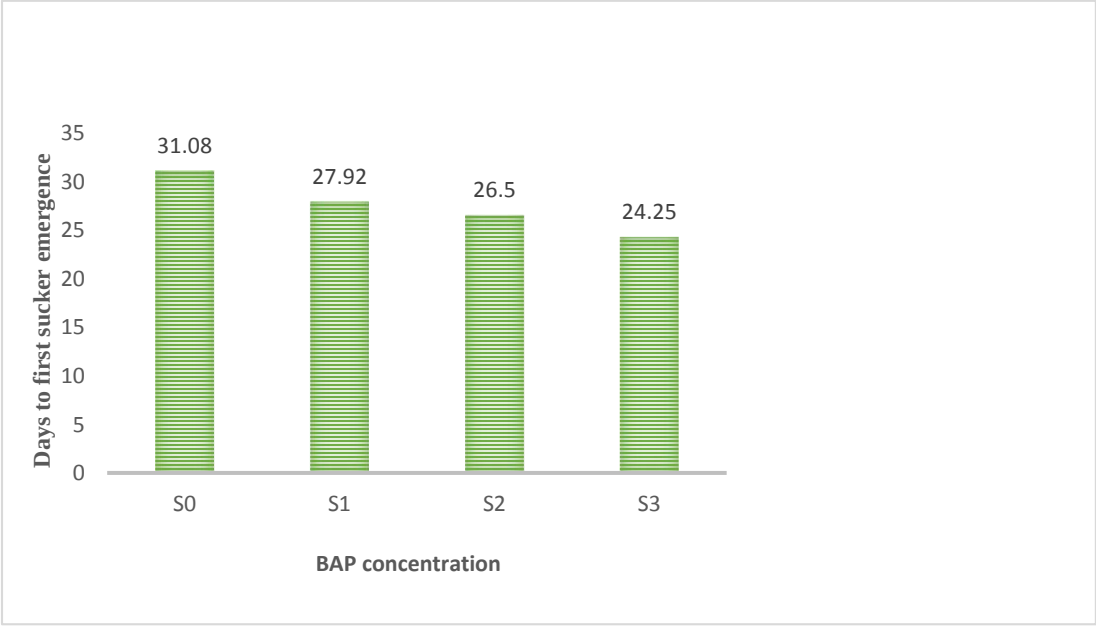
Application of different media and BAP concentrations showed significant difference in first banana buds sprouting or sucker emergence (Figure 2). Therefore in case of growing media shortest days required for bud sprouting was in Cocodust (M_3) and longest time period required in control condition Soil (Mo). The shortest and longest time (days) required for sprouting was recorded 24.17 days and 31.09 days respectively in the media. Other media's like Rice husk and Saw dust required 28.58 days and 25.92 days respectively for first banana sucker emergence.

Application of different levels of BAP also performed significant difference on first sucker emergence (Figure 3). However Shortest days required for bud sprouting was in S_3 (BAP 40 mg/l) and longest time period required in control condition So (No BAP application). The shortest and longest time (days) required for sprouting bud was recorded 24.25 days and 31.09 days respectively. In case of 20mg/L and 30mg/L BAP concentration, 29.92 days and 26.50 days respectively . Additionally it is observed that more concentration of BAP up to 40mg/L showed reverse impact of sucker emergence, i.e; in this solution days required less compare to other concentration.



Growing media, M_0=Soil, M_1=Rice husk, M_2= Saw dust, M_3 = Coco dust

Figure 2. Effect of media on days of first sucker emergence



BAP concentrations, S_0=0 mg/L(Control), S_1=20 mg/ L, S_2= 30 mg/ L, S_3= 40mg /L

Figure 3. Effect of BAP concentrations on days to first sucker emergence

In case of combined effect of different media and different BAP concentrations revealed no significant difference on first sucker emergence (Table 1). Hence the Shortest time (19.67 days) required in first sucker emergence in M_3S_3 combination and longest period (34.34 days) happened in MoSo combination. In other side M_3S_2 combination took 22.34 days to first sprouting which is second shortest time and M_2S_3 combination took third shortest time and it happened in 22.67 days. From the table it is noticeable that better combination occurred in Saw dust and Cocodust media and with 30mg/L and 40mg/L BAP concentration. Thungon *et al.(*2017) also found that, cocodust required shortest time and rice husk required longer period for emergence of primary and secondary suckers.

Table 1. Combined effect of media (M) and BAP concentrations (S) on first sucker emergence

Combined effect (M X S) of media (M) and BAP concentration(S)		Days to sucker emergence
M_0	S_0	34.34
	S_1	31.34
	S_2	30.34
	S_3	28.34
M_1	S_0	30.67
	S_1	29.00
	S_2	28.34
	S_3	26.34
M_2	S_0	29.34
	S_1	26.67
	S_2	25.00
	S_3	22.67
M_3	S_0	30.00
	S_1	24.67
	S_2	22.34
	S_3	19.67
Tukey HSD (0.05)		3.2042
CV %		3.84

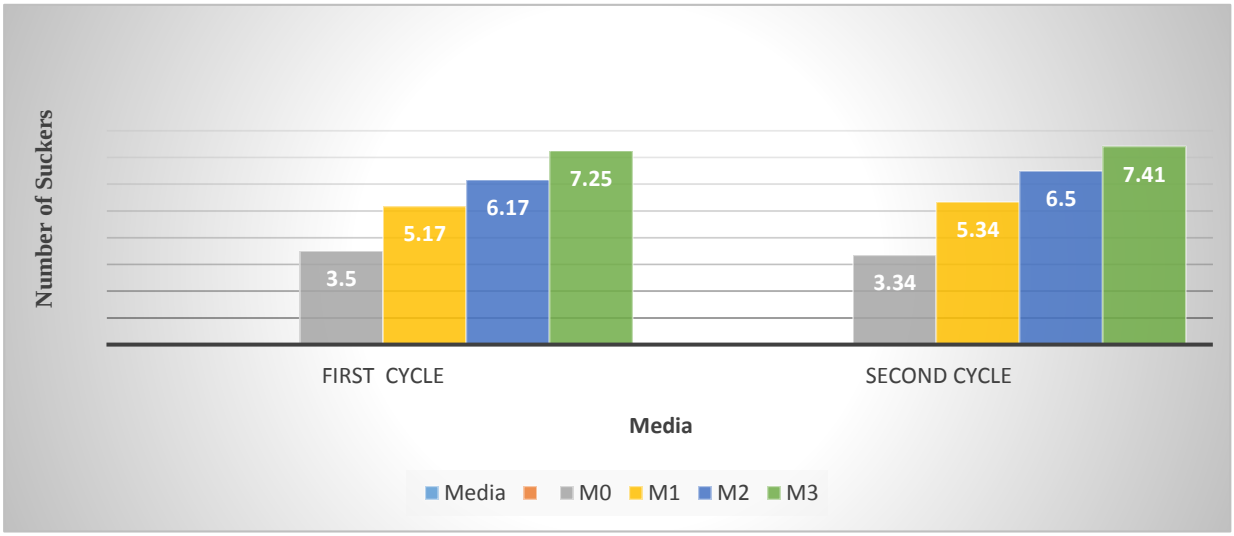
NS = Non Significant

Growing media, Mo = Control (Soil), M_1 = Rice husk, M_2 = Saw dust, M_3 = Coco dust

BAP concentration, S_0 = Control (0 mg/L), S_1 = 20 mg/L, S_2 = 30 mg/L, S_3 = 40 mg/L

4.2 Number of suckers produced in each corm

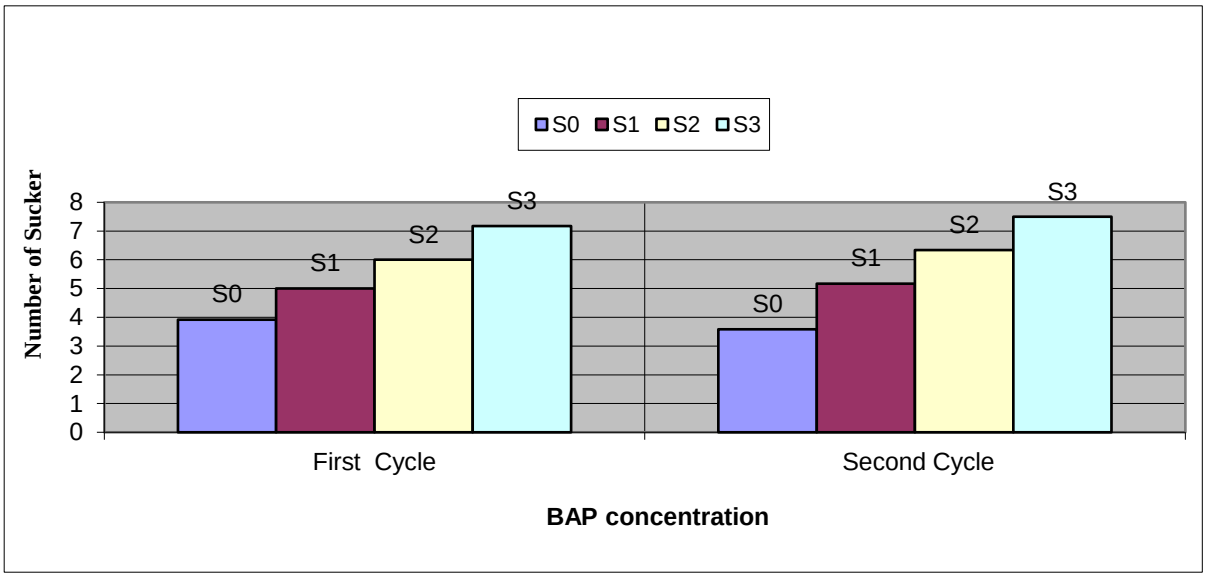
The number of suckers showed significant difference in both different media and different BAP Concentrations. The maximum suckers were produced in cocodust (M_3) media and minimum suckers produced in control (Figure 4), Mo (Soil) condition which was 7.25 and 3.50, respectively in first cycle. Moreover in this cycle rice husk media produced 5.17 and sawdust media produced 6.17 suckers. But in second cycle highest number of suckers were slightly increased 7.41 in as before in Cocodust (M_3) media and lowest also slightly decreased to 3.34 as before in controlled condition (Mo). In rice husk and saw dust media produced 5.34 and 6.5 suckers, respectively.



Growing media, M_0=Soil, M_1=Rice Husk, M_2= Saw dust, M_3 = Coco dust

Figure 4. Effect of media on number of sucker production.

Application of different BAP concentrations showed significant variations on number of sucker production. In first cycle highest (7.18) and lowest (3.92) suckers were produced in S_3 and So concentrations (Figure 5). At the same time S_1 and S_2 concentration produced 5.0 and 6.0 suckers, respectively. On the flip side in second cycle, highest (7.50) and lowest (3.58) number of suckers were produced in S_3 and So concentration. But in S_1 and S_2 condition suckers were produced 5.17 and 6.34, respectively.



BAP concentrations, S_0=0 mg/L(Control), S_1=20 mg/ L,S_2= 30 mg/ L,S_3= 40 mg /L

Figure 5. Effect of BAP concentrations on sucker production

The combined effect of media and BAP concentrations performed wide range of variation on number of sucker production (Table 2). The highest number of suckers was counted in M_3S_3 combination and lowest was recorded in MoSo combination and the number were 9.34 and 2.67, respectively in first cycle. However it is noted that simultaneously M_3S_2 and M_2S_3 produced 7.67 suckers, M_2S_2 , M_1S_3 and M_3S_1 combination generated 7.34, 7.0 and 6.67 and among the lower producer combination MoS_1, M_1So, MoS_2 produced 3.0, 3.34, 3.67 suckers . But in second cycle highest and lowest number of sucker were 10.00 and 2.34 in M_3S_3 and MoSo combination, respectively. Besides these M_2S_3 , M_3S_2, M_1S_3 , M_3S_1 produced remarkable suckers 8.67, 8.34, 7.34 and 6.67, respectively. Among lower producer combination MoS_1 and M_1So produced only 3.0 suckers; MoS_2 and MoS_3 produced 4.0 suckers, M_2So, M_1S_1 produced 4.34 and 5.0 suckers, respectively. Thungon *et al.* (2017) also found that higher number of suckers are produced in cocodust and lower in sawdust and rice husk. Additionally BAP (0.04%) showed higher number of suckers.

Table 2: Combined effect of different media (M) and different BAP concentrations (S) on number of sucker production

Combined effect of (M x S) media and BAP	Number of sucker production	
	First cycle	Second cycle
S_0	2.67	2.34
S_1	3.00	3.00
S_2	3.67	4.00
S_3	4.67	4.00
S_0	3.34	3.00
S_1	5.00	5.00
S_2	5.34	6.00
S_3	7.00	7.34
S_0	4.34	4.34
S_1	5.34	6.00
S_2	7.34	7.00
S_3	7.67	8.67
S_0	5.34	4.67
S_1	6.67	6.67
S_2	7.67	8.34
S_3	9.34	10.00
Tukey HSD (0.05)	1.7670	1.9102
CV%	10.52	10.91

Media ,

M_0=Soil

M_1=Rice husk

M_2= Saw dust

M_3 = Coco dust

BAP concentration ,

S_0=0 mg/L Water

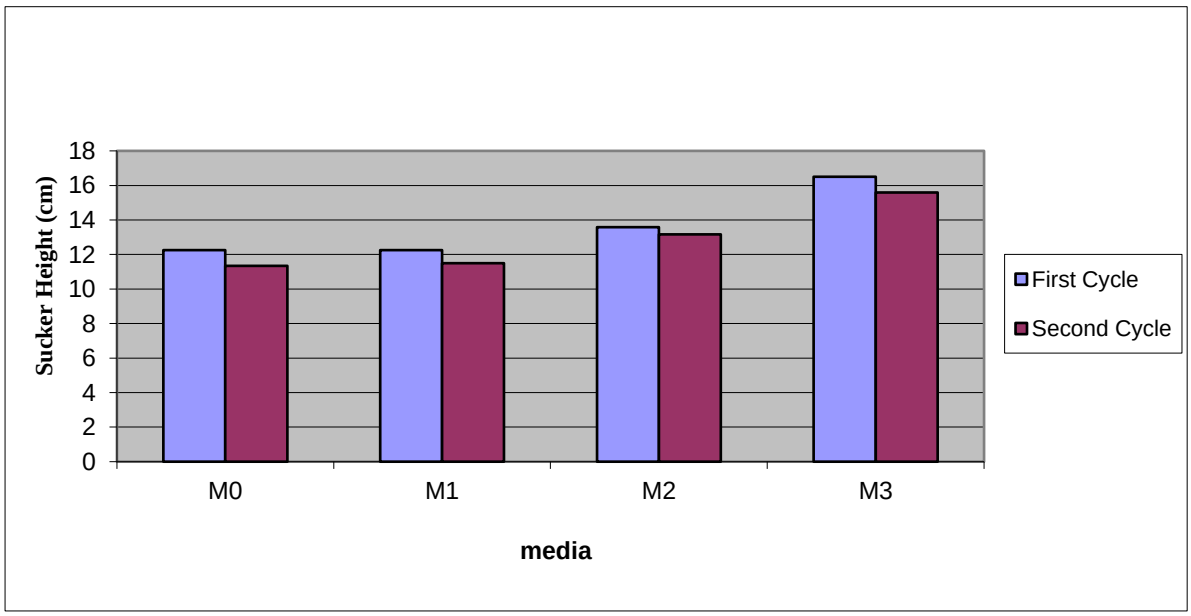
S_1=20 mg/ L Water

S_2= 30 mg/ L Water

S_3= 40 mg / L Water

4.3 Sucker height

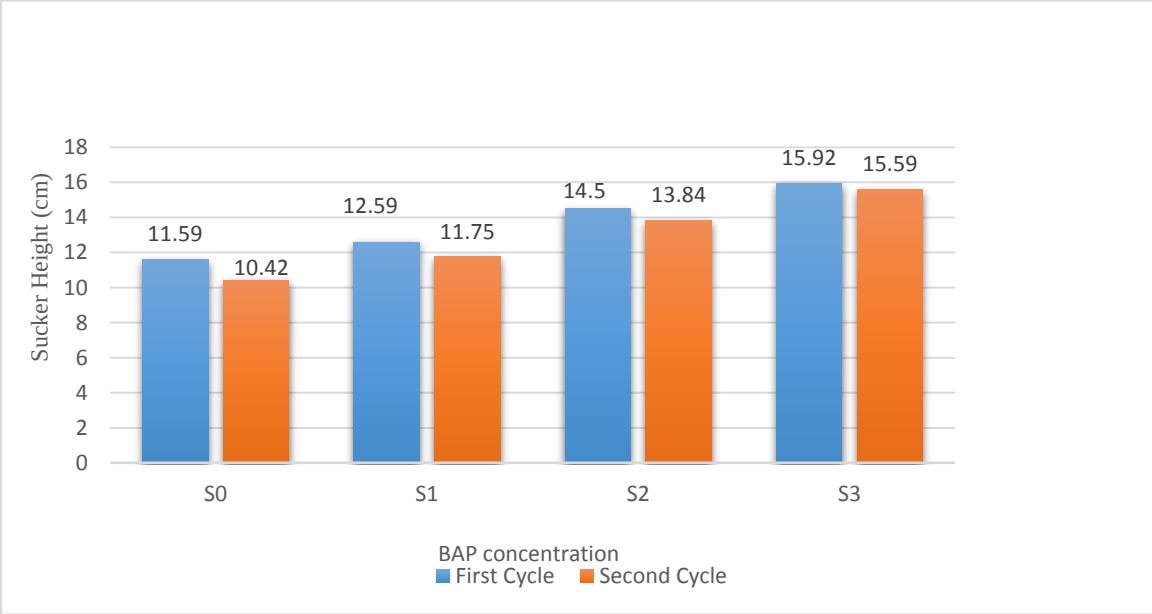
Application of different media shows no significant difference in sucker height (Figure 6). However tallest (16.50 cm) sucker was produced in cocodust (M_3) and smallest (12.25) sucker was produced in rice husk (M_1) and control (Mo) condition in first cycle. In addition to sawdust (M_2) media produced 13.59 cm sucker. When recorded in second cycle its tallest sucker was 15.59 cm and found in Cocodust (M_3) media and smallest was 11.34 cm and found in control (Mo) condition. Besides rice husk (M_1) and sawdust (M_2) produced 11.50 and 13.17 cm of sucker.



Growing media, M_0=Soil, M_1=Rice husk, M_2= Saw dust, M_3 = Coco dust

Figure 6. Effect of media on height of sucker

In case of BAP application, in first cycle tallest (15.92 cm) was recorded in S_3 and smallest (11.59) was recorded in control (So) condition where as in S_2 and S_1 height was 14.50 cm and 12.59 cm, respectively. On the other hand , in second cycle tallest (15.59 cm) and smallest (10.42 cm) sucker were found in S_3 and control (So) condition where as in S_1 and S_2 they are 11.75cm and 13.84 cm respectively (Figure 7).



BAP concentrations, S_0=0 mg/L (Control), S_1=20 mg/ L, S_2= 30 mg/ L, S_3= 40 mg / L

Figure 7. Effect of BAP concentrations on sucker height

In case of combined effect of media and BAP concentrations showed no significant difference on sucker height (Table 3). In first cycle tallest (20 cm) sucker was found in M_3S_3 combination and smallest (10.67) sucker was found MoS_1 and M_1So combination. Also first time collection in M_3S_2, M_2S_3, M_3S_1, M_2S_2, M_1S_3, MoS_3 produced 17.34 cm, 15.67 cm, 15.34 cm, 14.34 cm, 14cm, and 14cm height of suckers. In second cycle the result was found as tallest (19.34 cm) in M_3S_3 and smallest (9.67 cm) was found in MoSo and M1So as like as previous combination. In M_3S_2, M_2S_3, M_2S_2, M_1S_3 combination sucker heights were 17.34 cm, 16 cm, 14.34 cm and 14 cm in second cycle. Keeriol *et al.* (2018) found maximum shoot length in cultivar KM5 via shoot tip culture in BAP (0.03%). But Thungon (2017) found tallest sucker in cocodust growing media with BAP (0.04%), it supports this thesis findings.

Table 3. Combined effect of media and BAP on height of sucker (cm)

Combined (M x S) effect		Height of sucker (cm)	
		First cycle	Second cycle
M_0	S_0	11.00	9.67
	S_1	10.67	10.67
	S_2	13.34	12.00
	S_3	14.00	13.00
M_1	S_0	10.67	9.67
	S_1	11.34	10.67
	S_2	13.00	11.67
	S_3	14.00	14.00
M_2	S_0	11.34	10.34
	S_1	13.00	12.00
	S_2	14.34	14.34
	S_3	15.67	16.00
M_3	S_0	13.34	12.00
	S_1	15.34	13.67
	S_2	17.34	17.34
	S_3	20.00	19.34
Level of significant		3.1168	4.2293
CV%		7.51	10.78

NS = Non significant

Media ,

M_0=Soil

M_1=Rice husk

M_2= Saw dust

M_3 = Coco dust

BAP concentration ,

S_0=0 mg/L Water

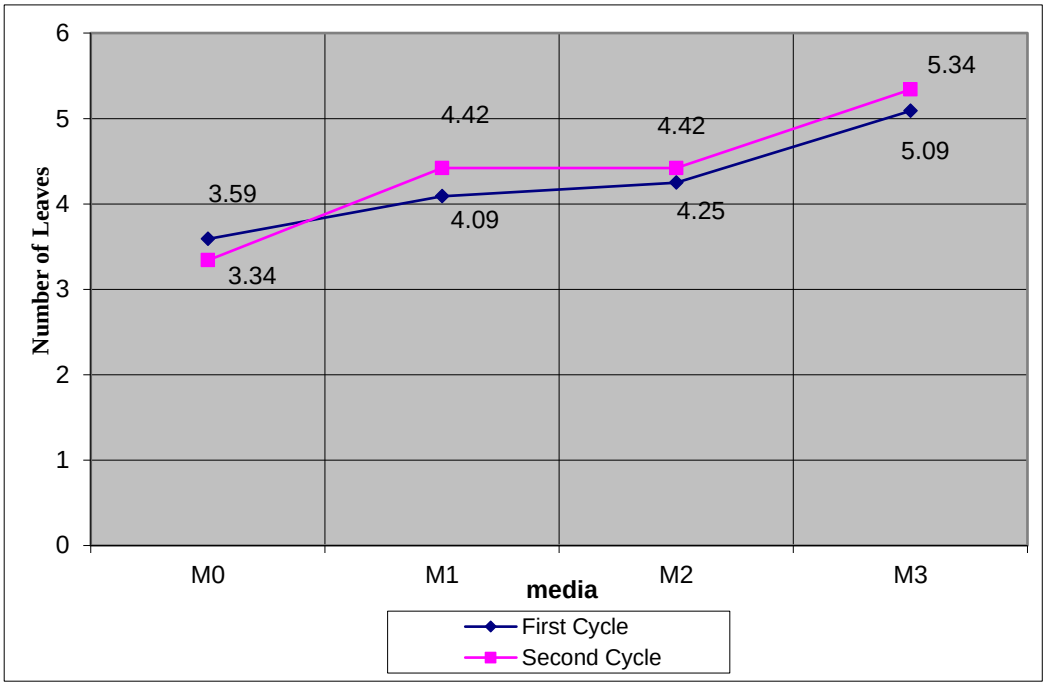
S_1=20 mg/ L Water

S_2= 30 mg/ L Water

S_3= 40 mg / L Water

4.4 Number of leaves in each sucker

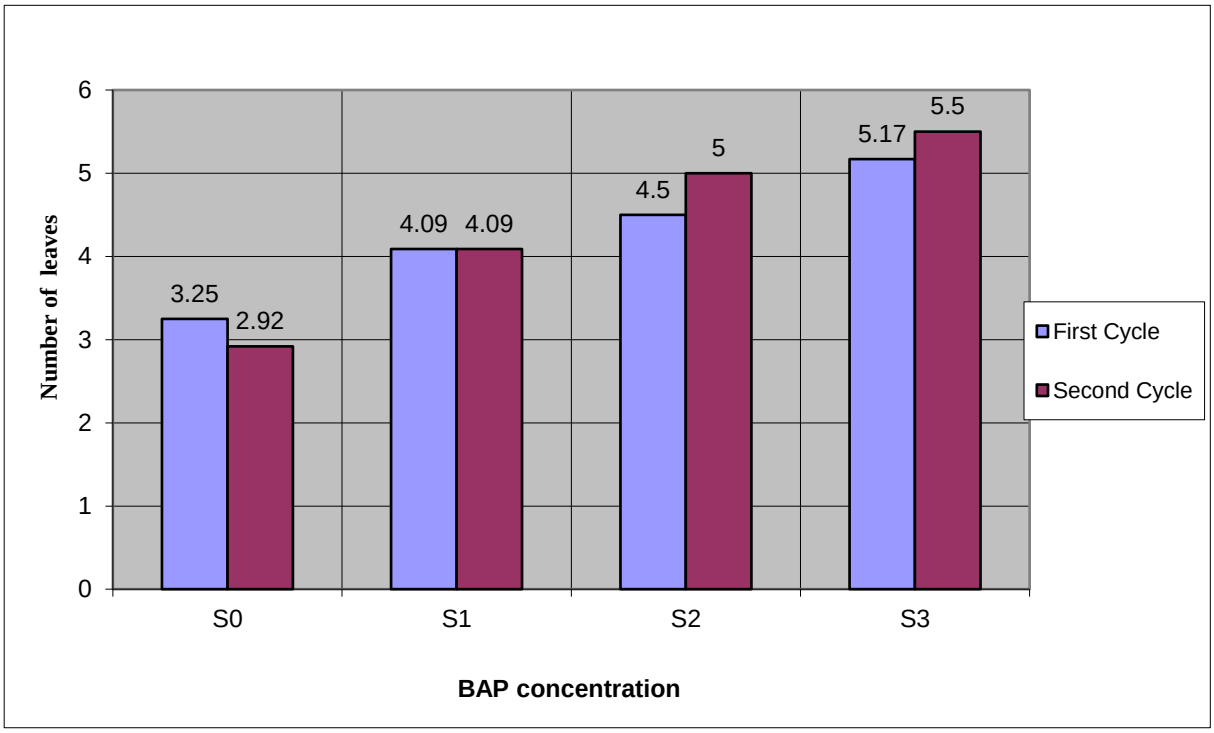
Due to application of different media and BAP concentrations there was no significance different in number of leaves per sucker (Figure 8) at all. However , in case of media cocodust (M_3) produced highest number leaves 5.09 and Control (Mo) condition produced lowest number of leaves 3.59 in first cycle where as rice husk and saw dust produced 4.09 and 4.25 leaves, respectively. But in second cycle highest (5.34)and lowest(3.34) number of leaves were in cocodust (M_3) and control(Mo) condition, respectively But in case of rice husk and saw dust leaves number is 4.42 only .



Growing media, M_0=Soil, M_1=Rice husk, M_2= Saw dust, M_3 = Coco dust

Figure 8. Effect of media on number of leaves production

The number of leaves in each produced sucker had no significant effect by BAP application (Figure 9). Due to use of BAP in different concentrations highest (5.09) leaves were found in cocodust (M_3) and lowest (3.25) were found in control (Mo) condition in first cycle and in rice husk and saw dust leaves were 4.09 and 4.50 . In case of second cycle maximum (5.50) number of leaves were from cocodust and minimum (2.92) number of leaves from control (Mo) condition. In rice husk and saw dust media leaves number were 4.09 and 5.0, respectively.



BAP concentrations, S_0=0 mg/L (Control), S_1=20 mg/ L, S_2= 30 mg/ L, S_3= 40 mg / L

Figure 9. Effect of BAP concentrations on number of leaves of banana

The combined effect of media and BAP concentrations in number of leaves produced in each sucker showed in (Table 4). It is showed that in first cycle highest number (6.34) of leaves recorded from M_3S_3 combination and lowest (2.67) number of leaves were found from MoSo combination. When we observe second cycle, maximum number of leaves were 6.67 and found from M_3S_3 combination but minimum number of leaves were 2.67 and found from MoSo combination.

Sajith *et al*. (2014) also stated that application of BAP with *Bacilus subtilis* produced higher number of primary, secondary and tertiary suckers leaves. Kindimba (2013) also found better growing media as coco dust and BAP (0.04%) reflects higher number of leaves in produced sucker.

Table 4. Combined effect of media and BAP concentrations on number of leaves in banana sucker

Interaction (M x S) effect of media and BAP		Number of leaves per sucker	
		First cycle	Second cycle
M_0	S_0	2.67	2.67
	S_1	3.34	3.00
	S_2	3.67	4.00
	S_3	4.67	3.67
M_1	S_0	3.34	3.00
	S_1	4.00	4.34
	S_2	4.34	4.67
	S_3	4.67	5.67
M_2	S_0	3.00	2.34
	S_1	4.00	3.67
	S_2	5.00	5.67
	S_3	5.00	6.00
M_3	S_0	4.00	3.67
	S_1	5.00	5.34
	S_2	5.00	5.67
	S_3	6.34	6.67
Tukey HSD (0.05)		1.5543	1.6947
CV%		12.02	12.78

NS=Non significant

Media ,

M_0=Soil

M_1=Rice husk

M_2= Saw dust

M_3 = Coco dust

BAP concentration ,

S_0=0 mg/L Water

S_1=20 mg/ L Water

S_2= 30 mg/ L Water

S_3= 40 mg / L Water

4.5 Sucker collar diameter

The sucker collar girth showed no significant difference due to different media use (Table 5). In first cycle sucker collar girth recorded 1.78 cm, 1.95 cm, 2.34 cm, 2.34 cm in Mo, M_1, M_2, M_3 media, respectively. In second cycle sucker collar girth was 2.04 cm, 2.17 cm, 2.50 cm, 2.55 cm in control (Mo), Rice husk (M_1), Saw dust (M_2), Coco dust (M_3), respectively.

Table 5. Effect of media on sucker collar diameter (cm)

Treatment (M)	Sucker collar diameter(cm)	
	First cycle	Second cycle
Media		
M_0	1.78	2.04
M_1	1.95	2.17
M_2	2.33	2.50
M_3	2.34	2.55
Tukey HSD (0.05)	0.0962	0.1953
CV%	4.12	7.60

Media, M_0=Soil, M_1=Rice husk, M_2= Saw dust, M_3 = Coco dust

Application of different BAP concentrations showed significant impact on sucker collar diameter (Table 6). In first cycle sucker collar diameter was the highest (2.42 cm) and the lowest (1.80 cm) was in S_3 (40 mg/L) and So (0 mg/L), respectively. In second cycle the highest (2.77 cm) and the lowest (1.90 cm) collar diameter was in S_3 (40 mg/L) and So (0 mg/L) concentrations.

Table 6. Effect of BAP concentration on sucker collar diameter

Treatment	Sucker collar diameter(cm)	
	First cycle	Second cycle
BAP concentrations		
S_0	1.80 d	1.90 d
S_1	1.98 c	2.20 c
S_2	2.22 b	2.40 b
S_3	2.42 a	2.77 a
Tukey HSD (0.05)	0.0962	0.1953
CV%	4.12	7.60

BAP concentration, S0=0 mg/L, S_1=20 mg/ L, S_2= 30 mg/L, S_3= 40 mg / L

The combined effect of media and BAP showed no significant difference in sucker diameter (Table 7 Appendix 9). In first cycle widest (2.90 cm) and least wide (1.54 cm) diameter was in M_3S_3 combination and MoSo combination respectively. Second (2.67cm) and third highest (2.47cm) sucker collar diameter were found in M_2S_3 and M_3S_2. But in second cycle it is slightly higher and collar diameter is highest (3.17 cm) and lowest (1.70 cm) in M_3S_3 and MoSo combination, respectively.

Table 7. Combined effect of media and BAP concentrations on sucker collar diameter of banana

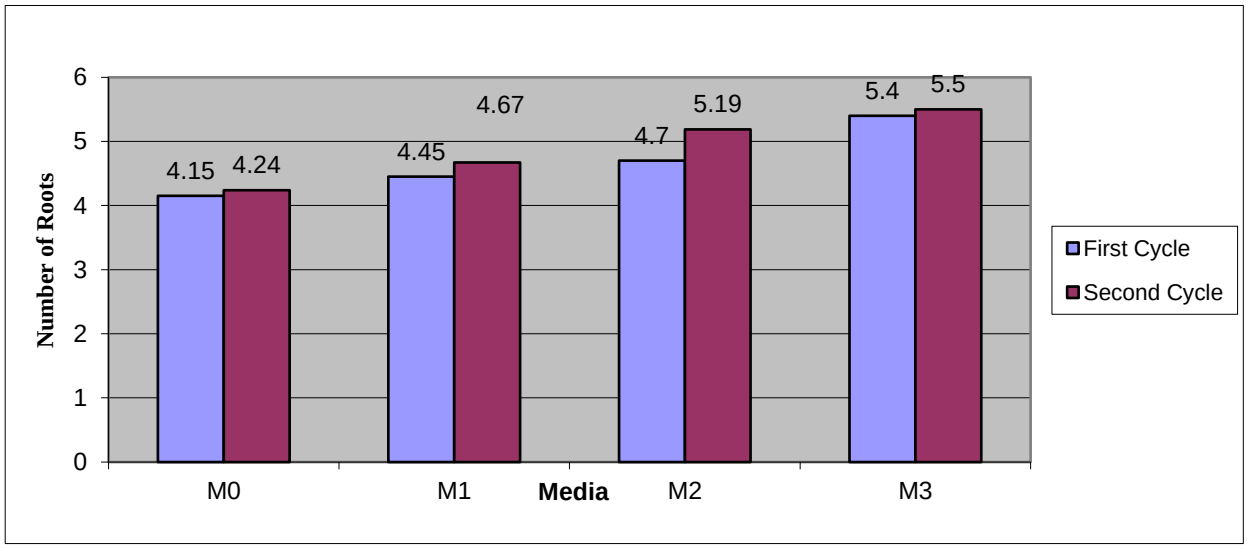
Interaction (M x S) of media and BAP		Sucker collar diameter (cm)	
		First cycle	Second cycle
M_0	S_0	1.54	1.70
	S_1	1.77	1.94
	S_2	1.90	2.00
	S_3	1.94	2.50
M_1	S_0	1.70	1.80
	S_1	1.84	2.14
	S_2	2.10	2.27
	S_3	2.17	2.47
M_2	S_0	2.10	2.17
	S_1	2.20	2.37
	S_2	2.40	2.57
	S_3	2.67	2.94
M_3	S_0	1.87	1.97
	S_1	2.10	2.34
	S_2	2.47	2.74
	S_3	2.90	3.17
Tukey HSD (0.05)		0.2637	0.5351
CV%		4.12	7.60

NS=Non significant

Media	BAP concentration
M0=Soil	H_0=0 mg/L Water
M1=Rice husk	H_1=20 mg/ L Water
M2= Saw dust	H_2= 30 mg/ L Water
M3 = Coco dust	H_3= 40 mg / L Water

4.6 Number of roots in banana sucker

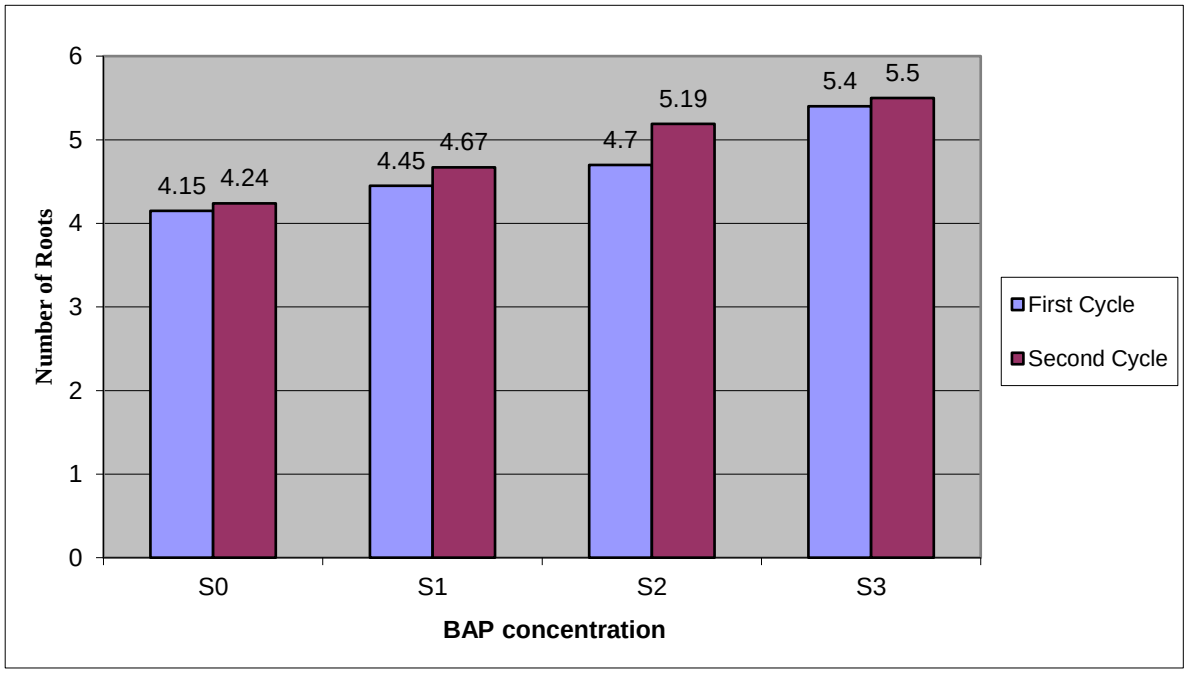
The number of roots showed significant difference in both different media and different BAP concentrations (Figure 10). The maximum roots were produced in cocodust (M_3) media and minimum was in control (Mo) Soil condition which was 5.40 and 4.15, respectively in first cycle. Furthermore in M_1 (rice husk) and M_2(saw dust) number of roots were 4.45 and 4.70 separately. But in second cycle highest number of roots were slightly increased 5.50 in as before in coco dust (M_3) media and lowest also slightly increased to 4.24 as before in controlled condition (Mo). Additionally in M_1 (rice husk) and M_2 (sawdust) media, 4.67 and 5.20 number of roots were found discretely. Thungon (2017) also found similar result of higher roots number in coco dust growing media and BAP(0.04%).



Growing media, M_0=Soil, M_1=Rice husk, M_2= Saw dust, M_3 = Coco dust

Figure 10. Effect of media on number of roots production

Application of different BAP concentrations showed significant variations on number of roots production. In first cycle highest (4.94) and lowest (4.42) roots were produced in S_3 (40 mg/L BAP) and So (0 mg/L BAP) concentrations in S_1 and S_2 concentration roots were 4.60 and 4.76 independently. On the other side in second cycle highest (5.27) and lowest (4.54) number of roots were produced in S_3 (40mg/L) and Control So (0 mg/L) condition (Figure 11), whereas in S_1 and S_2 concentration roots were 4.78 and 5.02, respectively. Thungon (2017) also found similar result of higher roots number in BAP (0.04%) application.



BAP concentrations, S_0=0 mg/L(Control),S_1=20 mg/ L,S_2= 30 mg/ L,S_3= 40mg /L

Figure 11. Effect of BAP concentrations on number of roots of banana sucker

The combined effect of media and BAP concentrations performed no significant variation on number of roots production (Table 8). The highest number of roots were counted in M_3S_3 combination and lowest was in MoSo combination and the number were 5.87 and 3.97 respectively in first cycle and second (5.54) and third (5.27) highest number of roots were found in M_3S_2 and M_3S_1 combination, respectively. But in second cycle highest and lowest number of roots were 5.97 and 3.94 in M_3S_3 and MoSo combination respectively. At the same time second (5.67) and third (5.37) highest number of roots were found in M_3S_2 and M_3S_1 combination, respectively. Thoungon (2017) found best combination of growing media as cocodust and BAP (0.04%) to produce higher number of roots.

Table 8. Combined effect of media (M) and BAP(S) concentrations on number of roots in banana

Interaction (M x S) effect of media and BAP		Number of roots per sucker	
		First cycle	Second cycle
M_0	S_0	3.97	3.94
	S_1	4.14	4.14
	S_2	4.24	4.40
	S_3	4.27	4.50
M_1	S_0	4.34	4.34
	S_1	4.40	4.57
	S_2	4.50	4.77
	S_3	4.57	5.00
M_2	S_0	4.40	4.87
	S_1	4.57	5.04
	S_2	4.77	5.27
	S_3	5.04	5.60
M_3	S_0	4.97	5.00
	S_1	5.27	5.37
	S_2	5.54	5.67
	S_3	5.87	5.97
Tukey HSD (0.05)		0.2786	0.5334
CV%		1.96	3.67

NS = Non significant

Media ,

M0=Soil

M1=Rice husk

M2= Saw dust

M3 = Coco dust

BAP concentration ,

H_0=0 mg/L Water

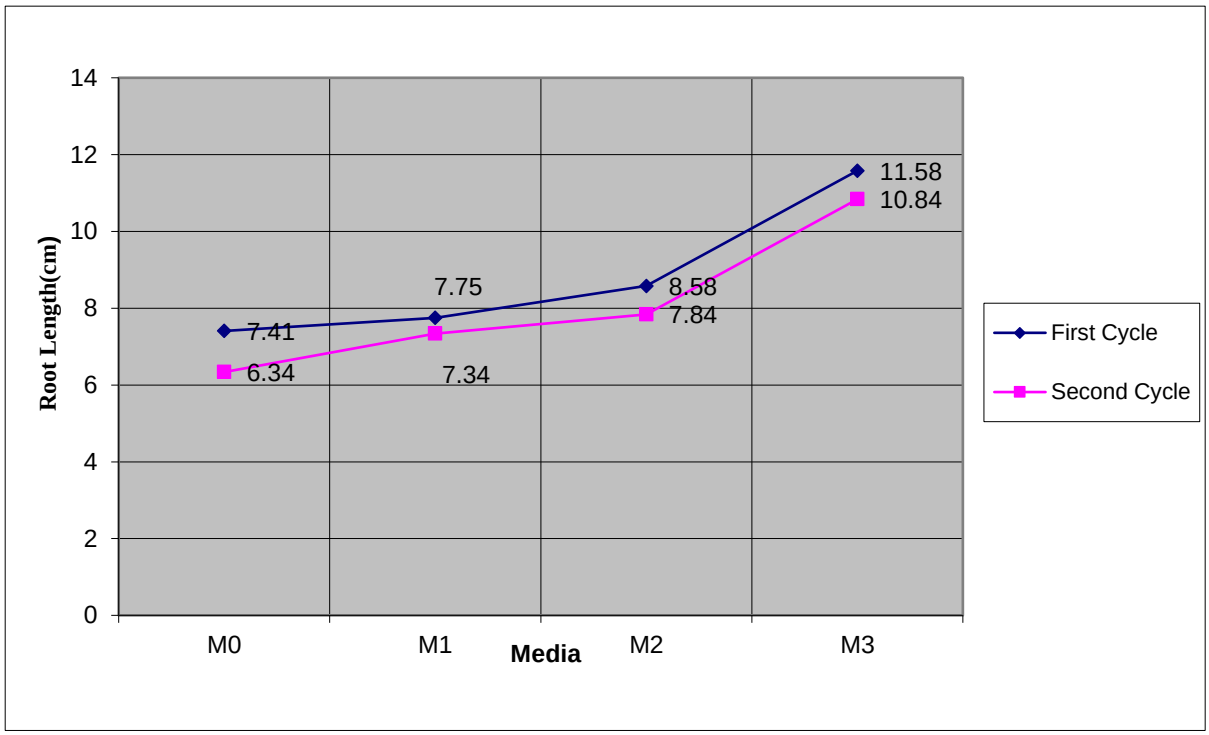
H_1=20 mg/ L Water

H_2= 30 mg/ L Water

H_3= 40 mg / L Water

4.7 Length of roots in banana sucker

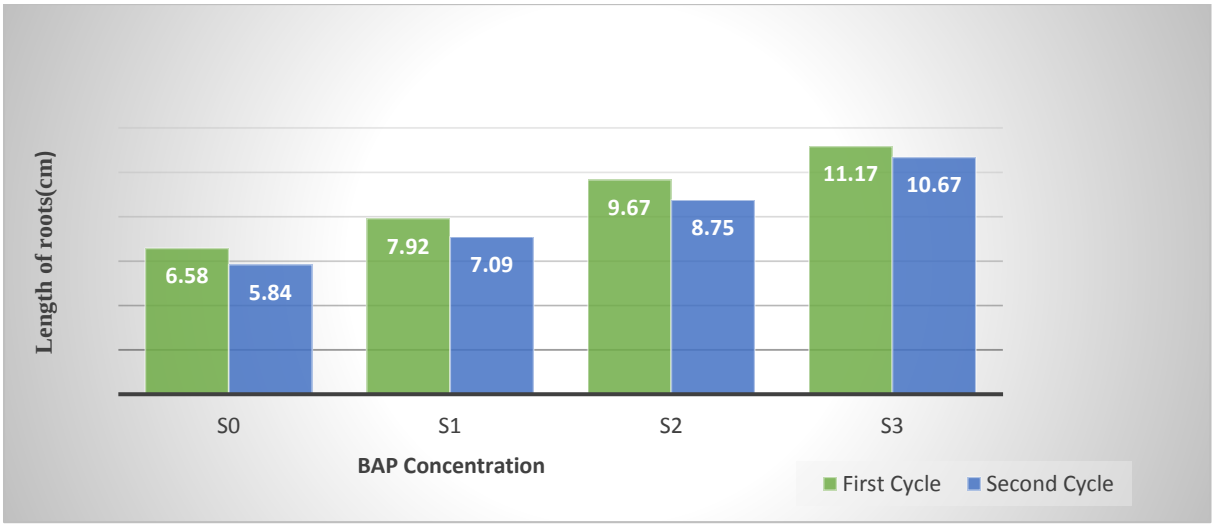
Application of different media shows no significant difference in root length (Figure 12). However the longest (11.60 cm) root was produced in cocodust (M_3) and smallest (7.41 cm) root was produced in control Mo (Soil) condition in first cycle. In the same cycle rice husk and saw dust produced 7.75 cm and 8.60 cm root length. When recorded in second cycle its longest root was 10.84 cm and found in coco dust (M_3) and smallest was 6.36 cm and found in control (Mo) condition. In rice husk (M_1) and sawdust (M_2) roots length were 7.34 cm and 7.84 cm, respectively.



Growing media, M_0=Soil, M_1=Rice husk, M_2= Saw dust, M_3 = Coco dust

Figure 12. Effect of media on roots length

In case of BAP application , in first cycle longest (11.17 cm) was recorded in S_3 and smallest (6.60 cm) was recorded in control (So) condition where as in S_1 (20 mg/L) and S_2 (BAP- 30 mg /L) height was 7.92 cm and 9.67 cm, respectively (Figure 13). On the other hand longest (10.67 cm) and smallest (5.84 cm) root was found in S_3 (40 mg/L) and control So (0 mg/L) condition in second cycle. Moreover in the same cycle, S_1 (20mg/L BAP) and S_2 (30mg/L BAP) produced 7.09 cm and 8.75 cm roots.



BAP concentrations, S_0=0 mg/L (Control), S_1=20 mg/ L, S_2= 30 mg/L,S_3= 40 mg /L

Figure 13. Effect of BAP concentrations on length of roots of banana sucker

In case of combined effect of media and BAP concentrations showed significant variation on root length (Table 9). In first cycle tallest (13.67 cm) root was found in M_3S_3 combination and smallest (5.00 cm) root was found MoSo combination. Roots length were 12.67 cm, 11.34 cm and 10.67 cm M_3S_2, M_2S_3 and M_1S_3 combination. In second cycle the result was found same combination as tallest (13.67 cm) in M_3S_3 and smallest (4.34 cm) was found in MoSo combination. But roots length were second (11.34 cm) and third (10.67 cm) highest in M_3S_2 and M_2S_3 combination. Thungon (2017) and Kindimba (2013) findings regarding macro propagation of banana support this results.

Table 9. Combined effect of media (M) and BAP (S) concentrations on length of roots (cm) of banana sucker

Combined (M x S) effect of media and BAP		Length of roots (cm) of banana sucker	
		First cycle	Second cycle
M_0	S_0	5.00	4.34
	S_1	6.67	5.34
	S_2	8.67	7.34
	S_3	9.34	8.34
M_1	S_0	6.00	5.34
	S_1	6.67	6.00
	S_2	8.00	8.00
	S_3	10.34	10.00
M_2	S_0	6.00	5.34
	S_1	7.67	7.00
	S_2	9.34	8.34
	S_3	11.34	10.67
M_3	S_0	9.34	8.34
	S_1	10.67	10.00
	S_2	12.67	11.34
	S_3	13.67	13.67
Tukey HSD (0.05)		2.2586	2.7928
CV%		9.12	11.36

BAP concentration ,

S_0=0 mg/L Water

S_1=20 mg/ L Water

S_2= 30 mg/ L Water

S_3= 40 mg / L Water

CHAPTER V

SUMMARY AND CONCLUSION

The experiment was conducted at the Horticulture Centre of Department of Agricultural Extension (DAE), Burirhat , Rangpur from February 2020 to May 2020 to study the growing media and BAP concentrations of macro propagation of banana, cv. Malbhog. The experiment comprised of two factors, Factor A: Different media i.e; Mo = Soil (Control), M_1= Rice husk, M_2= Sawdust, M_3=Cocodust; and four levels of BAP concentrations i.e; So= 0 mg/L, S_1= 20 mg/L, S_2=30 mg/L, S_3= 40 mg/L. The experiment was laid out RCBD with three replications. Data on different growth parameters were recorded and analyzed.

Number of days of first sucker emergence in shortest days required in M_3 (Cocodust) and longest time period was in control Mo (Soil) condition .The shortest and longest time (days) required for sprouting was recorded 24.17 days and 31.09 days respectively .On the other hand rice husk and saw dust required 28.58 days and 25.92 days respectively. The trend from shortest to longer period of media was soil, rice husk, saw dust and cocodust. Application of different levels of BAP also performed significant difference on first shoot emergence. However shortest days required for sucker emergence was in S_3 (BAP 40 mg/L) and longest time period required in control condition So (No BAP application). The shortest and longest time (days) required for sprouting bud was recorded 24.25 days and 31.09 days, respectively. For combine effect M_3S_3 combination resulted lowest spell (19.67 days) whereas MoSo combination took longest (34.34 days) period to sucker emergence

The produced number of suckers showed significant difference in both different media and different BAP concentrations. In Media Mo,M_1 , M_2,M_3 suckers were 3.50 , 5.17 , 6.17 and 7.25, respectively in first time sucker collection and 3.34, 5.34, 6.50 and 7.41 were in second time sucker collection. The maximum suckers were in cocodust (M_3) media and minimum were in control So (Soil) condition . Application of different BAP concentrations showed significant variations on number of sucker production. In first time 3.92, 5.0, 6.0, 7.18 suckers and in second time 3.58, 5.17, 6.34 and 7.50 suckers were produced in So, S_1, S_2, S_3 concentrations.

Highest (7.18) and lowest (3.92) suckers were produced in S_3 (40 mg/L BAP) and So (0 mg/L BAP) concentrations whereas in second time collection highest (7.50) and lowest (3.58) number of suckers were produced in S_3 (40 mg/L BAP) and control So(0 mg/L BAP) condition. The combined effect of media and BAP concentrations performed wide range of variation on number of sucker production. During first time collection the highest (9.34) number of suckers were recorded in M_3S_3 combination and lowest (2.67) were collected in MoSo combination . But in second time sucker collection highest (10.00) suckers were from M_3S_3 and lowest (2.34) suckers were collected from MoSo combination.

The sucker height ranges from 12.25 cm to 16.50 cm and 11.34 cm to 15.59 cm in first time and second time sucker collection, respectively. However tallest sucker was produced in coco dust (M_3) and smallest sucker was produced in control (Mo) condition in both time. In case of BAP application sucker height ranged from 11.59 cm to 15.92 cm during first time collection and 10.42 cm to 15.59 cm was recorded at second time sucker collection. In case of combined effect of media and BAP concentrations sucker height ranged from 10.67 cm to 20.00 cm. During first time collection tallest (20.00 cm) sucker was found in M_3S_3 combination and smallest (10.67 cm) sucker was found MoS_1 and M_1So combination. In second time collection the result was found in same combination as tallest (19.34 cm) in M_3S_3 combination and smallest (9.67 cm) was found in MoSo and M_1So combination.

The number of leaves per sucker in case of media cocodust (M_3) produced highest number leaves 5.09 and 5.34 and control (Mo) condition produced lowest number of leaves 3.59 and 3.34, respectively in first time and second time sucker collection. Firstly, in rice husk (M_1) and sawdust (M_2) media, leaves were 4.09 and 4.25, respectively. But in second time they were counted 4.42 leaves in both media. BAP application in S_3 and So concentrations produced highest 5.09 and 5.50 leaves and lowest 3.25 , 2.92 leaves during first time and second time sucker collection, respectively. If we consider combined, then M_3S_3 produced highest 6.34 leaves in first time and 6.67 leaves in second time collection. But lowest (2.67) number of leaves produced by MoSo combination in first time but during second time collection lowest (2.34) leaves were found in M_2So combination.

The sucker collar diameter showed no significant difference due to different media use. During first time collection sucker collar diameter was recorded 1.78 cm, 1.95 cm, 2.34 cm, 2.34 cm in Mo , M_1, M_2, M_3 media respectively. In second time collection sucker collar diameter was 2.04 cm, 2.17 cm, 2.50 cm, 2.55 cm in control Soil (Mo), rice husk(M_1), Saw dust (M_2), Coco dust (M_3), respectively. Though in media sucker collar diameter showed no significant difference but in BAP concentrations it showed significant difference. In case of BAP concentrations sucker collar diameter in first time collection was highest (2.42 cm) and lowest (1.80 cm) was in S_3 (BAP=40 mg/L) and So (BAP=o mg/L), respectively. In second time collection the highest (2.77 cm) and lowest (1.90 cm) collar diameter was in S_3 (BAP=40 mg/L) and So (BAP=0 mg/L) concentrations. The combined effect of media and BAP showed no significant difference in sucker diameter. When first time collection, widest (2.90 cm) and least wide (1.54 cm) diameter was in M_3S_3 combination and MoSo combination, respectively. But in second time collection, it is slightly higher and collar diameter is highest (3.17 cm) and lowest (1.70 cm) in M_3S_3 and MoSo combination, respectively.

In both media and BAP application, number of roots production showed significant difference. Due to different media used number of roots were produced from 4.15 to 5.40 in first time sucker collection while 4.24 to 5.50 in second time sucker collection. Highest number produced in coco dust (M_3) and lowest number produced in control soil (Mo) condition. At the same time in rice husk (M_1) and sawdust (M_2) roots were 4.45 and 4.70 during first time collection and 4.67 and 5.20 in second time sucker collection, respectively. But application of different BAP concentrations showed significant variations on number of roots production. In first time collection highest (4.94) and lowest (4.42) roots were produced in S_3 (40 mg/L BAP) and So (0mg/L BAP) concentrations. Whereas S_1 and S_2 produced 4.60 and 4.76 roots on average. On the other side when second time collection highest (5.27) and lowest (4.54) number of roots were also produced in S_3 (BAP= 40mg/L) and control So (BAP=0 mg/L) condition. The highest number of roots was counted in M_3S_3 combination and lowest was in MoSo combination and the number were 5.87 and 3.97, respectively in first time collection. But in second time collection highest and lowest number of roots were 5.97 and 3.94 in M_3S_3 and MoSo combination, respectively.

Roots length showed no significant difference in growing media. In first time sucker collection roots length ranged from 7.41 cm to 11.60 cm whereas in second time it ranged from 6.36 cm to 10.84 cm. In both time of sucker collection coco dust (M_3) produced highest roots and control soil (Mo) produced smallest roots. BAP concentrations showed significant difference in roots length. In first time sucker collection longest (11.17 cm) was recorded in S_3 (BAP=40 mg/L) and smallest (6.60 cm) was recorded in control So (BAP=0mg/L) condition. Whereas in S_1 (20 mg/L) and S_2 (30 mg /L) height was 7.92 cm and 9.67 cm, respectively. On the other hand during second time sucker collection longest (10.67 cm) and smallest (5.84 cm) root was found in S_3 (40 mg/L) and control So (0 mg/L) condition.

In case of combined effect of media and BAP concentrations showed no significant variation on root length. During first time sucker collection tallest (13.67 cm) root was found in M_3S_3 combination and smallest (5.00 cm) root was found MoSo combination. In second time sucker collection the result was found same combination as tallest (13.67 cm) in M_3S_3 and smallest (4.34 cm) was found in MoSo combination.

Recommendations

The present experiment was performed in only one season and in a single place. So, it is difficult to recommend this findings applicable in everywhere without further study. By considering the results of the present experiment further studies in the following areas are suggested below.

i. It was performed in open field where temperature, rainfall, humidity etcetera were not controlled. So, in future poly house shade or growth chamber may be used.
ii. Studies of similar nature could be carried out in different agro-ecological zones (AEZ's) in different seasons of Bangladesh for the evaluation of regional adaptability.
iii. In this study, four levels of BAP concentrations were used in cavity method. It is recommended to investigate corms soaking method in different BAP concentrations.

REFERENCES

Al-Amin, M., Karim , M.R., Amin, M.R., Rahman, S. and Mamun , A.N.M. (2009). In vitro micropropagation of banana(*Musa spp*.). *Bangladesh Journal of Agricultural Research*, **34**(4): 645-659.

Abad, M.,Noguera, P.,Puchades, R.,Maquieira , A. and Noguera, V.(2002). Physico-chemical and chemical properties of some coconut dusts for use as a peat substitute for containerized ornamental plants. *Bioresource Technology*, **82**: 241- 245.

Argo, W.R., 1998. Root medium physical properties. *Horticultural Technology*, **8**: 481- 485.

Awang , Y.,Shaharom, A.S., Mohammad , R.B. and Selamat, A.(2009). Chemical and Physical Characteristics of Cocopeat-Based Media Mixtures and Their Effects on the Growth and Development of Celosia cristata. *American Journal of Agricultural and Biological Sciences*, **4** (1): 63-71 .

Bunt, A.C., 1988. Media and Mixes for Container Grown Plants. 1st Edn. Springer,London.

Dagnew, A., Shibru, S., Debebe, A., Lemma, A., Dessalegn, L., Berhanu, B., Beyene, W., and Sierra, Y.M. (2012). Micro propagation of banana varieties (*Musa spp.)* using Shoot-tip culture. *Ethiopian Journal of Agricultural Science*. **22:** 14-25.

FAO (2019). Food and Agriculture Organization Production Year Book. Food Outlook.

Keeroi, I., Soomro, N.S.,Nizamani , G.S., Soomro, S.A., Khatri, A., Soomro, F.D.,Keerio, A.A., Sheikh, A.A., Manghwar, H., Rattar,Q.A. (2018).Effect of different phytohormones on micropropagation of banana (Musa sp.) cultivars and theirassessment through RAPD.*Bolan Society for Pure and Applied Biology*,**7**(3):1074-1084.

Kindimba, G. V. (2013). Effect of benzyl amino purine on *in vivo* multiplication and genetic stability of plantain (*musa* spp. Aab) cv. 'itoke sege'. MSc. Thesis, Sokoine University of Agriculture, Morogoro, Tanzania.

Kelta, A., Hajare, S.T. and Banjaw, A.(2018). Studies on in vitro micropropagation in banana. *International Journal of Current Microbiology and Applied Sciences.* **7**(7):3366-3375.

Lohidas , J. and Sujin, D.(2015). Effect of growth hormones in the micro propagation of banana cv. Matti. *Plant Archives,* **15**(1):307-314.

Muhammad, A., Hussain,I., Naqbi , S.M.S. and Rashid, H.(2004). Banana plantlet production through tissue culture. *Pakistan Journal of Botany*, **36**(3): 617-620.

Njau, N., Mwangi , M. and Gathu, R.K.(2010). Evaluation of the effectiveness of macro propagation method to produce healthy banana seedlings in Kenya. Proceedings of the 3rd International e-Conference on Agricultural Biosciences,Department of Agricultural Science and Technology, Kenyatta University, Nairobi,Kenya.

Njau, N., Mwangi, M., Gathu, R.,Mbaka, J. and Muasya, R. (2011). Potential challenges facing macropropagation technique in banana. Proceedings of the 3rd International e-Conference on Agricultural Biosciences, Department of Agricultural Science and Technology, Kenyatta University, Nairobi, Kenya.

Njau, N., Mwangi, M., Kahuthia-Gathu, R. and Muasya, R. (2012). Effectiveness of macro propagation technology in production of disease free banana seedlings incentral and eastern Kenya. . *Journal of Food Composition and Analysis*, **19**(2): 1327-1331.

Patel, M. K. and Rath, S. S. (2018). Standardization of Macro Propagation in Banana cultivars. *International Journal of Current Microbiology and Applied Sciences*, **7**(9): 390-400.

Ramirez, J.S., Batton, G.A. and Sacayanan, A.S. (2017). Production of saba banana using misting system and different growth enhancers under glasshouse condition. *European Journal of Advances in Engineering and Technology*. **4**(3): 189-193.

Rahman, S., Biswas, N., Hassan, M.M., Ahmed, M.G., Mamun, A., Islam, M.R., Moniruzzaman, M. and Haue, M.E. (2013). Micro propagation of banana (*musa* spp.) Cv. Agnishwar by *in vitro* shoot tip culture. *International Research Journal of Biotechnology*. **4**(4): 83-88.

Ramirez, J.S., Batoon, G.A. and Sacayanan, A.S. (2018). Macro propagation of saba banana using misting system and different plant growth enhancers. *MAYFEB Journal of Agricultural Science,* **4**:28-33.

Sajith, K. P., Uma, S., Saraswathi, M. S., Backiyarani, S. and Durai,p.(2014). Macropropagation of banana - Effect of bio-fertilizers and plant hormones. *Indian Journal of Horticulture,* **71**(3): 299-305.

Thungon, S. C., Hazarika, D. N., Langthasa, S., Goswami, R. K. and Kalita, M. K. (2017). Standardization of Growing Media for Macropropagation of Malbhog (AAB) Banana. *Journal of Food Composition and Analysis,* **12**(6): 270-277.

APPENDIX

Appendix I. Monthly recorded atmospheric temperature, rainfall, relative humidity and sunshine of the experimental site during the period from February, 2020 to July 2020.

Month	Air temperature(ºC)		Relative humidity(%)	Total rainfall(mm)	Sunshine(hr)
	Maximum	Minimum			
February, 2020	27	15	20	11	12
March, 2020	30	19	40	26	12
April, 2020	31	22	80	109	11
May, 2020	32	24	80	280	12
June, 2020	32	26	80	425	13
July, 2020	32	27	90	464	13

Source: Bangladesh Meteorological Department

Appendix II. Soil characteristics of Horticulture Centre, Department of Agricultural Extension (DAE), Burirhat, Rangpur , analyzed by Soil Resources Development Institute (SRDI), Dhaka.

A. Morphological characteristics

Morphological features	characteristics
Location	Horticulture center , Burirhat , Rangpur
AEZ	Active teesta flood plain (2)
Land type	High land
Flood level	Above flood level
Drainage	Well drained

B. Chemical analysis

Characteristics	Value
P^H	4.8-6.5
Organic matter(%)	1.55-1.82
Total N(%)	0.057-0.189
Available P(ppm)	4.21-92.55
Exchangeable K (me/100gm soil)	0.09 meq-0.40- meq
Available S (ppm)	0.64-68.61
Available Zn (ppm)	1.02-3.62
Available B (ppm)	0.001-1.40

Source: Soil Resources Development Institute

Appendix III. **Factorial ANOVA table for number of days of first sucker emergence**

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	21.375	10.688		
Media	3	331.396	110.465	99.54	0.0000
BAP	3	294.729	98.243	88.53	0.0000
Media*BAP	9	35.021	3.891	3.51	0.0045
Error	30	33.292	1.110		
Total	47	715.812			

Appendix IV. **Factorial ANOVA table for number of sucker (**first cycle)

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	0.542	0.2708		
Media	3	91.396	30.4653	90.27	0.0000
BAP	3	69.396	23.1319	68.54	0.0000
Media*BAP	9	6.521	0.7245	2.15	0.0565
Error	30	10.125	0.3375		
Total	47	177.979			

Appendix V. **Factorial ANOVA table for number of leaves sucker (**first cycle)

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	3.5000	1.75000		
Media	3	14.0000	4.66667	17.87	0.0000
BAP	3	23.1667	7.72222	29.57	0.0000
Media*BAP	9	2.5000	0.27778	1.06	0.4163
Error	30	7.8333	0.26111		
Total	47	51.0000			

Appendix VI. **Factorial ANOVA table for sucker height (**first cycle)

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	1.167	0.5833		
Media	3	144.562	48.1875	45.89	0.0000
BAP	3	135.229	45.0764	42.93	0.0000
Media*BAP	9	14.521	1.6134	1.54	0.1805
Error	30	31.500	1.0500		
Total	47	326.979			

Appendix VII. **Factorial ANOVA table for sucker collar diameter (**first cycle)

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	0.32792	0.16396		
Media	3	2.82729	0.94243	125.43	0.0000
BAP	3	2.63396	0.87799	116.85	0.0000
Media*BAP	9	0.49521	0.05502	7.32	0.0000
Error	30	0.22542	0.00751		
Total	47	6.50979			

Appendix VIII. **Factorial ANOVA table for number of roots per sucker (**first cycle)

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	0.1950	0.09750		
Media	3	10.3717	3.45722	412.12	0.0000
BAP	3	1.7683	0.58944	70.26	0.0000
Media*BAP	9	0.4833	0.05370	6.40	0.0000
Error	30	0.2517	0.00839		
Total	47	13.0700			

Appendix IX . **Factorial ANOVA table for length of roots (**first cycle)

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	2.542	1.2708		
Media	3	129.667	43.2222	66.64	0.0000
BAP	3	144.500	48.1667	74.26	0.0000
Media*BAP	9	4.500	0.5000	0.77	0.6437
Error	30	19.458	0.6486		
Total	47	300.667			

Appendix X. **Factorial ANOVA table for number of sucker (s**econd cycle)

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	3.292	1.6458		
Media	3	111.729	37.2431	98.22	0.0000
BAP	3	100.729	33.5764	88.55	0.0000
Media*BAP	9	11.854	1.3171	3.47	0.0048
Error	30	11.375	0.3792		
Total	47	238.979			

Appendix XI . **Factorial ANOVA table for number of leaves per sucker (second cycle)**

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	2.6250	1.3125		
Media	3	24.0833	8.0278	25.69	0.0000
BAP	3	46.4167	15.4722	49.51	0.0000
Media*BAP	9	8.7500	0.9722	3.11	0.0092
Error	30	9.3750	0.3125		
Total	47	91.2500			

Appendix XII. **Factorial ANOVA table for sucker height (second cycle)**

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	18.667	9.3333		
Media	3	140.229	46.7431	24.18	0.0000
BAP	3	186.729	62.2431	32.19	0.0000
Media*BAP	9	20.854	2.3171	1.20	0.3320
Error	30	58.000	1.9333		
Total	47	424.479			

Appendix XIII. **Factorial ANOVA table for sucker collar diameter (second cycle)**

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	0.73167	0.36583		
Media	3	2.32729	0.77576	25.07	0.0000
BAP	3	4.68563	1.56188	50.47	0.0000
Media*BAP	9	0.40688	0.04521	1.46	0.2076
Error	30	0.92833	0.03094		
Total	47	9.07979			

Appendix XIV. **Factorial ANOVA table for number of roots per sucker (second cycle)**

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	0.0037	0.00187		
Media	3	11.1950	3.73167	115.46	0.0000
BAP	3	3.6017	1.20056	37.15	0.0000
Media*BAP	9	0.1700	0.01889	0.58	0.7992
Error	30	0.9696	0.03232		
Total	47	15.9400			

Appendix XV. **Factorial ANOVA table for length of roots (second cycle)**

Source	**DF**	**SS**	**MS**	**F**	**P**
Replication	2	0.042	0.0208		
Media	3	135.000	45.0000	53.38	0.0000
BAP	3	158.167	52.7222	62.54	0.0000
Media*BAP	9	3.167	0.3519	0.42	0.9155
Error	30	25.292	0.8431		
Total	47	321.667			