

**GROWTH, SEX EXPRESSION AND FRUIT SETTING OF
BITTER GOURD AS INFLUENCED BY PGRs**

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**GROWTH, SEX EXPRESSION AND FRUIT SETTING OF
BITTER GOURD AS INFLUENCED BY PGRs**

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*This is to certify that thesis entitled, “**GROWTH, SEX EXPRESSION AND FRUIT SETTING OF BITTER GOURD AS INFLUENCED BY PGRs**” 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 in HORTICULTURE**, embodies the result of a piece of bona fide research work carried out by **MD. RAFIKUL ISLAM**, Registration No. **11-04610** 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 duly been acknowledged.*

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Dedicated To
*My Beloved Parents &
Respected Research
Supervisor*

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ABSTRACT

A field experiment was conducted to evaluate the effect of plant growth regulators for flowering, fruit setting and yield of bitter gourd during February to June 2016 at Sher-e-Bangla Agricultural University, Dhaka. The experiment was consisted of two factors. Factor A: Four levels of plant growth regulators: PGR₀- control, PGR₁- GA₃, PGR₂- NAA and PGR₃- MH and Factor B: Three levels of application stages: S₁-seed soaking, S₂- 4-leaf stage and S₃- flower budding stage, respectively. The experiment was laid out in Randomized Complete Block Design with four replications. Plant growth regulators and various application stages showed significant variations with most of the parameters. In case of plant growth regulators, the highest number of branch plant⁻¹ (5.83, 15.08 and 26.58) at 30 DAT, 60 DAT and harvest, highest number of female flower plant⁻¹ (25.95), highest number of fruit plant⁻¹ (22.37) and the maximum yield (21.50 t ha⁻¹) were recorded from PGR₃ and the lowest from PGR₀. For various application stages, the highest number of branch plant⁻¹ (6.43, 15.50, and 25.12) at 30 DAT, 60 DAT and harvest, number of female flower plant⁻¹ (24.77), number of fruits plant⁻¹ (22.50) and the maximum yield (21.77 t ha⁻¹) were recorded from S₂ and the lowest from S₁. For combined effect the highest number of branch plant⁻¹ (7.25, 17.00 and 28.00) at 30 DAT, 60 DAT and harvest, highest number of female flower plant⁻¹ (27.83), highest number of fruit plant⁻¹ (25.75) and the maximum yield (25.42 t ha⁻¹) were recorded from PGR₃S₂ and the lowest from PGR₀S₁. So the use of maleic hydrazide @ 100 ppm at 4-leaf stage would be the best option in growth, sex expression and yield of bitter gourd.

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LIST OF ACRONYMS

ACRONYMS	ELABORATIONS
AEZ	Agro-Ecological Zone
ANOVA	Analysis of variance
cm	Centimeter
CV	Coefficient of Variation
cv.	Cultivar (s)
DAT	Days after transplanting
d.f.	Degrees of freedom
DMRT	Duncan's Multiple Range Test
<i>et al.</i>	And others
FYM	Farmyard manure
g	Gram
LSD	Least Significant Difference
MOP	Muriate of Potash
ns	Non-significant
OM	Organic manure
PGR	Plant growth regulator
RH	Relative Humidity
SAU	Sher-e-Bangla Agricultural University
SE	Standard Error
TSP	Triple Super Phosphate
var.	Variety
Via	By way of
Viz.	Namely
WP	Wetable powder

CHAPTER I

INTRODUCTION

Bitter gourd (*Momordica charantia* L.) is a tendril bearing vine type herbaceous plant belongs to the cucurbitaceae family. Its distinct warty oblong fruit has high value in Bangladesh and widely cultivating throughout the country. Locally grown two types of bitter gourd called 'karala' and 'uchcheya' are cultivating during summer and winter respectively, and an average produced 54.54 thousand metric tons over 10.02 thousand hectares of land (BBS 2017). Fruits are highly nutritive and are relatively high in antibiotic, antimutagenic, antioxidant, antiviral, antidiabetic and immune enhancing properties (Grover and Yadav 2004). A compound called cucurbitacin present in the fruit which makes bitter and also reduce blood sugar levels (Shetty *et al.*, 2005). Like other cucurbits, maleness is one of the major problem in bitter gourd which significantly reduces the fruit yields. It may cause because of plant produce separate male and female flowers (Rashid, 2004). Flowering behavior may vary with cultivar, climatic conditions and cultural practices albeit this sex expression is a complex process and can be modified by environmentally and hormonal factors (Ghani *et al.*, 2013).

Plant growth regulators including both growth promoters and retardants are considered as the modifier of the plant growth usually by stimulating the part of natural growth regulatory system. Generally, they are used for enhancing flowering especially on sex expression and also, they enhance the source-sink relationship and stimulate the translocation of photo-assimilates thereby helping better fruit set (Ghani *et al.*, 2013). Gibberellic acid is an important growth regulator that may have many uses to modify the growth, yield and yield contributing characters of plant (Rafeekher *et al.*, 2002). Altering the sequence of flowering and sex ratio is the most important in sex modification of cucurbits. Maleic hydrazid can reduce the male and female flower ratio in lower (Kooner *et al.*, 2000). NAA also used for altering the sex ratio and sequence. NAA is an important growth regulator that can modify growth, sex ratio and yield-contributing characters in a plant (Shantappa *et al.*, 2007).

Different plant stages are the most prime consideration for PGR's application because of their sensitive receive. PGR's significantly enhance to early flowering and harvesting of fruit and also maximum fruit setting when applied at 2-leaf and flower initiation stage (Ghani *et al.*, 2013). Plant growth regulators had positive influence on vegetative, flowering, modification of sex expression and fruit traits in bitter gourd when sprayed twice at three leaf and tendril initiation stage (Nagamani *et al.*, 2015). So, PGR's have great potentialities to influence plant growth in terms of using in the suitable stage. Since very little information is available about real impact of growth regulators on bitter gourd at various stages.

In view of their wide spectrum effectiveness on every aspect of plant growth, the present experiment aimed that plant growth regulators might have a useful potentiality for increasing the yield with the following objectives:

1. To determine the sex ratio as affected by the PGR's.
2. To select the suitable PGR in terms of growth and yield of bitter gourd.
3. To find out the best stage of PGR's application regards to yield.

CHAPTER II

REVIEW OF LITERATURE

The role of plant growth regulators in various physiological and biological processes in plants is well known, which enables a rapid change in the phenotype of the plant. Growth regulators are known to affect seed germination, vegetative growth, sex expression, fruit setting (%), and yield. Various application stage of plant growth regulators helps the better expression of sex sequence, vegetative growth, fruit setting and yield attributes. There is a great deal of experimental evidence in the literature showing that endogenous growth substances are involved in many processes which lead to growth and development. Plants have also been shown to respond to exogenous application of plant growth regulators. Considering their role in plants, plant growth regulators have been designated as magic chemicals which bring about an unprecedented growth and help in removing and circumventing many of the barriers imposed genetically and environmentally. Crop yield is a complex heritable character influenced by many morphological and physiological characters of plant interacting with environment. An attempt has been made to present the impact of plant growth regulators on plant growth and development vis-à-vis physiological, biochemical and yield parameters. The literature on the use of growth regulators and stages in bitter melon is meager and hence the work on other closely related vegetable crops and also on other fruit crops and their effects on morphological, physiological, biochemical parameters and yield attributes are considered.

2. VEGETATIVE GROWTH CHARACTERS

2.1. Days to germination

Commonly PGRs improve seed germination capacity, increase biomass yield, confer resistance to diseases and adverse growth conditions, and produce yield earlier (Papadopoulos *et al.*, 2006).

The principle factors that influence seed dormancy include certain plant growth regulators (PGRs), and notably among them, the abscisic acid is involved in germination inhibition, while gibberellins participate in termination of seed dormancy (Halter *et al.*, 2005).

2.2. Plant height (cm)

Arun *et al.* (1982) revealed that the application of GA₃ @ 200 ppm resulted in maximum plant height followed by seed soaking with GA₃ @ 15 ppm in brinjal cv Pusa Purple long.

Experimented by Ram Asrey *et al.* (2001) the application of GA₃ at 500 ppm increased the length of the plant in muskmelon.

The application of GA₃ at 25 ppm and NAA at 50 ppm stimulated the elongation of main vine length in summer squash. Similarly, the application of GA₃ (25 ppm) at 2-4 true leaf stage resulted in the more vine length as compared to control in bottle gourd (Arora *et al.* 1985). It was noticed that the application of NAA at 2 and 4 true leaf stages increased the main vine length in watermelon cv. Sugar Baby (Shinde *et al.*, 1994).

2.3. Number of leaves plant⁻¹

Das and Swain (1977) reported that nitrogen and growth regulators increased leaf number as well as leaf area in pumpkin when the crop was sprayed with NAA (100 ppm), ethrel (200 ppm) and MH (200 ppm) at 10 and 20 days after planting.

Singh *et al.* (1991) reported that the foliar application of mixtalol (30 ml per 10 l) increased the number of leaves plant⁻¹ significantly in bottle gourd.

Seed soaking with 550 ppm GA₃ for 12 hrs increased the number of leaves plant⁻¹ in muskmelon (Ram Ashrey *et al.*, 2001).

2.4. Number of branch plant⁻¹

In bitter gourd, application of 75 ppm TIBA promoted an increase in the number of branches (Rahman *et al.*, 1992). The inhibited apical growth and increasing numbers of lateral branches may be associated with the polar transport of auxin that is the decisive force of apical dominance. Removal of the tip, the main auxin source, or inhibition of auxin transport leads to the outgrowth of axillary buds (Machakova *et al.*, 2008).

2.5. PHENOLOGICAL CHARACTERS

2.5.1. Days to first flowering

Application of MH at 150 mg L⁻¹ showed earliest appearance of first staminate and pistillate flowers; whereas, NAA at 50 mg L⁻¹ delayed the appearance of first staminate and pistillate flowers in cucurbitaceous crops (Arora *et al.*, 1985). The

application of NAA (50 ppm) produced the first male flower earlier (43 days) and was significantly superior to all other treatments in bitter gourd (Gedam *et al.*, 1998).

Arora *et al.* (1982) reported that the application of ethrel at 100 and 250 ppm was most effective in inducing early as well as increased number of female flowers than the male flowers in summer squash.

Dostogir *et al.* (2006) reported that the application of GA₃ at 85 ppm showed significant influence on days to first male flower (34.7) in bittergourd.

Mangal *et al.* (1981) reported that MH induced the female flowers much early at basal nodes than the number of male flowers in squash melon. Application of CCC at 250 ppm and 500 ppm recorded minimum number of days for the appearance of first female flower (48.4 to 49.5 days), which was about 13 days earlier to untreated control.

Pankaj *et al.* (2005) studied the effect of plant growth regulators in bottle gourd and recorded substantial variation in the number of days for first male and female flowers over control and the application of CCC at 200 ppm exhibited significantly lower values (50.94 days) for male flowers and 58.8 days for female flowers as against the control. Application of NAA at 50 ppm delayed the appearance of first male flower (48 days) than female flower (45.04 days) as compared to control in bitter gourd (Marbhal *et al.*, 2005).

Sidhu *et al.* (1981 & 1982) concluded that ethrel @ 100, 250 and 500 ppm induced the hermaphrodite flowers earlier at basal nodes than male flowers in muskmelon. Further they revealed that during both the seasons (summer & rainy), maximum number of days and nodes to first male and female flowers were recorded with the application of ethrel.

Sreeramulu (1987) found that ethrel 100 g L⁻¹ increased the number of pistillate flowers and also hastened the appearance of the female flower compared to the control in sponge gourd. It also delayed the appearance of the first staminate flower and also decreased the total number of male flowers.

The earliest (30.63 days) was obtained in control (The appearance of the first staminate flower is delayed and pistillate flower initiation is promoted by relatively low concentrations of GA₃ (Wang and Zeng, 1997; Akter and Rehman *et al.*, 2010).

Wang and Zeng (1996) reported that gibberellic acid at 25 to 100 ppm increased the number of female flowers up to 80 days.

2.5.2. Male and female flower ratio

Ahmad and Gupta (1981) found that the minimum ratio of male to female flower was reached at 1000 ppm of cycocel in case of smooth gourd and at 1500 ppm in bottle gourd and bitter gourd.

Negi *et al.* (2003) studied the effect of ethephon and row spacing on the growth and yield of bitter gourd. Treatments comprised: two ethephon levels (0 and 250 ppm) and three row spacing (1.0, 1.25 and 1.50 m). Ethephon (250 ppm) reduced the length of main vine and number of branches and delayed the appearance of the first male and female flowers.

Exogenous application of plant regulators can alter the sex ratio and sequence, if applied at 2 or 4 leaf stage, the critical stage at which the suppression or promotion of either sex is possible. Hence, modification of sex to desired direction has to be manipulated by exogenous application of plant regulators once, twice or even thrice, at different intervals (Devies, 1987).

The average ratio of staminate to pistillate flowers in monoecious lines throughout the flowering period is typically 50:1 (Rasco and Castillo, 1990), but ratios can vary dramatically 9:1 to 48:1 (Dey *et al.*, 2005).

It was reported that exogenous application of PGRs may shift the sex expression in cucurbits toward femaleness, increasing the number of pistillate flowers, number of fruits plant⁻¹, and individual fresh mass of fruit as well as yield (Mia *et al.*, 2014).

2.6. FRUIT CHARACTERISTICS AND YIELD COMPONENTS

Dostogir *et al.* (2006) stated that application of GA₃ at pre-flowering stage in bitter gourd plant significantly influenced flowering behavior and fruiting characteristics. They recorded highest fruit set plant⁻¹ (84.51%) in plants sprayed with GA₃ at 70 ppm while it was lowest (63.41) with GA₃ (20 ppm) in bitter gourd.

Hidayatullah *et al.* (2012) recorded maximum number of fruits in bottle gourd by exogenous application GA₃ @ 30 µmol.

Prabhu and Natarajan (2006) recorded maximum fruit length in Ivy gourd when GA₃ and NAA were applied @ 100 and 400 ppm respectively. Department of Vegetable Crops, Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore - 641 003, India.

Gedam *et al.* (1998) and Dostogir *et al.* (2006) that both GA₃ and NAA significantly increased fruit diameter in bitter gourd.

Yasuyoshi and Yoshiyuki (1995) opined that the application of NAA (150 ppm) at 2 and 4 true leaf stages increased the average fresh mass of fruit and also the combined effect of both hand pollination and cytokinin increased the fresh mass of fruit in watermelon. The foliar application of NAA (50 ppm) and boron (4 ppm) recorded an increase in fruit diameter and fresh mass of fruit in bitter gourd (Gedam *et al.*, 1998).

Arora *et al.* (1987) reported that MH at 150 mg L⁻¹ was most effective in producing the maximum fresh mass of fruit plant⁻¹ and ultimately the yield.

Foliar spray of ethephon (100- 500 mg L⁻¹), GA₃ (10 mg L⁻¹), MH (50 – 150 mg L⁻¹) and TIBA (25-50 mg L⁻¹) increased the yield in most of the cucurbits (Sonkar 2003; Jatoi *et al.*, 2010).

Hossain *et al.* (2006) recorded the maximum number of fruit plant⁻¹ when GA₃ (25 ppm) was applied in bitter gourd.

Marbhal *et al.*, (2005) that the fruit yield in bitter gourd was increased by the application of NAA @ 50 ppm as compared to control.

Plant growth regulators have significant effect on yield and fruit characteristics in cucurbitaceous crops (Akter and Rehman, 2010).

Rahman and Karim (1997) reported that fresh weight of fruit and yield plant⁻¹ were highest when a combination of 100 ppm NAA, 75 ppm TIBA and 50 ppm GA₃ was applied.

Sure *et al.* (2012) revealed that PGRs and planting method had significant effects on vegetative, flowering fruit and seed yield. They concluded that GA₃ @ 25 ppm in four

leaf stage at trellis method could be a suitable treatment for enhancing growth and yield of medicinal pumpkin

The foliar application of GA₃ (5, 10, 20 ppm) and MH (50, 100, 200 ppm) at 2, 4 and 6 leaf stages resulted in increase in fruit diameter of summer cucumber; whereas, GA₃ was inferior to MH (Rafeekar *et al.*, 2002). The foliar application of NAA at 50 ppm showed increase in fresh mass of fruit by 34 per cent, as compared to 100 ppm MH (19%) and 13% with 50 ppm ethephon (Marbhal *et al.*, 2005). The application of GA₃ at 40 ppm showed the maximum fruit diameter and fresh mass of fruit and it was lowest with GA₃ (85 ppm) in bitter gourd (Dostogir *et al.*, 2006).

The maximum average length (6.0 cm) and average diameter (5.7 cm) was observed in squash melon with the application of 20 ppm & 10 ppm triaccontanol (Mahajan and Sharma, 2000). Ram Asrey *et al.*, (2001) studied seed soaking with 400 ppm GA₃ solution for 12 hours and showed increase in fresh mass of fruit in muskmelon.

According to Jadav *et al.*, (2010) revealed that Ethrel @ 200 ppm was most effective in converting femaleness, producing more number of branches and increasing the yield in cucumber.

Al-Masoum and Al-Masri (1999) reported that Cucumber cv. Beit Alpha was grown in a greenhouse in 1996-97 and ethephon applied at 250 ppm, 350 ppm and 450 ppm at the seedling stage (2-4 true leaves). Ethephon induced femaleness (pistillate flowers) on the main stem that led to greater fruit production.

An investigation was done by (Mangal *et al.*, 1981) to study the influence of various chemicals (Ethrel, NAA, Cycocel, MH, PCPA, Ascorbic acid and Boron) on the growth, flowering and yield of bitter gourd was conducted. PCPA at 100 ppm improved plant growth significantly. The treatment of CCC at 250 and 500 ppm produced female flowers about 12 days earlier in comparison to control plant. Maximum fruit yield plant⁻¹ (3123g) was produced under Cycocel 250 ppm followed by Ascorbic acid 25 ppm and Cycocel 250 ppm

Arora *et al.* (1988) stated that in 2 season field trials with cv. Lagenaria cylindrical (*Lagenaria aegyptiaca* L.) PusaChikni, the plants were sprayed with 5 different growth regulators at the 2 and 4 true leaf stages. The total yield (2.39 kg plant⁻¹) was

the highest in plants treated with Ethrel (ethephon) at 100 ppm. The average control yield was 0.69 kg plant⁻¹.

Baruah and Das (1997) observed that plants sprayed with NAA at 25 ppm and MH at 50 ppm produced the best yields (5.48 and 4.86 kg plant⁻¹, respectively) in *Lagenaria siceraria* L. Yield decreased with late sowing dates from 5.49 to 2.62 kg plant⁻¹.

Gedam *et al.* (1998) reported that a significant increase in fruit yield plant⁻¹ and per ha was due to the application of NAA (50 ppm) as compared to other treatments in bitter gourd.

Saimbhi and Thakur (2006) applied single aqueous sprays of 2- chloroethyl-phosphonic acid (CEPA) -1 250, 500 and 1000 mg L⁻¹ ; TIBA 25, 50 -1 and 100 mg L⁻¹ ; and (2-chloroethyl) trimethyl-ammonium chloride (CCC) -1 250, 500 and 1000 mg L⁻¹ were applied to squash melon (*Citrullus vulgaris* Schrad. var. *fistulosus* Stocks.) at the 2-3 leaf stage. The CEPA decreased -1 while both TIBA @ 25 and 50 mg L⁻¹ and CCC 500 mg L⁻¹ increased the number of fruits plant-1 and the yield.

Saleh and Abdul (1980) conducted an experiment with GA₃ (25 and 50 ppm), which were applied 3 times in June to early July. They reported that GA₃ stimulated plant growth. It reduced the total number of flowers plant-1, but increased the total yield compared to the control. GA₃ also improved fruit quality.

Seed soaking with GA₃ (400 ppm) for 24 hours increased the number of fruits and yield in maskmelon (Ram Asrey *et al.*, 2001). Maximum number of fruits and yield per ha were observed in the order of ethrel > MH > NAA > GA₃ and optimum concentration was 100 ppm ethrel in summer cucumber (Rafeekar *et al.*, 2002).

Sidhu *et al.* (1981) reported that the foliar application of GA₃ (10 ppm) and NAA (100 ppm) at two and four true leaf stages increased the fruit yield per ha by increasing the average fresh mass of fruit plant⁻¹ in squash melon. The highest fruit yield per ha was recorded with the application of ethrel (500 ppm) in comparison to other treatments in muskmelon (Sidhu *et al.*, 1982).

The application of ethrel (400 ppm) in cucumber cv. Poinsettee was found to be superior with respect to yield with maximum number of fruits plant⁻¹ (12.65) and yield (25.83 t ha⁻¹) than Beigaum Local (Vadigeri *et al.*, 2001).

The application of MH (150 ppm) in summer squash significantly enhanced fruit yield followed by 25 ppm GA₃ (Arora *et al.*, 1982). Foliar spray of MH (150 ppm) at 2 and 4 true leaf stages at 7 days interval recorded highest total yield (376.3 qha⁻¹) by number and weight in bottle gourd (Arora *et al.*, 1985).

Tomar and Ramgiry (1997) conducted an experiment and found that plants treated with GA₃ showed significantly greater plant height, number of branches/plant, number of fruit plant⁻¹ and yield than untreated controls. GA₃ treatment at the seedling stage offered valuable scope for obtaining higher commercial tomato yields.

2.7. BIOCHEMICAL CHARACTERS

Apart from morphological and physiological alterations, growth regulators also influence various biochemical parameters, thereby bringing alterations in the quality characters in various crops. It was found that oblong fruited cultivars were rich in large amounts of total phenols, glycoalkaloid and crude protein (Bajaj *et al.*, 1979).

Foliar application of GA₃ to tomato increased the sugar content in fruits (Adhlakha and Verma 1984). While, Siddareddy (1988) noted that the foliar application of mixtalol (1-2 ppm) increased the contents of reducing, non-reducing, total sugars and protein content in potato tubers.

CHAPTER III

MATERIALS AND METHODS

3.1. Experimental site

The experiment was conducted during February-June 2016 in open field provision at Horticulture Farm of Sher-e-Bangla Agricultural University, Bangladesh in 24.09⁰N and 90.26⁰E longitude with an elevation of 8.20 m from sea level (Appendix i).

3.2. Characteristics of soil

The soil in the experimental field was classified in loam textural class. Soil samples of the experimental plot were collected from a depth of 5-30 cm for analyzed before the onset of the experiments and data recorded. Soil was analyzed in the Soil Resources Development Institute (SRDI), Soil Testing Laboratory, Khamarbari, Dhaka (Appendix ii).

3.3. Climate and weather

The experimental site was under the subtropical climate characterized by three distinct seasons; winter season from November to February and the pre-monsoon or hot season from March to April and the monsoon period from May to October. Details of the meteorological data during the experimentation were collected from the Bangladesh Meteorological Department, Agargaon, Dhaka and has been presented in (Appendix iii).

3.4. Agro-ecological region

The experimental field belongs to the agro-ecological region of the Madhapur Tract (AEZ-28). The landscape comprises level upland, closely or broadly dissected terraces associated with either shallow or broad, deep valleys.

3.5. Experimental details

3.5.1. Planting material

In this research work, seeds of “BARI Karala-1” a variety of bitter gourd were collected from the Horticulture Research Centre, Bangladesh Agriculture Research Institute, Gazipur and used as planting material. It is a summer variety, fruits are dark green and medium sized (17-18 cm long), number of fruits plant⁻¹ 35-40 where average fresh mass of fruit 100g and, yield 24-27 t ha⁻¹ (Azad *et al.*, 2017).

3.5.2. Treatment of the experiment

The experiment consisted of two factors *viz.*, four levels of plant growth regulator and three application stages.

Factor A: Four levels of plant growth regulator (PGR) were studied as follows:

- i) PGR₀: control @ 0 ppm
- ii) PGR₁: GA₃ @ 100 ppm
- iii) PGR₂: NAA @ 100 ppm
- iv) PGR₃: MH @ 100 ppm

Factor B: Three levels of application stages (S) where PGR_s were applied during

- i) S₁: Seed soaking stage
- ii) S₂: 4-leaf stage
- iii) S₃: flower budding stage

There were 12 (4 × 3) treatments combination such as PGR₀S₁, PGR₀S₂, PGR₀S₃, PGR₁S₁, PGR₁S₂, PGR₁S₃, PGR₂S₁, PGR₂S₂, PGR₂S₃, PGR₃S₁, PGR₃S₂ and PGR₃S₃.

3.5.3. Design and layout of the experiment

The two factors experiment were laid out in a Randomized Complete Block Design (RCBD) with four replications. The total area of the experimental plot was 1.5 m² with length 1.5 m and width 1 m. The total area was divided into four equal blocks. Each block was divided into 12 plots where 12 treatments combination were distributed randomly. There were 48-unit plots altogether in the experiment. The size of each plot was 1.5 m × 1 m and were raised up to 15 cm. The distance maintained between two blocks and two plots were 1 m and 0.5 m, respectively.

3.6. Cultural practices

3.6.1. Land, bed and pit preparation

The experimental plot was opened in the 2nd week of February 2017 with a power tiller and exposed to the sun for a week. Then the land was harrowed and cross-ploughed 3 times followed by laddering. Weeds and stubbles were removed. Pits of 45 x 45 x 40 cm sized were prepared with 1.5 m apart in a single row along the bed. Centre of the pits were kept 45 cm apart from the bottom side along their irrigation channel. 60 cm irrigation channel and 30 cm drainage channel were kept alternatively between the plots.

3.6.2. Application of manure and fertilizers

Inorganic fertilizers- N, P, K, S, Zn and B @ 120, 40, 35, 3.5, 3, 3.5 g pit⁻¹ and organic fertilizer @10 kg pit⁻¹ were used for the commercial production in the form of urea, triple superphosphate, muriate of potash, gypsum, zinc sulphate, boric acid and cow dung, respectively (Chowdhury & Hassan, 2013). All doses of fertilizers were applied ten days prior to transplant except N. On the other hand, N was applied in 6 installments at 12 days interval starting from transplanting.

3.6.3. Seed treatment

Seeds were soaked as per treatments for 15 hours over night for easy germination. Seeds were treated with vitavax @ 2 g kg⁻¹ seeds before sowing to avoid seed borne diseases and get vigorous seedlings.

3.6.4. Raising the seedlings and transplanting

Seeds were sown in 6 x 8 cm polybags on 21 February 2017 having the growing media was prepared by mixing well decomposed compost and soil in a 50:50 ratio. Two seeds were sown in each polybag. The polybags were kept in shady place and they were watered regularly for profound germination during their raising period. Seedlings of 20 days old when they attained 4 leaves and hard enough, were transplanted in the prepared pit on 2nd week of March, 2017. Seedlings were watered immediately after transplanting and continued every afternoon till seedling establishment. Among the two seedlings in any pit, comparatively vigorous one was finally allowed to grow in the field for crop production.

3.7. Intercultural operations

After transplanting of seedlings in the well prepared main field various intercultural operations were furnished for proper growth and development of the crop.

3.7.1. Gap filling

Dead, injured and weak seedlings were replaced by new vigor seedling from the same stock of the experiment.

3.7.2. Weeding

The field was kept free from weeds during the crop period. Hand hoeing was performed to keep the plots free from weeds whenever it was necessary.

3.7.3. Irrigation

Bitter gourd plant was irrigated whenever it was necessary during vegetative, flowering and fruit setting stage.

3.7.4. Vine management and trellis preparation

Stormy weather may cause the tendering vine of the plants fell down from the supports (Trellis). For proper growth and development of the plants the vines were managed upward with the help of supports. Bamboo poles were set slantingly keeping 5 feet high from the ground level in every plot. The poles were connected to one another tightly by iron rope in such a way that they make opposite “V” shaped. Nylon nets were placed on the supports. Thus, a trellis for each plot was made for creeping the vines of bitter gourd.

3.7.5. Plant protection

There was a plan to protect the plant from the attack of insects-pests specially fruit flies and fruit borer by spraying of insecticides. There was also fungicide applied in the crop field during the experimental period to avoid any sort of diseases.

3.7.6. General observation

The field was frequently observed to notice any changes in plants, pest and disease attack and necessary action was taken for normal plant growth.

3.8. Harvesting

Bitter gourd fruits were harvested at green edible stage when fruits looking shiny, bright and standard size but not over matured.

3.9. Data collection

Data were collected on days to germination, plant height, branch (primary and secondary) number, days to first flowering, number of male and female flower ratio, sex ratio, fruit setting%, length and diameter of fruit, single fresh mass of fruit, fresh and dry matter content of 100g fruit, number of fruit plant⁻¹, fruit yield t ha⁻¹, fresh mass of plant, root length, biochemical characters (reducing sugar, non reducing sugar, total sugar, total phenols). Water content in fruit was simply recorded by the difference in mass of fresh and dry fruit and calculated as percentage.

3.9.1. Days to germination

Days to germination was recorded on the basis of first emergence of seedlings in poly bags out of total seeds sown per bag.

3.9.2. Plant height

Plant height was measured in centimeter (cm) from the ground level to the tip of the growing point. It was recorded at 30 DAT, 60 DAT and during harvesting of fruits for all treatments.

3.9.3. Number of leaves plant⁻¹

Number of leaves was counted each of plant in every plot by the manual way through vegetative stage to last harvest stage.

3.9.4. Number of branches plant⁻¹

Number of branches were counted by including primary and secondary branches plant⁻¹ through vegetative stage to last harvest stage.

3.9.5. Days to first flowering

Days after transplanting, total time required to first flowering was recorded for every plant and the average was calculated.

3.9.6. Number of male and female flowers

Total number of male and female flowers were counted regularly after first flowering to ensure last flowers.

3.9.7. Sex ratio (male : female)

Ratio of male and female flower was counted by following the formula:

Sex ratio: Number of male flower/ number of female flower.

3.9.8. Fruit setting (%)

The total number of fruits and number of female flowers produced plant⁻¹ was recorded and fruit setting (%) was calculated using following formula:

Fruit setting (%) = (Number of fruits plant⁻¹ / number of female flowers plant⁻¹) x 100

3.9.9. Length and diameter of fruit

The length of 10 randomly selected fruits per plot was measured after each harvest and then the average was taken. Diameter of the same 10 randomly selected fruits as harvested was measured and the average was calculated in cm.

3.9.10. Fresh mass of fruit⁻¹ (g)

After each harvest, the weight of randomly selected 10 fruits per plant was recorded and then the average weight per fruit was calculated in (g).

3.9.11. Dry matter content of fruit (%)

After harvesting, randomly selected 100 gram of fruit sample previously sliced in to very thin pieces. The fruits were then dried in the sun for one day and placed in oven maintaining at 70^oc for 72 hrs. The sample was then transferred into desiccators and allowed to cool down to the room temperature. The final weight of the sample was taken.

3.9.12. Number of fruits plant⁻¹

The number of fruits in every plant of bitter gourd was counted at every harvest and thus the total number of fruits plant⁻¹ was recorded and average number of fruits was calculated.

3.9.13. Fresh mass of plant (g)

Fresh mass of plant was taken by using a balance from the plant which was uprooted for taking data from each plot and data recorded in gram (g). It was calculated from summation of leaves and stem excluding roots weights. Data was taken at harvest.

3.9.14. Root length at harvest (cm)

Root length at harvest was taken by using a scale from the plant which was uprooted for taking data from each plot and data recorded in cm.

3.9.15. Fruit yield ha⁻¹

To estimate yield, all the six plants in every plot and all the fruits in every harvest were considered. Thus, the average yield per plot was measured. The yield ha⁻¹ was calculated considering the area covered by the six plants.

The gross yield of fruits per hectare was calculated from the m⁻² yield data and was recorded in tones.

3.10. BIOCHEMICAL COMPOSITIONS

All biochemical properties was determined at applied nutrient research section, Institute of Food Science and Technology (IFST), Bangladesh Council of Scientific and Industrial Research (BCSIR), Dhaka-1207, Bangladesh.

3.10.1. Estimation of reducing sugars by Nelson method (1941)

Reducing sugar content in freshly harvested fruit samples was estimated at the time of harvest.

3.10.1.1. Sample extraction

1. Leaves were cut in small pieces and 1.0 g of fresh plant sample was weighed and immersed in 10 ml of boiling ethanol, allowed to boil for 5-10 minutes on a steam bath.
2. The contents were cooled and the tissue was crushed thoroughly in a pestle and mortar and filtered through cheese cloth.
3. Repeated the extraction procedure to ensure the complete removal of alcohol soluble substances.
4. Pooled both the extracts and filtered through Whatman No. 41 filter paper. The volume of the extract was reduced by evaporating on hot water bath to represent 5-10 ml of the extract for every gram of tissue.
5. The extract was dried on hot water bath to remove the traces of alcohol; the volume was made up to 10 ml with distilled water and this was used for estimating sugar content.

3.10.1.2. Reagents

Alkaline copper reagent

Solution A: Dissolved 2.5 g of anhydrous sodium carbonate, 2.0 g of sodium bicarbonate, 2.5 g of potassium sodium tartrate and 20 g of anhydrous sodium sulphate in 80 ml water and made up the volume to 100 ml.

Solution B: Dissolved 15 g of copper sulphate in a small volume of distilled water, added one drop of Sulphuric acid and the volume was made up to 100 ml. Mixed 96.0 ml of solution A and 4.0 ml of solution B just before use.

Arsenomolybdate reagent: Dissolved 2.5 g of ammonium molybdate in 4.0 ml of water, to which 2.5 ml of sulphuric acid was added and mixed well. Then, 0.3 g of disodium hydrogen arsenate was added.

3.10.1.3. Procedure

Pipetted out 0.4 ml of sample aliquot in a test tube. Similarly, pipetted out 0.2, 0.4, 0.6, 0.8 and 1.0 ml of working standard solutions into different test tubes. Made up the volume to 1.0 ml with distilled water. Blank was maintained taking 1.0 ml of

distilled water in a separate test tube. Added 1.0 ml of alkaline copper reagent to each tube and placed them on a boiling water bath for 20 minutes. Then cooled the tubes under running tap and added 1.0 ml of arsenomolybdate reagent to each tube with a regular stirring to get blue color. The volume was made up to 10 ml with distilled water and the absorbance was measured at 510 nm in spectrophotometer. Dissolved 100 mg of glucose in little quantity of water and made up the volume to 100 ml to get a stock solution. Diluted 10 ml of stock solution to 100 ml with distilled water to get a working standard of 100 mg/ml concentration. The other procedure followed was similar to that used for plant samples.

3.10.2. Estimation of total sugars by enthrone method

Total sugar content was estimated in fresh fruit samples at the time of harvest.

3.10.2.1. Enthrone reagent

Dissolved 0.2 g of enthrone in 100 ml of concentrated sulphuric acid. Fresh solution was prepared just before use.

3.10.2.2. Procedure

One ml of the aliquot was taken in a test tube. The volume was made up to 2.5 ml with distilled water. All the test tubes were kept in the ice bath and to which, 5.0 ml of enthrone reagent was added slowly. Contents were stirred gently with a glass rod and heated on boiling water bath exactly for 7.5 minutes and cooled immediately on ice bath. After cooling, the absorbance of the solutions was measured at 630 nm against the blank in a spectrophotometer and the sugar content was calculated from the standard curve.

3.10.3. Non-reducing sugars

Non-reducing sugars was estimated by subtracting the reducing sugar from total sugar content of the sample.

3.10.4. Estimation of total phenols

Estimation of total phenols present in plant samples was done by following Folic Cocteau Reagent method (Saadian and Manikam, 1992).

3.10.4.1. Reagent

1. Folic – Cocteau Reagent (FCR) 1% 1:1 of (FCR + Wd)
2. Sodium carbonate (2%)

3.10.4.2. Procedure

One ml of the alcohol extract was taken in a test tube, to which one ml of Folic – Cocteau reagent followed by 2.0ml of sodium carbonate solution (2%) were added. The tubes were shaken well and heated on a boiling water bath for exactly one minute and then cooled under running tap water. The blue color developed was diluted to 25ml with distilled water and its absorbance was read at 650 nm in a Spectrophotometer. The amount of phenols present in the sample was calculated from a standard curve prepared from catechol.

3.10.5 Water content (%)

3.11. Statistical analysis

Analysis of variance was performed in order to evaluate the significance of the effect of plant growth regulators and their application stage in bitter melon for seed germination, plant growth, sex expression, fruit setting%, no. of fruit plant⁻¹, fruit yield t ha⁻¹, dry matter content of 100g fruit (%), fresh mass of plant, fresh weight of root, dry weight of root and chemical parameters. LSD test was used to determine variances among the treatments where $P < 0.05$ considered as significant.

CHAPTER IV

RESULTS AND DISCUSSION

A field experiment was conducted during February-June 2016 to evaluate the effect of different plant growth regulators *viz.*, GA₃, NAA and MH applied at three stages of seed soaking, 4-leaf stage and flower budding stage with the variety of bitter gourd 'BARI Karala-1' on growth, flowering and yield. The results obtained from the investigation are presented in this chapter.

4.1. Days to germination

The required days for germination was found non-significant in terms of plant growth regulators (Figure 1 and appendix iv). Thereafter, the maximum (6.92) days was required in germinating the seedlings from untreated seeds which was statistically analogous to others. The similar results regarding germination were found by all plant growth regulators because of they might have same activity for short duration. Although MH was needed more time than others due to its inhibiting function on cell division. This results also a good agreement with the findings of (Banker, (1987); Stino *et al.*, (1996); Pawshe *et al.*, (1997); Ratan and Reddy, (2004) they reported that GA₃ from 150-500 ppm is helpful for getting better germination of custard apple seeds.

There was also no significant variation in germination with all stages. Among the stages all the treatments, the maximum (6.93) days to germination was recorded at seed soaking stage (Figure 2 and appendix iv).

Although, plant growth regulators were applied at three stages of plant thereafter they showed non-significant effect on germination. The maximum days (7.50) to germination was recorded in the treatment combination of (PGR₀S₁) and the minimum days (6.00) to germination was recorded from the treatment combination of PGR₀S₂ (Figure 3 and appendix iv).

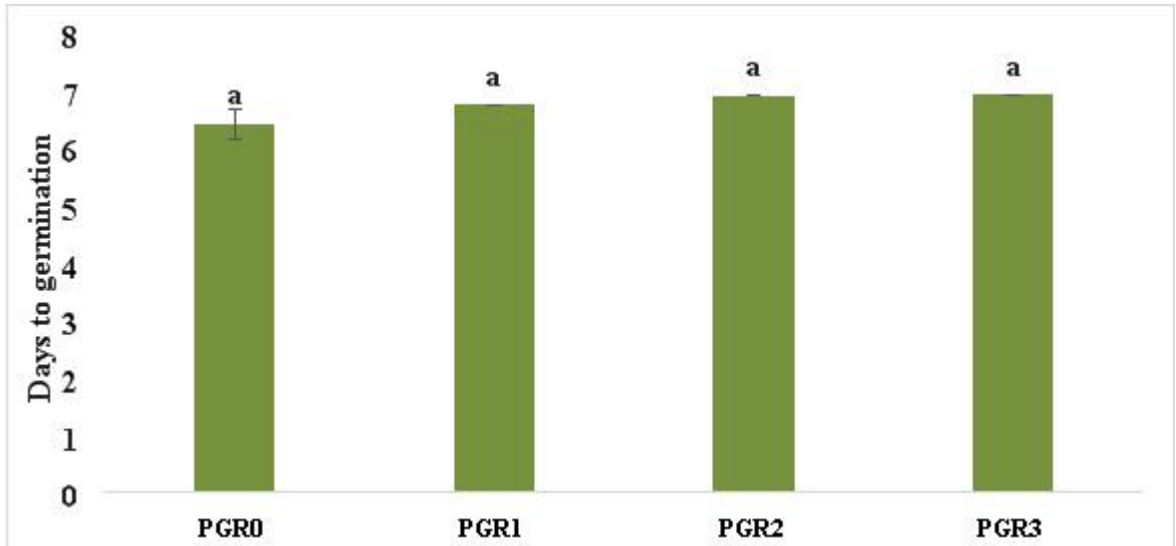


Figure 1. Required days to germination of bitter gourd as influenced by various plant growth regulators. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively.

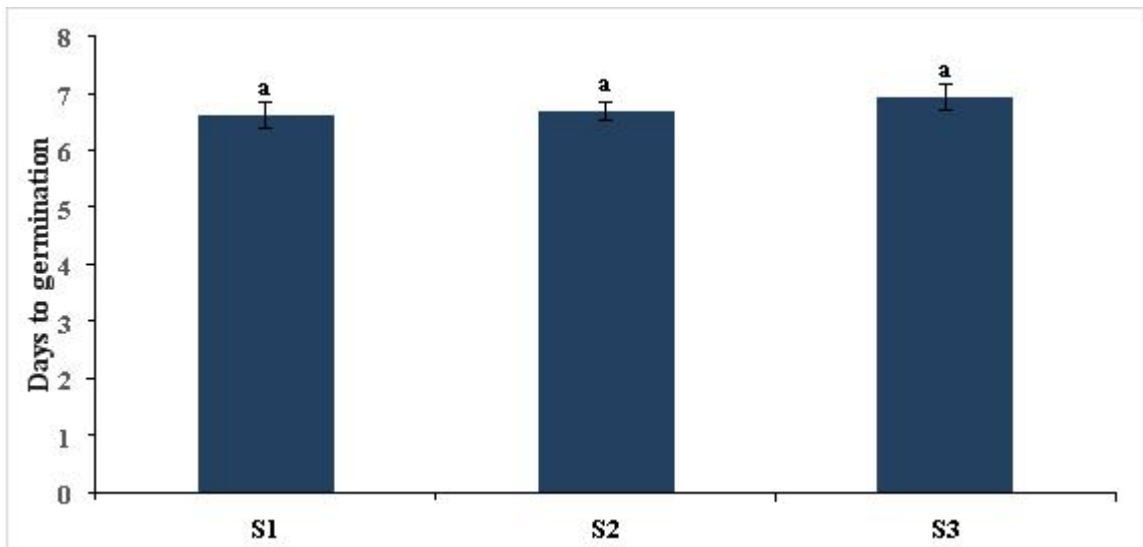


Figure 2. Effect of application stage of plant growth regulators on days to germination of bitter gourd. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; Seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage.

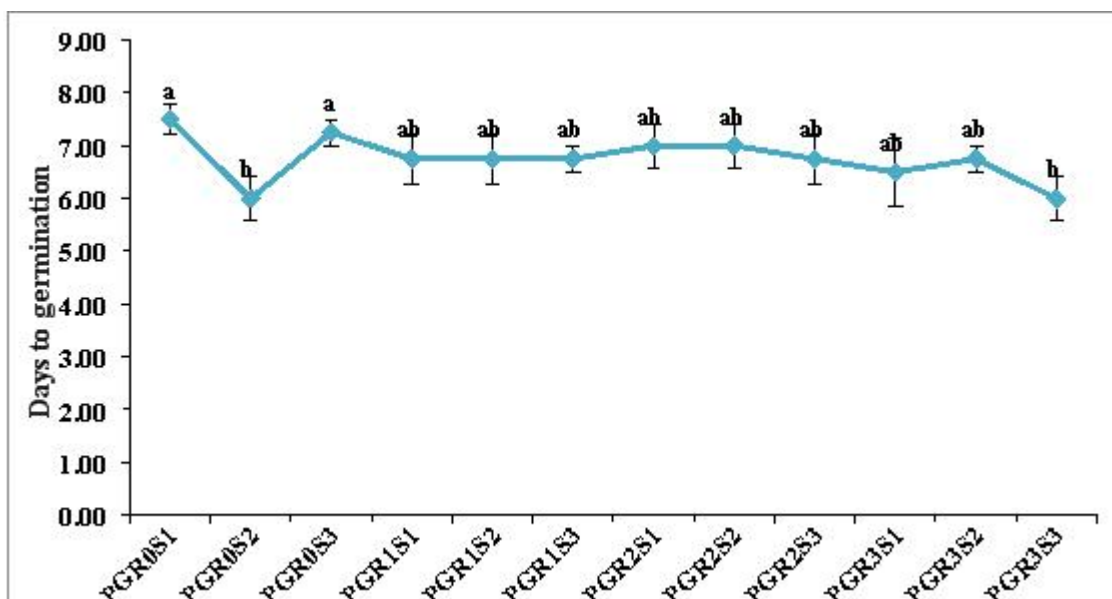


Figure 3: Combined effect of plant growth regulators and application stage on days to germination in bitter gourd. Vertical bar represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; Seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage.

4.2. Plant height (cm)

A significant result by using GA₃, NAA and MH on bitter gourd was observed for plant height (Figure 4 and Appendix v), with the doses of GA₃, NAA and MH. The maximum plant height (125.00, 233.50 and 456.50 cm) was recorded under the effect of 100 ppm GA₃ at 30 DAT and 60 DAT and harvest respectively. Plants grown up without GA₃ application were shorter than those grown with plant growth regulators specially GA₃. The lowest plant height (95.00 cm) remarked on the control, which means without GA₃ or NAA and MH. Maleic hydrazide has a mutagenic effect on the cells and prevents cell division in tubers as well as an inhibitory effect on biosynthetic activity. That result was an agreement with Sarkar *et al.*, (2014). They stated that plant height significantly decreased by MH.

A significant result by using GA₃, NAA and MH on bitter gourd was observed for plant height with the doses of GA₃, NAA and MH for different application stages. The maximum plant height (115.75, 219.06 and 423.68 cm) at 30 DAT, 60 DAT and harvest were recorded in spray at 4-leaf stage (S₂) and the lowest plant height (99.93, 191.25 and 394.87cm) were recorded in seed soaking stage (S₁) respectively. Application of PGRs like GA₃ @ 25 ppm and NAA at 2-4 true leaf stage has the positive effect on plant height. These result was agreement with Arora *et al.*, (1985).

They stated that application of PGRs like GA₃ @ 25 ppm at 2-4 true leaf stage increased the main vine length in bottle gourd (Figure 5 and Appendix v).

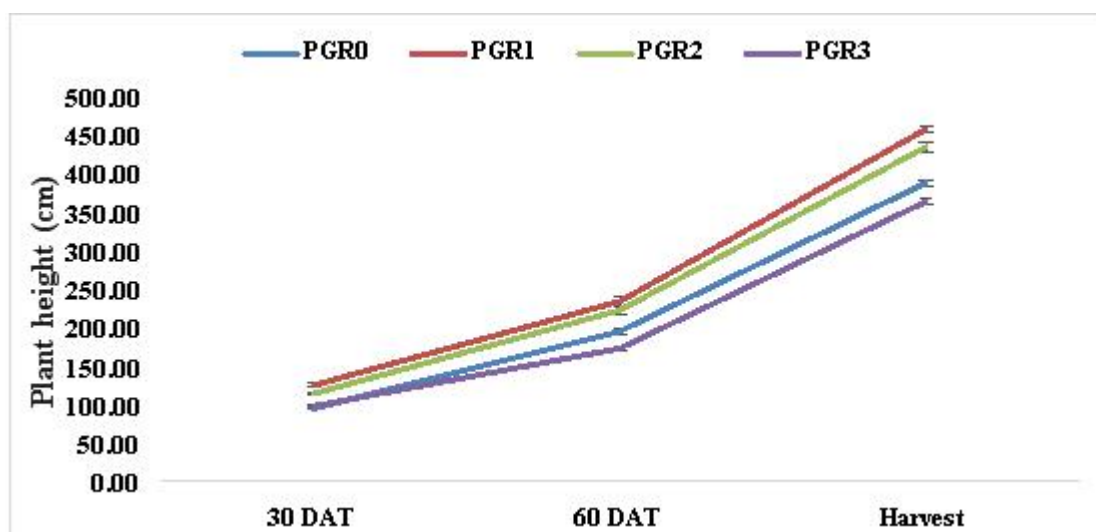


Figure 4. Effect of plant growth regulators on plant height in bitter gourd. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively.

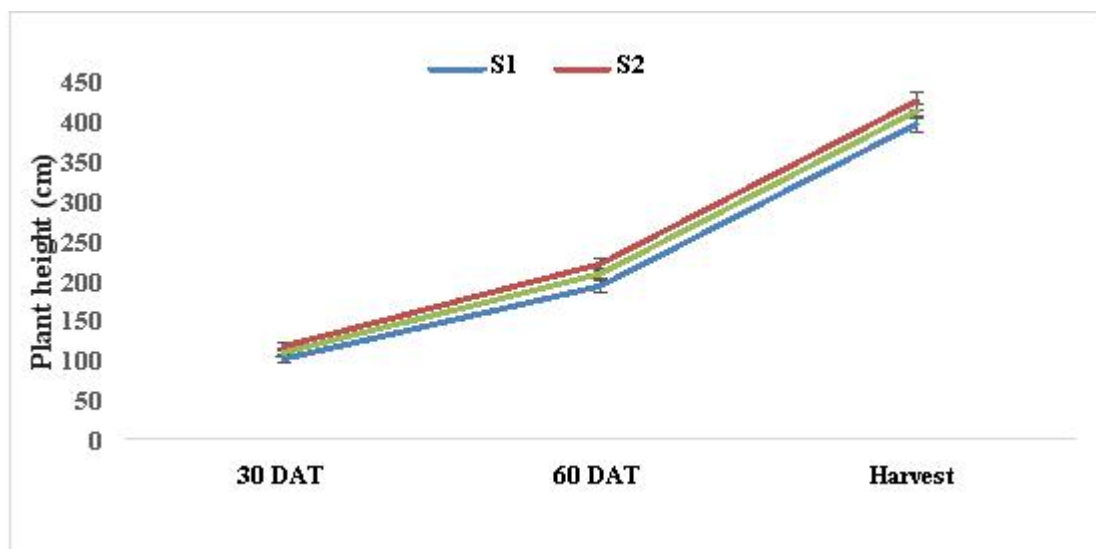


Figure 5. Effect of application stage on plant height of bitter gourd. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; Seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage.

Significant variation was recorded due to interaction effect of plant growth regulators and their application stage at 30 DAT. In case of 60 DAT and harvest interaction effect of plant growth regulators and their application stage was found to be non-

significant. The tallest (135.50, 248.25 and 467.75 cm) plant at 30 DAT, 60 DAT and harvest was recorded in the treatment combination of GA₃ and spray at 4-leaf stage (PGR₁S₂) and in case of 30 DAT the shortest (87.00 cm) was recorded in the treatment combination of control condition and seed soaking stage whereas at 60 DAT and harvest the shortest (171.50 and 360.50 cm) was recorded in the treatment combination of MH and spray at flower budding stage (PGR₃S₃) (Table 1 and Appendix v).

Table 1: Interaction effect of plant growth regulators and application stage on plant height of bitter gourd

Treatments	Plant height (cm) at 30 DAT	Plant height (cm) at 60 DAT	Plant height (cm) at harvest
PGR ₀ S ₁	87.00±0.91 ^h	178.00±1.63	371.25±4.31
PGR ₀ S ₂	101.75±0.48 ^e	211.75±1.65	401.25±3.59
PGR ₀ S ₃	96.25±0.63 ^f	194.25±3.42	388.25±0.48
PGR ₁ S ₁	113.50±0.65 ^c	213.00±0.91	445.25±2.56
PGR ₁ S ₂	135.50±0.96 ^a	248.25±13.55	467.75±3.22
PGR ₁ S ₃	126.00±1.83 ^b	239.25±13.58	456.50±2.90
PGR ₂ S ₁	106.00±0.91 ^d	206.00±2.52	409.75±4.33
PGR ₂ S ₂	123.50±1.85 ^b	237.75±4.39	451.50±6.86
PGR ₂ S ₃	112.75±0.48 ^c	222.00±2.68	439.25±6.29
PGR ₃ S ₁	93.25±2.29 ^g	168.00±1.47	353.25±4.64
PGR ₃ S ₂	102.25±0.63 ^e	178.50±2.02	374.25±1.11
PGR ₃ S ₃	97.75±0.85 ^f	171.50±2.90	360.50±4.35
LSD (.05)	2.69	17.21	11.64
CV%	1.73	5.82	1.97
P-value	0.00	0.36	0.12

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. DAT= days after transplanting. Values are mean ± SE.

4.3. Number of leaves plant⁻¹

A statistically significant variation was recorded in terms of leaves plant⁻¹ at 30 DAT, 60 DAT and last harvest for different plant growth regulators. The highest (70.17 and 241.67) number of leaves plant⁻¹ at 30 DAT and 60 DAT was recorded from the application of MH (PGR₃) which was statistically identical with PGR₁ (55.92 and 185.17) at 30 DAT and 60 DAT respectively (Table 2 and Appendix v) while the lowest (48.58 and 159.08) number of leaves plant⁻¹ was recorded in control condition at 30 DAT and 60 DAT where no plant growth regulator was applied.

At last harvest highest (413.67) number of leaves was recorded for the application of MH (PGR₃) that was closely followed by NAA (378.08) and the lowest (340.58) number of leaves plant⁻¹ at harvest was recorded in control condition (PGR₀). The results indicated that highest leaves plant⁻¹ at 30 DAT, 60 DAT and last harvest was produced by the application of plant growth regulators compared to control. This result is also in agreement with the findings of (Wankhede *et al.*, 2002b, Sharma *et al.*, 2004, Rana *et al.*, 2005, Bhalla and Kumar 2008, Kumar *et al.*, 2008, Awasthi *et al.*, 2012, Chopde *et al.*, 2012, Dogra *et al.*, 2012, Sudhakar and Kumar 2012 and Sarkar *et al.*, 2014) where they reported that, the growth parameters of gladiolus plants were significantly altered due to the application of growth regulators where plant growth regulators leads to increased vegetative growth of plants with vigorous shoots, with increased number of leaves.

Table 2: Effect of plant growth regulators on number of leaves plant⁻¹ in bitter gourd

Treatments	Number of leaves at 30 DAT	Number of leaves at 60 DAT	Number of leaves at harvest
PGR ₀	48.58±0.82 ^c	159.08±3.04 ^d	340.54±4.46 ^c
PGR ₁	55.92±1.62 ^b	185.17±3.34 ^b	366.33±3.39 ^b
PGR ₂	47.83±0.52 ^c	170.08±5.04 ^c	378.08±4.11 ^b
PGR ₃	70.17±2.75 ^a	241.67±0.89 ^a	413.67±8.80 ^a
LSD (.05)	1.39	1.33	14.75
CV%	3.00	0.98	4.75
P-value	0.00	0.00	0.00

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. DAT=days after transplanting. Values are mean ± SE.

In terms of leaves plant⁻¹ at 30 DAT, 60 DAT and last harvest in relation with different application stage were varied significantly under the trial. The highest (58.43 and 192.25) number of leaves plant⁻¹ at 30DAT and 60 DAT was recorded from the application in spray at 4-leaf stage and seed soaking stage respectively (Table 3 and Appendix v) which was closely followed by (54.93 and 181.06) in spray at flower budding stage and 4-leaf stage respectively and the lowest (53.50 and 181.06) number of leaves plant⁻¹ at 30 DAT and 60 DAT was recorded in seed soaking stage (S₁) and spray at 4-leaf stage. At last harvest the highest (379.93) no. of leaves plant⁻¹ was recorded in spray at flower budding stage (S₃) that was statistically identical with spray at 4-leaf stage (S₂) and seed soaking stage(S₁) while the lowest (371.50) number of leaves plant⁻¹ was recorded in spray at 4-leaf stage (S₂).

Table 3: Effect of different application stage on number of leaves plant⁻¹ in bitter gourd

Treatments	Number of leaves at 30 DAT	Number of leaves at 60 DAT	Number of leaves at harvest
S ₁	53.50±2.56 ^c	192.25±8.70 ^a	372.56±10.45
S ₂	58.43±2.70 ^a	181.06±8.76 ^b	371.50±7.48
S ₃	54.93±2.88 ^b	193.68±8.64 ^a	379.93±6.44
LSD _(.05)	1.20	1.33	12.80
CV%	3.00	0.98	4.75
P-value	0.00	0.00	0.36

Means with the same letter did not significantly differ from each other at p < 0.05. Abbreviations are as follows, S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. DAT= days after transplanting. Values are mean ± SE.

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation in consideration of leaves plant⁻¹ at 30 DAT, 60 DAT and harvest. The highest (78.00, 243.50) number of leaves plant⁻¹ at 30 and 60 DAT that was recorded in the treatment combination of MH and spray at 4-leaf stage (PGR₃S₂) and the lowest (46.50, 146.25) number of leaves plant⁻¹ was recorded in the treatment combination from PGR₀S₃) and (PGR₀S₂) respectively.

In case of harvest the highest (426.00) number of leaves plant⁻¹ was recorded from PGR₃S₁ and the lowest (325.25) number of leaves plant⁻¹ was recorded from PGR₀S₁. The results indicated that combination of plant growth regulators and different application stage ensures the optimum condition for the growth and development of bitter gourd and the ultimate result is the highest number of leaves plant⁻¹ at 30 DAT, 60 DAT and last harvest (Table 4 and Appendix v).

Table 4: Combined effect of plant growth regulators and different application stage on number of leaves plant⁻¹ in bitter gourd

Treatments	Number of leaves at 30 DAT	Number of leaves at 60 DAT	Number of leaves at harvest
PGR ₀ S ₁	47.25±0.75 ^{fg}	160.25±0.48 ^g	325.25±7.26 ^f
PGR ₀ S ₂	52.00±0.41 ^f	146.25±0.85 ⁱ	344.75±1.89 ^{ef}
PGR ₀ S ₃	46.50±0.87 ^g	170.75±0.48 ^f	351.75±6.06 ^{de}
PGR ₁ S ₁	61.75±0.85 ^c	172.00±0.82 ^f	365.00±7.63 ^{cde}
PGR ₁ S ₂	56.75±0.63 ^d	184.75±0.95 ^e	361.25±2.78 ^{de}
PGR ₁ S ₃	49.25±1.11 ^f	198.75±1.38 ^c	372.75±6.13 ^{bcd}
PGR ₂ S ₁	47.50±1.04 ^{fg}	193.25±1.18 ^d	374.00±9.16 ^{bcd}
PGR ₂ S ₂	47.00±0.71 ^{fg}	155.00±0.91 ^h	388.75±3.45 ^{bc}
PGR ₂ S ₃	49.00±0.82 ^f	162.00±0.82 ^g	371.50±5.73 ^{bcd}
PGR ₃ S ₁	57.50±0.65 ^d	243.50±0.65 ^a	426.00±22.00 ^a
PGR ₃ S ₂	78.00±0.71 ^a	238.25±1.38 ^b	391.25±8.20 ^b
PGR ₃ S ₃	75.00±0.82 ^b	243.25±0.75 ^a	423.75±6.72 ^a
LSD _(.05)	2.40	2.66	25.59
CV%	3.00	0.98	4.75
P-value	0.00	0.00	0.04

Means with the same letter did not significantly differ from each other at p < 0.05. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. DAT=days after transplanting. Values are mean ± SE.

4.4. Number of branch plant⁻¹

A significant result by using 100 ppm GA₃, 100 ppm NAA and 100 ppm MH on bitter gourd was observed for the number of branch plant⁻¹ at 30 DAT, 60 DAT and last harvest. The highest (5.83, 15.08 and 26.58) number of branch plant⁻¹ at 30, 60 DAT and harvest was recorded from PGR₃ whereas the lowest (4.50, 12.92 and 20.17) number of branch plant⁻¹ at 30 DAT, 60 DAT and harvest was recorded from PGR₀ where no plant growth regulator was applied (Figure 6 and Appendix v). The mechanism of increasing the number of branch due to application of maleic hydrazide @100 ppm that lead to slowing down of cell division and reduction in cell expansion as well as reduce plant height but partially increases the number of branches. This result was also an agreement with Rahman *et al.* (1992). He stated that application of @ 75 ppm TIBA promoted an increase in the number of branches.

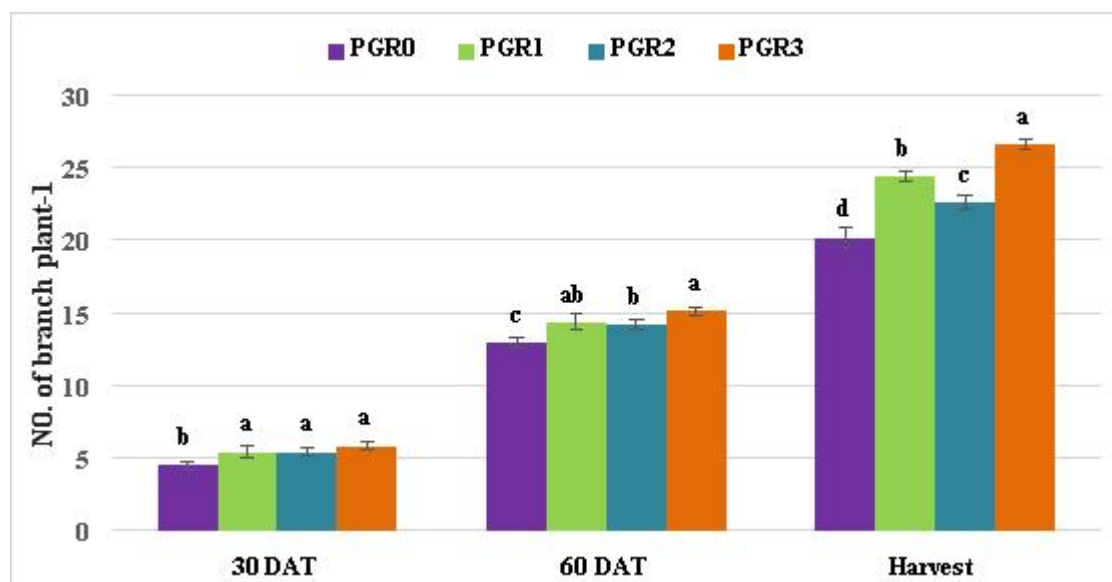


Figure 6. Effect of plant growth regulators on branch plant⁻¹ in bitter gourd. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively.

In case of different application stage, the number of branch plant⁻¹ at 30 DAT, 60 DAT and last harvest showed the statistically significant result. Different application stage showed different number of branches plant⁻¹. The highest (6.43, 15.50, and 25.12) number of branch plant⁻¹ at 30 DAT, 60 DAT and harvest was recorded from S₂ whereas the lowest (5.31, 14.00, and 21.68) number of branch plant⁻¹ was recorded from S₁ (Figure 7 and Appendix v).

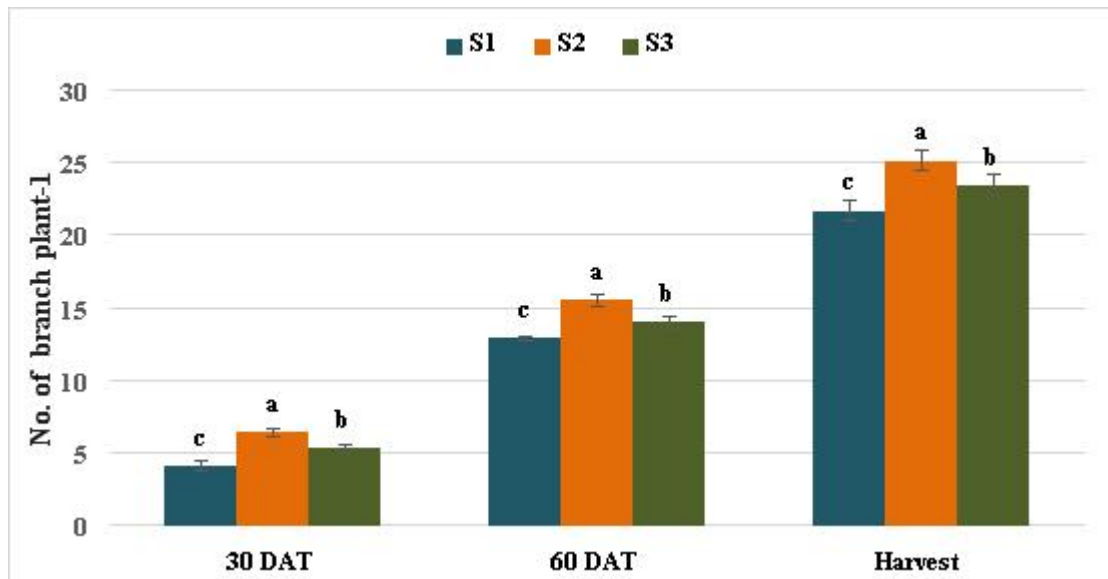


Figure 7. Effect of application stage on branch plant⁻¹ in bitter gourd. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage.

Combined effect of different plant growth regulators and different application stage showed statistically non-significant difference on number of branches plant⁻¹. From (Table 5 and Appendix v) it was found that maximum number of branches plant⁻¹ in regards to 30, 60 DAT and harvest (7.25, 17.00 and 28.00) was recorded from PGR₃S₂ while the minimum number of branches plant⁻¹ (3.50, 11.75 and 17.75) in regards to 30, 60 DAT and harvest was recorded from PGR₃S₂.

Table 5: Interaction effect of plant growth regulators and different application stage on branch plant⁻¹ in bitter gourd

Treatments	Number of branch at 30 DAT	Number of branch at 60 DAT	Number of branch at harvest
PGR ₀ S ₁	3.50±0.29	11.75±0.25	17.75±0.63
PGR ₀ S ₂	5.50±0.29	14.25±0.25	24.25±0.25
PGR ₀ S ₃	4.50±0.29	12.75±0.25	22.75±0.48
PGR ₁ S ₁	4.50±0.29	13.50±0.65	25.25±0.48
PGR ₁ S ₂	6.50±0.29	15.50±0.29	22.75±0.63
PGR ₁ S ₃	5.75±0.25	14.75±0.75	26.50±0.29
PGR ₂ S ₁	4.25±0.25	13.00±0.41	23.00±0.41
PGR ₂ S ₂	6.50±0.29	15.25±0.25	25.50±0.29
PGR ₂ S ₃	5.50±0.29	14.25±0.48	24.50±0.29
PGR ₃ S ₁	4.25±0.25	13.25±0.25	20.75±0.25
PGR ₃ S ₂	7.25±0.25	17.00±0.41	28.00±0.41
PGR ₃ S ₃	5.50±0.29	14.25±0.25	20.00±0.41
LSD _(.05)	0.81	1.14	1.24
CV%	10.53	5.59	3.67
P-value	0.89	0.65	0.13

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. DAT=days after transplanting. Values are mean ± SE.

4.5. Days to first flowering

The data on days to first flowering was found to be significant in terms of plant growth regulators. The maximum (42.92) days to first flowering was recorded from (PGR₀) while the minimum (39.09) days to first flowering was recorded from PGR₃ (Table 6 and Appendix vi). This is might be due to regulating effect of exogenous application of PGRs that influences early floral initiation. It was also reported by Hasanuzzaman *et al.* (2007), Deadname *et al.* (2007), Dogra *et al.* (2012), Sudhakar and Kumar (2012) and Sarkar *et al.* (2014a) where they stated that, PGRs promotes vegetative growth, increases the photosynthetic and metabolic activities causing more transport and utilization of photosynthetic products resulting early flowering in bitter gourd.

The data on days to germination presented in (Table 7 and Appendix vi) was found to be non-significant in terms of different application stage. The maximum (40.56) days to first flower was recorded in control condition where no plant growth regulators was applied while the minimum (39.87) days to first flower was recorded from (S₂).

Interaction effect between plant growth regulators and application stage was found to be non-significant. The maximum (43.75) days to first flower was recorded in the treatment combination of (PGR₀S₁) whereas the minimum (38.50) days to first flower was recorded in the treatment combination of (PGR₃S₂) (Table 8 and Appendix vi).

4.6. Number of male flower plant⁻¹

The data on number of male flower was found to be significant in terms of plant growth regulators. The maximum (61.67) number of male flower was recorded from PGR₁ which is statistically identical to PGR₂, and PGR₃ while the minimum (56.09) number of male flower was recorded from PGR₀ (Table 6 and Appendix vi).

The data on number of male flower presented in (Table 7 and Appendix vi) was found to be significant in terms of different application stage. The maximum (60.38) number of male flower was recorded from S₂ which is statistically identical to other treatment.

Interaction effect between plant growth regulators and different application stage was found to be non-significant variation in consideration of male flower in number. The maximum (62.25) number of male flower was recorded from PGR₁S₃ (Table 8 and Appendix vi) while the minimum (55.25) number of male flower from PGR₀S₁.

4.7. Number of female flower plant⁻¹

The data on number of female flower was found to be significant in terms of plant growth regulators. The maximum (25.95) number of female flower was recorded from PGR₃ while the minimum (20.55) number of female flower in number was recorded from PGR₀. The result showed that highest number of female flower produced from MH due to the highest number of branch plant⁻¹ (Table 6 and Appendix vi).

The data on number of female flower presented in (Table 7 and Appendix vi) was found to be significant in terms of different application stage. The maximum (24.77) number female flower was recorded from spray S₂ while the minimum (22.03) number female flower was recorded from S₁. These result was an agreement with Devies *et al.* (1987). He stated that application of PGRs at 2 or 4 leaf stage it can alter

the sex ratio and sequence at which the suppression or promotion of either sex is possible.

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation in consideration of female flower in number. The maximum (27.83) number of female flower was recorded from the treatment combination of PGR₃S₂ (Table) while the minimum (19.50) number of female flower was recorded from combination treatment of PGR₀S₁ (Table 8 and Appendix vi).

4.8. Ratio of male and female flower

The data on ratio of male and female flower presented in (Table 6 and Appendix vi) was found to be significant in terms of plant growth regulators. The maximum (2.74) ratio of male and female flower was recorded in control condition whereas the minimum (2.32) ratio of male and female flower was recorded for the application of MH (PGR₃). The result was revealed that application of NAA @100 ppm, IAA @100 ppm and 200 ppm and MH 50 ppm and 200 ppm were equally effective in suppressing the male flowers and increasing the number of female flowers in cucumber Chudhury *et al.* (1990).

The data on ratio of male and female flower was found to be non-significant in terms of different application stage. The maximum (2.69) ratio of male and female flower was recorded in seed soaking stage whereas the minimum (2.46) ratio of male and female flower was recorded from S₂ (Table 7 and Appendix vi).

Interaction effect between plant growth regulators and different application stage showed non-significant variation in consideration of ratio of male and female flower. The maximum (2.84) was recorded in the treatment combination of (PGR₀S₁) while the minimum (2.19) ratio of male and female flower was recorded in the treatment combination of (PGR₃S₂) (Table 8 and Appendix vi).

Table 6: Effect of plant growth regulators on days to first flowering, number of male flower, number of female flower and sex ratio (male: female) in bitter gourd

Treatments	Days to first flowering	Number of male flower	Number of female flower	Sex Ratio
PGR ₀	42.92±0.40 ^a	56.09±0.63 ^b	20.55±0.29 ^c	2.74±0.04 ^a
PGR ₁	39.42±0.34 ^b	61.67±0.60 ^a	23.92±0.32 ^b	2.59±0.03 ^b
PGR ₂	39.75±0.28 ^b	61.09±0.67 ^a	23.93±0.44 ^b	2.57±0.05 ^b
PGR ₃	39.09±0.36 ^b	59.75±1.01 ^a	25.95±0.53 ^a	2.32±0.07 ^c
LSD (.05)	1.03	2.95	0.76	0.21
CV%	3.07	4.49	2.92	5.80
P-value	0.00	0.00	0.00	0.00

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively. Values are mean ± SE.

Table 7: Effect of application stage on days to first flowering, number of male flower, number of female flower and sex ratio (male: female) in bitter gourd

Treatments	Days to first flowering	Number of male flower	Number of female flower	Sex ratio
S ₁	40.56±0.47	58.94±0.63	22.03±0.54 ^c	2.69±0.05
S ₂	39.87±0.52	60.38±0.98	24.77±0.61 ^a	2.46±0.16
S ₃	40.43±0.49	59.63±0.86	23.96±0.65 ^b	2.50±0.06
LSD (.05)	0.89	2.32	0.60	0.17
CV%	3.07	4.49	2.92	5.80
P-value	0.26	0.33	0.00	0.46

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; Seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. Values are mean ± SE.

Table 8: Combined effect of plant growth regulators and different application stage on days to first flowering, number of male flower, number of female flower and sex ratio (male: female) in bitter gourd

Treatments	Days to first flowering	Number of male flower	Number of female flower	Sex ratio
PGR ₀ S ₁	43.75±0.48	55.25±1.18	19.50±0.20 ⁱ	2.84±0.04
PGR ₀ S ₂	42.25±0.85	56.75±1.03	21.32±0.26 ^{gh}	2.67±0.07
PGR ₀ S ₃	42.75±0.63	56.25±1.25	20.83±0.49 ^{hi}	2.70±0.08
PGR ₁ S ₁	39.75±0.85	61.00±1.08	22.60±0.35 ^{efg}	2.70±0.41
PGR ₁ S ₂	39.25±0.48	61.75±1.18	24.40±0.2 ^{cd}	2.53±0.05
PGR ₁ S ₃	39.25±0.48	62.25±1.10	24.75±0.25 ^{bcd}	2.52±0.04
PGR ₂ S ₁	40.00±0.41	59.50±1.04	22.27±0.50 ^{fgh}	2.68±0.08
PGR ₂ S ₂	39.50±0.65	62.00±1.41	25.50±0.29 ^{bc}	2.44±0.07
PGR ₂ S ₃	39.75±0.48	61.75±0.85	24.00±0.36 ^{cde}	2.58±0.06
PGR ₃ S ₁	38.75±0.63	60.00±1.77	23.75±0.33 ^{def}	2.53±0.11
PGR ₃ S ₂	38.50±0.65	61.00±1.58	27.83±0.26 ^a	2.19±0.08
PGR ₃ S ₃	40.00±0.41	58.25±2.05	26.25±0.45 ^{ab}	2.22±0.07
LSD (.05)	1.78	6.61	1.70	0.47
CV%	3.07	4.49	2.92	5.80
P-value	0.63	0.80	0.01	0.65

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. Values are mean ± SE.

4.9. Fruit setting (%)

The data on fruit setting% presented in (Table 9 and Appendix vii) was found to be non-significant in terms of plant growth regulators. All the treatment showed the statistically identical whereas the percent fruit setting was found to be maximum (90.86) in GA₃ (PGR₁) and the minimum (86.10) percent fruit set was found to be in MH (PGR₃). This might be occurs due to application of auxin at the time of flowering and resulted lower flowers drop that enhance fruit setting and contributed higher percentage of fruit setting. This result is in agreement with the findings of Hasanuzzaman *et al.* (2007). This result also is in agreement with the findings of Deka and Shadeque (1996) obtained the fruit set of bell pepper with cycocel at 500, 1000 of 1500 ppm.

All the treatment showed the statistically identical in terms of different application stage. Among the treatments, the percent fruit setting was maximum (88.46) in spray at 4-leaf stage while the minimum percent fruit setting (83.72) was recorded in spray at flower budding stage (Table 9 and Appendix vii).

Interaction effect between plant growth regulators and different application stage was found to be non-significant of fruit set percent. Among the combination treatments, the percent fruit setting was maximum (88.5) in the treatment combination PGR₃S₂ while the percent fruit setting was minimum (88.5) in the treatment combination of PGR₃S₃ Table 10 and Appendix vii).

4.10. Fruit length (cm)

The data on fruit length was found to be non-significant in terms of plant growth regulators. Maximum (16.69 cm) fruit length was recorded for the application of GA₃ (PGR₁) while the minimum (12.83 cm) fruit length was recorded in control treatment (PGR₀). The longer fruits under GA₃ might be due increased promotes cell division and cell elongation which would have favored uptake of water and nutrients. A similar effect with gibberellic acid application was reported by Singh *et al.* (1998). NAA increased the fruit length possibly by activating cell division, enlarging the cell and increasing the metabolic activity. Similar findings were reported by Dubey (1983). The result was revealed that maximum fruit length in Ivy gourd obtained when GA₃ and NAA were applied @ 100 and 400 ppm respectively Prabu and Natarajan (2006) (Table 9 and Appendix vii).

The data on fruit length was found to be non-significant in terms of different application stage. Maximum (16.95 cm) fruit length was recorded from (S₂) (Table) while the minimum (13.08 cm) fruit length was recorded in seed soaking stage (S₁) (Table 9 and Appendix vii).

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation for fruit length compare to control. The maximum (19.52 cm) fruit length was recorded in the treatment combination of GA₃ and spray at 4-leaf stage (PGR₁S₂) while the minimum (9.97cm) was recorded in the treatment combination of control and seed soaking stage (PGR₀S₁) (Table 10 and Appendix vii).

4.11. Fruit diameter (cm)

The data on fresh mass of fruit indicated significant differences among the treatment of plant growth regulators. The maximum (4.48 cm) fruit diameter was recorded for the application of GA₃ (PGR₁) while the minimum (3.86 cm) fruit diameter was recorded in control treatment (PGR₀) (Table 9 and Appendix vii). Fruit diameter increase under GA₃ treatment was observed in the present study and might be attributed to cell growth and cell elongation. This finding are in contrast with other studies which noted that GA₃ can affect the growth and development of fruit as well as promoting diameter (Hye *et al.*, 2002; Islam *et al.*, 2007; Nagwa *et al.*, 2013), in tomato (Choudhury *et al.*, 2013) and in gladiolus (Sarkar *et al.*, 2014).

The data on fruit diameter was found to be non-significant in terms of different application stage. The maximum (4.56 cm) fruit diameter was recorded in application of spray at 4-leaf stage and the minimum (3.82 cm) fruit diameter was recorded in seed soaking stage (S₁) (Table 9 and Appendix vii). The result was revealed that foliar application of GA₃ (5, 10, 20 ppm) and MH (50, 100, 200 ppm) at 2, 4 and 6 leaf stages resulted in increase in fruit diameter of summer cucumber; whereas, GA₃ was inferior to MH (Rafeekar *et al.*, 2002).

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation for fruit diameter. The maximum (4.99 cm) fruit diameter was recorded in the treatment combination of GA₃ and spray at 4-leaf stage (PGR₁S₂) and the minimum (3.36 cm) was recorded in the treatment combination of control and seed soaking stage (PGR₀S₁) (Table 10 and Appendix vii).

4.12. Fresh mass of fruit (g)

The data on fresh mass of fruit indicated significant differences among the treatment of plant growth regulators. The maximum (147.38g) fresh mass of fruit was recorded for the application of GA₃ (PGR₁) (Table 9 and Appendix vii) while the minimum (114.90g) fresh mass of fruit was recorded in control treatment (PGR₀). The effect of different treatments of growth regulators on individual fresh mass of fruit of Ivy gourd was found to be significant. The individual fresh mass of fruit varied from 8.03g to 13.25g. The highest fresh mass of fruit (13.25g) was obtained with GA₃ 100 ppm, followed by GA₃ 200 ppm (12.75g) and these were significantly superior over control (8.03g). NAA 400 ppm (12.36g) NAA 300 ppm (10.90g), NAA 200 ppm

(10.37g) and GA₃ 50 ppm (9.95g) treatments produced higher fresh mass of fruit than control. Similar observations were recorded by Vijay and Jalikop (1980) Das *et al.* (2001) and Sarkar *et al.* (1989). Prasad and Kumar, (2003) stated that plant growth regulators promote the cell wall loosening processes providing a state of extensive flexibility within the cell leading ultimately in plant growth.

The data on fresh mass of fruit was found to be non-significant in terms of different application stage. The maximum (144.22g) fresh mass of fruit was recorded in application of spray at 4-leaf stage while the minimum (128.02g) fresh mass of fruit was recorded in seed soaking stage (S₁) (Table 9 and Appendix vii). These result was an agreement with Yasuyoshi and Yoshiyuki (1995). They opined that application of NAA @ 150 ppm at 2 and 4 true leaf stages increased the average fresh mass of fruit.

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation for fresh mass of fruit. The maximum (156.26g) fresh mass of fruit was recorded in the treatment combination of GA₃ and spray at 4-leaf stage (PGR₁S₂) while the minimum (102.30g) was recorded in the treatment combination of control and seed soaking stage (PGR₀S₁) (Table 10 and Appendix vii).

4.13. Dry matter content of fruit (%)

The data on dry matter content of fruit indicated significant differences among the treatment of plant growth regulators. The maximum (9.02 %) dry matter content of fruit was recorded for the application of GA₃ (PGR₁) that was statistically identical to NAA(G₂) (9.01 %) while the minimum (7.4 %) dry matter content content of fruit was recorded in control (PGR₀) (Table 9 and Appendix vii).

The data on dry matter content of fruit was found to be non-significant in terms of different application stage. The maximum (9.24 %) dry matter content of fruit was recorded in application of spray at 4-leaf stage that was statistically identical to spray at flower budding stage (S₃) (8.09 %) while the minimum (7.65 %) dry matter content of fruit was recorded in seed soaking stage (Table 9 and appendix vii)

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation for dry matter content of fruit. The maximum (10.37 %) dry matter content of fruit % was recorded in the treatment combination of PGR₁S₂ while the minimum (6.80 %) dry matter content of fruit was recorded in the treatment combination of control and seed soaking stage (PGR₀S₁) (Table 10 and Appendix vii).

Table 9: Effect of plant growth regulators and various application stage on fruit setting, fruit length, fruit diameter, fresh mass of fruit, dry matter content of fruit and number of fruit plant⁻¹ in bitter gourd

Treatments	Fruit setting (%)	Fruit length (cm)	Fruit diameter (cm)	Fresh mass of fruit (g)	Dry matter content of fruit (%)	Number of fruit plant ⁻¹
PGR ₀	87.40±1.19	12.83±0.68 ^c	3.86±0.11 ^b	114.90±2.77 ^c	7.40±0.24 ^b	17.98±0.42 ^b
PGR ₁	90.86±1.38	16.69±0.70 ^a	4.48±0.14 ^a	147.38±2.83 ^a	9.02±0.29 ^a	21.71±0.37 ^a
PGR ₂	89.82±1.63	15.20±0.34 ^b	4.38±0.07 ^a	140.81±2.64 ^b	9.01±0.10 ^a	21.50±0.53 ^a
PGR ₃	86.10±1.97	15.35±0.44 ^b	3.92±0.09 ^b	143.92±1.21 ^{ab}	7.80±0.22 ^b	22.37±0.78 ^a
LSD (.05)	5.39	0.61	0.11	3.11	0.16	1.18
CV%	7.12	4.88	3.18	2.74	2.29	5.13
P-value	0.09	0.00	0.00	0.00	0.00	0.00
Stages						
S ₁	87.74±1.36	13.08±0.66 ^c	3.82±0.13 ^c	128.02±4.42 ^c	7.65±0.26 ^c	19.34±0.64 ^c
S ₂	90.86±1.08	16.95±0.54 ^a	4.56±0.11 ^a	144.22±3.67 ^a	9.24±0.27 ^a	22.50±0.67 ^a
S ₃	87.08±1.80	15.02±0.56 ^b	4.10±0.09 ^b	138.01±3.54 ^b	8.09±0.25 ^b	20.83±0.61 ^b
LSD (.05)	4.24	0.53	0.10	2.70	0.14	0.93
CV%	7.12	4.88	3.18	2.74	2.29	5.13
P-value	0.08	0.00	0.00	0.00	0.00	0.00

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively, S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. Values are mean ± SE.

Table 10: Effect of plant growth regulators and various application stage interaction on fruit setting, fruit length, fruit diameter, fresh mass of fruit, dry matter content of fruit and number of fruit plant⁻¹ in bitter gourd

Treatments	Fruit setting (%)	Fruit length (cm)	Fruit diameter (cm)	Fresh mass of fruit (g)	Dry matter content of fruit (%)	Number of fruit plant ⁻¹
PGR ₀ S ₁	84.91±1.70	9.97±0.20 ^f	3.36±0.01 ^h	102.30±0.80 ^h	6.80±0.09 ^h	16.50±0.28 ^c
PGR ₀ S ₂	91.19±1.92	15.08±0.43 ^{cd}	4.18±0.01 ^d	122.73±0.93 ^g	8.56±0.05 ^d	19.45±0.53 ^{cd}
PGR ₀ S ₃	86.47±1.24	13.46±0.51 ^e	4.06±0.01 ^e	119.70±1.41 ^g	7.09±0.0 ^g	18.00±0.40 ^{de}
PGR ₁ S ₁	91.85±1.25	14.14±0.43 ^{de}	4.02±0.03 ^{ef}	137.19±3.19 ^{ef}	8.25±0.07 ^e	20.75±0.25 ^{bc}
PGR ₁ S ₂	88.18±2.05	19.52±0.31 ^a	4.99±0.16 ^a	156.26±3.33 ^a	10.37±0.05 ^a	21.50±0.28 ^{bc}
PGR ₁ S ₃	92.56±3.39	16.42±0.52 ^b	4.15±0.02 ^{de}	148.72±2.29 ^{bc}	8.50±0.13 ^d	22.90±0.80 ^b
PGR ₂ S ₁	89.84±3.04	14.07±0.24 ^{de}	4.32±0.07 ^{cd}	132.47±3.75 ^f	8.66±0.01 ^d	20.00±0.71 ^{cd}
PGR ₂ S ₂	91.56±3.33	16.60±0.24 ^b	4.76±0.02 ^b	149.98±1.99 ^b	9.38±0.14 ^b	23.32±0.67 ^{ab}
PGR ₂ S ₃	88.06±2.57	14.95±0.28 ^{cd}	4.38±0.06 ^c	139.10±2.69 ^{de}	8.99±0.09 ^c	21.20±0.52 ^{bc}
PGR ₃ S ₁	84.67±3.14	14.18±1.00 ^{de}	3.62±0.03 ^g	140.16±1.34 ^{de}	6.96±0.04 ^{gh}	20.10±0.72 ^{cd}
PGR ₃ S ₂	92.53±0.94	16.60±0.26 ^b	4.31±0.06 ^{cd}	147.96±1.51 ^{bc}	8.67±0.10 ^d	25.75±0.25 ^a
PGR ₃ S ₃	81.10±3.02	15.30±0.35 ^c	3.85±0.11 ^f	143.67±1.35 ^{cd}	7.81±0.20 ^{hf}	21.25±0.47 ^{bc}
LSD _(.05)	12.08	1.06	0.19	5.39	0.28	2.65
CV%	5.53	4.88	3.18	2.74	2.29	5.13
P-value	0.08	0.00	0.00	0.00	0.00	0.00

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. Values are mean ± SE.

4.14. Number of fruits plant⁻¹

The data on number of fruits plant⁻¹ indicated significant differences among the treatment of plant growth regulators. The maximum number of fruits plant⁻¹ (22.37) was recorded in PGR₃ and it was significantly superior over PGR₁, PGR₂ and PGR₀. The minimum number of fruits plant⁻¹ (17.98) was recorded in control compared to all other treatments which was significantly lower with all other treatments (Table 9 and Appendix vii). These result was an agreement with Sonkar (2003) and Jatoi et al., (2010). They stated that foliar spray of ethephon (100-500 mg l⁻¹), GA3 910 mg l⁻¹, MH (50-150 mg l⁻¹) and TIBA (25-50 mg l⁻¹) increased the number of fruit yield plant⁻¹ in most of the cucurbits.

Different application stage showed statistically significant variation on fruit number plant⁻¹. The maximum (22.50) fruit number plant⁻¹ was recorded in application of spray at 4-leaf stage (S₂) while the minimum (19.34) fruit number of fruits plant⁻¹ was recorded in seed soaking stage (S₁) (Table 9 and Appendix vii). These result was an agreement with Arora *et al.*, (1988). He stated that plants sprayed with 5 different plant growth regulators at 2 and 4-leaf stage. The total yield (2.39 kg plant⁻¹) was the highest in plants treated with ethereal ethephone at @ 100 ppm compare to control (0.69 kg plant⁻¹) in case of (*Leganaria aegyptiaca* L).

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation for fruit number plant⁻¹. The maximum (25.75) fruit number plant⁻¹ was recorded in the treatment combination of MH and spray at 4-leaf stage (PGR₃S₂) while the minimum (16.50) was recorded in the treatment combination of control and seed soaking stage (PGR₀S₀) (Table 10 and Appendix vii).

4.15. Fruit yield (t ha⁻¹)

The data on fruit yield (t ha⁻¹) was showed significant differences among the plant growth regulators. Among the treatments, the maximum fruit yield (21.50 t ha⁻¹) was recorded from PGR₃ (MH @ 100 ppm) while the minimum fruit yield (13.85 t ha⁻¹) was recorded from PGR₀ (Figure 8 and Appendix vii). Application of 4-CPA has positive effect on yield of bell pepper. This result is in agreement with the findings of Hasanuzzaman *et al.* (2007) and Appireddy *et al.* (2008) where they reported that, plant growth regulators increase the yield of bell pepper. Baliyan *et al.* (2013) and

Sarkar *et al.* (2014a) also reported that plant growth regulators increases has great potentiality to facilitate the flower and fruit setting as well as yield $t\ ha^{-1}$ of summer tomato.

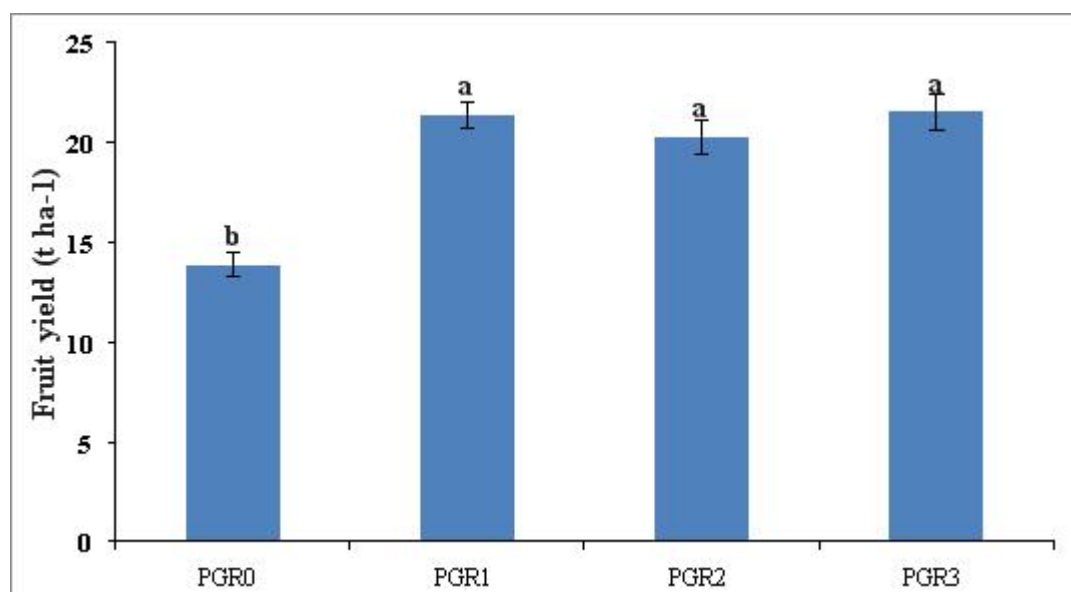


Figure 8: Effect of plant growth regulators on fruit yield in bitter gourd.

Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively

The data on fruit yield ($t\ ha^{-1}$) at harvest indicated non-significant differences in terms of different application stage. The maximum fruit yield ($21.77\ t\ ha^{-1}$) was registered in spray at flower budding stage and the minimum fruit yield ($16.68\ t\ ha^{-1}$) was recorded in 4-leaf stage (Figure 9 and Appendix vii). This result is agreement with (Arora *et al.* 1985) who reported that foliar spray of MH (150 ppm) at 2 and 4 true leaf stages at 7 days interval recorded highest total yield ($376.3\ t\ ha^{-1}$) by number and weight in bottle gourd.

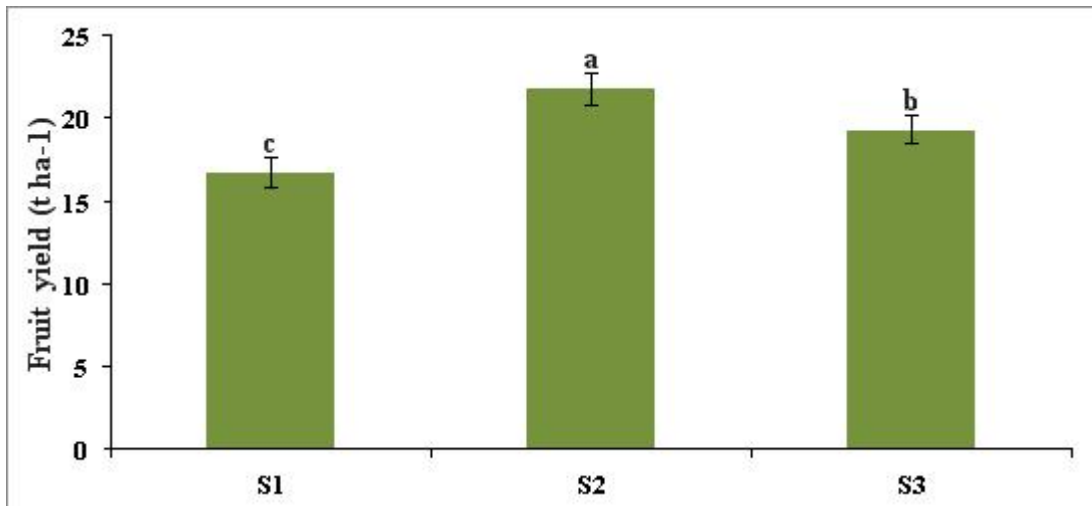


Figure 9: Effect of application stage on fruit yield in bitter gourd. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; Seed soaking stage, S₂; Spray at 4-leaf stage and S₃; Spray at flower budding stage

Interaction effect between plant growth regulators and different application stage was found to be non-significant of fruit yield (t ha⁻¹). Among the combination treatments, the maximum (25.42 t ha⁻¹) fruit yield was recorded from PGR₃S₂ while the minimum (11.26 t ha⁻¹) fruit yield was recorded from the treatment combination of PGR₀S₁ (Figure 10 and Appendix vii).

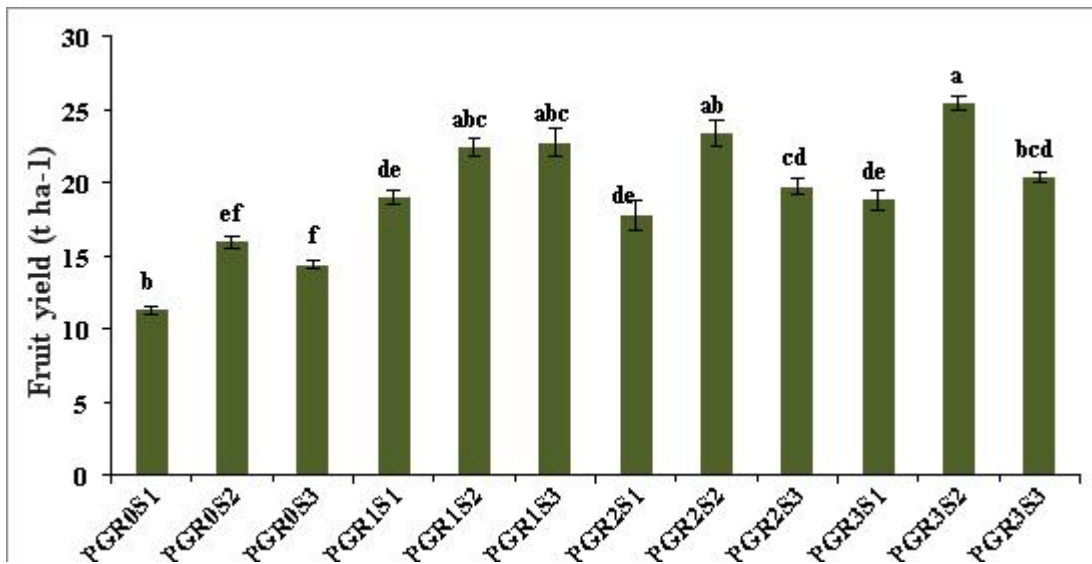


Figure 10: Interaction effect of plant growth regulators and application stage on fruit yield in bitter gourd. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage.

4.16. Root lengths at harvest (cm)

Plant growth regulators showed a statistically non-significant variation on root length. The maximum (24.12 cm) root length was recorded for the application of PGR₃ while the minimum (25.62 cm) root length was recorded in PGR₂ (Figure 11 and Appendix viii). The increased root length due to GA₃ might have resulted from the cell growth and cell elongation and thus developed an elongated root system. This result is consistent with the previous findings for onion (Hye *et al.* 2002).

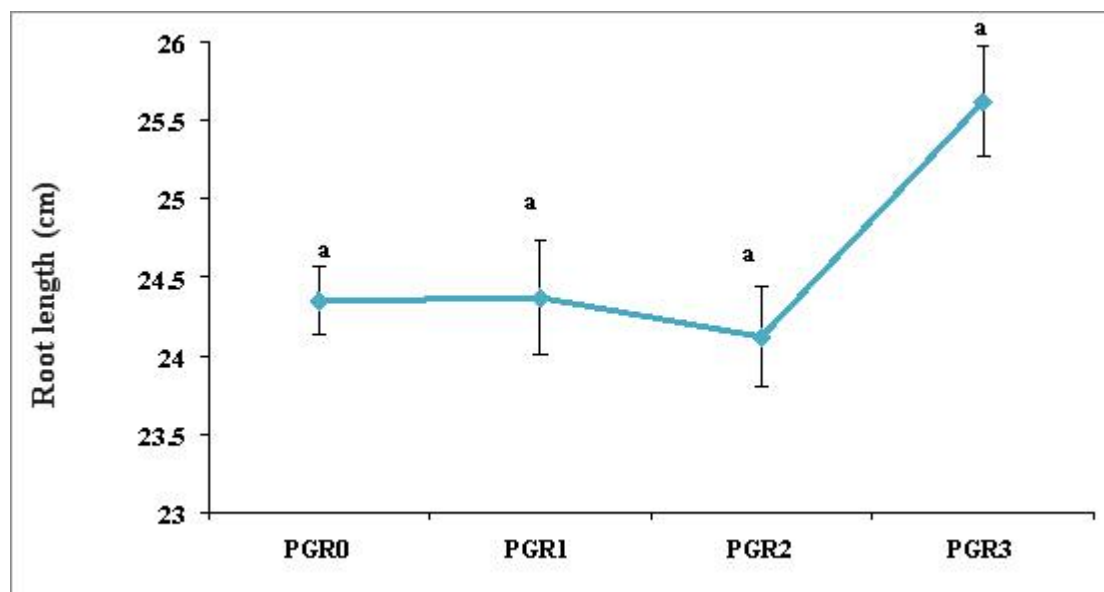


Figure 11. Effect of plant growth regulators on root length at harvest in bitter gourd. Vertical bar represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively

Different application stage showed statistically significant variation on root length. The maximum (25.30 cm) root length was recorded in spray at flower budding stage (S₃) while the minimum (23.56 cm) root length was recorded in application of spray at 4-leaf stage (Figure 12 and Appendix viii).

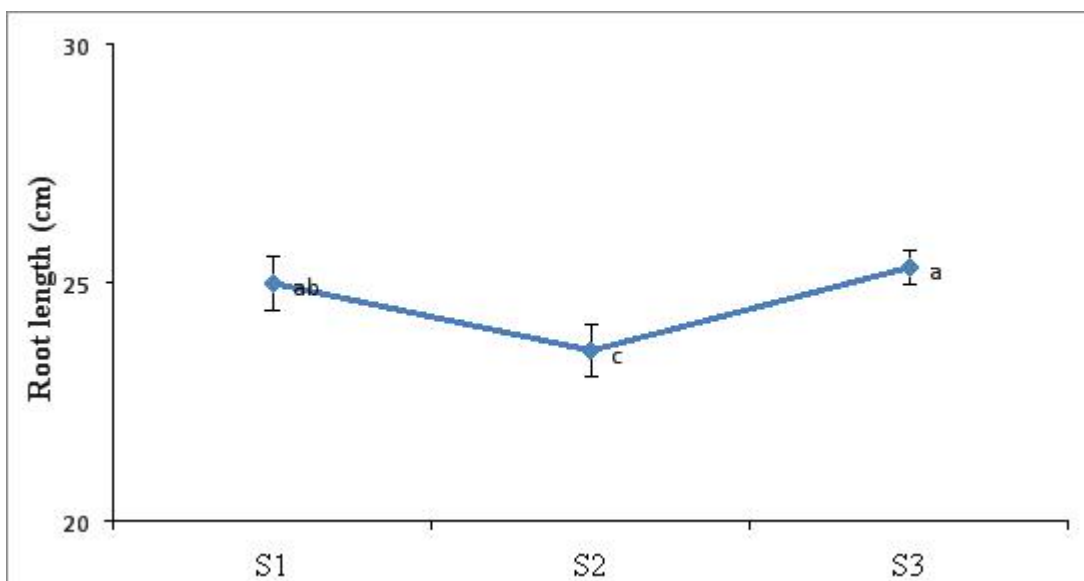


Figure 12. Effect of application stage on root length at harvest in bitter gourd. Vertical bar represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation for root length. The maximum (27.25 cm) root length was recorded in the treatment combination of (PGR₃S₃) while the minimum (22.25 cm) was recorded in the treatment combination of (PGR₁S₂) (Figure 13 and Appendix viii)

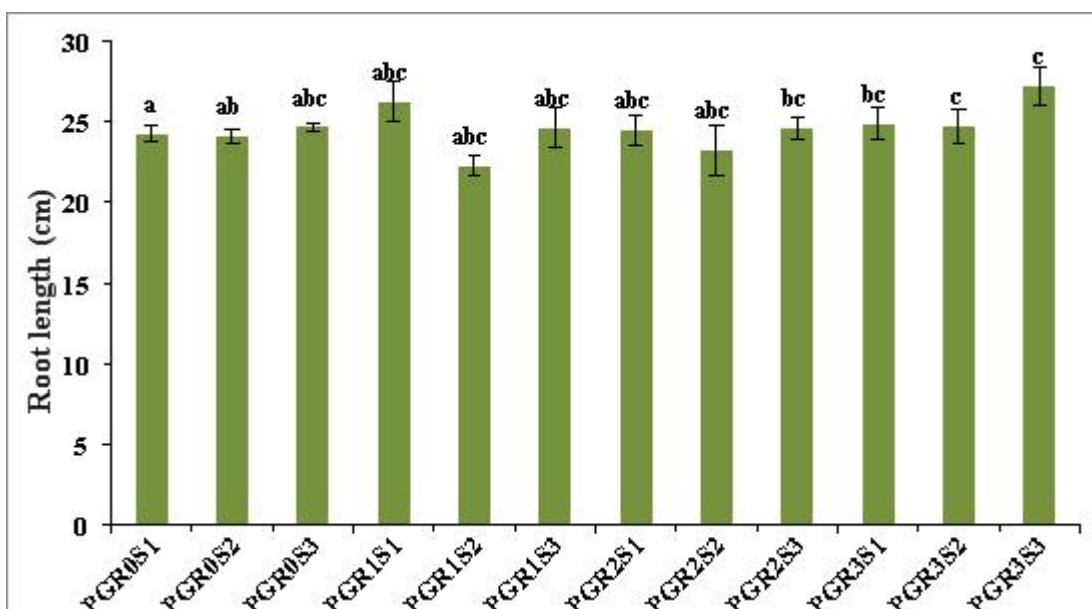


Figure 13. Combined effect of plant growth regulators and application stage on root length at harvest in bitter gourd. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; Seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage.

4.17. Plant fresh mass at harvest (g)

Plant growth regulators showed a statistically significant variation on plant fresh mass. The maximum (11.16g) plant fresh mass was recorded from the application of (PGR₁) that was statistically identical to control condition (PGR₀) (9.58g) while the minimum (8.71g) plant fresh mass was recorded from PGR₃ (Table 12 and Appendix ix).

Different application stage showed statistically significant variation on plant fresh mass. The maximum (10.62g) plant fresh mass was recorded in application of spray at 4-leaf stage (Table 13 and Appendix ix) that was statistically identical to spray at flower budding stage (S₃) (9.31g) while the minimum (9.07g) plant fresh mass was recorded in seed soaking stage (S₁).

Interaction effect between plant growth regulators and different application stage showed a statistically significant variation for plant fresh mass. The maximum (12.20g) plant fresh mass was recorded in the treatment combination of GA₃ and spray at 4-leaf stage (PGR₁S₂) while the minimum (8.22g) was recorded in the treatment combination of MH and seed soaking stage (PGR₃S₁) (Table 14 and Appendix ix).

4.18. Root fresh mass at harvest (g)

Plant growth regulators did not show a statistically significant variation on root fresh mass. The maximum (9.80g) root fresh mass at harvest was recorded for the application of MH (PGR₃) while the minimum (9.73g) root fresh mass was recorded in NAA (PGR₂) (Table 11 and Appendix ix).

Different application stage did not show statistically significant variation on plant fresh mass at harvest. The maximum (10.17g) root fresh mass was recorded in seed soaking stage (S₁) while the minimum (9.39g) root fresh mass was recorded in flower budding stage (S₃) (Table 12 and Appendix ix).

Interaction effect between plant growth regulators and different application stage did not show a statistically significant variation for plant fresh mass at harvest. The maximum (10.97g) root fresh mass was recorded in the treatment combination of PGR₁S₁ and the minimum (8.55g) root fresh mass was recorded in the treatment combination of PGR₂S₃ (Table 13 and Appendix ix).

4.19. Root dry matter content at harvest (%)

Plant growth regulators showed a statistically significant variation on root dry matter content at harvest. The maximum (2.50 %) dry matter content of root was recorded for the application of MH (PGR₃) while the minimum (1.45 %) dry matter content of root was recorded in NAA (PGR₂) (Table 11 and Appendix ix).

Different application stage showed statistically significant variation on dry matter content of root at harvest. The maximum (2.14 %) dry matter content of root at harvest was recorded in seed soaking stage (S₁) while the minimum (1.72 R) dry matter content of root was recorded in flower budding stage (S₃) (Table 12 and Appendix ix).

Table 11: Effect of plant growth regulators on fresh mass of plant, fresh mass of root and root dry matter content at harvest in bitter gourd

Treatments	Fresh mass of plant (g)	Root fresh mass (g)	Root dry matter content (%)
PGR ₀	9.58±0.29 ^b	9.75±0.17	2.07±0.09 ^b
PGR ₁	11.16±0.26 ^a	9.77±0.55	1.45±0.02 ^c
PGR ₂	9.19±0.22 ^c	9.73±0.44	2.50±0.06 ^a
PGR ₃	8.71±0.15 ^d	9.80±0.33	1.53±0.05 ^c
LSD (.05)	1.15	1.09	0.08
CV%	1.34	13.40	3.81
P-value	0.00	0.10	0.00

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively. Values are mean ± SE.

Interaction effect between plant growth regulators and different application stage did not showed a statistically significant variation for root dry matter content at harvest. The maximum (2.75 %) dry matter content of root was recorded in the treatment combination of NAA and spray at 4-leaf stage PGR₂S₂ while the minimum (1.34 %) dry matter content of root was recorded in the treatment combination of GA₃ and seed soaking stage PGR₁S₁ (Table 13 and Appendix ix).

Table 12: Effect of different application stage on fresh mass of plant, fresh mass of root and root dry matter content at harvest in bitter gourd

Treatments	Fresh mass of plant (g)	Root fresh mass (g)	Root dry matter content (%)
S ₁	9.07±0.29 ^c	10.17±0.43	1.72±0.12 ^c
S ₂	10.62±0.32 ^a	9.73±0.34	2.14±0.13 ^a
S ₃	9.31±0.31 ^b	9.39±0.20	1.80±0.12 ^b
LSD _(.05)	0.12	0.95	0.07
CV%	1.34	13.40	3.81
P-value	0.00	0.26	0.00

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; Seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. Values are mean ± SE.

Table 13: Combined effect of plant growth regulators and different application stage on fresh mass of plant, fresh mass of root and root dry matter content at harvest in bitter gourd

Treatments	Fresh mass of plant (g)	Root fresh mass (g)	Root dry matter content (%)
PGR ₀ S ₁	8.60±0.00 ^f	9.70±0.24	1.90±0.00 ^d
PGR ₀ S ₂	10.90±0.00 ^b	9.65±0.28	2.50±0.00 ^b
PGR ₀ S ₃	9.24±0.00 ^e	9.93±0.42	1.80±0.00 ^d
PGR ₁ S ₁	10.10±0.00 ^c	10.97±1.32	1.34±0.00 ^{ef}
PGR ₁ S ₂	12.20±0.00 ^a	9.12±0.51	1.54±0.00 ^e
PGR ₁ S ₃	11.20±0.00 ^b	9.24±0.74	1.45±0.00 ^{ef}
PGR ₂ S ₁	9.35±0.00 ^e	10.59±0.58	2.25±0.00 ^c
PGR ₂ S ₂	9.99±0.00 ^d	10.09±0.48	2.75±0.00 ^a
PGR ₂ S ₃	8.25±0.00 ^{gh}	8.55±0.89	2.50±0.00 ^b
PGR ₃ S ₁	8.22±0.00 ^h	9.44±0.33	1.36±0.00 ^f
PGR ₃ S ₂	9.38±0.00 ^e	10.09±0.71	1.76±0.00 ^d
PGR ₃ S ₃	8.55±0.00 ^{fg}	9.88±0.72	1.45±0.00 ^{ef}
LSD _(.05)	0.32	1.89	0.18
CV%	1.34	13.40	3.81
P-value	0.00	0.27	0.00

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. Values are mean ± SE.

4.20. BIOCHEMICAL COMPOSITION

4.20.1. Reducing sugar (mg. g fr.wt.⁻¹) in fruits

The data on reducing sugars in fruits at harvest indicated non-significant differences among the plant growth regulators. Among the treatments, the maximum reducing sugars (0.42 mg) was registered in MH while the minimum reducing sugar (0.39 mg) was recorded in NAA (Table 14 and Appendix x).

The data on reducing sugar in fruits at harvest indicated non-significant differences in terms of different application stage. The maximum reducing sugars (0.41 mg) was registered in spray at 4-leaf stage that while the minimum reducing sugar (0.40 mg) was recorded in seed soaking stage (Table 15 and Appendix x).

Interaction effect between plant growth regulators and different application stage was found to be non-significant of reducing sugars. Among the combination treatments, the reducing sugar was maximum (0.43 mg) in MH and seed soaking stage (PGR₃S₁) while the reducing sugar was minimum (0.38 mg) in the treatment combination of NAA and seed soaking stage (PGR₂S₁) (Table 16 and Appendix x).

4.20.2. Non-reducing sugar (mg. g fr.wt.⁻¹) in fruits

The data on non-reducing sugar in fruits at harvest indicated significant differences among the plant growth regulators. Among the treatments, the maximum non-reducing sugar (4.07 mg) was registered in MH and the minimum reducing sugar (3.86 mg) was recorded in control condition (Table 14 and Appendix x).

The data on non-reducing sugar in fruits at harvest indicated non-significant differences in terms of different application stage. The maximum non-reducing sugar (4.04 mg) was registered in spray at flower budding stage that and the minimum non-reducing sugars (3.93 mg) was recorded in seed soaking stage (Table 15 and Appendix x).

Interaction effect between plant growth regulators and different application stage was found to be non-significant of non-reducing sugar. Among the combination treatments, the non-reducing sugar was maximum (4.10 mg) in MH and spray at flower budding stage (PGR₃S₃) and the non-reducing sugar was minimum (3.73 mg) in the treatment combination of control and seed soaking stage (PGR₀S₁) (Table 16 and Appendix x).

4.20.3. Total sugar (mg. g fr.wt.⁻¹) in fruits

The data on total sugar in fruits at harvest indicated significant differences among the plant growth regulators. Among the treatments, the maximum total sugar (4.50 mg) was registered from (PGR₃) while the minimum total sugars (4.26 mg) was recorded from (PGR₀) (Table 14 and Appendix x).

The data on total sugar in fruits at harvest indicated non-significant differences in terms of different application stage. The maximum total sugar (4.44 mg) was registered in spray at flower budding stage while the minimum total sugar (4.34 mg) was recorded in seed soaking stage (Table 15 and Appendix x).

Interaction effect between plant growth regulators and different application stage was found to be non-significant of total sugar. Among the combination treatments, the total sugar was maximum (4.51 mg) was found from PGR₀S₁ and the total sugar was minimum (4.12 mg) in the treatment combination of PGR₀S₂ (Table 16 and Appendix x).

4.20.4. Total phenols (mg. g fr.wt.⁻¹) in fruits

The data on total phenols in fruits at harvest indicated non-significant differences among the plant growth regulators. Among the treatments, the maximum total phenols (9.90 mg) was registered from (PGR₂) while the minimum total phenols (9.52 mg) was recorded from (PGR₀) (Table 14 and Appendix x).

The data on total phenols in fruits at harvest indicated non-significant differences in terms of different application stage. The maximum total phenols (9.83 mg) was registered in spray at 4-leaf stage and the minimum total phenols (9.69 mg) was recorded in seed soaking stage (Table 15 and Appendix x).

Interaction effect between plant growth regulators and different application stage was found to be non-significant of total phenols. Among the combination treatments, the total phenols was maximum (10.12 mg) from (PGR₀S₁) while the total phenols was minimum (9.41 mg) from (PGR₃S₃) (Table 16 and Appendix X).

Table 14: Effect of plant growth regulators on reducing sugar (mg. g fr.wt⁻¹), non-reducing sugar (mg. g fr.wt⁻¹), total sugar (mg. g fr.wt⁻¹) and total phenol (mg. g fr.wt⁻¹) in bitter gourd

Treatments	Reducing sugar	Non-reducing sugar	Total sugar	Total phenol
PGR ₀	0.40±0.00	3.86±0.03 ^b	4.26±0.03 ^b	9.52±0.10
PGR ₁	0.40±0.00	4.03±0.02 ^a	4.43±0.02 ^a	9.84±0.12
PGR ₂	0.39±0.00	4.02±0.04 ^a	4.43±0.04 ^a	9.90±0.03
PGR ₃	0.42±0.00	4.07±0.03 ^a	4.50±0.04 ^a	9.85±0.12
LSD _(.05)	0.03	0.14	0.15	0.42
CV%	5.91	3.25	3.05	3.92
P-value	0.10	0.00	0.00	0.07

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively. Values are mean ± SE.

Table 15: Effect of different application stage on reducing sugar (mg. g fr.wt⁻¹), non-reducing sugar (mg. g fr.wt⁻¹), total sugar (mg. g fr.wt⁻¹) and total phenol (mg. g fr.wt⁻¹) in bitter gourd

Treatments	Reducing sugar	Non-reducing sugar	Total sugar	Total phenol
S ₁	0.40±0.00	3.93±0.03	4.34±0.03	9.69±0.09
S ₂	0.41±0.00	4.02±0.02	4.43±0.03	9.83±0.10
S ₃	0.41±0.00	4.04±0.04	4.44±0.04	9.81±0.09
LSD _(.05)	0.02	0.13	0.12	0.33
CV%	5.91	3.25	3.05	3.92
P-value	0.80	0.07	0.06	0.54

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. Values are mean ± SE.

Table 16: Combined effect of plant growth regulators and different application stage on reducing sugar (mg. g fr.wt⁻¹), non-reducing sugar (mg. g fr.wt⁻¹), total sugar (mg. g fr.wt⁻¹) and total phenol (mg. g fr.wt⁻¹) in bitter gourd

Treatments	Reducing sugar	Non-reducing sugar	Total sugar	Total phenol
PGR ₀ S ₁	0.39±0.00	3.73±0.02	4.51±0.03	9.50±0.21
PGR ₀ S ₂	0.40±0.00	3.90±0.04	4.12±0.04	9.41±0.18
PGR ₀ S ₃	0.40±0.02	3.95±0.03	4.29±0.03	9.55±0.21
PGR ₁ S ₁	0.40±0.02	4.00±0.04	4.35±0.06	9.57±0.17
PGR ₁ S ₂	0.39±0.00	4.02±0.06	4.40±0.06	9.83±0.29
PGR ₁ S ₃	0.41±0.01	4.05±0.02	4.42±0.02	9.87±0.08
PGR ₂ S ₁	0.38±0.00	3.95±0.06	4.46±0.06	9.90±0.07
PGR ₂ S ₂	0.41±0.00	4.07±0.11	4.33±0.10	9.94±0.07
PGR ₂ S ₃	0.40±0.00	4.04±0.08	4.48±0.08	9.77±0.03
PGR ₃ S ₁	0.43±0.01	4.05±0.09	4.44±0.10	9.84±0.08
PGR ₃ S ₂	0.42±0.01	4.08±0.04	4.47±0.04	9.94±0.06
PGR ₃ S ₃	0.41±0.01	4.10±0.07	4.50±0.06	10.12±0.40
LSD _(.05)	0.06	0.32	0.33	0.947
CV%	5.91	3.25	3.05	3.92
P-value	0.61	0.81	0.72	0.73

Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage. Values are mean ± SE.

4.20.5. Water content (%)

The data on water content% in fruits at harvest indicated significant differences among the plant growth regulators. Among the treatments, the maximum water content (94.58 %) was registered in (PGR₃). The minimum water content (93.48 %) was recorded from (PGR₀) (Figure 14 and Appendix x).

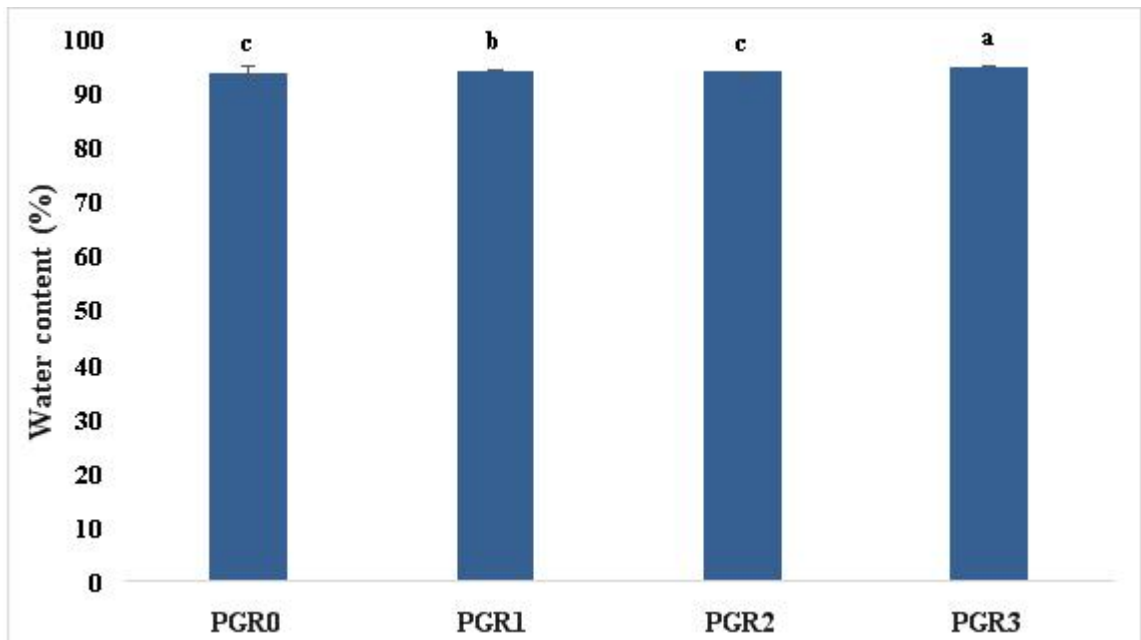


Figure 14. Effect of plant growth regulators on water content of fruit. Vertical bars represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH respectively

The data on water content% in fruits at harvest indicated significant differences in terms of different application stage. The maximum water content (94.12%) was registered from (S₃) and the minimum water content (93.56%) was recorded from (S₂) (Figure 15 and Appendix x).

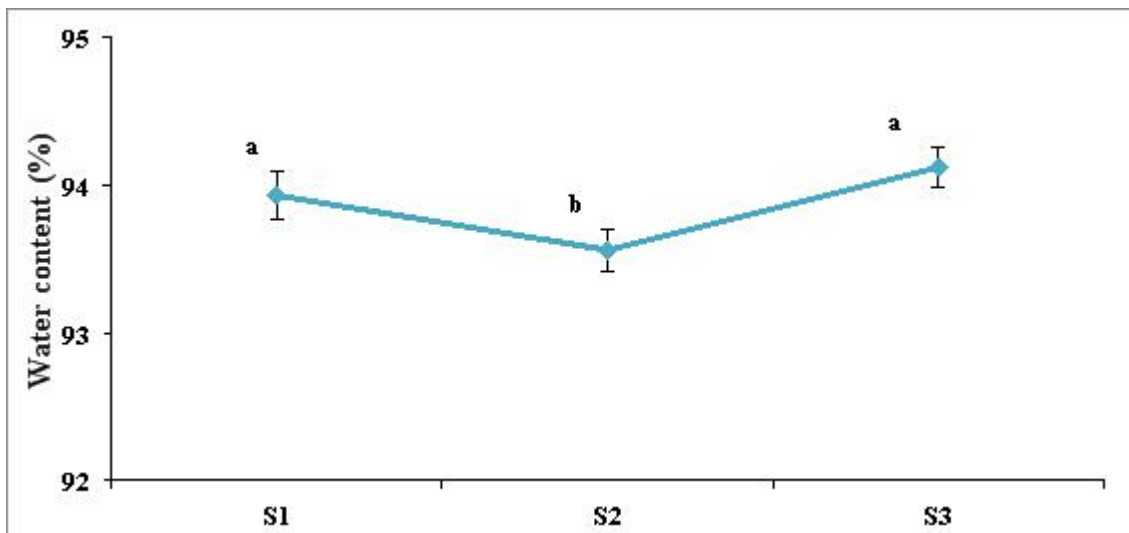


Figure 15. Effect of application stage on water content of fruit. Vertical bar represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows, S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage

Interaction effect between plant growth regulators and different application stage was found to be significant of water content (%). Among the combination treatments, the water content was recorded maximum (95.04 %) from PGR₀S₁ while the minimum (93.02 %) water content was recorded in the treatment combination from PGR₃S₃ (Figure 16 and Appendix x).

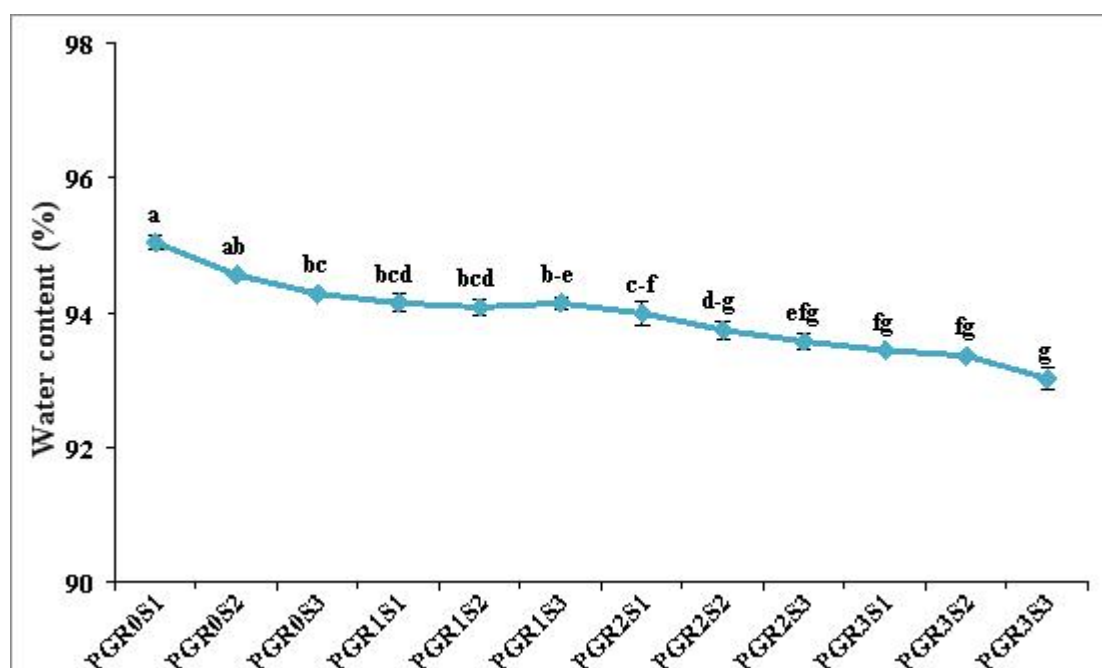


Figure 16. Combined effect of plant growth regulators and application stage on water content of fruit. Vertical bar represent means \pm standard error mean. Means with the same letter did not significantly differ from each other at $p < 0.05$. Abbreviations are as follows PGR₀; 0, PGR₁; 100 ppm GA₃, PGR₂; 100 ppm NAA and PGR₃; 100 ppm MH. S₁; seed soaking stage, S₂; 4-leaf stage and S₃; flower budding stage.

CHAPTER V

SUMMARY AND CONCLUSION

The effect of different doses of plant growth regulator at three application stages on the growth, sex expression and yield of bitter melon were studied during February 2016 to June 2016. The experimental site belongs to Tejgaon series under AEZ No.28 soil having clay loam in texture a 0.68% organic carbon in top soil. Four levels of plant growth regulators ((PGR₀: Control @ 0 ppm), PGR₁: GA₃ @ 100 ppm), PGR₂: NAA @ 100 ppm), PGR₃: MH @ 100 ppm) applied at three application stages of (seed soaking, 4-leaf stage and flower budding stage) in the study. Levels of these two factors made 12 treatment combinations. The experiment was carried out in a Randomized Complete Block Design with four replications.

At harvesting plant growth regulators had a significant effect on plant height. Plants grown with GA₃ showed a gradual increase in plant height. The tallest plant (456.50 cm) was produced by using PGR₁ (GA₃@ 100 ppm), while the shortest (362.67 cm) plant was observed from PGR₃ (Maleic hydrazide). In case of application stage, the tallest plant (423.68 cm) was produced by S₂ (spray at 4-leaf stage) and the shortest plant (394.87cm) was shown by S₁ (seed soaking). The treatment combinations demonstrated significant variation in plant height at 30, 60DAT and at harvesting. At harvesting the tallest plant (467.75 cm) was produced by PGR₁S₂ (GA₃ and spray at 4-leaf stage) while the shortest (360.50 cm) was shown from PGR₃S₃ (MH and spray at flower budding stage).

The highest (70.17 and 241.67) number of leaves plant⁻¹ at 30 DAT and 60 DAT was recorded from the application of MH (PGR₃) while the lowest (48.58 and 159.08) number of leaves plant⁻¹ was recorded in control condition at 30 DAT and 60 DAT. At last harvest highest (413.67) number of leaves was recorded for the application of MH (PGR₃) and the lowest (340.58) number of leaves plant⁻¹ at harvest was recorded in control condition (PGR₀). The highest (58.43 and 192.25) number of leaves plant⁻¹ at 30 DAT and 60 DAT was recorded from the application in spray at 4-leaf stage and seed soaking stage respectively and the lowest (53.50 and 181.06) number of leaves plant⁻¹ at 30 DAT and 60 DAT was recorded in seed soaking stage (S₁) and spray at 4-leaf stage. At last harvest the highest (379.93) number of leaves plant⁻¹ was recorded in spray at flower budding stage (S₃) while the lowest (371.50) number of leaves

plant⁻¹ was recorded in spray at 4-leaf stage (S₂). The highest (78.00, 243.50) number of leaves plant⁻¹ at 30 and 60 DAT that was recorded in the treatment combination of MH and spray at 4-leaf stage (PGR₃S₂) and the lowest (46.50, 146.25) number of leaves plant⁻¹ was recorded in the treatment combination from PGR₀S₃) and (PGR₀S₂) respectively. In case of harvest the highest (426.00) number of leaves plant⁻¹ was recorded from (PGR₃S₁) and the lowest (325.25) number of leaves plant⁻¹ was recorded from PGR₀S₁.

The maximum number of branches plant⁻¹ was found at PGR₃ (15.08) and the minimum value (12.92) was obtained from PGR₀. On the other hand, this parameter was also significantly influenced by application stages. It was the maximum 15.50 in S₂ when the minimum was in PGR₀. The maximum total number of branches plant⁻¹ (28.00) was given by the PGR₃S₂. The minimum number of branches plant⁻¹ (17.75) was recorded from combination of PGR₀S₁.

The maximum (25.95) number of female flower was recorded from MH (PGR₃) while the minimum (20.55) number of female flower in number was recorded from control condition (PGR₀). The maximum (24.77) number female flower was recorded from spray at 4-leaf stage (S₂) while the minimum (22.03) number female flower was recorded from seed soaking stage (S₁). The maximum (27.83) number of female flower was recorded from the treatment combination of (PGR₃S₂) while the minimum (19.50) number of female flower was recorded from combination treatment of (PGR₀S₁).

The maximum (2.74) ratio of male and female flower was recorded in control condition whereas the minimum (2.32) ratio of male and female flower was recorded for the application of MH (PGR₃). The maximum (2.69) ratio of male and female flower was recorded in seed soaking stage whereas the minimum (2.46) ratio of male and female flower was recorded in spray at 4-leaf stage (S₂). The maximum (2.84) was recorded in the treatment combination of (PGR₀S₁) while the minimum (2.19) ratio of male and female flower was recorded in the treatment combination of (PGR₃S₂).

Maximum (16.6 cm) fruit length was recorded for the application of GA₃ (PGR₁) while the minimum (12.83 cm) fruit length was recorded in control treatment (PGR₀). Maximum (16.95 cm) fruit length was recorded from (S₂) while the minimum (13.08

cm) fruit length was recorded in seed soaking stage (S_1). The maximum (19.52 cm) fruit length was recorded in the treatment combination of GA_3 and spray at 4-leaf stage (PGR_1S_2) while the minimum (9.97cm) was recorded in the treatment combination of control and seed soaking stage (PGR_0S_1).

The maximum (147.38g) fresh mass of fruit was recorded for the application of GA_3 (PGR_1) while the minimum (114.90g) fresh mass of fruit was recorded in control treatment (PGR_0). The maximum (144.22g) fresh mass of fruit was recorded in application of spray at 4-leaf stage while the minimum (128.02g) fresh mass of fruit was recorded in seed soaking stage (S_1). The maximum (156.26g) fresh mass of fruit was recorded in the treatment combination of GA_3 and spray at 4-leaf stage (PGR_1S_2) while the minimum (102.30g) was recorded in the treatment combination of control and seed soaking stage (PGR_0S_1).

The maximum (4.48 cm) fruit diameter was recorded for the application of GA_3 (PGR_1) while the minimum (3.86 cm) fruit diameter was recorded in control treatment (PGR_0). The maximum (4.56 cm) fruit diameter was recorded in application of spray at 4-leaf stage and the minimum (3.82 cm) fruit diameter was recorded in seed soaking stage (S_1). The maximum (4.99 cm) fruit diameter was recorded in the treatment combination of GA_3 and spray at 4-leaf stage (PGR_1S_2) and the minimum (3.36 cm) was recorded in the treatment combination of control and seed soaking stage (PGR_0S_1).

The maximum (9.02g) dry weight content of 100g fruit was recorded for the application of GA_3 (PGR_1) while the minimum (7.4g) dry weight content of 100g fruit was recorded in control (PGR_0). The maximum (9.24 g) dry weight content of 100g fruit was recorded in application of spray at 4-leaf stage while the minimum (7.65 g) dry weight content of 100g fruit was recorded in seed soaking stage (S_1). The maximum (10.37g) dry weight content of 100g fruit was recorded in the treatment combination of GA_3 and spray at 4-leaf stage (PGR_1S_2) while the minimum (6.80g) dry weight content of 100g fruit was recorded in the treatment combination of control and seed soaking stage (PGR_0S_1).

The maximum number of fruits $plant^{-1}$ (22.37) was recorded in maleic hydrazide while the minimum number of fruits $plant^{-1}$ (17.98) was recorded in control. The maximum (22.50) fruit number $plant^{-1}$ was recorded in application of spray at 4-leaf

stage (S_2) (Table) while the minimum (19.34) fruit number plant⁻¹ was recorded in seed soaking stage (S_1). The maximum (25.75) fruit number plant⁻¹ was recorded in the treatment combination of MH and spray at 4-leaf stage (PGR_3S_2) while the minimum (16.50) was recorded in the treatment combination of control and seed soaking stage (PGR_0S_0).

The maximum fruit yield (21.50 t ha⁻¹) was recorded from MH while the minimum fruit yield (13.85 t ha⁻¹) was recorded from control condition. The maximum fruit yield (21.77 t ha⁻¹) was registered in spray at flower budding stage and the minimum fruit yield (16.68 t ha⁻¹) was recorded in 4-leaf stage. The maximum (25.42 t ha⁻¹) fruit yield was recorded from PGR_3S_2 while the minimum (11.26 t ha⁻¹) fruit yield was recorded from the treatment combination of PGR_0S_1 .

The maximum non-reducing sugar (4.07 mg) was registered in MH and the minimum reducing sugar (3.86 mg) was recorded in control condition. The maximum non-reducing sugar (4.04 mg) was registered in spray at flower budding stage that and the minimum non-reducing sugars (3.93 mg) was recorded in seed soaking stage. The non-reducing sugar was maximum (4.10 mg) in the treatment combination of (PGR_3S_3) and the non reducing sugar was minimum (3.73 mg) in the treatment combination of control and seed soaking stage (PGR_0S_1).

The maximum total sugar (4.50 mg) was registered from (PGR_3) while the minimum total sugar (4.26 mg) was recorded in control condition. The maximum total sugar (4.44 mg) was registered in spray at flower budding stage while the minimum total sugar (4.34 mg) was recorded in seed soaking stage. The total sugar was maximum (4.51 mg) in the treatment combination of (PGR_0S_1) and the total sugar was minimum (4.12 mg) in the treatment combination of control and spray at 4-leaf stage (PGR_0S_1).

The maximum water content% (94.58%) was registered in (PGR_3). The minimum water content (93.48 %) was recorded from (PGR_0). The maximum water content (94.12%) was registered from (S_3) and the minimum water content (93.56%) was recorded from (S_2). The water content was recorded maximum (95.04%) from the treatment combination of (PGR_0S_1) while the minimum (93.02%) water content was recorded in the treatment combination from (PGR_3S_3).

As plants treated with various plant growth regulators at the vegetative stage potentially perform better on sex altering and fruit yield of bitter melon along with

nutritional values. Thereafter in comparing with the others, it could be concluded that maleic hydrazide @ 100 ppm possibly be used at 4-leaf stage in bitter gourd to get desirable fruit setting as well as fruit yield.

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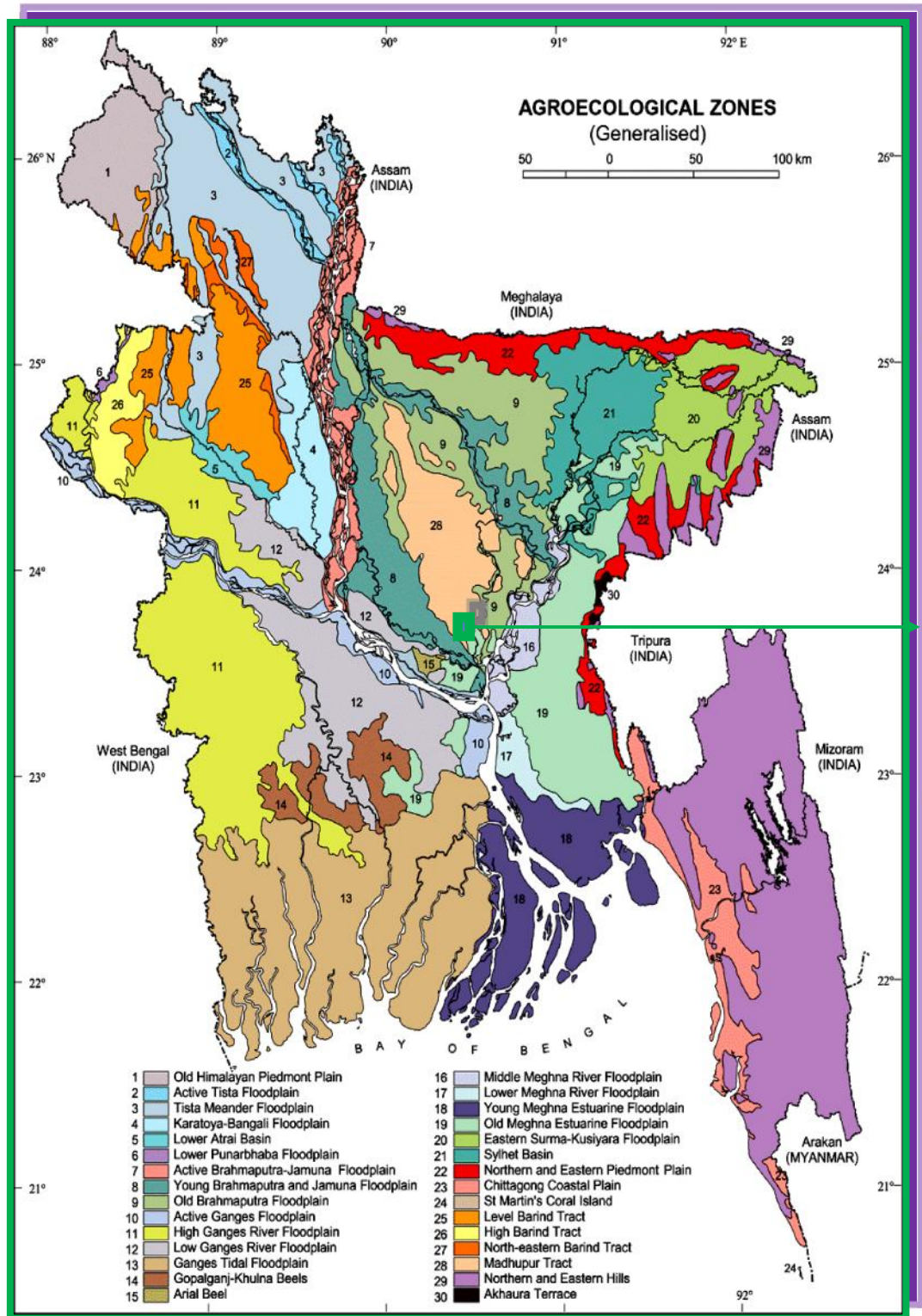
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APPENDICES

Appendix-i. Map showing the experimental site



The experimental site

Appendix -ii. Analytical data of soil sample of the experimental plot

A. Morphological Characteristics

Morphological features	Characteristics
Location	Horticulture Garden, SAU, Dhaka
AEZ	Modhupur Tract (28)
General Soil Type	Shallow red brown terrace soil
Land Type	Medium high land
Soil Series	Tejgaon
Topography	Fairly leveled
Flood Level	Above flood level
Drainage	Well drained

B. Mechanical analysis

Constituents	Percent
Sand	27
Silt	43
Clay	30

C. Chemical analysis

Soil properties	Amount
Soil pH	5.8
Organic carbon (%)	0.45
Total nitrogen (%)	0.03
Available P (ppm)	20
Exchangeable K (%)	0.1
Available S (ppm)	45

Source: Soil Resource Development Institute (SRDI)

Appendix-iii. Monthly records of air temperature, relative humidity, rainfall and sunshine during the period from January 2016 to May 2016.

Time	** Air temperature (⁰C)			**Relative humidity (%)	*Rainfall (mm)
	Maximum	Minimum	Mean		
January, 2016	25.18	18.26	21.72	64.23	25
February, 2016	28.79	22.54	25.66	65.53	32.1
March, 2016	32.32	24.40	28.36	84.06	60.00
April, 2016	35.77	28.17	31.97	87.65	92.12
May, 2016	34.77	25.49	30.13	89.21	120.10
June, 2016	33.67	24.34	29.00	88.12	220.23

*Monthly total, ** Monthly average, Source: Bangladesh Meteorological Department (Climate & weather division) Agargoan, Dhaka, Bangladesh

Appendix -iv. Analysis of variance on data with the effect of plan growth regulators and application stage on days to germination of bitter gourd.

Source of variation	Degrees of freedom	Mean square of days to germination
Replication	3	2.16667
Factor A	3	0.66667 ^{ns}
Factor B	2	0.43750 ^{ns}
AB	6	0.93750 ^{ns}
Error	33	0.54545

ns: Non-significant

Appendix-v. Analysis of variance on data with the effect of plant growth regulators and application stage on height of plant⁻¹ (cm), number of leaves plant⁻¹ and number of branch plant⁻¹ of bitter gourd at different days after transplanting (DAT).

Source of variation	Degrees of freedom	Mean square of								
		Height of plant ⁻¹ at			Number of leaves plant ⁻¹ at			Number of branch plant ⁻¹ at		
		30 DAT	60 DAT	Harvest	30 DAT	60 DAT	Harvest	30 DAT	60 DAT	Harvest
Replication	3	29.47	158.52	96.4	0.08	4.1	212.2	0.2500	1.1389	0.3889
Factor A	3	2400.25**	8995.02*	21951.0**	1287.36**	1616.53**	11055.2**	3.8056**	9.6944**	88.5000**
Factor B	2	1000.77**	3107.69*	3338.8**	103.19**	764.3**	337.9 ⁿ	21.3958**	27.7500**	47.2708**
AB	6	33.69**	165.35 ^{ns}	123.4 ⁿ	195.13**	750.1**	804.7*	0.1181 ^{ns}	0.4444 ⁿ	1.3542 ^{ns}
Error	33	3.49	143.16	65.5	2.78	3.4	316.3	0.3106	0.6237	0.7374

** : at <0.01 level of probability, ns: non-significant * : at <0.05 level of probability

Appendix -vi. Analysis of variance on data with the effect of plant growth regulators and application stage on days to first flowering, number of male flower, number of female flower and sex ratio (male: female) of bitter gourd.

Source of variation	Degrees of freedom	Days to first flowering	Mean square of		
			Male flower	Female flower	Sex ratio
Replication	3	0.038	—	—	0.36451
Factor A	3	151.076**	75.4097**	60.0100**	0.23550**
Factor B	2	11.459**	8.2708**	31.5431**	0.01342**
AB	6	1.246**	3.6597 ^{ns}	1.5675*	0.01342 ^{ns}
Error	33	0.256	7.1736	0.4734	0.01835

** : at <0.01 level of probability, ns: non-significant * : at <0.05 level of probability

Appendix- vii. Analysis of variance on data with the effect of plant growth regulators and application stage on fruit setting, fruit length, fruit diameter, fresh mass of fruit⁻¹ , dry matter content of fruit, number of fruit plant⁻¹ and yield t ha⁻¹ of bitter gourd.

Source of variation	Degrees of freedom	Mean square of						
		Fruit setting (%)	Fruit length (cm)	Fruit diameter (cm)	Fresh mass of fruit ⁻¹ (g)	Dry matter content of Fruit (%)	Number of fruit Plant ⁻¹	Yield t ha ⁻¹
Replication	3	29.47	3.7455	0.01734	90.38	0.0627	–	–
Factor A	3	56.8472 ^{ns}	30.8173**	1.18551**	2633.43**	7.7605**	46.6586**	159.172**
Factor B	2	66.0685 ^{ns}	59.7340**	2.18003**	1069.48**	10.6989**	40.2206**	103.599**
AB	6	50.2621 ^{ns}	3.0987**	0.15294**	51.93**	0.5832**	6.7834**	5.607**
Error	33	23.9752	0.5378	0.01749	14.03	0.0365	1.1476	1.582

** : at<0.01 level of probability, ns: non-significant *: at<0.05 level of probability

Appendix -viii. Analysis of variance on data with the effect of plant growth regulators and application stage on root length at harvest of bitter gourd.

Source of variation	Degrees of freedom	Mean square of root length at harvest (cm)
Replication	3	0.9602
Factor A	3	5.5519 ^{ns}
Factor B	2	13.0300 ^{ns}
AB	6	4.5800 ^{ns}
Error	33	4.0011

ns: Non-significant

Appendix -ix. Analysis of variance on data with the effect of plant growth regulators and application stage of fresh mass of plant at harvest, fresh mass of root at harvest, dry matter content of root (%) at harvest of bitter gourd.

Source of variation	Degrees of freedom	Mean square of		
		Fresh mass of plant at harvest	Root fresh weight at harvest	Dry matter content of root (%)
Replication	3	3.7455	2.62630	1.54630
Factor A	3	13.5236**	0.00886 ^{ns}	2.94836**
Factor B	2	11.1223**	2.42479 ^{ns}	0.80583**
AB	6	1.15163**	2.31206 ^{ns}	0.07792**
Error	33	4.04431	1.71403	1.63132

** : at<0.01 level of probability, ns: non-significant * : at<0.05 level of probability

Appendix -x. Analysis of variance on data with the effect of plant growth regulators and application stage on reducing sugar, non-reducing sugar, total sugar, total phenol and water content of bitter gourd.

Source of variation	Degrees of freedom	Mean square of				Water content (%)
		Reducing sugar	Non-reducing sugar	Total sugar	Total phenol	
Replication	3	0.00044	3.7455		2.62630	0.0627
Factor A	3	5.55938 ^{ns}	0.11003 ^{ns}	0.12768**	0.37842 ^{ns}	2.92749**
Factor B	2	5.84904 ^{ns}	0.04873 ^{ns}	0.05456 ^{ns}	0.08981 ^{ns}	1.30441**
AB	6	2.71518 ^{ns}	0.00817 ^{ns}	0.01086 ^{ns}	0.08658 ^{ns}	0.54960**
Error	33	0.00035	0.01688	0.01799	0.14733	0.05940

** : at<0.01 level of probability * : at<0.05 level of probability ns: non-significant