

## Optimizing Sponge Gourd Cultivation: Effects of CHAs on Morphological Traits and Flower Development

Commented [k1]: Avoid Abbreviation in Title of the Article

### Abstract

The study investigates the effects of various chemical hybridizing agents (CHAs) on the morphological traits of Sponge gourd (*Luffa cylindrica*) over two growing seasons (2023-2024 and 2024-2025) at the Vegetable Research Centre, JNKVV, Jabalpur. The research primarily focuses on vine length, number of primary branches, internodal length, nodes per vine, and flower development stages. Statistical analysis of the collected data reveals that the application of CHAs significantly enhances vine length at 30, 60, and 90 days after transplanting (DAT), with the highest growth observed in treatments involving GA3 (200 ppm) and Sulphonyl Urea (0.4 ml/L). Notably, vine lengths reached up to 6.88 m at final harvest for the most effective treatments. Furthermore, the number of primary branches and nodes per vine also increased substantially with CHA application, particularly with GA3 and Sulphonyl Urea. The timing of flower appearance was influenced by these treatments as well, with earlier flowering noted in Ethrel-treated plants. The study concludes that strategic application of CHAs can significantly improve growth parameters and flowering dynamics in Sponge gourd cultivation, suggesting potential for enhanced yield and agricultural practices. This research contributes valuable insights for horticulturists aiming to optimize growth conditions for this economically important crop.

Commented [k2]: italics

**Keywords:** *Luffa cylindrica*, Vine Morphology, Growth Regulators, Flowering Phenology, Agronomic Performance.

### 1. Introduction

Sponge gourd (*Luffa cylindrica* (L.) Roem.), a member of the Cucurbitaceae family, is an important vegetable crop cultivated extensively in tropical and subtropical regions. The crop holds significant economic importance due to its dual-purpose utility – both as a vegetable in its tender form and as a natural sponge when mature. In recent years, there has been growing interest in understanding and manipulating the growth, development, and sex expression patterns of this crop to enhance its commercial viability and productivity.

Sex expression in cucurbitaceous vegetables, including sponge gourd, is a complex physiological process influenced by various genetic, environmental, and hormonal factors. The predominant expression of male flowers over female flowers in most cultivars often results in reduced fruit set and yield. Plant growth regulators (PGRs) and chemical hybridizing agents (CHAs) have emerged as powerful tools for modifying plant architecture, sex expression, and reproductive biology in various crops.

Recent research by Soni et al. (2015) demonstrated that foliar application of gibberellic acid (GA3) significantly enhanced vine length and primary branch formation in bottle gourd, leading to improved vegetative growth characteristics. Their findings showed a 23% increase in vine length

Commented [k3]: italics

with GA3 application at 200 ppm. Similarly, Matsumara [et al., \(2020\)](#) reported that GA3 application in bitter gourd not only promoted vegetative growth but also influenced the timing of flower initiation and sex ratio.

Commented [k4]: italics

The role of plant growth regulators in modifying internodal length has been well-documented. A comprehensive study by Zhang [et al. \(2022\)](#) in cucumber revealed that ethrel application significantly reduced internodal length while promoting female flower formation. They observed that ethrel at 1000 ppm reduced internodal length by 18% compared to control plants. This architectural modification has important implications for plant management and yield potential.

Commented [k5]: italics

The manipulation of sex expression using chemical hybridizing agents has gained considerable attention in cucurbit breeding programs. Research conducted by Arora [et al., \(1982\)](#) showed that applications of maleic hydrazide and ethrel effectively increased female flower production in Bottle gourd. Their work demonstrated a 40% increase in female flowers following ethrel application at the 2-4 leaf stage.

Commented [k6]: italics

The timing of CHA application plays a crucial role in determining its effectiveness. Barcnas [et al. \(2023\)](#) investigated the stage-specific responses of cucumber to various growth regulators and found that applications during the early vegetative phase (2-4 leaf stage) produced optimal results in terms of growth modification and sex expression. These findings align with earlier work by Vidyullatha [et al. \(2022\)](#), who reported enhanced efficacy of naphthalene acetic acid when applied at similar growth stages in watermelon.

Commented [k7]: italics

Commented [k8]: italics

The interaction between different plant growth regulators and environmental conditions significantly influences their effectiveness. A detailed study by Lai [et al \(2022\)](#) revealed that temperature and photoperiod conditions during CHA application could significantly affect their absorption and subsequent physiological responses in cucurbits. They found that applications during morning hours under optimal temperature conditions (25-28°C) showed maximum effectiveness.

Commented [k9]: italics

Recent advances in understanding the molecular mechanisms underlying sex expression in cucurbits have provided new insights into the mode of action of various CHAs. Research by Li [et al. \(2019\)](#) demonstrated that ethrel application influences the expression of sex-determining genes through ethylene-mediated pathways, leading to modified flower sex ratios. Their work has opened new avenues for targeted manipulation of sex expression in cucurbitaceous crops.

Commented [k10]: italics

Additionally, Priyadarshi [et al \(2022\)](#) reported that the combined application of growth regulators at different stages could produce synergistic effects on plant growth and development. Their studies in bitter gourd showed that sequential applications of GA3 and ethrel resulted in optimal plant architecture and improved fruit set compared to single applications.

Commented [k11]: italics

## 2. Material and methods

### 2.1 Experimental Site

The present study was conducted at the Vegetable Research Centre, Maharajpur, under the Department of Horticulture, Jawaharlal Nehru Krishi Vishwa Vidyalaya (JNKVV), Jabalpur,

Madhya Pradesh, during the 2023-24 growing season. The experimental site is geographically located at 23°22'34.93" N latitude and 79°96'35.26" E longitude, at an altitude of 300 meters above mean sea level. The soil utilized for the experiment is classified as laterite, characterized by high drainage capacity and a homogeneous texture. It exhibits moderate water retention and rich in iron and aluminum, typically forming in hot, humid subtropical climates.

## 2.2 Experimental Details

The experimental design employed in this study was a Randomized Block Design (RBD) with three replications, aimed at evaluating the effects of various treatments on sponge gourd (*Luffa cylindrica*). The specific variety used was Jawahar Gilki 14, and the experiment was conducted over two growing seasons: 2023-2024 and 2024-2025. A total of 13 distinct treatments were applied across the experimental plots, which were organized into blocks to control for variability. Each block consisted of plots measuring 15.00 m<sup>2</sup>, resulting in a total of 39 plots (13 treatments multiplied by 3 replications). To ensure optimal growth conditions, basal nutrients were applied in the form of 10 tons of Farm Yard Manure, along with 45 kg of phosphorus (P<sub>2</sub>O<sub>5</sub>) and 50 kg of potassium (K<sub>2</sub>O) per hectare. This structured approach allows for a comprehensive analysis of treatment effects while minimizing the influence of environmental variability. A detailed list of treatments is provided in Table 1.

**Table 1.** Description of treatments involving different Chemical hybridizing agents and their concentrations applied at two growth stages (cotyledon stage: 10-12 DAS; true leaf stage: 24-28 DAS) in sponge gourd.

| Treatment      | Chemical                   | Concentration | Application Timing          |
|----------------|----------------------------|---------------|-----------------------------|
| T <sub>1</sub> | Maleic Hydrazide           | 200 ppm       | Cotyledon stage (10-12 DAS) |
| T <sub>2</sub> | Gibberellic acid           | 200 ppm       | Cotyledon stage (10-12 DAS) |
| T <sub>3</sub> | 2,3,5-Triiodo Benzoic acid | 25 ppm        | Cotyledon stage (10-12 DAS) |
| T <sub>4</sub> | 1-Naphthalene acetic acid  | 100 ppm       | Cotyledon stage (10-12 DAS) |
| T <sub>5</sub> | Ethrel                     | 1250 ppm      | Cotyledon stage (10-12 DAS) |
| T <sub>6</sub> | Sulphonyl urea             | 0.4 ml/L      | Cotyledon stage (10-12 DAS) |
| T <sub>7</sub> | Maleic Hydrazide           | 200 ppm       | True leaf stage (24-28 DAS) |
| T <sub>8</sub> | Gibberellic acid           | 200 ppm       | True leaf stage (24-28 DAS) |

**Commented [k12]:** mention the season in respective years

| Treatment       | Chemical                   | Concentration | Application Timing          |
|-----------------|----------------------------|---------------|-----------------------------|
| T <sub>9</sub>  | 2,3,5-Triiodo Benzoic acid | 25 ppm        | True leaf stage (24-28 DAS) |
| T <sub>10</sub> | 1-Naphthalene acetic acid  | 100 ppm       | True leaf stage (24-28 DAS) |
| T <sub>11</sub> | Ethrel                     | 1250 ppm      | True leaf stage (24-28 DAS) |
| T <sub>12</sub> | Sulphonyl urea             | 0.4 ml/L      | True leaf stage (24-28 DAS) |
| T <sub>13</sub> | Control                    | -             | -                           |

### 2.3 Morphological Parameters

This section details the methodologies used to assess various morphological parameters of the vines throughout their growth period, with measurements taken at specified intervals and at final harvest.

**2.3.1 Vine Length Measurements:** Vine length was measured at 30, 60, and 90 days after transplanting (DAT), as well as at final harvest. Each vine's length was recorded in centimeters from the ground to the tip, and mean values were calculated for each time point.

**2.3.2 Number of Primary Branches per Vine:** At final harvest, the total number of primary branches per vine was counted for each treatment group, and mean values were calculated for comparison.

**2.3.3 Internodal Length:** Internodal length was measured using a centimeter scale to determine the distance between nodes on the vines. The average internodal distance was computed for each treatment.

**2.3.4 Number of Nodes per Vine:** The total number of nodes per vine was counted at final harvest for each treatment group, with mean values calculated to evaluate overall productivity.

**2.3.5 Nodes at Which First Female Flower Appeared:** The node number where the first female flower appeared was recorded for each vine across treatments, and mean values were calculated to assess flowering initiation.

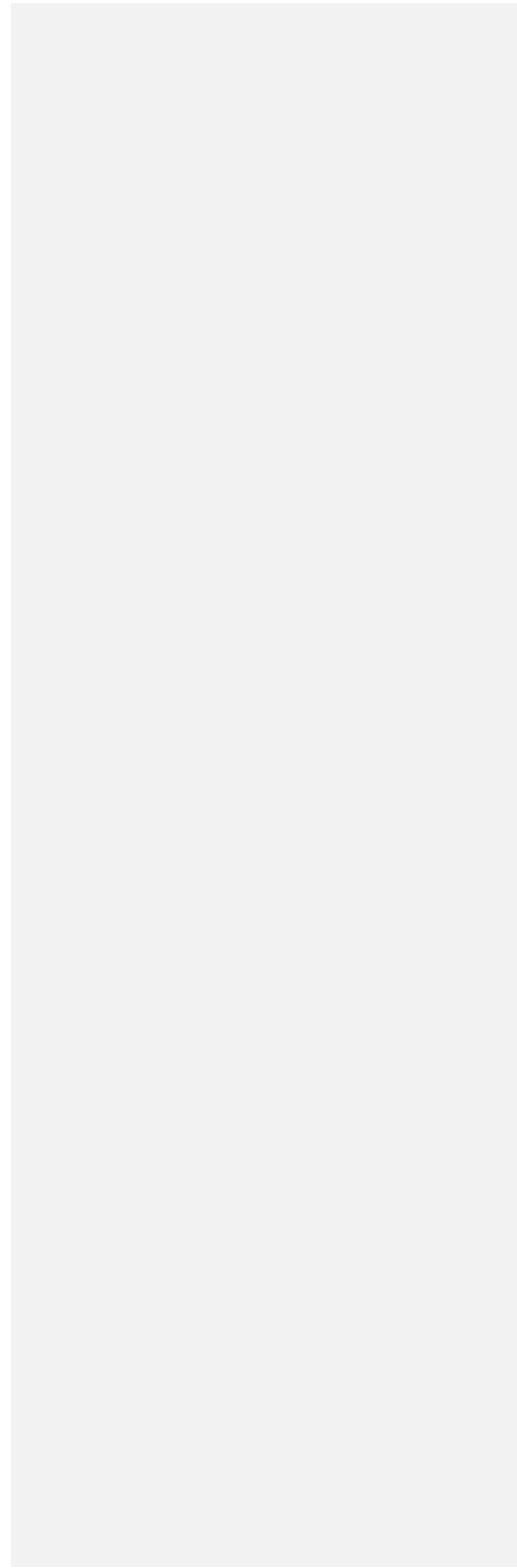
**2.3.6 Nodes at Which First Male Flower Appeared:** The node number for the first male flower appearance was documented for each vine, with mean values calculated to evaluate male flowering timing relative to females.

**2.3.7 Number of Female Flowers per Vine:** The count of female flowers began with the first appearance and continued at regular intervals until final harvest, providing an accurate assessment of female flowering.

**2.3.8 Number of Male Flowers per Vine:** The number of male flowers was recorded from the emergence of the first male flower up to final harvest, with counts taken at regular intervals.

**2.3.9 Sex Ratio:** The sex ratio was determined by comparing the total numbers of male and female flowers, computed as the ratio of male to female flowers, offering insights into sexual expression across treatments.

UNDER PEER REVIEW



### 3. RESULT

The results obtained from the investigation of Sponge gourd (*Luffa cylindrica* (L.) Roem.)” was conducted at Vegetable Research Centre (VRC) Maharajpur, Department of Horticulture, JNKVV, Jabalpur, during summer season of the year 2023-24 and 2024-25. The data collected during experimental period on various characters were analysed statistically and the results are presented below.

With regards to morphological traits vine length (m) at (30, 60, 90 DAS and final harvest), Internodal length, Number of primary branches, Number of nodes per vine and other parameters affected by application of chemicals at different stages described in details:

#### 3.1 Vine length at 30 DAT

The data presented in table 2 and Fig.1 showed that application of CHAs at different stages significantly promoted the vine length at 30 DAT in both the years. The maximum vine length was found to result in T<sub>8</sub> - GA<sub>3</sub> 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) 3.88 m, 4.27 m and 4.07 m, respectively, in first year, second year and pooled data which was at par with T<sub>12</sub> - Sulphonyl Urea 0.4 ml/L - Spray at 10 and 24 DAS (2-4 Leaf Stage) with vine length 3.62 m, 4.18 m and 3.90 m respectively, in first year, second year and pooled data followed by T<sub>10</sub> -1-Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with vine length 3.09 m, 3.75 m and 3.42 m. The minimum vine length was observed in T<sub>9</sub> -2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) i.e.1.91 m,1.75 m and 1.83 m respectively, in first year, second year and pooled data. The vine length increased significantly increase with stages of application.

#### 3.2 Vine length at 60 DAT

According to the data in the table 2 and Fig.1, the vine length at 60 DAT in both years was considerably increased by the use of CHAs at various stages. The longest vine length was found to produce T<sub>8</sub>-GA<sub>3</sub>-200 ppm-Spray at 10 and 24 DAS (2-4 Leaf Stage) 5.61 m, 4.92 m, and 5.27 m, respectively, in the first year, second year and pooled data. This was at par with T<sub>12</sub>-1-Sulphonyl Urea 0.4 ml/L-Spray at 10 and 24 DAS (2-4 Leaf Stage) with vine lengths of 5.08 m, 4.86 m, and 4.97 m, respectively, in the first year, second year and pooled data followed by T<sub>2</sub> -GA<sub>3</sub> 200 ppm - Spray at 10 DAS (Cotyledon Stage) in first year and pooled data (4.78 m and 4.80 m) while T<sub>10</sub>-1-Naphthalene Acetic Acid 100 ppm-Spray at 10 and 24 DAS (2-4 Leaf Stage) in second year (4.84 m). In the first, second and pooling data, the shortest vine length was found in T<sub>9</sub>-2,3,5-Triiodo Benzoic Acid 25 ppm-Spray at 10 and 24 DAS (2-4 Leaf Stage) with 3.76 m, 2.99 m and 3.38 m, respectively. With each application stage, the vine length rose noticeably.

#### 3.3 Vine length at 90 DAT

The data in the table 2 and Fig.1 indicates that the use of CHAs at different stages significantly increased the vine length at 90 DAT in both years. In the first year, second year and pooled data, the maximum vine length produced in T<sub>8</sub>-GA<sub>3</sub>-200 ppm-Spray at 10 and 24 DAS (2-4 Leaf Stage) 6.61 m, 6.01 m and 4.98 m, respectively. This was at par with T<sub>12</sub>-1-Sulphonyl Urea 0.4 ml/L-Spray at 10 and 24 DAS (2-4 Leaf Stage) with vine lengths of 6.23 m and 6.02 m, respectively, in

**Commented [k13]:** in methodology it is mentioned that vine length is expressed in Cm but here m, use most appropriate unit, uniformly in whole manuscript

the first year and pooled data while 5.97 m in T<sub>10</sub> - 1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) in second year followed by T<sub>2</sub>-GA<sub>3</sub>- 200 ppm-Spray at 10 DAS (Cotyledon Stage) in the first year and pooled data (5.84 m and 5.88 m) while T<sub>7</sub> - Maleic Hydrazide 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) in the second year (5.93 m). T<sub>9</sub>-2,3,5-Triiodo Benzoic Acid 25 ppm-Spray had the shortest vine length in the first, second, and pooled data at 10 and 24 DAS (2-4 Leaf Stage), measuring 5.00 m, 4.90 m, and 4.98 m, respectively. The vine length increased significantly with each application stage.

### 3.4 Vine length at final harvest

The data in the table 2 and Fig.1 showed that the vine length at final harvest in both years was considerably boosted by the application of CHAs at various stages. The maximum vine length generated in T<sub>8</sub>-GA<sub>3</sub>-200 ppm-Spray at 10 and 24 DAS (2-4 Leaf Stage) was 6.88 m and 6.66 m in first year and pooled data, respectively while 6.74 m in T<sub>10</sub> - 1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) in second year. This was at par with T<sub>12</sub>-1-Sulphonyl Urea 0.4 ml/L-Spray at 10 and 24 DAS (2-4 Leaf Stage) with vine lengths of 6.50 m, 6.56 m and 6.53 m, respectively, in the first year, second year and pooled data followed by T<sub>5</sub> - Ethrel 1250 ppm - Spray at 10 DAS (Cotyledon Stage) with 6.41 m in first year, T<sub>2</sub>-GA<sub>3</sub>- 200 ppm-Spray at 10 DAS (Cotyledon Stage) with 6.51 m in second year and T<sub>10</sub> - 1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 6.36 m in pooled data. In the first year T<sub>2</sub> -Sulphonyl Urea 0.4 ml/L - Spray at 10 DAS (Cotyledon Stage) with 5.58 m in first year while in second year and pooled data T<sub>9</sub>-2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) had the shortest vine length, measuring 5.47 m and 5.55 m, respectively. With every stage of application, the vine's length considerably grew.

Commented [k14]: express in cm or m

**Table 2. Effect of chemical hybridizing agents on Vine length (m) at 30, 60, 90 and Final harvest**

Commented [k15]: unit m / cm

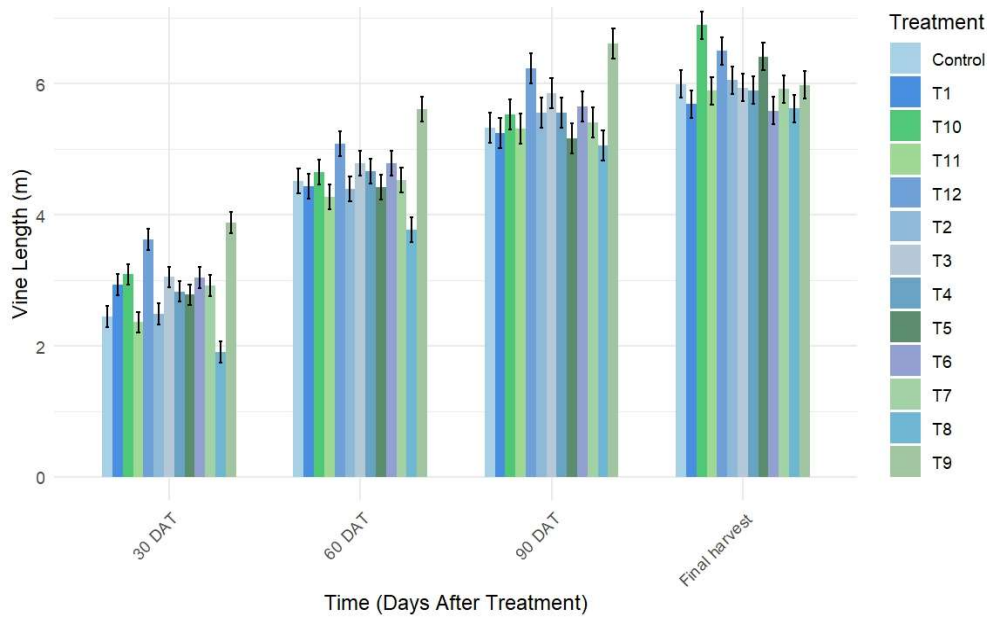
| S.No.          | Treatments   | Vine length (m) |           |           |                  |
|----------------|--|-----------------|-----------|-----------|------------------|
|                |  | At 30 DAT       | At 60 DAT | At 90 DAT | At Final harvest |
| T <sub>1</sub> | Maleic Hydrazide 200 ppm - Spray at 10 DAS (Cotyledon Stage)           | 2.93            | 4.43      | 5.25      | 5.69             |
| T <sub>2</sub> | 2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 DAS (Cotyledon Stage)  | 2.49            | 4.39      | 5.56      | 6.05             |
| T <sub>3</sub> | GA 3 200 ppm - Spray at 10 DAS (Cotyledon Stage)                       | 3.05            | 4.79      | 5.85      | 5.94             |
| T <sub>4</sub> | 1- Naphthalene Acetic Acid 100 ppm - Spray at 10 DAS (Cotyledon Stage) | 2.83            | 4.67      | 5.56      | 5.90             |
| T <sub>5</sub> | Ethrel 1250 ppm - Spray at 10 DAS (Cotyledon Stage)                    | 2.78            | 4.42      | 5.16      | 6.41             |
| T <sub>6</sub> | Sulphonyl Urea 0.4 ml/L - Spray at 10 DAS (Cotyledon Stage)            | 3.04            | 4.79      | 5.65      | 5.59             |
| T <sub>7</sub> | Maleic Hydrazide 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage)     | 2.92            | 4.53      | 5.41      | 5.92             |

Commented [k16]: unit

|                 |  |      |      |      |      |
|-----------------|--|------|------|------|------|
| T <sub>8</sub>  | 2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 DAS (Cotyledon Stage)        | 1.91 | 3.77 | 5.06 | 5.62 |
| T <sub>9</sub>  | GA 3 200 ppm - Spray at 10 DAS (Cotyledon Stage)                             | 3.88 | 5.61 | 6.61 | 5.98 |
| T <sub>10</sub> | 1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) | 3.09 | 4.65 | 5.53 | 6.89 |
| T <sub>11</sub> | Ethrel 1250 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage)                    | 2.36 | 4.27 | 5.31 | 5.89 |
| T <sub>12</sub> | Sulphonyl Urea 0.4 ml/L - Spray at 10 and 24 DAS (2-4 Leaf Stage)            | 3.62 | 5.08 | 6.23 | 6.50 |
| T <sub>13</sub> | Control  | 2.45 | 4.51 | 5.32 | 5.99 |
|                 | C.D. at 5%   | 0.48 | 0.57 | 0.67 | 0.63 |
|                 | SE(m) ±  | 0.16 | 0.19 | 0.23 | 0.21 |
|                 | C.V.   | 9.89 | 7.29 | 7.14 | 6.2  |

**Commented [k17]:** since it is a two seasons experiment the data table should include season 1 avg, season 2 avg and pooled data, applies for all the tables in the manuscript

**Fig. 1 :** Effect of chemical hybridizing agents on Vine length (m) at 30, 60, 90 and Final harvest





### 3.5 Number of primary branches

The data depicted in Table 3 and Fig.2, application of CHAs considerably enhanced the number of primary branches per vine in both years. The maximum number of primary branches per vine was observed with the application of treatment T<sub>8</sub>-GA<sub>3</sub>-200 ppm-Spray at 10 and 24 DAS (2-4 Leaf Stage) of 10.07, 10.67 and 10.37 branches first year, second year and pooled data, respectively at par with T<sub>12</sub>-1-Sulphonyl Urea 0.4 ml/L-Spray at 10 and 24 DAS (2-4 Leaf Stage) with 9.00 in first year and T<sub>4</sub>- 1- Naphthalene Acetic Acid 100 ppm - Spray at 10 DAS (Cotyledon Stage) with 10.17 in second year and 8.85 in pooled data. It was observed that all the treatments differed significantly from one another. In the first year, second year and pooled data. The T<sub>1</sub>-Maleic Hydrazide 200 ppm - Spray at 10 DAS (Cotyledon Stage) with 6.20 in first year and 6.82 in pooled data while control (T<sub>13</sub>) group with 7.17 had the smallest number of primary branches per vine.

### 3.6 Internodal length (cm)

Application of CHAs considerably extended the internodal length in both years, according to data in Table 3 and Fig.2. The internodal length grew as the number of sprays increased and it was evident that all of the treatments differed significantly from one another. The application of T<sub>8</sub>-GA<sub>3</sub>-200 ppm-Spray at 10 and 24 DAS (2-4 Leaf Stage) resulted in the maximum internodal lengths of 15.53 cm, 15.58 cm and 15.69 cm in the first, second and pooled data, respectively which is at par with T<sub>12</sub>-1-Sulphonyl Urea 0.4 ml/L-Spray at 10 and 24 DAS (2-4 Leaf Stage) of 13.75 cm in first year and T<sub>2</sub> -Sulphonyl Urea 0.4 ml/L - Spray at 10 DAS (Cotyledon Stage) 15.80 cm and 14.72 cm in second year and pooled data. In first year, second year and pooled data, T<sub>3</sub> - 2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 DAS (Cotyledon Stage) were shown to have the lowest internodal lengths of 10.48 cm, 11.05 cm and 10.77 cm, respectively.

Commented [k18]: replace with growing seasons

### 3.7 Number of nodes per vine

The data framed in table 3 and Fig.2 indicates that the number of nodes per vine were significantly influenced by application of different CHAs treatment at different growing stages. The maximum number of nodes per vine was recorded in T<sub>8</sub>-GA<sub>3</sub>-200 ppm-Spray at 10 and 24 DAS (2-4 Leaf Stage) with 62.53, 61.00 and 61.77 in the first, second and pooled data which is at par with 59.66 at T<sub>12</sub>-1-Sulphonyl Urea 0.4 ml/L-Spray at 10 and 24 DAS (2-4 Leaf Stage) in second year followed by T<sub>12</sub>-1-Sulphonyl Urea 0.4 ml/L-Spray at 10 and 24 DAS (2-4 Leaf Stage) of 54.46 and 57.07 in first year and pooled data while T<sub>2</sub> -Sulphonyl Urea 0.4 ml/L - Spray at 10 DAS (Cotyledon Stage) of 58.66 in second year. The minimum nodes per vine was observed in T<sub>3</sub> - 2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 DAS (Cotyledon Stage) with 43.87, 52.16 and 48.02 in the first, second and pooled data, respectively.

### 3.8 Node at which first male flower appeared

The data depicted in table 3 and Fig.2 showed significant variance among the different levels of CHAs application on the node at which first male flower appeared. The earliest node for the first male flower appeared was recorded in T<sub>11</sub> -Ethrel 1250 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 15.26 and 13.22 in first year and pooled data while T<sub>7</sub> Maleic Hydrazide 200 ppm -

Spray at 10 and 24 DAS (2-4 Leaf Stage) with 10.50 in second year which is at par with T<sub>5</sub> -Ethrel 1250 ppm - Spray at 10 DAS (Cotyledon Stage) of 17.73 in first year, T<sub>1</sub> - Maleic Hydrazide 200 ppm - Spray at 10 DAS (Cotyledon Stage) and T<sub>11</sub> -Ethrel 1250 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 11.17 in second year while T<sub>1</sub> - Maleic Hydrazide 200 ppm - Spray at 10 DAS (Cotyledon Stage) with 14.92 in pooled data. However, T<sub>7</sub> -Maleic Hydrazide 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 23.46 in first year, T<sub>10</sub> -1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 19.83 in second year and T<sub>13</sub> - Control with 20.57 in pooled data was depicted as maximum number of node for first male flower appeared.

### 3.9 Node at which first female flower appeared

According to the data presented in the table 3 and Fig.2, the administration of various CHAs treatments at various growth phases had a substantial impact on the node at which first female flower appeared. The earliest node for first female flower appeared was found in T<sub>11</sub> -Ethrel 1250 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 18.20, 15.17 and 16.68 in first, second and pooled data, which is at par with T<sub>10</sub> -1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) of 19.60 in first year, T<sub>1</sub> - Maleic Hydrazide 200 ppm - Spray at 10 DAS (Cotyledon Stage) of 18.83 in second year. However, the maximum number of nodes for the first female flower appearance was shown in T<sub>7</sub> -Maleic Hydrazide 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 27.40 in first year and T<sub>13</sub> - Control with 32.83 and 29.55 in second year and pooled data, respectively.

### 3.10 Number of male flowers per vine

The data presented in table 3 and Fig.2 showed significant fluctuations were recorded for the number of male flowers among the different levels of CHAs application at different stages. During the assessment, the maximum number of male flowers were observed in T<sub>11</sub> -Ethrel 1250 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 214.93 in first year and 322.33, 267.57 in second year and pooled data which is at par with T<sub>7</sub> -Maleic Hydrazide 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) of 213.93 in first year while T<sub>11</sub> -Ethrel 1250 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 302.83, 258.88 second year and pooled data. The minimum numbers of male flowers was recorded in control i.e. 166.06, 223.50 and 194.78 in first year, second year and pooled data, respectively.

### 3.11 Number of female flowers per vine

The data presented in table 3 and Fig.2 reveals that the number of female flowers was significantly influenced by different levels of CHAs application at different stages. During the experiment, the maximum number of female flowers were observed in T<sub>10</sub> -1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 20.80, 25.33 and 23.07 in first year, second year and pooled data, respectively which is at par with T<sub>11</sub> -Ethrel 1250 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 19.07, 25.17 and 22.12 in first year, second year and pooled data, respectively. The analysis of data revealed that T<sub>3</sub> -2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 DAS (Cotyledon Stage) with 13.67, 16.83 and 15.42 in first year, second year and pooled data was spotted to be minimum.

### 3.12 Sex ratio

The effect of use of CHAs at different stages was the functional effect on sex ratio. During the investigation, the data presented in table 3 and Fig.2 showed in relation to CHAs application at different stages. The minimum sex ratio was recorded in T<sub>8</sub>-GA<sub>3</sub> 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 10.12 in first year, T<sub>13</sub>-Control with 11.62 and T<sub>10</sub>-1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 10.90 which is at par with T<sub>10</sub>-1- Naphthalene Acetic Acid 100 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) of 10.17 and 11.64 in first year and second year while T<sub>8</sub>-GA<sub>3</sub> 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 11.11 in pooled data. The maximum sex ratio was recorded in T<sub>3</sub> -2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 DAS (Cotyledon Stage) with 14.17 and 14.36 in first year and pooled data while T<sub>9</sub> -2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage) with 16.63 in second year.

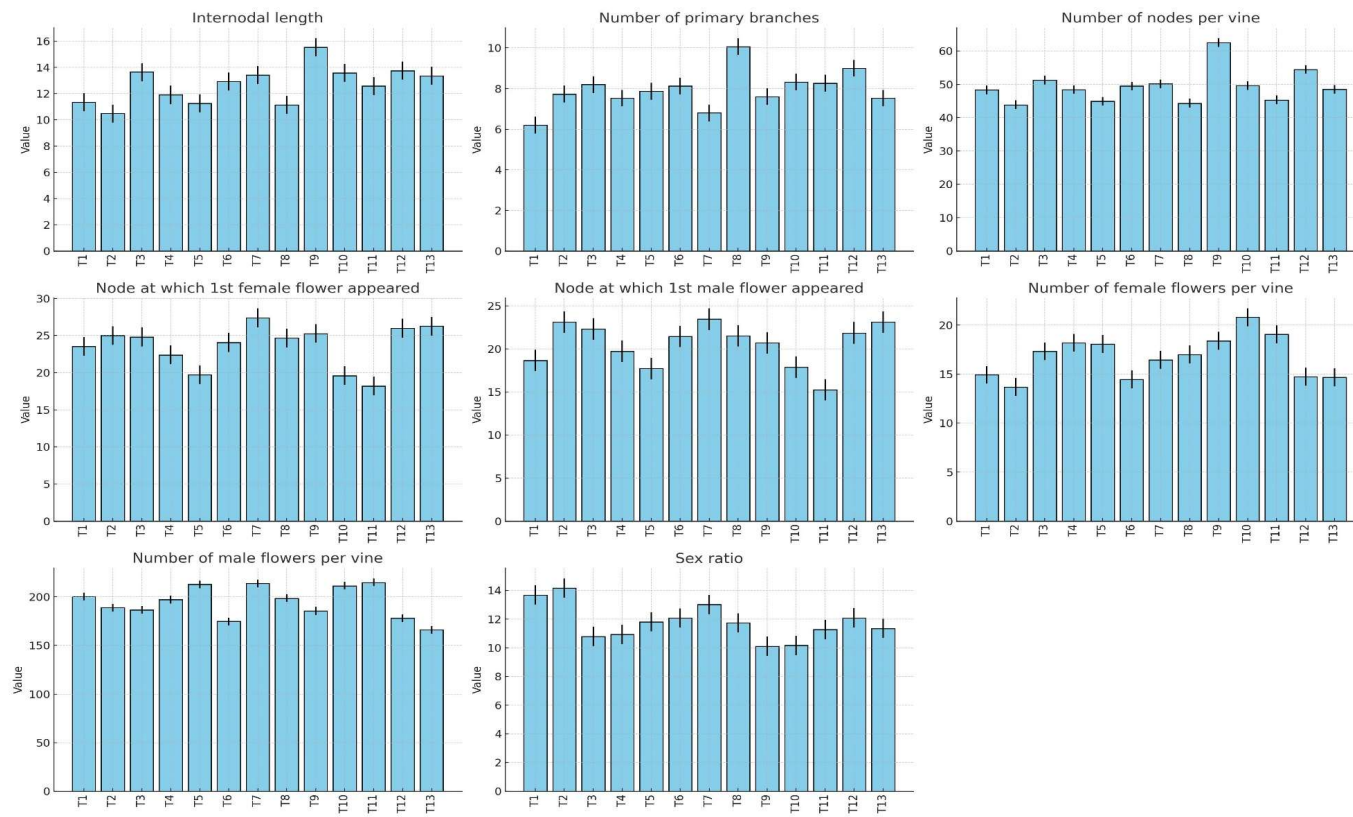
UNDER PEER REVIEW

**Table 3. Effect of chemicals on morphological parameters**

| S.No.          | Treatments   | Internodal length | Number of primary branches | Number of nodes per vine | Node at which 1st female flower appeared | Node at which 1st male flower appeared | Number of female flowers per vine | Number of male flowers per vine | Sex ratio |
|----------------|--|-------------------|----------------------------|--------------------------|--|--|-----------------------------------|---------------------------------|-----------|
| T <sub>1</sub> | Maleic Hydrazide 200 ppm - Spray at 10 DAS (Cotyledon Stage)           | 11.34             | 6.20                       | 48.33                    | 23.53                                    | 18.67                                  | 14.93                             | 200.33                          | 13.69     |
| T <sub>2</sub> | 2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 DAS (Cotyledon Stage)  | 10.48             | 7.73                       | 43.87                    | 25.00                                    | 23.13                                  | 13.67                             | 188.93                          | 14.17     |
| T <sub>3</sub> | GA 3 200 ppm - Spray at 10 DAS (Cotyledon Stage)                       | 13.64             | 8.20                       | 51.27                    | 24.80                                    | 22.33                                  | 17.33                             | 186.67                          | 10.80     |
| T <sub>4</sub> | 1- Naphthalene Acetic Acid 100 ppm - Spray at 10 DAS (Cotyledon Stage) | 11.91             | 7.53                       | 48.40                    | 22.40                                    | 19.73                                  | 18.20                             | 197.20                          | 10.94     |
| T <sub>5</sub> | Ethrel 1250 ppm - Spray at 10 DAS (Cotyledon Stage)                    | 11.26             | 7.87                       | 44.93                    | 19.73                                    | 17.73                                  | 18.07                             | 212.80                          | 11.82     |
| T <sub>6</sub> | Sulphonyl Urea 0.4 ml/L - Spray at 10 DAS (Cotyledon Stage)            | 12.92             | 8.13                       | 49.47                    | 24.07                                    | 21.47                                  | 14.47                             | 174.80                          | 12.09     |
| T <sub>7</sub> | Maleic Hydrazide 200 ppm - Spray at 10 and 24 DAS (2-4 Leaf Stage)     | 13.42             | 6.80                       | 50.13                    | 27.40                                    | 23.47                                  | 16.47                             | 213.93                          | 13.02     |
| T <sub>8</sub> | 2,3,5-Triiodo Benzoic Acid 25 ppm - Spray at 10 DAS (Cotyledon Stage)  | 11.13             | 10.07                      | 44.33                    | 24.67                                    | 21.53                                  | 17.00                             | 198.53                          | 11.75     |
| T <sub>9</sub> | GA 3 200 ppm - Spray at 10 DAS (Cotyledon Stage)                       | 15.53             | 7.60                       | 62.53                    | 25.27                                    | 20.73                                  | 18.40                             | 185.60                          | 10.12     |

|                 |  |             |             |             |             |             |             |              |             |
|-----------------|--|-------------|-------------|-------------|-------------|-------------|-------------|--------------|-------------|
| T <sub>10</sub> | 1- Naphthalene Acetic Acid<br>100 ppm - Spray at 10 and<br>24 DAS (2-4 Leaf Stage) | 13.59       | 8.33        | 49.60       | 19.60       | 17.87       | 20.80       | 211.47       | 10.17       |
| T <sub>11</sub> | Ethrel 1250 ppm - Spray at<br>10 and 24 DAS (2-4 Leaf<br>Stage)                    | 12.57       | 8.27        | 45.33       | 18.20       | 15.27       | 19.07       | 214.93       | 11.28       |
| T <sub>12</sub> | Sulphonyl Urea 0.4 ml/L -<br>Spray at 10 and 24 DAS (2-<br>4 Leaf Stage)           | 13.75       | 9.00        | 54.47       | 26.00       | 21.87       | 14.73       | 177.93       | 12.10       |
| T <sub>13</sub> | <b>Control</b>   | 13.36       | 7.53        | 48.53       | 26.27       | 23.13       | 14.67       | 166.07       | 11.35       |
|                 | <b>C.D. at 5%</b>  | <b>1.99</b> | <b>1.19</b> | <b>3.81</b> | <b>3.7</b>  | <b>3.67</b> | <b>2.65</b> | <b>11.69</b> | <b>1.95</b> |
|                 | <b>SE(m) ±</b>   | <b>0.69</b> | <b>0.41</b> | <b>1.29</b> | <b>1.26</b> | <b>1.25</b> | <b>0.91</b> | <b>3.98</b>  | <b>0.67</b> |
|                 | <b>C.V.</b>  | <b>9.25</b> | <b>8.89</b> | <b>4.55</b> | <b>9.29</b> | <b>9.53</b> | <b>9.39</b> | <b>3.54</b>  | <b>9.83</b> |

**Fig 2: Effect of chemicals on morphological parameters**



#### 4. DISCUSSION

The present study investigated the significant effects of various chemical hybridizing agents (CHAs) on morphological traits and sex expression in sponge gourd (*Luffa cylindrica* (L.) Roem.). The research, conducted during the summer seasons of 2023-24 and 2024-25 at the Vegetable Research Centre (VRC), Maharajpur, JNKVV, Jabalpur, revealed critical insights into the complex interplay between growth regulators and plant development across different growth stages.

The application of gibberellic acid (GA3) at 200 ppm at the 2-4 leaf stage consistently produced maximum vine length across all growth stages (30, 60, 90 days after sowing (DAS), and at final harvest). This finding aligns with Chovatia et al. (2010), who observed a 35% increase in vine length with GA3 application in bitter gourd, and Impana et al. (2024), who reported enhanced vegetative growth in bitter gourd. The synergistic effect of split applications at 10 and 24 DAS proved more effective than single applications, supporting similar findings in bottle gourd by Saha et al. (2007). In contrast, treatments involving 2,3,5-triiodo benzoic acid consistently resulted in shorter vine lengths, highlighting the growth-inhibiting properties of certain chemicals.

Maximum primary branches were observed with GA3 treatment at the 2-4 leaf stage, followed by sulphonyl urea application. This is consistent with Sapkota et al. (2020), who reported a 42% increase in lateral branch formation in cucumber with GA3 application. The timing of CHA application played a critical role, as early vegetative stage treatments resulted in optimal branching patterns (Kumawat et al., 2020). Increased branching not only improves light interception but also enhances fruit set and overall yield potential (Paponov et al., 2023).

GA3 treatment significantly increased internodal length, corroborating findings by Gebremeskel et al. (2019) in watermelon and Shan et al. (2021), who noted that gibberellins promote cell elongation and internodal growth. Internodal length plays a vital role in optimizing space utilization in high-density planting systems, where elongated internodes can enhance yield per unit area.

Ethrel application at the 2-4 leaf stage resulted in the earliest male and female flower appearance, consistent with Syed (2019), who highlighted the role of ethylene-releasing compounds in influencing sex determination pathways. Increased female flower production with naphthalene acetic acid (NAA) aligns with Khatoon et al. (2019), who observed similar effects in bitter gourd. Ma et al. (2014) explained this feminization mechanism as a modification of endogenous hormone balance. The improved sex ratio (reduced male-to-female flower ratio) with GA3 and NAA treatments corroborates findings by Rajashree & Deepanshu (2022) in ridge gourd, suggesting these growth regulators influence sex-determining genes.

The variation in node number and flowering position observed with different CHA treatments aligns with Hirpara et al. (2023) in bitter gourd. Ethrel treatment led to earlier female flower appearance at lower nodes, supporting Singh & Choudhury (1983), who reported similar effects in cucumber. Such modifications are critical for improving reproductive efficiency and yield potential.

Commented [k19]: italics  
observe and correct this mistake in whole manuscript

The statistical analysis, using analysis of variance (ANOVA), validated the significant effects of CHA treatments on various parameters. The consistency of results across two growing seasons underscores the robustness of the experimental design and the reliability of the findings. Repeated trials ensure that the recommendations derived from this study are applicable under diverse agricultural practices.

This research highlights the profound impact of CHAs on the growth, sex expression, and yield of sponge gourd. The findings provide valuable insights for optimizing CHA application timing and dosages to enhance productivity. By tailoring CHA treatments to specific growth stages, growers can improve both vegetative and reproductive performance, ultimately contributing to sustainable and efficient horticultural practices.

## 5. CONCLUSION

The investigation into the effects of various chemical hybridizing agents (CHAs) on the morphological traits of Sponge gourd (*Luffa cylindrica*) has yielded significant insights into optimizing its cultivation. The results clearly demonstrate that the application of CHAs, particularly GA3 and Sulphonyl Urea, markedly enhances vine length, number of primary branches, internodal length, and nodes per vine across multiple growth stages. Specifically, treatments involving GA3 at 200 ppm resulted in the longest vine lengths and increased branching, underscoring its effectiveness as a growth regulator. Furthermore, the timing of flower development was positively influenced by these treatments, with earlier flowering observed in Ethrel-treated plants.

The findings suggest that strategic application of CHAs can lead to improved growth parameters and flowering dynamics, which are crucial for maximizing yield potential in Sponge gourd cultivation. This research contributes valuable knowledge for horticulturists and agricultural practitioners seeking to enhance productivity through targeted chemical applications. Overall, the study advocates for the integration of CHAs into standard agricultural practices for Sponge gourd to achieve optimal growth and flowering outcomes, thereby supporting sustainable agricultural development in this economically important crop.

## 6. FUTURE WORK AND SUGGESTIONS

The future scope of this study on the effects of chemical hybridizing agents (CHAs) on Sponge gourd (*Luffa cylindrica*) presents numerous opportunities for further exploration and application. One significant area for future research is the investigation of the long-term effects of CHAs on soil health and plant resilience, providing insights into sustainable agricultural practices. Additionally, expanding the range of CHAs tested could uncover new compounds that may further enhance growth and flowering dynamics. A promising avenue for future exploration lies in the application of metabolomics to unravel the biochemical pathways modulated by CHAs. Metabolomics has proven effective in understanding plant responses to various treatments and stressors, as demonstrated by Gautam et al. (2024) in their study on rice against root-knot nematodes (RKN) and by Gautam et al. (2025), who explored the metabolomic changes in rice induced by *Arthrobotrys oligospora* against RKN. Similar metabolomic studies on Sponge gourd could provide detailed insights into the molecular mechanisms through which CHAs influence

Commented [k20]: italics



growth, development, and stress tolerance, enabling more precise and efficient CHA applications. Conducting large-scale field trials across diverse environmental conditions will also be essential to validate the efficacy of these treatments in varying agricultural settings. Furthermore, integrating CHAs with organic farming practices could enhance growth while maintaining ecological balance, appealing to a market increasingly focused on sustainability. Lastly, a thorough economic analysis assessing the cost-effectiveness of CHA applications in Sponge gourd production would be beneficial for farmers considering these treatments as part of their cultivation strategies. By pursuing these avenues, particularly through the lens of metabolomics, future research can significantly contribute to improving Sponge gourd cultivation practices, ultimately enhancing yield and profitability for growers.

#### Data availability statement

Data will be made available on request.

#### REFERENCES

Barcenas Jr., G.A., & Barcenas, L.M. (2023). Vegetative Growth Characteristics and Flower Sex Expression of *Cucumis sativus* Affected by Exogenous Application of Plant Growth Regulators. *Sarhad Journal of Agriculture*. <https://doi.org/10.17582/journal.sja/2023/39.4.815.822>

Chovatia, R.S., Ahlawat, T.R., Kavathia, Y.A., Jivani, L.L., & Kaila, D. (2010). Effect of plant growth regulators on vegetative growth, flowering and yield of bitter gourd cv. Priya. *Indian Journal of Horticulture*, 67, 254-258.

Gautam, V., Garg, V., Nagar, R., Meena, N., Kumari, S., Dashrath, H. A., Singh, S., Patel, S., Mukesh, Singh, H., Masurkar, P., & Singh, R. K. (2025). Characterizing metabolic changes in rice roots induced by *Meloidogyne graminicola* and modulated by *Arthrobotrys oligospora*: A pathway-based approach. *Physiological and Molecular Plant Pathology*, 136, 102541. <https://doi.org/10.1016/j.pmpp.2024.102541>

Gautam, V., Nagar, R., Barai, P., Garg, V., Singh, S., Singh, H., Patel, S., Mukesh, Kumar, A., & Singh, R. K. (2024). Exploring the rice root metabolome to unveil key biomarkers under the stress of *Meloidogyne graminicola*. *Plant Stress*, 14, 100620. <https://doi.org/10.1016/j.stress.2024.100620>

Gebremeskel, H., Dou, J., Li, B., Zhao, S., Muhammad, U., Lu, X., He, N., & Liu, W. (2019). Molecular Mapping and Candidate Gene Analysis for GA3 Responsive Short Internode in Watermelon (*Citrullus lanatus*). *International Journal of Molecular Sciences*, 21. <https://doi.org/10.3390/ijms21010290>

Hirpara, A.J., Vaddoria, M.A., & Polara, A.M. 2015. Effects of Plant Growth Regulators on Flowering, Fruiting and Fruit Yield in Bitter Gourd (*Momordica charantia* L.). *Journal of Pure and Applied microbiology*. Vol. 9(4), p. 3099-3102.

Commented [k21]: italics

Commented [k22]: page no.

Commented [k23]: italics

Commented [k24]: italics

Commented [k25]: DOI

Impana, M., Sonkamble, A.M., Wagh, A.P., Bawkar, S.O., & Kankal, D.S. (2024). GROWTH AND YIELD OF BITTER GOURD AS INFLUENCED BY PLANT GROWTH REGULATORS. *Plant Archives*. <https://doi.org/10.51470/plantarchives.2024.v24.no.1.148>

Commented [k26]: Sentence case  
Volume no and page no.

Khatoon, R., & Moniruzzaman, M. (2019). Effect of foliar spray of GA3 and NAA on sex expression and yield of bitter gourd. *Bangladesh Journal of Agricultural Research*. <https://doi.org/10.3329/BJAR.V44I2.41818>

Commented [k27]: Volume and page no.

Kumawat, K., Raja, W., Singh, D.B., Chand, L., Mir, J.I., Rai, K.M., & Kirmani, S.N. (2020). Effects of plant growth regulators applications on induction of lateral branching in Oregon Spur apple nursery trees. *Indian Journal of Horticulture*, 77, 72. <https://doi.org/10.5958/0974-0112.2020.00030.4>

Commented [k28]: Volume no. page no.

Lai, Y., Shen, D., Zhang, W., Zhang, X., Qiu, Y., Wang, H., Dou, X., Li, S., Wu, Y., Song, J., Ji, G., & Li, X. (2018). Temperature and photoperiod changes affect cucumber sex expression by different epigenetic regulations. *BMC Plant Biology*, 18. <https://doi.org/10.1186/s12870-018-1490-3>

Commented [k29]: Page no.

Ma, B.M., Ms, I., & Shamsuddin, Z.H. (2014). Altered sex expression by plant growth regulators: An overview in medicinal vegetable bitter gourd (*Momordica charantia* L.). *Journal of Medicinal Plants Research*, 8, 361-367. <https://doi.org/10.5897/JMPR10.032>

Matsumura, H., Gautam, K. K., Urasaki, N., & Pandey, S. (2020). Sex Determination in Bitter Gourd (pp. 73–81). Springer. [https://doi.org/10.1007/978-3-030-15062-4\\_6](https://doi.org/10.1007/978-3-030-15062-4_6)

Commented [k30]: Volume no

Paponov, M., Verheul, M.J., Dobrev, P.I., & Paponov, I.A. (2023). Additive effects of light and branching on fruit size and chemical fruit quality of greenhouse tomatoes. *Frontiers in Plant Science*, 14. <https://doi.org/10.3389/fpls.2023.1221163>

Commented [k31]: Page no

Priyadarshi, S., Verma, R.B., Babu, L.D., & Singh, V.K. (2022). Effect of different PGRs and stages of spray on growth attributes of bitter gourd. *Vegetable Science*. <https://doi.org/10.61180/vegsci.2022.v49.i1.17>

Commented [k32]: Volume and page no

Rajashree, V., & Deepanshu, .. (2022). Growth and Yield of Bitter Gourd as Influenced by Gibberellic Acid and Naphthalene Acidic Acid (*Momordica charantia* L.). *International Journal of Plant & Soil Science*. <https://doi.org/10.9734/ijpss/2022/v34i2131335>

Commented [k33]: italics

Saha, S., Mori, H., & Hattori, K. (2007). Synergistic Effect of Kinetin and Benzyl Adenine Plays a Vital Role in High Frequency Regeneration from Cotyledon Explants of Bottle Gourd (*Lagenaria siceraria*) in Relation to Ethylene Production. *Breeding Science*, 57, 197-202. <https://doi.org/10.1270/JSBBS.57.197>

Commented [k34]: italics

Sapkota, B., Dhital, M., Shrestha, B., & Tripathi, K.M. (2020). Effect of plant growth regulators on flowering and fruit yield of cucumber (*Cucumis sativus* cv. Malini) in Chitwan, Nepal. *Journal of Agriculture and Forestry University*. <https://doi.org/10.3126/jafu.v4i1.47065>

Commented [k35]: italics

Shan, F., Zhang, R., Zhang, J., Wang, C., Lyu, X., Xin, T.R., Yan, C., Dong, S., Ma, C., & Gong, Z. (2021). Study on the Regulatory Effects of GA3 on Soybean Internode Elongation. *Plants*, 10. <https://doi.org/10.3390/plants10081737>

Singh, R.K., & Choudhury, B. (1983). Differential Response of Chemicals on Sex Modification of Three Genera of Cucurbits. *Indian Journal of Horticulture*, 45, 89-99. <https://doi.org/10.17660/ACTAHORTIC.1983.137.41>

Soni, S., Singh, K. V., Malik, S., Singh, M. K., Singh, B., & Singh, S. K. (2015). Impact of Foliar Application of Plant Bioregulators on Morphological, Floral and Yield Traits in Bottle Gourd [*Lagenaria Siceraria*(Molina) Standl.] CV. Pusa Naveen. *Progressive Agriculture*, 15(2), 268. <https://doi.org/10.5958/0976-4615.2015.00016.2>

Syed, S. (2019). INFLUENCE OF PLANT GROWTH REGULATORS AND STAGE OF APPLICATION ON SEX EXPRESSION OF BITTER GOURD (*MOMORDICA CHARANTIA* L.) CV. VK-1-PRIYA. *Plant Archives* (09725210), 19(2).

Vidyullatha, L.M., & Topno, S.E. (2022). Effect of Naphthalene Acetic Acid & Indole Acetic Acid on Growth, Yield and Quality of Muskmelon (*Cucumis melo* L.). *International Journal of Plant & Soil Science*. <https://doi.org/10.9734/ijpss/2022/v34i2231519>

Commented [k36]: italics

Commented [k37]: Italics and BN format

Commented [k38]: Sentence case

Commented [k39]: italics

Commented [k40]: quote the references in standard format (APA format)