MATERIALS AND METHODS
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Chicory (Chicorium intybus (L.) Family: Asteraceae) was selected for the study, Indole acetic acid (IAA), gibberellic acid (GA) and kinetin (KIN) were selected as PGRs. Chart-1 represents the layout of experiments for the study on response of Chicory to PGRs. The details are as follows:

EXPT.-I: EFFECT OF DIRECT APPLICATION OF PGRs ON PERFORMANCE OF CHICORY

IAA,+: STUDY ON GROWTH AND METABOLISM OF CHICORY SEEDLINGS GROWN WITH PGRs

The seeds of Chicory were germinated in sterilized petriplates lined with sterilized Whatmann filter paper no.1. The media were
1) DW (Control)
2) IAA (10^-5 M)
3) GA (10^-5 M)
4) KIN (10^-5 M)
5) IAA+GA (10^-5 M)
6) IAA+KIN (10^-5 M)
7) GA+KIN (10^-5 M)
8) IAA+GA+KIN (10^-5 M)

DW grown seeds were control. Experiment was conducted at 28±2°C under laboratory conditions up to 120h. The response of seedlings to PGRs was studied as follows:

(i) Study on growth:
Ten seedlings from each treatment were studied for growth on completion of 72h, 96h, and 120h of germination. The elongation of root and shoot was measured; mean was calculated and expressed as cm/seedling. The fresh weight of seedlings was recorded and expressed as mg/seedling, then dried at 80°C for 48h; dry weight was recorded and expressed as mg/seedling. The growth response of seedlings to PGRs was evaluated with the help of percent change in the growth of PGRs grown seedlings over growth of DW (control) seedlings.

Percent change:
Percent change was calculated using the following formula:

\[
\text{Percent change} = \frac{\text{Value of particular parameter in PGR treated plant}}{\text{Value of particular parameter in control plant}} \times 100
\]

(ii) Study on metabolism:
72h, 96h and 120h old DW and PGRs grown seedlings in replicate were analyzed for the following metabolism:

Carbohydrate Metabolism
1. α-amylase and β-amylase activities
2. Invertase Activity
3. Reducing sugar and Nonreducing sugar contents

**Protein Metabolism**

4. Protease Activity
5. Protein Content
6. Total Amino Acid Content

**Antioxidant Enzyme**

7. Peroxidase Activity

**Phenolic Substances**

8. Total Phenol content

For enzymatic activities, seedlings were crushed in ice cold DW and centrifuged at 2000 rpm. Supernatant was the source of enzymes. The enzymatic activities were expressed on the basis of protein. Following is the method for enzyme protein determination.

**Enzyme protein:**

Enzyme protein was estimated by the method of Lowry et al., (1951). To 1ml aliquot, 4ml 12.5% sodium carbonate and 1ml 0.1% copper sulphate were added, incubated for 30 minutes then 0.5% folin-phenol reagent was added. After incubating for 10 minutes optical density (OD) was recorded at 660nm on Systronics 106 spectrophotometer. The following regression formula was calculated by using known concentrations of casein.

\[ X = 236.60Y - 35.22 \]

Where, \( Y \) = optical density (OD)

The protein content was expressed as \( \mu g/mg \) fresh weight.

**Carbohydrate Metabolism**

1. \( \alpha \)-amylase and \( \beta \)-amylase activities:

These activities were determined by estimating total amylase activity and \( \alpha \)-amylase activity. Total amylase activity was determined using the method of Paleg et al., (1962). 1.0 ml enzyme aliquot was mixed with 1.0 ml citrate buffer (pH 5) and 1.0 ml of 1% starch solution and incubated for 30 minutes at room temperature. The blank had 1.0 ml of DW instead of the enzyme extract.

The enzyme activity was stopped by adding 0.5 ml of cold iodine solution (2.5 g of iodine and 25 g of potassium iodide in 1 l of 0.05 N HCl). The final volume was made up to 20ml and optical density (OD) was read at 600nm. The difference in OD between the blank and reaction i.e. starch hydrolyzed was calculated using the following regression formula prepared from known concentration of starch.

\[ X = 331.49 Y - 2.34 \]

Where, \( Y \) = optical density (OD)

The amylase activity was expressed as \( \mu g \) starch hydrolysed/h/\( \mu g \) protein. \( \alpha \)-amylase activity was determined by killing the \( \beta \)-amylase. For this, enzyme aliquot was heated at 100°C for 20 min. and then it was used for determination of \( \alpha \)-amylase. Same procedure was used for the determination and calculation of enzyme activity. The value of \( \beta \)-amylase activity was calculated by subtracting the value of \( \alpha \)-amylase activity from the value of total amylase activity.
2. Invertase Activity:
Invertase activity was determined by the method of Hatch and Glasziou, (1963). To 1ml enzyme aliquot, 1ml acetate buffer (pH 4.8) and 1ml 0.25% sucrose (250mg sucrose in 100ml acetate buffer pH 4.8) was added. Blank had 1ml acetate buffer instead of 0.25% sucrose. This was incubated for one hour at room temperature. Enzyme activity was stopped by adding 2ml 5% perchloric acid and volume was made up to 10ml. 1ml of this mixture was taken to develop colour by Nelson-Somogyi method (Wharton and McCarty, 1972). Method is mentioned under the method for reducing and non-reducing sugars. OD was taken at 540nm on Systronics 106 spectrophotometer, by using the following regression equation prepared from known concentration of glucose,

\[ X = 426.67 Y - 15.25 \]

amount of reducing sugar released by invertase activity was calculated and expressed as µg glucose liberated/h/µg protein.

3. Reducing sugar and Nonreducing sugar contents:
Weighed plant material (oven dried) was boiled in 80% ethanol for 4 to 5 minutes, homogenized with sand and centrifuged. Residue was again extracted with 5ml ethanol. After centrifugation, the ethanol was evaporated and the residual sugars were dissolved in a fixed volume (20ml) of DW. From the 20ml of the extract, 10ml each was taken for reducing sugar and total sugar. To the extract of total sugar, 3ml IN HCl (8.75ml of conc. HCl + 91.25ml DW) was added and kept in boiling water bath for 20 minutes to hydrolyse non-reducing sugar. It was cooled and neutralized by adding 3ml 1N NaOH. 1ml of 25% lead acetate and 1ml of 25% sodium carbonate were added in both the sets and volume was made up to 20ml, then it was filtered, 1ml of the aliquot was taken for sugar estimation by the method of Nelson-Somogyi (Wharton and McCarty, 1972).

To 1ml of the above aliquot was added 1ml of Nelson-Somogyi reagent (Nelson reagent A : 12.5g sodium carbonate, 12.5g sodium potassium tartarate, 10g sodium bicarbonate and 100g sodium sulphate were dissolved and volume was made up to 500ml with DW and Nelson reagent B : 15g copper sulphate in 100ml DW and 2 drops of conc. H₂SO₄). Nelson reagent was prepared by mixing 50 parts of A with 2 parts of B. Tubes were capped with glass marbles and heated in a water-bath at 100°C for 20 minutes then cooled rapidly. 1ml arsenomolybdate reagent (25g ammonium molybdate dissolved in 400ml DW. To this was added 21ml conc. H₂SO₄ + 3g sodium arsenate in 25ml DW and volume was made to 500ml) was added and shaken thoroughly for 5 minutes to dissolve the red precipitates. Final volume was made to 25ml with DW. OD was taken at 540nm on Systronics 106 spectrophotometer. By referring to the following regression equation of glucose,

\[ X = 426.67 Y - 15.25 \]
Amount of reducing sugar and total sugar were calculated and expressed as mg/g dry weight. Nonreducing sugar was calculated from the data of total and reducing sugar.

**Protein Metabolism**

**4. Protease Activity:**
The method described by Penner and Ashton, (1967) was followed for protease activity. The reaction mixture containing 2ml enzyme extract, 1ml of 0.5% casein solution (pH 7.0) and 3ml of 0.1M phosphate buffer (pH 7.0) was incubated for one hour at room temperature. Then 2ml of the reaction mixture was mixed with 2ml of 15% TCA. After 20 minutes, the precipitated protein was discarded by centrifugation. To 1ml of enzyme aliquot, 1ml of DW, 4ml of 0.5N NaOH and 1ml of folin-phenol reagent were added. After 30 minutes, the OD was recorded at 660nm on spectrophotometer. The following regression formula was made by reacting known amounts of tyrosine with folin-phenol reagent and activity was expressed in terms of μg tyrosine liberated/h/μg protein.

\[ X = 232.14 Y - 0.71 \]

**5. Protein Content:**
Protein was determined using the method of Lowry et al., (1951). Weighed amount of oven-dried plant material was ground in 80% ethanol and extracted twice. The residue was first washed with cold 5% perchloric acid (to remove sugars and soluble protein fractions), centrifuged and secondly, it was washed with mixture of ethanol: ether: chloroform in the ratio of 2:1:1 (to remove acid soluble fractions and lipids) and centrifuged. The protein fraction was dissolved in 1N sodium hydroxide and kept for one hour and centrifuged. The supernatant was made upto 5ml with sodium hydroxide and used as an aliquot. To 2ml of the above aliquot was added 5ml of Lowry reagent C (prepared by mixing 50ml reagent A which is 2% sodium carbonate in 0.1N NaOH and 1ml of reagent B which is 0.5% copper sulphate in 1% sodium potassium tartarate) and incubated at room temperature for 30 minutes. The colour was developed by adding 0.5ml folin phenol reagent. After 10 minutes, OD was read at 600nm on Systronics 106 spectrophotometer. The following regression equation was prepared by using known concentrations of casein.

\[ X = 236.6 Y - 35.22 \]

The protein content was expressed as mg/g dry weight.

**6. Total Amino Acid Content:**
The content of total amino acid was determined following the method of Harding and McClean, (1916). The reaction system containing 0.5ml of ethanol extract from the dried material, 1ml 10% pyridine and 1 ml of 2% ninhydrin reagent was stoppered and heated in water-bath at 100°C for 30 minutes. Violet blue colour was developed. Later it was cooled and diluted with DW to a final volume of 10ml. OD of the violet blue colour was read at 570nm on spectrophotometer. The following regression formula was prepared using isoleucine as standard.
\[ X = 413.42Y - 19.23 \]

The total amino acid content was expressed as mg/g dry weight.

**Antioxidant Enzyme**

7. Peroxidase Activity:

George, (1953) and Maehly, (1954) method was employed to assay peroxidase activity. The reaction mixture having 2ml enzyme aliquot, 2ml 0.2M phosphate buffer (pH 7.0) and 2ml 20mM guaiicol reagent (0.22ml guaiicol in 100ml DW, was prepared 24 hours before carrying out the estimation) was taken in a cuvette. The cuvette was placed in the spectrophotometer. The wavelength was adjusted to 470nm and OD was noted. 2 drops of 10µM hydrogen peroxide (0.4ml H_2SO_4 of 20 volumes in 9.6ml DW) was added to the reaction mixture in the cuvette and again the OD was noted after 30 seconds. The difference in OD before and after adding H_2O_2 to the reaction mixture was used to calculate peroxidase activity. Peroxidase activity was expressed as difference in OD/min/µg protein.

8. Total phenol content:

It was determined using the method of Farks and Kiraly, (1962). The reaction mixture containing 0.5ml of the ethanol extract, 1ml of 20% sodium carbonate and 0.5ml folin phenol reagent was heated in a water-bath for 10 minutes, cooled and diluted to a fixed volume of 5ml with DW. It was filtered using Whatmann filter paper No. 1 to remove the precipitates. The OD of the blue coloured filtrate was read at 660nm. Phenol content was expressed as mg/g dry weight. The following regression formula was prepared using gallic acid as standard.

\[ X = 96.05Y + 10.03 \]

**IB: - STUDY ON GROWTH, PHOTOSYNTHETIC PIGMENTS AND LEAF METABOLISM OF CHICORY PLANTS GROWN WITH FOLIAR SPRAY OF PGRs**

The seeds of Chicory were sown in plots (1 M x 1 M). The plants were raised using normal agricultural practice under field conditions. On completion of 40 and 80 days of sowing (40 DAS and 80 DAS), the plants were sprayed with DW, IAA, GA, KIN, IAA+GA, IAA+KIN, GA+KIN and IAA+GA+KIN (10^{-5} M each). DW spray was control of spray treatment. The plants without any spray were control. The tween 20 was used as a surfactant. The following were the treatments:

**40 DAS**

1) Control
2) DW
3) IAA (10^{-5} M)
4) GA (10^{-5} M)
5) KIN (10^{-5} M)
6) IAA+GA (10^{-5} M)
7) IAA+KIN (10^{-5} M)
8) GA+KIN (10^{-5} M)
9) IAA+GA+KIN (10^{-5} M)
5 plots were kept for each treatment, each plot had 10 plants. Hand spray was used and spray was given till both the sides of leaf became wet, three such sprays were given within a week. Response of Chicory plants to PGRs was studied as follows:

(i) Study on growth:
Following parameters were studied.
- Root length - cm/plant
- Shoot length - cm/plant
- Leaf number - no/plant
- Fresh weight of root and shoot - g/plant
- Dry weight of root and shoot - g/plant

As per normal agricultural practices plants were harvested before flowering thus vegetative growth was recorded at regular intervals. Methods of Gregory, (1921, 1926) and Hunt, (1978) were used to study the growth of 60, 90 120 and 150 days (40 DAS) and 120, 150 days (80 DAS)) old plants. Ten plants at random were selected from each treatment. These plants were carefully uprooted (minimizing the damage to the root) and brought to the laboratory, thoroughly washed with water and gently pressed against blotting sheets to remove moisture from their surface. Root length and shoot length of each plant were measured and expressed as cm/plant, leaf number was also noted and expressed as no/plant. Plant parts viz. root and shoot were separated and fresh weight (g/plant) of each part was recorded. These were then placed in a paper bag and transferred to oven at 80°C for a period of one week for complete drying. Dry weight (g/plant) was recorded.

Percent Allocation
The partitioning of dry matter in root and shoot were calculated. The following formula was used.

\[
\text{Percent Allocation} = \frac{\text{Dry matter of particular organ}}{\text{Dry matter of whole plant}} \times 100
\]

Growth Indices viz. Relative Growth Rate (RGR), Leaf Weight Ratio (LWR) and Net Assimilation Rate (NAR) are important parameters for the growth study in plants (Blackman, 1919, Gregory, 1921, Nilsen and
Orcutt, 1996). RGR, LWR and NAR were calculated as follows from the data of dry weight.

**RGR:** It was calculated at thirty days interval as the difference between Naperian logarithms of dry weight of successive samples. The formula for RGR is as follows:

\[ RGR = \frac{\log_e W_1 - \log_e W_0}{t} \]

Where, \( W_0 \) = initial dry weight of the plant
\( W_1 \) = dry weight of the plant on succeeding sampling date
\( t \) = time- days

**LWR:** The following formula was used for determining the Leaf Weight Ratio (LWR):

\[ LWR = \frac{L_1 - L_0}{\log_e L_1 - \log_e L_0} / \frac{W_1 - W_0}{\log_e W_1 - \log_e W_0} \]

Where \( L_0 \) and \( L_1 \) represent the successive dry weights of leaves. \( W_0 \) and \( W_1 \) represent the successive dry weight of the whole plants.

**NAR:** From the data of dry matter production of whole plant and leaf, Net Assimilation Rate (NAR) was calculated as follows:

\[ NAR = \frac{W_1 - W_0}{L_1 - L_0 / \log L_1 - L_0} \]

Where, \( W_0 \) and \( W_1 \) represent the successive dry weights of the whole plant, \( L_0 \) and \( L_1 \) represent dry weights of the leaves of corresponding samples.

The growth response of Chicory to PGRs was evaluated with the help of percent change in various parameters of growth. The formula for percent change is given in Expt:IA(i).

(ii) **Study on Photosynthetic pigments:**

The leaf in replicate from control, DW and PGRs sprayed 60, 90, 120, and 150 days (40 DAS) and 120, 150 days (80 DAS) old plants were estimated for photosynthetic pigments using the method of Arnon, (1949). Weighed fresh leaf material was crushed in 80% acetone (80ml acetone + 20ml DW) with a pinch of sand. The homogenate was filtered using Whatmann filter paper No. 1 and the filtrate was made up to a specific volume. The absorbance of the chlorophyll suspension was read on Systronics 106 spectrophotometer at 480, 510, 645 and 663nm wavelength. The following formulae were used to calculate the quantity of photosynthetic pigments:

- Chlorophyll 'a' (mg / g fr wt) = 12.7 (D663) - 2.69 (D645)
- Chlorophyll 'b' (mg / g fr wt) = 22.9 (D645) - 4.68 (D663)
- Total Chlorophyll (mg / g fr wt) = 20.2 (D645) + 8.02 (D663)
Carotenoids (mg / g Fr wt) = 7.6 (D480) – 1.49 (D510)
Where, D = Optical Density.

(iii) Study on metabolism:
The leaf in replicate from control, DW and PGRs sprayed 60, 90, 120, and 150 days (40 DAS) and 120, 150 days (80 DAS) old plants were analyzed for following metabolisms.

(I) Carbohydrate Metabolism
1. α-amylase and β-amylase activities
2. Invertase Activity
3. Reducing sugar and Nonreducing sugar contents

(II) Protein Metabolism
4. Protease Activity
5. Protein Content
6. Total Amino Acid Content

(III) Antioxidant Enzyme
7. Peroxidase Activity

(IV) Phenolic Substances
8. Total Phenol content

The methods were the same as mentioned in Expt- IA (ii).

IC.-: EFFECT OF PGRs FOLIAR SPRAY ON GROWTH OF LATE SOWN CHICORY PLANTS
The seeds of Chicory were sown on 16th Nov. and 16th Dec. (2007) in plots (1 M x 1 M). On completion of 40 days of sowing (40 DAS), late sown i.e. 16th Dec. sown plants were sprayed with IAA, GA, KIN, IAA+GA, IAA+KIN, GA+KIN and IAA+GA+KIN (10⁻⁵ M). DW was also sprayed as control of spray treatment. Three sprays were given within a week. Tween 20 was used as a surfactant. The plants without any spray were the control. Following were the treatments.
1) D1 Control- Normal date (16th Nov.) sown plants
2) D2 Control- Late date sown (16th Dec.) sown plants
3) D2- DW
4) D2- IAA (10⁻⁵ M)
5) D2- GA (10⁻⁵ M)
6) D2- KIN (10⁻⁵ M)
7) D2- IAA+GA (10⁻⁵ M)
8) D2- IAA+KIN (10⁻⁵ M)
9) D2- GA+KIN (10⁻⁵ M)
10) D2- IAA+GA+KIN (10⁻⁵ M)

5 plots (10 plants/plot) were kept for each treatment. The effects of PGRs foliar spray on growth of late sown Chicory were studied as follows:

(i) Study on growth:
10 plants of normal sown, late sown control, DW and PGRs sprayed plants were studied for root length, shoot length, leaf number, fresh weight and dry weight of root and shoot. The observations were recorded on 60, 90, 120 and 150 days of growth. (Expt.-1B (i)) Percent
allocation and growth indices (Expt.-IB (i)) were also calculated. The impact of PGR foliar spray on growth performance of late sown Chicory was evaluated by calculating percent change in growth of late sown chicory plants over growth of normal date sown plants. The formula is given in Expt. IB (i).

**Expt.-II: EFFECT OF PRESOASKING SEED TREATMENT WITH PGRs ON PERFORMANCE OF CHICORY**

**IIA.:- EFFECT OF PRESOASKING SEED TREATMENT WITH PGRs ON GROWTH AND METABOLISM OF CHICORY SEEDLINGS**

Chinoy et. al., (1969a) method was used for pretreatment of chicory seeds. The uniform graded seeds of Chicory were soaked in IAA, GA, KIN, IAA + GA, IAA + KIN and IAA + GA+ KIN (10^{-5} M), for four hours and then dried under laboratory conditions at room temperature. Seeds were also soaked in DW for four hours then dried. The unsoaked seeds were control. The unsoaked, DW and PGRs soaked i.e. pretreated seeds were kept on sterilized petriplates lined with sterilized Whatmann filter paper no.1 and germinated in DW. The following were the treatments.

1) Control – untreated
2) DW– Pretreatment
3) IAA (10^{-5} M) – Pretreatment
4) GA (10^{-5} M) – Pretreatment
5) KIN (10^{-5} M) – Pretreatment
6) IAA+GA (10^{-5} M) – Pretreatment
7) IAA+KIN (10^{-5} M) – Pretreatment
8) GA+KIN (10^{-5} M) – Pretreatment
9) IAA+GA+KIN (10^{-5} M) – Pretreatment

Experiment was conducted at 28±2°C under laboratory conditions up to 120h. The response of Chicory seedlings to presoaking treatments was studied as follows:

**(i) Study on growth:**
10 seedlings grown from control, DW and PGRs pretreated seeds were studied for root length, shoot length, fresh weight and dry weight. The data were recorded on completion of 72h, 96h and 120h of germination. (Expt. IA(i)) The growth response of Chicory seedlings to PGRs pretreatment was evaluated (Expt. IA(i)).

**(ii) Study on metabolism:**
72h, 96h and 120h old control, DW and PGRs pretreated seedlings in replicate were analyzed for enzymes and metabolites. The biochemical parameters and methods were the same as mentioned in Expt.-IA (ii)

**IIB.:- EFFECT OF PRESOASKING SEED TREATMENT WITH PGRs ON GROWTH, PHOTOSYNTHETIC PIGMENTS AND LEAF METABOLISM OF CHICORY PLANTS**

The untreated i.e. control, DW and PGRs pretreated (Expt.-IIA) were sown in the plots of 1 M x 1 M size. Following were the treatments.
1) Control  
2) DW - Pretreatment  
3) IAA (10^{-5} M) - Pretreatment  
4) GA (10^{-5} M) - Pretreatment  
5) KIN (10^{-5} M) - Pretreatment  
6) IAA+GA (10^{-5} M) - Pretreatment  
7) IAA+KIN (10^{-5} M) - Pretreatment  
8) GA+KIN (10^{-5} M) - Pretreatment  
9) IAA+GA+KIN (10^{-5} M) - Pretreatment

Total nine treatments and five plots (10 plants/plot) were kept for each treatment. Necessary irrigation was given. Plants were grown using normal agriculture practices. The response of Chicory plants to PGRs presoaking treatments was studied as follows:

(i) Study on growth:
10 plants from each treatment were regularly studied for different parameters of growth. The parameters and methods were the same as mentioned in Expt. IB (i). 30, 60, 90 and 120 days old plants were used for study purpose. Percent allocation of dry matter, growth indices and percent change in growth were also calculated (Expt. IB (i)).

(ii) Study on photosynthetic pigments:
The photosynthetic pigments from leaf (in replicate) of 30, 60, 90, and 120 days old control, DW and PGRs pretreated plants were estimated as per the method mentioned in Expt. IB (ii).

(iii) Study on metabolism:
The enzymes and metabolites listed in Expt. IA (ii) were estimated in replicate from the leaf of control, DW and PGRs pretreated plants. The biochemical estimations were carried out from 30, 60, 90 and 120 days old plants.

Expt. - III: INTERACTIVE EFFECT OF ASCORBIC ACID AND PGRs ON GROWTH PERFORMANCE OF CHICORY
III A. -- INTERACTIVE EFFECT OF ASCORBIC ACID AND PGRs ON GROWTH PERFORMANCE OF CHICORY SEEDLINGS
The uniform graded seeds of Chicory were germination in sterilized petriplates lined with sterilized Whatmann filter paper no.1. The media for germination were DW, AA (Ascorbic acid), PGRs (IAA, GA, and KIN) and mixtures of AA with PGRs. The following were the treatments:
1) DW (Control) 
2) AA (10^{-5} M) 
3) IAA (10^{-5} M) 
4) AA+IAA (10^{-5} M) 
5) GA (10^{-5} M) 
6) AA+GA (10^{-5} M) 
7) KIN (10^{-5} M) 
8) AA+KIN (10^{-5} M)
The experiment was conducted under laboratory conditions up to 120h. Interactive effects of AA with PGRs on performance of Chicory seedlings were studies as follows.

(i) Study on growth:
72h, 96h and 120h old control, AA, PGRs and AA+PGRs grown seedlings were selected for growth, 10 seedlings from each treatment were used for growth study. The parameters and methods were the same as mentioned in Expt. -IA (i). The percent changes in growth of treated seedlings over control seedlings were also calculated with the help of formula given in Expt IA (i).

IIIB.-: INTERACTIVE EFFECT OF ASCORBIC ACID AND PGRs ON GROWTH PERFORMANCE OF CHICORY PLANTS
The seeds of Chicory were sown in the plots (1 M x 1 M). The necessary irrigation was given and on completion of 45 days of sowing (45 DAS) plants were sprayed with DW, AA, PGRs and mixtures of AA with PGRs. The following were the treatments.

1) Control
2) DW
3) AA (10\(^{-5}\) M)
4) IAA (10\(^{-5}\) M)
5) AA+IAA (10\(^{-5}\) M)
6) GA (10\(^{-5}\) M)
7) AA+GA (10\(^{-5}\) M)
8) KIN (10\(^{-5}\) M)
9) AA+KIN (10\(^{-5}\) M)
10) IAA+GA (10\(^{-5}\) M)
11) AA+IAA+GA (10\(^{-5}\) M)
12) IAA+KIN (10\(^{-5}\) M)
13) AA+IAA+KIN (10\(^{-5}\) M)
14) GA+KIN (10\(^{-5}\) M)
15) AA+GA+KIN (10\(^{-5}\) M)
16) IAA+GA+KIN (10\(^{-5}\) M)
17) AA+IAA+GA+KIN (10\(^{-5}\) M)

Three such sprays were given in the week and five plots (10 plants/plot) were kept for each treatment. The growth performance of Chicory plants sprayed with DW, AA, PGRs and mixtures of AA with PGRs was studied as follows:
(i) Study on growth:
10 plants from each treatment at regular intervals were used for growth study. The parameters were the same as mentioned in Expt. -IB (i). The percent allocation of dry matter, growth indices and percent change in growth were calculated (Expt IB (i)).

Expt. -IV: INTERACTIVE EFFECT OF SEED TREATMENT WITH ASCORBIC ACID AND PGRs ON GROWTH PERFORMANCE OF CHICORY
IVA.-: INTERACTIVE EFFECT OF SEED TREATMENT WITH ASCORBIC ACID AND PGRs ON GROWTH PERFORMANCE OF CHICORY SEEDLINGS

The seeds of Chicory were soaked in DW, AA, PGRs and AA+PGRs (10^{-5} M each) for four hours, then dried at room temperature under laboratory conditions (Chinoy et al., 1969a). The control i.e. untreated, DW, AA, PGRs and AA+PGRs pretreated seeds were germinated in petriplates lined with Whatmann filter paper no.1. DW was used for germination. The experiment was conducted up to 120h.

The following were the treatments.
1) DW (Control) (10^{-5} M) - untreated
2) AA (10^{-5} M) - pretreatment
3) IAA (10^{-5} M) - pretreatment
4) AA+IAA (10^{-5} M) - pretreatment
5) GA (10^{-5} M) - pretreatment
6) AA+GA (10^{-5} M) - pretreatment
7) KIN (10^{-5} M) - pretreatment
8) AA+KIN (10^{-5} M) - pretreatment
9) IAA+GA (10^{-5} M) - pretreatment
10) AA+IAA+ GA (10^{-5} M) - pretreatment
11) IAA+KIN (10^{-5} M) - pretreatment
12) AA+IAA+KIN (10^{-5} M) - pretreatment
13) GA+KIN (10^{-5} M) - pretreatment
14) AA+GA+KIN (10^{-5} M) - pretreatment
15) IAA+GA+KIN (10^{-5} M) - pretreatment
16) AA+IAA+GA+KIN (10^{-5} M) - pretreatment

The response of Chicory seedlings to interactive effects of seed treatment with AA and pretreatment of AA with PGRs was studied as follows:

(i) Study on growth:
10 seedlings from each treatment at regular intervals were studied for various growth parameters (Expt. IA (i)). The percent change in growth of treated over control seedlings were also calculated (Expt. IA (i)).

IVB.-: INTERACTIVE EFFECT OF SEED TREATMENT WITH ASCORBIC AND PGRs ON GROWTH PERFORMANCE OF CHICORY PLANTS

The untreated, DW, AA, PGRs and AA+PGRs pretreated seeds (Chinoy et al., 1969), were sown in plots (5 for each treatment, 10 plants/plot)
and standard agricultural practices were used for cultivation of Chicory plants. The following were the treatments.

1) Control - untreated
2) DW - pretreatment
3) AA (10^{-5} M) - pretreatment
4) IAA (10^{-5} M) - pretreatment
5) AA+IAA (10^{-5} M) - pretreatment
6) GA (10^{-5} M) - pretreatment
7) AA+GA (10^{-5} M) - pretreatment
8) KIN (10^{-5} M) - pretreatment
9) AA+KIN (10^{-5} M) - pretreatment
10) IAA+GA (10^{-5} M) - pretreatment
11) AA+IAA+GA (10^{-5} M) - pretreatment
12) IAA+KIN (10^{-5} M) - pretreatment
13) AA+IAA+KIN (10^{-5} M) - pretreatment
14) GA+KIN (10^{-5} M) - pretreatment
15) AA+GA+KIN (10^{-5} M) - pretreatment
16) IAA+GA+KIN (10^{-5} M) - pretreatment
17) AA+IAA+GA+KIN (10^{-5} M) - pretreatment

The response of Chicory plants to interactive effects of seed treatment with AA and PGRs was studied as follows:

(i) Study on growth:
10 plants at regular intervals from each treatment were selected for the study. The growth parameters were the same as mentioned in Expt. -IB (i). Percent allocation of dry matter, growth indices and percent change in growth of treated over control plants were calculated (Expt IB (i)).

Statistical analysis
The growth data, data on photosynthetic pigments, enzymes and metabolites (Expt. -I, II, III and IV) were analyzed statistically using ANOVA (Fisher 1954).