

# Interactive Effect of Simulated Acid Rain and Auxin on Carbohydrate Content in the Leaves of *Capsicum Frutescens* Var. Sweet Magic

## Abstract

Acid rain has detrimental effect on plants and its contents. In order to counter effect of acid rain and enrich *Capsicum frutescens* var. Sweet magic in carbohydrate content, experiments were carried out by treatment of plants with acid water of pH3, pH 4 and pH 5 concentration; Auxin solution of  $1 \times 10^{-5}$ ,  $1 \times 10^{-6}$  and  $1 \times 10^{-7}$  ppm concentration as well as with combination of acid rain and auxin measuring pH  $3.0+1 \times 10^{-5}$ , pH  $3.0+1 \times 10^{-6}$ , pH  $3.0+1 \times 10^{-7}$ , pH  $4.0+1 \times 10^{-5}$ , pH  $4.0+1 \times 10^{-6}$ , pH  $4.0+1 \times 10^{-7}$ , pH  $5.0+1 \times 10^{-5}$ , pH  $5.0+1 \times 10^{-6}$ , pH  $5.0+1 \times 10^{-7}$ , concentration: Maximum carbohydrate content was observed at the plant age of 105 days treated with Auxin.

**Keywords:** Acid Rain, Auxin, Carbohydrate.

## Introduction

Carbohydrates are among five basic constituents required by living beings to lead a healthy life. Work has been done in recent past on different aspects of carbohydrates in plants. Function and dynamics of auxins and carbohydrates during early wood / latewood transition in scotspine have been studied by Uggla et.al.<sup>1</sup>. Gupta et.al.<sup>2</sup> studied sugar signaling and gene expression in relation to carbohydrate metabolism under abiotic stress in plants. Biological effects of acid rains and its implications were studied by Ferenbaugh<sup>3</sup>. Studies have also been done on the regulation by auxins of carbohydrate metabolism involved in cell wall synthesis by Pea stem tissue<sup>4</sup>. Andrea et.al.<sup>5</sup> investigated sugar signaling and plant development. Majid Ghorbani Javid et. al. did investigations on effects of exogenous applications of auxins and cytokinins on carbohydrate accumulation in rice grains<sup>6</sup>. Effects of auxins on soluble carbohydrates, starch and soluble protein on *Aechmea blalchetiana* have also been studied<sup>7</sup>. Kausar et.al. investigated response of simulated acid rain on morphological, biochemical and leaf characters of wheat<sup>8</sup>. Impact of simulated acid rain on carbohydrate yield has also been revealed by Kumar and Tomar<sup>9</sup>. Kaur et.al. carried out studies on effect of GA<sub>3</sub>, kinetin and indole acetic acid on carbohydrate metabolism in chick pea seedling germination<sup>10</sup>. Shripal et.al. also studied effects of acid rain on carbohydrate yield but in green pepper. Effect of phtoharmones on carbohydrate and nitrogen metabolism has also been studied<sup>12</sup>. Effect of Indole acetic acid on carbohydrate in beans was studied in 1938 by Taylor<sup>13</sup>.

Enrichment of vegetables, fruits and cereals with carbohydrates is important to improve the quality of food and therefore, experiments were conducted to increase carbohydrate content in *Capsicum frutescens*. Accordingly in one group, a set of plants of *Capsicum frutescens* were treated with normal water, second set of plants with acid water of pH3, third set with pH 4 and fourth with pH 5 concentration. Similarly, in third group sets of plants were treated with Auxin solution of  $1 \times 10^{-5}$ ,  $1 \times 10^{-6}$  and  $1 \times 10^{-7}$  ppm concentration. In fourth group sets of plants were treated with different concentrations of acid water of pH3, pH 4 and pH 5 concentration and Auxin solution of  $1 \times 10^{-5}$ ,  $1 \times 10^{-6}$  and  $1 \times 10^{-7}$  ppm concentration to assess the interactive effect of Acid rain and Auxin solution of different concentrations. The quantification was done by ANTHRONE method. The tests were done after 45, 60, 75, 90 and 105 days respectively.



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**Aim of the Study**

The objective behind experimentation being presented in this paper was to compensate detrimental effect of acid rain through treatment of plants with auxin solution of different concentration during studies. It was also aimed to enhance concentration of carbohydrate content in the leaves of *Capsicum frutescens* var. 'sweet magic' as this will give carbohydrate enriched fruits.

**Material and methods****Estimation of Carbohydrate**

Quantification of carbohydrate content is done as per protocol developed by Anthrone method (Hedge et.al., 1962)<sup>14</sup>. Carbohydrates are first hydrolysed in to simple sugars using dilute hydrochloric acid. In hot acidic medium glucose is dehydrated to hydroxymethyl furfural. This compound forms a green coloured product with anthrone with an absorption maximum at 630 nm. 100 mg oven dried powdered leaves of the control and treated plants are taken. Sample was transferred in to eppendorf tube. Now added 1 ml of 2.5 N HCL. Kept eppendorf tubes in stand. Placed it in the boiling water bath for 3 hour. Cooled the mixture to room temperature. Now added a pinch of sodium bicarbonate slowly to it until the colour disappears and no CO<sub>2</sub> releases. Now Centrifuged at 13000 rpm for 15 minutes. Taking 10µl of supernatant in a test tube and added 990 µl distilled water in test tube. Added 4 ml of anthrone (ice cold). Vertixed it (mix well). Kept it in water bath for 8 to 10 minutes. Cooled it to room temperature and O.D. at 630 nm with U.V. spectrophotometer is recorded. Carbohydrate content is calculated with the help of calibration curve.

**Results and discussion**

Table-1 shows the effect of simulated acid rain of different concentrations (pH 3.0, 4.0 and 5.0) on carbohydrate content of *Capsicum frutescens* var. *Sweet magic*. When the plants were treated with acid rain (pH 3.0), the carbohydrate content were 89.22%, 87.43%, 124.88%, 91.83% and 85.31% , with acid rain (pH 4.0), the amount of carbohydrate were 80.09%, 64.92%, 91.41%, 91.07% and 96.54% of control, while at the treatment with acid rain (pH 5.0), the carbohydrate content were 71.05%, 101.88%, 95.65%, 97.68% and 97.34% of control at the plant age of 45, 60, 75, 90 and 105 days, respectively.

Table-2 represents the effect of treatment of plants with 1x10<sup>-5</sup>, 1x10<sup>-6</sup>, 1x10<sup>-7</sup> M auxin on carbohydrate content in the leaves of *Capsicum frutescens* var. *Sweet magic*. When the plants were treated with 1x10<sup>-5</sup> M, the carbohydrate content is 118.03%, 281.99%, 187.59%, 111.10% and 138.46% of the control, while at the treatment of 1x10<sup>-6</sup> M, the carbohydrate content were 87.16%, 249.16%, 110.47%, 112.53% and 122.13% of control and when the plants were treated with 1x10<sup>-7</sup> M auxin the carbohydrate content were 78.83%, 254.97%, 104.00%, 147.36% and 159.47% of the control at the plant age of 45, 60, 75, 90 and 105 days, respectively.

Table-3 shows the Interactive effect of treatment of simulated acid rain and auxin on carbohydrate content in the leaves of *Capsicum frutescens* var. *Sweet magic*. It is noticed that when

the plants were treated with acid rain and auxin (pH 3.0+1x10<sup>-5</sup> M), the carbohydrate content were 108.07%, 78.05%, 99.35%, 107.86% and 117.33%, at the treatment of acid rain and auxin (pH 3.0+1x10<sup>-6</sup> M), the carbohydrate content were 106.23%, 121.95%, 129.24%, 123.19% and 103.91% and at the treatment of acid rain and auxin (pH 3.0+1x10<sup>-7</sup> M), the carbohydrate content were 114.06%, 131.52%, 149.76%, 142.91% and 105.08% of the control at the age of 45, 60, 75, 90 and 105 days respectively.

At the treatment of acid rain and auxin (pH 4.0+1x10<sup>-5</sup> M), the values of carbohydrate content were 121.80%, 244.65%, 277.65%, 207.14% and 127.43%, when the treatment of acid rain and auxin (pH 4.0+1x10<sup>-6</sup> M), were given , carbohydrate content were 194.93%, 369.98%, 113.24%, 110.82% and 106.35% and the carbohydrate content were 108.70%, 121.95%, 121.94%, 129.67% and 117.55% at the treatment of acid rain and auxin (pH 4.0+1x10<sup>-7</sup> M) at the age of 45, 60, 75, 90 and 105 days respectively.

When the plants were treated with acid rain and auxin (pH 5.0+1x10<sup>-5</sup> M), the carbohydrate content were 130.84%, 256.47%, 261.93%, 136.92% and 162.73%, at the treatment of acid rain and auxin (pH 5.0+1x10<sup>-6</sup> M), the carbohydrate content were 109.62%, 103.75%, 175.24%, 175.88% and 129.30% and when the treatment of acid rain and auxin (pH 5.0+1x10<sup>-7</sup> M) was given , the carbohydrate content were 140.79%, 156.66%, 129.00%, 130.55% and 110.87% of the control at the plant age of 45, 60, 75, 90 and 105 days, respectively.

The maximum value was observed at 105th day of all the treatments, while minimum was observed at the 60th day of all the treatments. The overall pattern at all the pH is variable. However, a notable fact is sudden rise in carbohydrate content from 90-105 days. Rise in chlorophyll content in this period may be responsible for it, which increases the rate of photosynthesis leading to more carbohydrate content. The decline in carbohydrate content may be attributed to the fact that at higher acidity carbohydrate may degrade. The finding of this investigation on carbohydrate contents is supported by the results of Ferrenbough (1976)<sup>15</sup>, Forshine (1983)<sup>16</sup>, Zhang et. al. (2005)<sup>17</sup> and Shaukat and Khan (2008)<sup>18</sup>.

Wager. et. al. (1954)<sup>19</sup> has recorded significant increase in carbohydrate contents in maturing green pea and attributed it to increase in leaf area. Increase in carbohydrate content is also explainable on the basis of increase in number of stomata which leads to increased photosynthetic activity. Ahmed et. al. (2012)<sup>20</sup> has recorded highest value of total sugar in the lowest rate (100 ppm) when compared with 200 and 300 ppm. Significant and insignificant increase in total sugar concentration has been reported in the leaves of tree sprayed with any of the three different rates of IAA. Altman and Wareing (1975)<sup>21</sup> reported that there is a close relationship between IAA treatment and sugar accumulation. Combination for which carbohydrate content is significant and maximum is pH 5.0+1x10<sup>-5</sup> M, which occurs at the 105th day. Combination for

which carbohydrate content is least among all significant values is pH  $5.0+1 \times 10^{-7}$  M, which occurs at 60th day. The content is lesser than control.

Increase in carbohydrate contents is through increase in number of chloroplasts (Wager et. al., 1954)<sup>19</sup>. A relationship between enhancement of sugar accumulation and IAA treatment has been reported by Altman and Wereing also (1975)<sup>21</sup>.

#### Conclusion

Carbohydrate content is maximum at 105th day for all the concentrations. Most significant rate is  $1 \times 10^{-7}$  M and most important day is 105th day. Minima are seen at 60 days at all the concentrations.

#### End Notes

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**Table 1 : Effect of simulated acid rain (pH 3.0, 4.0, 5.0) on Carbohydrate content(mg/100 mg  $\pm$ SD dry wt.) in the leaves of *Capsicum frutescens* var. *sweet magic***

| Treatment | PLANT AGE (in Days) |                     |                    |                    |                     |
|-----------|---------------------|---------------------|--------------------|--------------------|---------------------|
|           | 45                  | 60                  | 75                 | 90                 | 105                 |
| Control   | 0.96<br>$\pm$ 0.08  | 0.25<br>$\pm$ 0.05  | 0.79<br>$\pm$ 0.05 | 0.84<br>$\pm$ 0.04 | 2.36<br>$\pm$ 0.03  |
| 3.0       | 0.85<br>$\pm$ 0.19  | 0.22<br>$\pm$ 0.03  | 0.98<br>$\pm$ 0.18 | 0.77<br>$\pm$ 0.06 | 2.01*<br>$\pm$ 0.14 |
| 4.0       | 0.77*<br>$\pm$ 0.07 | 0.16*<br>$\pm$ 0.02 | 0.72<br>$\pm$ 0.04 | 0.77<br>$\pm$ 0.04 | 2.28<br>$\pm$ 0.09  |
| 5.0       | 0.68*<br>$\pm$ 0.13 | 0.25<br>$\pm$ 0.04  | 0.75<br>$\pm$ 0.05 | 0.82<br>$\pm$ 0.05 | 2.3<br>$\pm$ 0.10   |

NB \*\*=.01 level of significance \*=.05 level of significance

**Table 2 : Effect of Auxin (1x10-5,1x10-6,1x10-7 M) on Carbohydrate content (mg/100 mg. dry wt.,  $\pm$ SD.) in the leaves of *Capsicum frutescens* var. *sweet magic***

| Treatment | Plant Age (in Days) |                      |                      |                      |                      |
|-----------|---------------------|----------------------|----------------------|----------------------|----------------------|
|           | 45                  | 60                   | 75                   | 90                   | 105                  |
| Control   | 0.96<br>$\pm$ 0.08  | 0.25<br>$\pm$ 0.05   | 0.79<br>$\pm$ 0.05   | 0.84<br>$\pm$ 0.04   | 2.36<br>$\pm$ 0.03   |
| 1x10-5 M  | 1.13<br>$\pm$ 0.14  | 0.70**<br>$\pm$ 0.08 | 1.48**<br>$\pm$ 0.16 | 0.94<br>$\pm$ 0.09   | 3.27**<br>$\pm$ 0.09 |
| 1x10-6 M  | 0.83<br>$\pm$ 0.02  | 0.61**<br>$\pm$ 0.11 | 0.87<br>$\pm$ 0.12   | 0.95<br>$\pm$ 0.06   | 2.88*<br>$\pm$ 0.22  |
| 1x10-7 M  | 0.76<br>$\pm$ 0.11  | 0.63**<br>$\pm$ 0.05 | 0.82<br>$\pm$ 0.07   | 1.24**<br>$\pm$ 0.13 | 3.76**<br>$\pm$ 0.17 |

N.B. \*\*=.01 level of significance \*=.05 level of significance

**Table 3 : Interactive Effect of simulated acid rain (pH 3.0, 4.0, 5.0) and Auxin (1x10-5,1x10-6,1x10-7 M) on Carbohydrate content(mg/100 mg dry wt. $\pm$ SD ) in the leaves of *Capsicum frutescens* var. *sweet magic***

| Treatment  | PLANT AGE (in Days)  |                      |                      |                      |                      |
|------------|----------------------|----------------------|----------------------|----------------------|----------------------|
|            | 45                   | 60                   | 75                   | 90                   | 105                  |
| Control    | 0.96<br>$\pm$ 0.08   | 0.25<br>$\pm$ 0.05   | 0.79<br>$\pm$ 0.05   | 0.84<br>$\pm$ 0.04   | 2.36<br>$\pm$ 0.03   |
| 3.0+1x10-5 | 1.04<br>$\pm$ 0.04   | 0.19<br>$\pm$ 0.09   | 0.78<br>$\pm$ 0.11   | 0.91<br>$\pm$ 0.03   | 2.77*<br>$\pm$ 0.17  |
| 3.0+1x10-6 | 1.02<br>$\pm$ 0.12   | 0.30<br>$\pm$ 0.06   | 1.02*<br>$\pm$ 0.13  | 1.04*<br>$\pm$ 0.09  | 2.45<br>$\pm$ 0.16   |
| 3.0+1x10-7 | 1.09<br>$\pm$ 0.09   | 0.32<br>$\pm$ 0.05   | 1.18**<br>$\pm$ 0.12 | 1.20*<br>$\pm$ 0.15  | 2.48<br>$\pm$ 0.07   |
| 4.0+1x10-5 | 1.17*<br>$\pm$ 0.06  | 0.60**<br>$\pm$ 0.05 | 2.19**<br>$\pm$ 0.19 | 1.75**<br>$\pm$ 0.06 | 3.01**<br>$\pm$ 0.04 |
| 4.0+1x10-6 | 1.87**<br>$\pm$ 0.16 | 0.91**<br>$\pm$ 0.07 | 0.89*<br>$\pm$ 0.03  | 0.93*<br>$\pm$ 0.03  | 2.51<br>$\pm$ 0.13   |
| 4.0+1x10-7 | 1.04<br>$\pm$ 0.03   | 0.30<br>$\pm$ 0.04   | 0.96**<br>$\pm$ 0.03 | 1.09**<br>$\pm$ 0.03 | 2.77**<br>$\pm$ 0.07 |
| 5.0+1x10-5 | 1.25*<br>$\pm$ 0.10  | 0.63**<br>$\pm$ 0.04 | 2.06**<br>$\pm$ 0.45 | 1.15**<br>$\pm$ 0.05 | 3.84**<br>$\pm$ 0.18 |
| 5.0+1x10-6 | 1.05<br>$\pm$ 0.02   | 0.26<br>$\pm$ 0.01   | 1.38**<br>$\pm$ 0.04 | 1.48**<br>$\pm$ 0.13 | 3.05**<br>$\pm$ 0.09 |
| 5.0+1x10-7 | 1.35**<br>$\pm$ 0.05 | 0.39*<br>$\pm$ 0.07  | 1.02**<br>$\pm$ 0.03 | 0.10**<br>$\pm$ 0.07 | 2.61**<br>$\pm$ 0.04 |

N.B. \*\*=.01 level of significance \*=.05 level of significance