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# Effect of Gibberellic Acid ($Ga_3$) on Morphological and Physiological Attributes of Ispaghol (*Plantago ovata* L.)

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**Abstract:** This experiment involved the study of effect of gibberellic acid on Ispaghol. Ispaghol is a medicinal plant that belongs to the familyPlantaginaceae. The plants were grown in pots of equal size and diameter in loamy soil which were placed in research field. The pots were of equal diameter of 22cm that were used to sow seeds. The treatments of $GA_3$ (0, 20 and 40ppm) were given after 14 days of germination. The hormone was applied by foliar application. Two types of observations were taken after 21 and 42 days of treatment. The morphological parameters for $GA_3$were studied which showed that the $GA_3$ increased the length and growth of the plants. The maximum growth was observed in plants treated with $GA_3$ (40ppm). The transpiration rate was also increased by $GA_3$ treatment. The chemical analysis of the treated plants revealed that $GA_3$ had no effects on sodium and potassium ion concentration.

**Key words:** $GA_3$ · GA · Ispaghol · Effect of Hormones · Plant Hormone · Phytohormonal response

## INTRODUCTION

Ispaghol (*Plantago ovata*) is classified in family Plantaginaceae. It is found in temperate regions and in tropics. Ispaghol is a medicinal species of great significance cultivated in India, Pakistan and Iran. The seeds contain 20 to 30% mucilage, utilized by the pharmaceutical industry [1]. About 800 species of Plantago are found across the world. Out of these 800 about 10-14 are native to India. In different regions of the world there is multiplicity of names of plant such as ashwagolam, aspaghol, aspagol, bazarqutuna, blond psyllium, ch'-ch'ientzu, ghoda, grappicol, Indian plantago, Indische Psylli-samen, isabgul, isabgul gola, ispaghula, isphagol, vithai, issufgul, jiru, obeko, psyllium, plantain, spogel seeds[2-5].Gibberellic acid is naturally occurring plant hormone that arouses and controls plants growth. Called as $GA_3$, is responsible for cell division and cell elongation that results in an increase in plant length [6]. This hormone was discovered by Japaneses and this was found in a fungus *Gibberella fujikuroii.* [7]. It is since then cultivated and extracted into liquid [8] and is used to control plant growth which ultimately increases the yield of plants.Gibberellic acids belong to a large family of diterpene acids. Over 136 gibberellic acids are found naturally which are still discovered [9]. These are shortened as GA with a subscript such as GA1, GA2, GA3, GA4 and so on, of which GA3, commonly known as gibberellic acid, is the most significant GA in plants. GAs and compounds like GA are found in every class of plant kingdom from lower fungi to higher angiosperms. They are formed in roots and younger leaves, but they are found in higher concentration in seeds of higher plants. $GA_3$ is the leading broadly existing active form of commercial GAs. Gibberellins (GAs) play a vital role in plants and their processes which are seed germination, endosperm utilization, stem elongation, leaf extension, flower and fruit set and their structure and contents are usually associated with plant growth and progress [10]. GA is identified to disturb plant morphology [11]. $GA_3$ rises shoot length by increasing its rate of elongation in most of the plants, including *Brassica campestris* [12]. Root length was also detected to increase by $GA_3$ treatment in *Lupinus albus* [13].The seed germination proportion raised by the use of gibberellins that result in rise of the amino acid content in embryo and which are source to discharge hydrolytic enzyme needed for digestion of endospermic starch when seeds restart growth at germination. GA performs synergistically with auxins, cytokinins and might be with the other plant

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hormones; what might be called as synergism. The whole growth of plant is controlled by the growth hormones, nutrient and abiotic elements. They also fluctuate in their germination condition. Gibberellic acid ($GA_3$) is also responsible for controlling plant responses to abiotic factors [14]. $GA_3$ has also been revealed to lessen the effects of salt stress on water use efficiency [15]. Das Gupta *et al*. [16] documented that foliar use of plant growth regulators like IAA and GA aided the plant to reestablish obstruction in water content in Mungbean plants exposed to water stress.

## MATERIALS AND METHODS

The research was conducted in 2012-2013. The plants of ispaghol (*Plantago ovata*) were grown in research field of department of Botany, University of Gujrat. The objective of this research was to evaluate the effect of $GA_3$ on morpho-physiological attributes of ispaghol. The seeds were obtained from an agriculture seed shop located in Sialkot. The seeds were grown in pots of loamy soil 22cm deep. About 8-10 seeds were sown in each pot having soil weight approximately 5 Kg with 4 replicates for each.The experimental design used for the experiment was CRD (Complete Randomized Design).

**Treatments**

To (control) = Distill Water

T1= $GA_3$ 20 ppm

T2= $GA_3$ 40 ppm

Hormone treatment was applied for 14 days after germination. The first data was taken after 21 days of treatment and second data was taken 42 days of treatment. Different parameters of morphological, physiological and biochemical nature were studied.

**Morphological Studies:** Plants, roots and shoots lengths were measured with scale meter. Roots were separated from shoots and fresh and dry weights of shoots and roots were recorded with electric balance.

**Chlorophyll and Carotenoids Test:** The chlorophyll and Carotenoids tests were taken on spectrophotometer model UV 300, Company O.R.I., Reinbeker Weg 75 Hamburg, Germany. The readings of samples were taken on different wavelengths i.e. 480, 510, 645, 663. Following formula was applied on the spectrophotometer reading.

Chl.a (mg $g^{-1}$ f.wt) = [12.7 (O.D 663) - 2.69 (O.D 645)] x V/1000 x W

Chl.b (mg $g^{-1}$ f.wt) = [22.9 (O.D 645) - 4.68 (O.D 663)] x V/1000 x W

Carotenoids (mg $g^{-1}$ f.wt) = [7.6 (O.D 480) - 1.69 (O.D 510)] x V/1000 x W

W= weight of the fresh leaf (g)
V= volume of the extract (ml)

**Gaseous Exchange:** Gaseous exchange parameters including photosynthetic rate, transpiration rate, stomatal conductance, sub stomatal $CO_2$concentration etc., using open photosynthetic measurement system IRGA.

**Minerals Analysis of $K^+$ and $Na^+$:** The chemical analysis was carried out by the Digestion test. This procedure was given by WOLF in 1982. $Na^+$ and $K^+$ cations were determined with flame photometer graded series of standard curve for each element was drawn. Then the minerals tests were taken on flame photometer model SPECTROLAB S20-4 Company Spectrolab systems Ltd. Wiltshire, U.K. And the readings were multiplied with correction factor.

$$Correction\ factor = \frac{\text{total end volume (after digestion) x 100}}{\text{Sample dry weight}}$$

**Statistical Analysis:** Completely randomized design (analysis of variance technique) of the data was computed for all attributes by using COSTAT for DOS computer program version 3.03. Bar graph values were drawn by using Microsoft Excel software.

## RESULTS AND DISCUSSION

The plants treated with $GA_3$ showed a significant increase in the shoot length. It is responsible for the increase in shoot length and the maximum increase in the shoot length was observed in the plants treated with 40 ppm solution of $GA_3$. The use of gibberrellic acid ($GA_3$) to the shoot promotes an increase in both shoot and root elongation [17]. Sharma and Kumar [18] also conducted an experiment on the *Chlorophytum tuberosum* and *Pergularia daemia* to study the effects of growth regulators on these plants. The higher concentration 100ppm of $GA_3$ increased the growth in C.*tuberosum*. For root elongation, it was found that $GA_3$ has significant effect on the root elongation as it promotes cell division

First Harvest:

Table 1: Means of analysis of variance forShoot length, Root length and Shoot fresh weight

| Source | df | M.s of Shoot length | M.s of Root length | M.s of Shoot fresh weight |
|---|---|---|---|---|
| $GA_3$ | 2 | 19.360*** | 1.115ns | 0.002ns |
| Error | 9 | 0.536 | 0.288 | 0.001 |
| Total | 11 | | | |

Table 2: Means of analysis of variance for Root fresh weight, Shoot dry weight andRoot dry weight

| Source | df | M.s of Root fresh weight | M.s of Shoot dry weight | M.s of Root dry weight |
|---|---|---|---|---|
| $GA_3$ | 2 | 1.155ns | 1.75ns | 1.908ns |
| Error | 9 | 3.338 | 2.305 | 9.416 |
| Total | 11 | | | |

Table 3: Means of analysis of variance for Chlorophyll a,Chlorophyll b and Carotenoids

| Source | df | M.s of Chlorophyll a | M.s of Chlorophyll b | M.s of Carotenoids |
|---|---|---|---|---|
| $GA_3$ | 2 | 7.453ns | 5.292ns | 1.485ns |
| Error | 9 | 7.056 | 9.914 | 1.855 |
| Total | 11 | | | |

Table 4: Means of analysis of variance for CO2 assimilation rate, Transpiration rate,Stomatal conductance and Sub-Stomatal CO2 concentration

| Source | df | M.s of CO2 assimilation rate | M.s of Transpiration rate | M.s of Stomatal conductance | M.s of Sub-Stomatal CO2 concentration |
|---|---|---|---|---|---|
| $GA_3$ | 2 | 5.800ns | 174.207*** | 0.314ns | 34797.25*** |
| Error | 9 | 9.265 | 1.770 | 0.073 | 1159.527 |
| Total | 11 | | | | |

Table 5: Means of analysis of variance for K+ ion conc.in Shoot, K+ ion conc.in Root, Na+ ion conc.in Shoot and Na+ ion conc.in Root

| Source | df | M.s of K+ ion conc.in Shoot | M.s of K+ ion conc.in Root | M.s of Na+ ion conc.in Shoot | M.s of Na+ ion conc.in Root |
|---|---|---|---|---|---|
| $GA_3$ | 2 | 10.775ns | 1.75ns | 1.725ns | 0.067ns |
| Error | 9 | 16.026 | 4.081 | 0.739 | 0.069 |
| Total | 11 | | | | |

Second Harvest:

Table 6: Means of analysis of variance for Shoot length, Root length and Shoot fresh weight

| Source | df | M.s of Shoot length | M.s of Root length | M.s of Shoot fresh weight |
|---|---|---|---|---|
| $GA_3$ | 2 | 19.360*** | 1.925** | 0.013ns |
| Error | 9 | 0.536 | 0.202 | 0.008 |
| Total | 11 | | | |

Table 7: Means of analysis of variance for Root fresh weight, Shoot dry weight and Root dry weight

| Source | df | M.s of Root fresh weight | M.s of Shoot dry weight | M.s of Root dry weight |
|---|---|---|---|---|
| $GA_3$ | 2 | 5.833ns | 0.003ns | 4.3ns |
| Error | 9 | 1.055 | 0.002 | 3.311 |
| Total | 11 | | | |

Table 8: Means of analysis of variance for Chlorophyll a,Chlorophyll b and Carotenoids

| Source | df | M.s of Chlorophyll a | M.s of Chlorophyll b | M.s of Carotenoids |
|---|---|---|---|---|
| $GA_3$ | 2 | 0.006ns | 0.002ns | 7.75ns |
| Error | 9 | 0.001 | 0.002 | 6.611 |
| Total | 11 | | | |

Table 9: Means of analysis of variance for CO2 assimilation rate, Transpiration rate,Stomatal conductance and Sub-Stomatal CO2 concentration

| Source | df | M.s of CO2 assimilation rate | M.s of Transpiration rate | M.s of Stomatal conductance | M.s of Sub-Stomatal CO2 concentration |
|---|---|---|---|---|---|
| $GA_3$ | 2 | 134.101*** | 66.536** | 0.0314ns | 7105.583ns |
| Error | 9 | 2.991 | 5.140 | 0.073 | 1801.944 |
| Total | 11 | | | | |

Table 10: Means of analysis of variance for K+ ion conc.in Shoot, K+ ion conc.in Root, Na+ ion conc.in Shoot and Na+ ion conc.in Root

| Source | df | M.s of K+ ion conc.in Shoot | M.s of K+ ion conc.in Root | M.s of Na+ ion conc.in Shoot | M.s of Na+ ion conc.in Root |
|---|---|---|---|---|---|
| $GA_3$ | 2 | 12.460ns | 17.550* | 1.307ns | 0.270ns |
| Error | 9 | 7.346 | 4.029 | 0.672 | 0.082 |
| Total | 11 | | | | |

Fig. 1-17: Shows the effect of $GA_3$ on different Morphological and Physiological Attributes of Ispaghol (*Plantago ovata* L.)

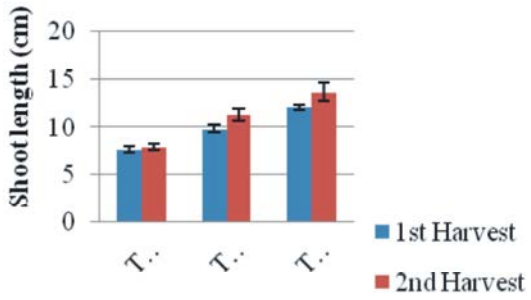


Fig. 1: Effect of $GA_3$ on Shoot length (cm) of Ispaghol

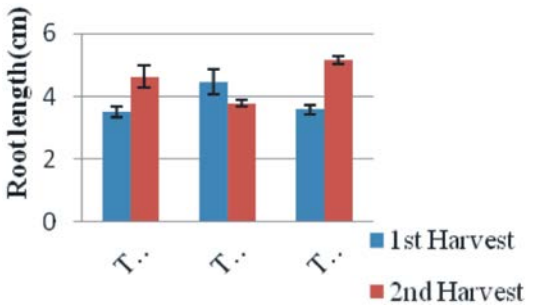


Fig. 2: Effect of $GA_3$ on Shoot length (cm) of Ispaghol

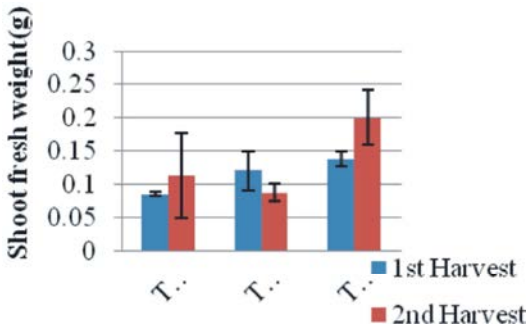


Fig. 3: Effect of $GA_3$ on Shoot Fresh weight (g) of Ispaghol

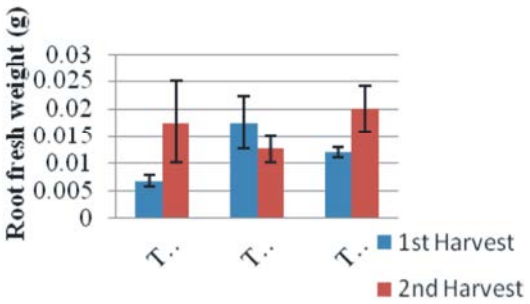


Fig. 4: Effect of $GA_3$ on Root fresh weight (g) of Ispaghol

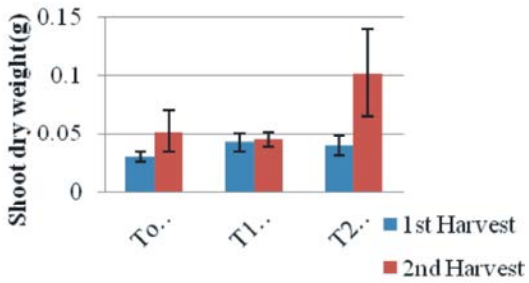


Fig. 5: Effect of $GA_3$ on Shoot dry weight (g) of Ispaghol

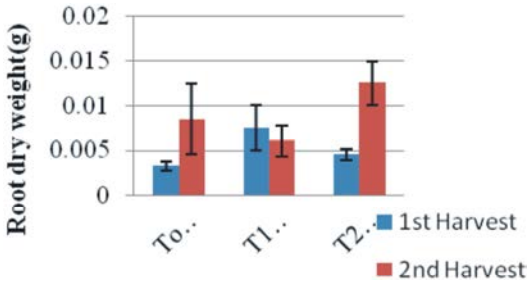


Fig. 6: Effect of $GA_3$ on Root dry weight (g) of Ispaghol

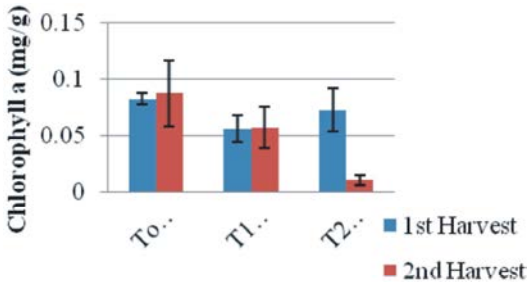


Fig. 7: Effect of $GA_3$ on ChlorophyII a (mg/g) of Ispaghol

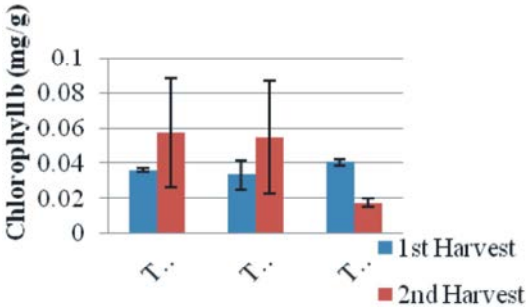


Fig. 8: Effect of $GA_3$ on ChlorophyII b (mg/g) of Ispaghol

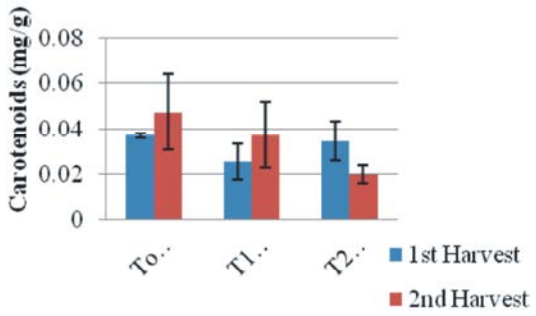


Fig. 9: Effect of $GA_3$ on ChlorophyII b (mg/g) of Ispaghol

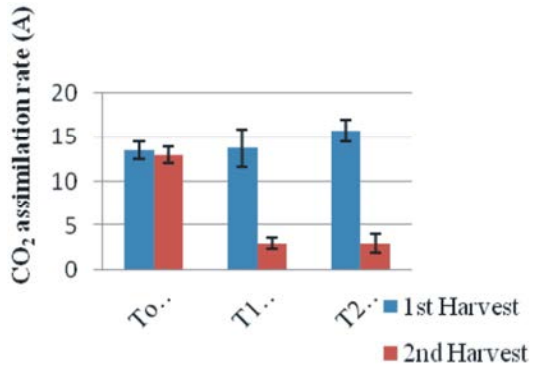


Fig. 10: Effect of $GA_3$ on $CO_2$ assimilation rate (A) of Ispaghol

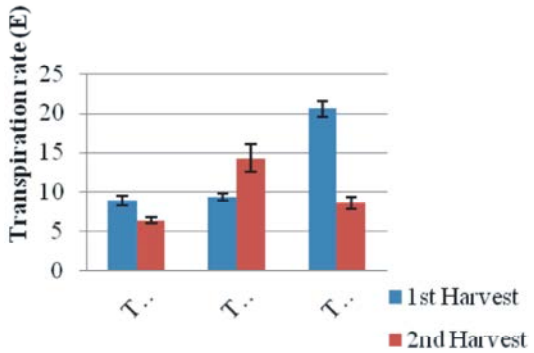


Fig. 11: Effect of $GA_3$ on Transpiration Rate (E) of Ispaghol

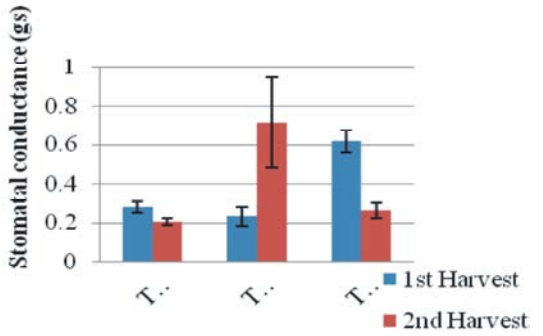


Fig. 12: Effect of $GA_3$ on Stomatal Conductance (gs) of Ispaghol

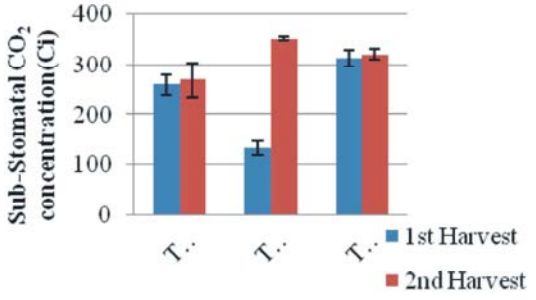


Fig. 13: Effect of $GA_3$ on Sub-Stomatal CO2 concentration (Ci) of Ispaghol

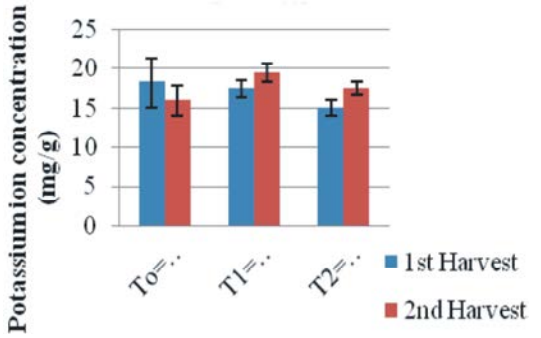


Fig. 14: Effect of $GA_3$ on Potassiumion concentration in Shoot (mg/g) of Ispaghol

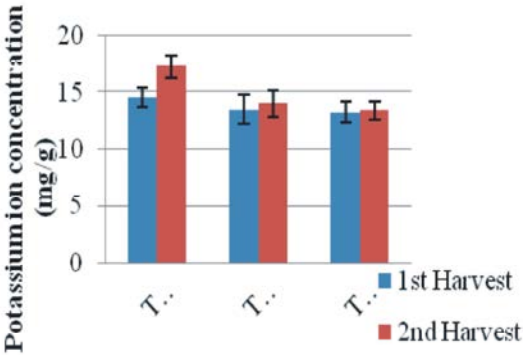


Fig. 15: Effect of $GA_3$ on Potassiumion concentration in Root (mg/g) of Ispaghol

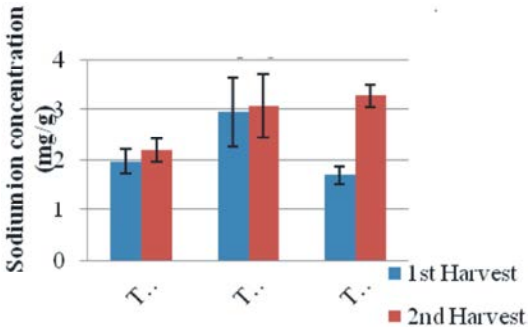


Fig. 16: Effect of $GA_3$ on Sodiumion concentration in Shoot (gm/g) of Ispaghol

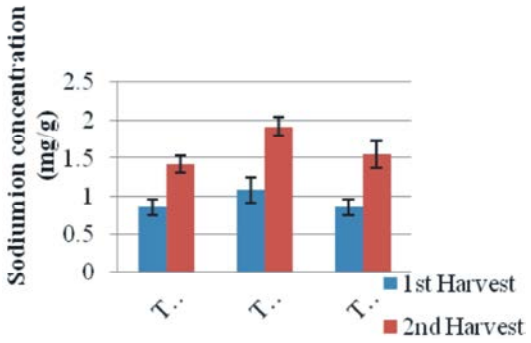


Fig. 17: Effect of $GA_3$ on Sodiumion concentration in Root (mg/g) of Ispaghol

and cell elongation. $GA_3$ increase the length of roots in various plants. But regarding this experiment the root length was not much increased by application of $GA_3$.

The results for fresh weight and dry weight of ispaghol for this experiment were non-significant. They were not affected by the application of the hormone $GA_3$.Marie *et al*. [19] conducted an experiment, concerning with the effect ofIAA & $GA_3$ on Okra *(Abelmoschus esculentus* L.M.), showed an increase in the fresh & dryweight of plant and plant height as compared with control.

The amount of chlorophyll contents was not effected by treatments given in this experiment to the plants. The other investigators found contrary results to results of this experiment. Sharaf *et al*. [20] studied the effect of $GA_3$ on growth characteristics, earliness, yield, carbohydrates and protein in *Cynara cardunculus* and investigated that highest quantity of chlorophyll content was detected in 60 µM applications (17.5 mg/ g of fresh weight) and lowest was detected in 15 µM applications of gibberellic acid (12.2 mg/g of fresh weight). Almost 28 % improvement in chlorophyll content was detected in GA3 treated plants as related to control. It is expected that the increase in the chlorophyll content may be due to phytochromes because they have relations and interactions with gibberellins. El-Abagy *et al*. [21] worked on Artichoke (*Cynara Scolymus* L.) for studying physiological and biochemical parameters to increase the productivity of the plants. The results for their experiment were different than the outcomes of this experiment. Their results showed that the treatment of $GA_3$ enhanced the chlorophyll contents and carotenoids. The most significant results were obtained by $GA_3$ at the concentration of 50 ppm and 100pm.Work on growth regulators of certain investigators can show results in accordance with this experiment. Ouzounidou *et al*. [22] worked on effects of growth regulators on onion and garlic. In their results it was found that $GA_3$ did not affect the chlorophyll contents of onion and garlic when applied in 100 µM. The stomatal conductance (gs) was also studied in ispaghol plants. The results showed that $GA_3$ has not much effect on stomatal conductance of ispaghol plants. But the transpiration rate in both the harvest was increased and effected significantly by the treatment of $GA_3$ (Table.4 &9).

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