

# Synergistic Effects of Gibberellic Acid and Triacontanol on Growth, Physiology, Enzyme Activities and Essential Oil Content of *Coriandrum sativum* L.

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## ABSTRACT

Two well known growth regulators, triacontanol (TRIA) and gibberellic acid ( $GA_3$ ), were applied on economically as well as medicinally important plant coriander (*Coriandrum sativum* L.) and their effects were noted in the present study. The plants were sprayed with either TRIA or different combinations of TRIA and  $GA_3$  at 30 days after sowing. Among the treatments, a foliar spray of  $10^{-6}$  T +  $10^{-6}$  G significantly promoted the values for most of the growth (shoot and root lengths, fresh and dry weights), physiological and biochemical (total chlorophyll and carotenoid, nitrate reductase activity, carbonic anhydrase activity, leaf nitrogen and leaf phosphorus content except leaf potassium content) parameters, including the essential oil content and yield characteristics (number of umbels per plant, number of fruits per umbel, 100-seed weight and seed yield). On the basis of data obtained from the present work, it may be concluded that a combined spray of TRIA and  $GA_3$  ( $10^{-6}$  T +  $10^{-6}$  G) on coriander plant is highly effective for productivity with increased essential oil content.

**Keywords:** coriander, carbonic anhydrase activity, leaf nitrogen, nitrate reductase activity, phosphorus and potassium content

## INTRODUCTION

Coriander (*Coriandrum sativum* L.; Umbelliferae family) is widely adapted to a variety of climate and soil types in India. It occupies 0.42 million ha with an annual production of 0.25 million tonnes of seed and is mainly grown during the winter season on the northwestern plains of the country. The productivity of coriander seed is 595 kg ha<sup>-1</sup> in India, which is very low. One of the main reasons for the low productivity is that this crop is grown in areas characterized by light soils with medium fertility (Diederichsen 1996; Kumar *et al.* 2008). Among a large number of cultured plants representing the Caucasus flora, a special position is occupied by coriander. The flowering stem, which is slender and smooth, reaches a height of 20-120 cm. Hermaphrodite and staminate flowers occur in each umbel. The fruits are nearly globular, 3-4 mm in diameter and are yellow-brown when ripe. The fruits consist of two halves, single-seeded mericarps (The Wealth of India 2001). The fruits of coriander produce a normalizing action, and the related preparations containing ethereal oils are used to improve the appetite and digestion. Decoctions of coriander fruits and leaves are used for the treatment of neurasthenia and some liver and gall-bladder disorders (Ceska *et al.* 1988). Coriander fruits were also recommended as an antiseptic, expectorant and pain-relieving remedy in cases of gastritis and gastric ulceration. These fruits enter into the compositions of well-known bile-expelling and laxative herbal teas. The coriander plant yields two primary products that are used for flavoring purposes: the fresh green herb and the spice (Pruthi 1980; The Wealth of India 2001).

Triacontanol (TRIA; Fig. 1) is a synthetic growth regulator and naturally synthesizes as a plant epicuticular wax. TRIA is a long chain primary alcohol (C<sub>30</sub>H<sub>61</sub>OH) known to be a potent plant growth promoting substance for many agricultural and horticultural crops (Ries 1985, 1991). Its efficiency has been proved essential for high yield in a number of horticultural crops like barley, rice, tomatoes, maize, lettuce, cucumber, potatoes, cauliflower, brinjal,

chillies, opium and hyacinth bean (Nagoshi and Kawashima 1996; Muthuchelian *et al.* 1997; Borowski *et al.* 2000; Kumaravelu *et al.* 2000; Khan *et al.* 2006, 2007; Naeem *et al.* 2009). TRIA raises plant productivity by improving photosynthesis and cell division (Haugstad *et al.* 1983).

On the other hand,  $GA_3$  (Fig. 1), a hormone found in plants, is a simple gibberellin, which promotes growth and elongation of cells. Since  $GA_3$  regulates growth, applications of very low concentrations have a profound effect while too much have the opposite effect but eventually plants develop tolerance to it (Sadowska *et al.* 1984).  $GA_3$

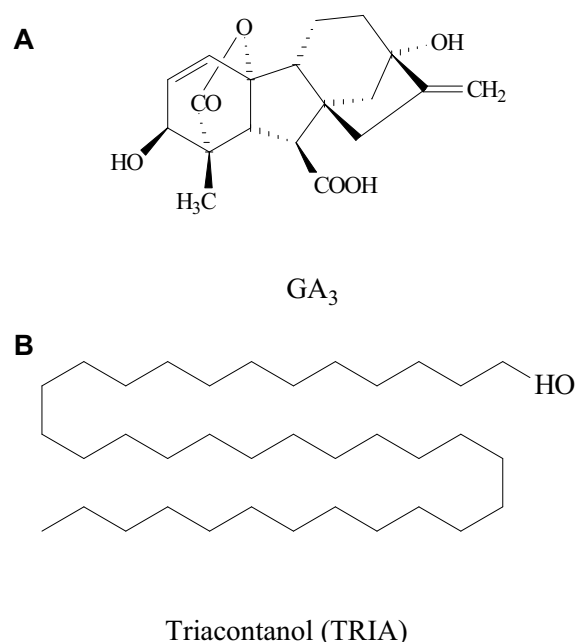


Fig. 1 Structural formulae. (A) Gibberellic acid ( $GA_3$ ); (B) triacontanol (TRIA).

have also a number of effects on plant development including stimulate rapid stem growth, induce mitotic division in the leaves of some plants and increase seed germination rate (Riley 1987; Tipirdamaz and Gomurgan 2000).

Previous studies (Ohlsson and Berglund 2001; Srivastava and Srivastava 2007) suggested that GA<sub>3</sub> enhances the production of active constituents in all plant parts but minimize the total biomass production which result in decreased overall production of secondary metabolites such as alkaloid and essential oil. However, TRIA increased rapidly fresh and dry weight in contrary to GA<sub>3</sub> (Kumaravelu *et al.* 2000). Keeping the economical as well as medicinal importance of coriander in mind, a hypothesis was designed to investigate whether the TRIA alone or in combination with GA<sub>3</sub> could promote physiological and biochemical attributes and plant productivity including essential oil content.

## MATERIALS AND METHODS

### Plant material and growth conditions

The seeds of coriander (*Coriandrum sativum* L. var. CO-2) were obtained from the Indian Agricultural Research Institute, New Delhi. Healthy seeds of uniform size were selected and their viability was tested by keeping the overnight soaked seeds in 1% tetrazolium salt solution for 1 hr. It was found that 90% of seeds were viable. Thereafter, the seeds were surface sterilized with 95% ethanol followed by repeated washing with distilled water. Prior to seed sowing, 5 kg homogenous mixture of soil and organic manure in the ratio of 4: 1 was filled in the earthen pots (25 cm diameter × 25 cm height). A uniform basal dose of nitrogen, phosphorus and potassium was given at a rate of 15, 25 and 25 kg ha<sup>-1</sup>, respectively. The soil was maintained at proper moisture to ensure better growth of the plants. The seeds were sown directly at a depth of 2 cm in each earthen pot.

### Experimental design, growth and yield analyses

The experiment was conducted in the naturally illuminated environmental conditions during 2008-2009 in the net-house of the Department of Botany, AMU, Aligarh (27° 53'N latitude, 78° 51' E longitude, and 187.45 masl). The design of the experiment was simple randomized. The position of each pot was randomized at 4-day intervals to minimize spatial effects in the net-house. The plants were sprayed with deionized water (control), 10<sup>-6</sup> M TRIA (T) and with different combinations of GA<sub>3</sub> (G) viz. 10<sup>-6</sup> T + 10<sup>-8</sup> G, 10<sup>-6</sup> T + 10<sup>-6</sup> G and 10<sup>-6</sup> T + 10<sup>-4</sup> G at 30 days after sowing (DAS). Molar concentrations were used for all spray treatments. The treatments were given regularly as foliar spray in 10-day intervals. For each treatment three replicates were used. The sampling was done at the flowering stage (60 DAS). At this stage, three plants from each treatment were uprooted carefully and washed with tap water to remove all adhering foreign particles. They were dried thereafter using blotting paper. Then, the plant fresh weight was recorded. The plants were dried at 80°C for 24 hr using a hot air oven, and the plant dry weight was recorded. Umbels were threshed and cleaned. The number of fruits per umbel was recorded. Afterward, 100 seed weight and seed-yield per plant was calculated accordingly. Yield parameters were recorded at the time of harvest (90 DAS).

### Physiological and biochemical analyses

The fresh leaves were used for the analysis of various physiological and biochemical attributes except leaf N, P and K content.

#### Estimation of total chlorophyll and carotenoid content

Total chlorophyll and carotenoid content in fresh leaves were determined according to the methods of Lichtenthaler and Buschmann (2001). The method consisted of repeated acetone extraction until obtained colourless residue, using a pestle and mortar and filtered over cotton pad. The extracts were made up to 50 ml with acetone. The concentration of chlorophyll and carotenoid contents

were measured using a spectrophotometer (Shimadzu UV-1700, Tokyo, Japan). Total chlorophyll and carotenoid content were expressed as mg g<sup>-1</sup> FW.

#### Determination of nitrate reductase (NR) activity

The enzyme activity was estimated by the intact tissue method developed by Jaworski (1971), which is based on the reduction of nitrate to nitrite as per the following biochemical reaction:



The nitrite formed was determined spectrophotometrically, where 200 mg of fresh chopped leaves were weighed and transferred to a plastic vial. Each vial contained 2.5 ml phosphate buffer (pH 7.5), 0.5 ml potassium nitrate solution and 5% isopropanol. After incubation of this mixture, 1% sulphanilamide and 0.02% N-(1-naphthyl) ethylenediamine dihydrochloride (NED-HCl) were added. The test tubes were kept for 20 min at room temperature for colour development. The OD of colour developed was read at 540 nm using a spectrophotometer. The NR activity was expressed as nM NO<sub>2</sub><sup>-</sup> g<sup>-1</sup> FW h<sup>-1</sup>.

#### Determination of carbonic anhydrase (CA) activity

The carbonic anhydrase (CA) activity was measured in fresh leaves, using the method described by Dwivedi and Randhawa (1974). 200 mg of fresh leaf pieces were weighed and transferred to Petri dishes. The leaf pieces were dipped in 10 ml of 0.2 M cysteine hydrochloride solution for 20 min at 48°C. Thereafter, 4 ml of 0.2 M sodium bicarbonate solution and 0.2 ml of 0.002% bromothymol blue were added. The reaction mixture was titrated against 0.05 N HCl using methyl red as an indicator. CA was expressed as μM CO<sub>2</sub> kg<sup>-1</sup> leaf FW s<sup>-1</sup>.

#### Nutrient analysis

Leaf samples of each treatment were digested for the estimation of leaf N, P and K content. The leaves were dried in a hot air oven at 80°C for 24 h. Dried leaves were powdered and the powder was passed through a 72 mesh. The sieved powder was used for N, P and K content. 100 mg of oven-dried leaf powder was carefully transferred to a digestion tube and 2 ml of sulphuric acid was added to it. It was heated on a temperature controlled assembly for about 2 h. After heating, the contents of the tube turned black. It was cooled for about 15 min at room temperature and then 0.5 ml 30% hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) was added drop by drop. The addition of H<sub>2</sub>O<sub>2</sub> followed by heating was repeated until the contents of the tube turned colourless. The prepared aliquot (peroxide-digested material) was used to estimate N, P and K content. Leaf N, P and K content were expressed in terms of percent dry weight.

#### Estimation of nitrogen content

Leaf-N content was estimated according to the method of Lindner (1944). A 10 ml aliquot (peroxide-digested material) was poured into a 50 ml volumetric flask. Then 2 ml of 2.5 N NaOH and 1 ml of 10% sodium silicate solutions were added to neutralize the excess of acid and to prevent turbidity. In a graduated test tube, 5 ml aliquot of this solution was taken and then 0.5 ml Nessler's reagent was added. The contents of the test tubes were allowed to stand 5 min for maximum colour development. The OD of the solution was recorded at 525 nm, using a spectrophotometer. The reading of each sample was compared with the standard calibration curve of ammonium sulphate to estimate the percent N content.

#### Estimation of phosphorus content

The method of Fiske and Subba Row (1925) was used to estimate the leaf-P content in the digested material. The same aliquot was used to determine the leaf-P content. A 5 ml aliquot was poured into a 10 ml graduated test tube. One ml molybdic acid (2.5%) was added, followed by addition of 0.4 ml 1-amino-2-naphthol-4-sulphonic acid. When the color of the tube contents became blue, the

volume was increased to 10 ml with the addition of double distilled water. The solution was shaken for 5 min. The OD of the solution was recorded at 620 nm using the spectrophotometer.

### Estimation of potassium content

Potassium content was analyzed using a flame-photometer as suggested by Hald (1946). The solution (peroxide-digested material) is discharged through an atomizer in the form of a fine mist into a chamber, where it is drawn into a flame. Combustion of the elements produces light of a particular wavelength [ $\lambda$  max for K=767 nm (violet)]. The light produced was conducted through the appropriate filter to impinge upon a photoelectric cell that activates a galvanometer. Potassium content was estimated in the aliquot with the help of emission spectra using specific filter in a flame-photometer.

### Estimation of essential oil content

Dried fruits were powdered using a mortar and pestle. The samples of the powdered fruits were steam distilled for four hours using a Clevenger-type apparatus and the essential oil was extracted and determined gravimetrically.

### Statistical analysis

Each pot was treated as one replicate and all the treatments were repeated three times. The data was analyzed statistically with SPSS-17 statistical software (SPSS Inc., Chicago, IL, USA). Means were statistically compared by Duncan's multiple range test at  $P < 0.05\%$  using different letters. Data were presented as mean ( $n = 4$ )  $\pm$  SE.

## RESULTS

Data revealed that the effects of TRIA alone and its combined application with GA<sub>3</sub> on all parameters except leaf potassium content was found to be significant (Figs. 2-4, Tables 1, 2).

## Growth parameters

The treatment  $10^{-6}$  T +  $10^{-6}$  G proved the best for most of the growth parameters including shoot and root lengths, fresh and dry weights. However, treatment  $10^{-6}$  T +  $10^{-6}$  G, showed parity with that of treatment  $10^{-6}$  T +  $10^{-8}$  G in its effect for shoot length, root length and dry weight and producing 37.91, 34.78 and 33.3% higher values respectively in comparison to their respective controls (Fig. 2A-D). The effect of TRIA alone at concentration of  $10^{-6}$  M was significant on all growth attributes (Fig. 2A-D). Treatment  $10^{-6}$  T +  $10^{-6}$  G also proved superior and registered 40% higher value for fresh weight than that of control (Fig. 2C). However, treatment  $10^{-6}$  T +  $10^{-4}$  G showed inhibitory response in comparison to other treatments tested on the plants (Fig. 2A-D).

## Physiological and biochemical parameters

Of the five treatments,  $10^{-6}$  T +  $10^{-6}$  G followed by  $10^{-6}$  T +  $10^{-8}$  G proved to be best, enhancing the total chlorophyll content by 49.01%. The value of control recorded was minimum (Fig. 3A-D). The total carotenoid content was also increased by the combination of treatment  $10^{-6}$  T +  $10^{-6}$  G followed by treatment  $10^{-6}$  T +  $10^{-8}$  G being 26.69% exceeding the control (Fig. 3B) Carbonic anhydrase activity was found maximum in plants treated with  $10^{-6}$  T +  $10^{-6}$  G followed by treatment  $10^{-6}$  T +  $10^{-8}$  G and registered 27.49 and 21.51% higher values for CA activity than that of control (Fig. 3D). However, treatments  $10^{-6}$  T and  $10^{-6}$  T +  $10^{-4}$  G were also found significant. Of the five levels, treatment  $10^{-6}$  T +  $10^{-6}$  G proved to be best and generated the maximum NR activity (29.69% over the control) followed by treatments  $10^{-6}$  T +  $10^{-8}$  G and  $10^{-6}$  T that registered 22.67 and 10.14% higher values, respectively for NR activity than the control (Fig. 3C). However, treatment  $10^{-6}$  T +  $10^{-4}$  G exhibited a value equal to that of the control (Fig. 3C).

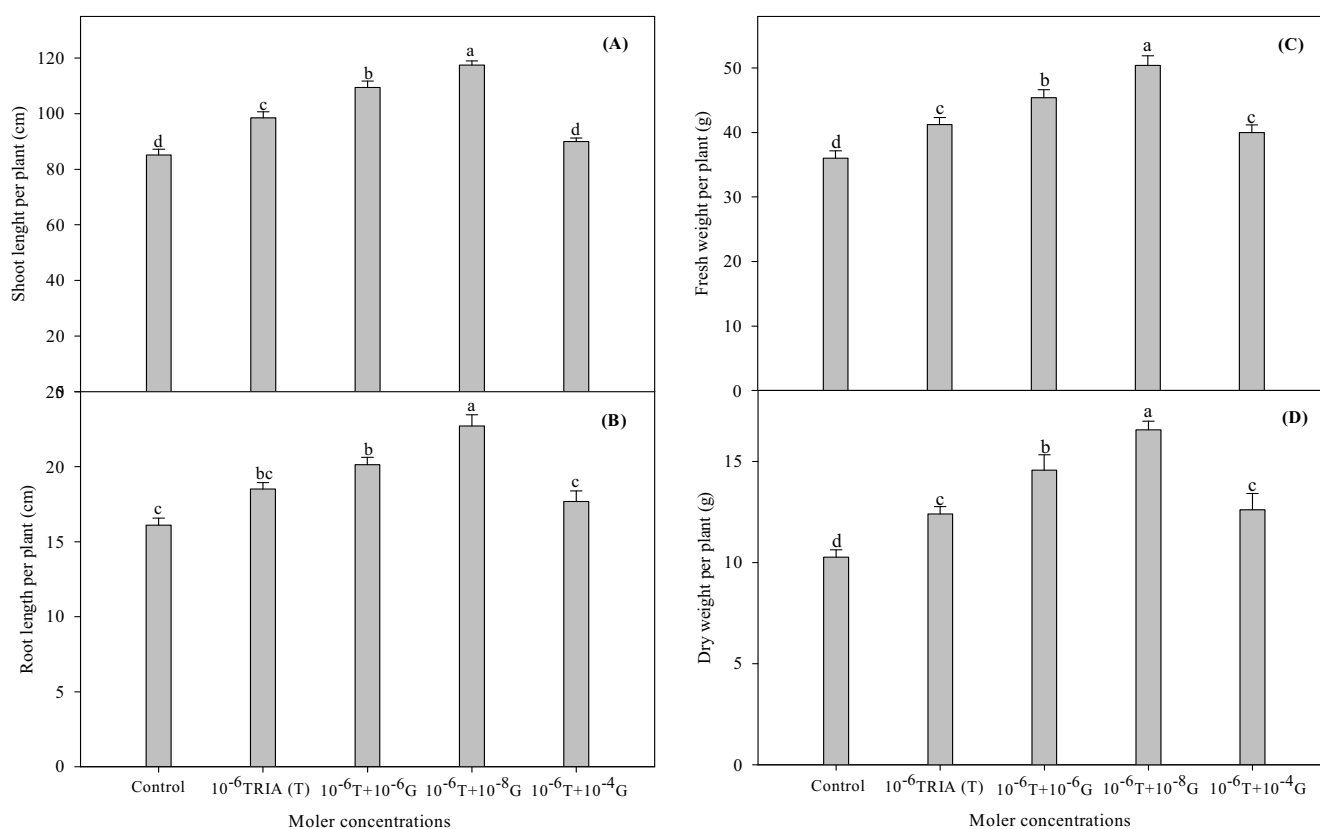


Fig. 2 Effect of spray of combined application of TRIA and GA<sub>3</sub> on shoot length (A), root length (B), fresh weight (C) and dry weight (D).

**Table 1** Effect of spray of TRIA and the combined application of TRIA and GA<sub>3</sub> on leaf N, P and K content. Means within a column followed by the same letter are not significantly different ( $P \leq 0.05$ ). The data shown are means of four replicates  $\pm$  SE.

Treatments	Leaf-N content (%)	Leaf-P content (%)	Leaf-K content (%)
Control	2.094 $\pm$ 0.009 d	0.259 $\pm$ 0.002 c	2.296 $\pm$ 0.003 a
10 <sup>-6</sup> TRIA (T)	2.450 $\pm$ 0.009 c	0.303 $\pm$ 0.005 b	2.278 $\pm$ 0.005 a
10 <sup>-6</sup> T+10 <sup>-8</sup> G	2.652 $\pm$ 0.056 b	0.321 $\pm$ 0.006 ab	2.299 $\pm$ 0.009 a
10 <sup>-6</sup> T+10 <sup>-6</sup> G	2.931 $\pm$ 0.072 a	0.349 $\pm$ 0.011 a	2.285 $\pm$ 0.004 a
10 <sup>-6</sup> T+10 <sup>-4</sup> G	2.153 $\pm$ 0.028 d	0.299 $\pm$ 0.005 b	2.283 $\pm$ 0.009 a

### Leaf-N, -P and -K content

The effect of treatments 10<sup>-6</sup> T + 10<sup>-6</sup> G, 10<sup>-6</sup> T + 10<sup>-8</sup> G and 10<sup>-6</sup> T were found to be significant for leaf-N and leaf-P content. The same treatments produced 39.9, 26.6 and 17.0% higher values for leaf-N content and 35.0, 21.0 and 17.0% for leaf-P content than the control (Table 1). However, leaf-K content was not significant by applying all treatments (Table 1).

### Essential oil content

The effect of treatment 10<sup>-6</sup> T + 10<sup>-6</sup> G proved to be best followed by 10<sup>-6</sup> T + 10<sup>-8</sup> G and producing 15.66 and 9.03% higher values for essential oil content in comparison to control (Fig. 4). However, the effect of treatment 10<sup>-6</sup> T and 10<sup>-6</sup> T + 10<sup>-6</sup> G was found non significant for this parameter.

### Yield characteristics

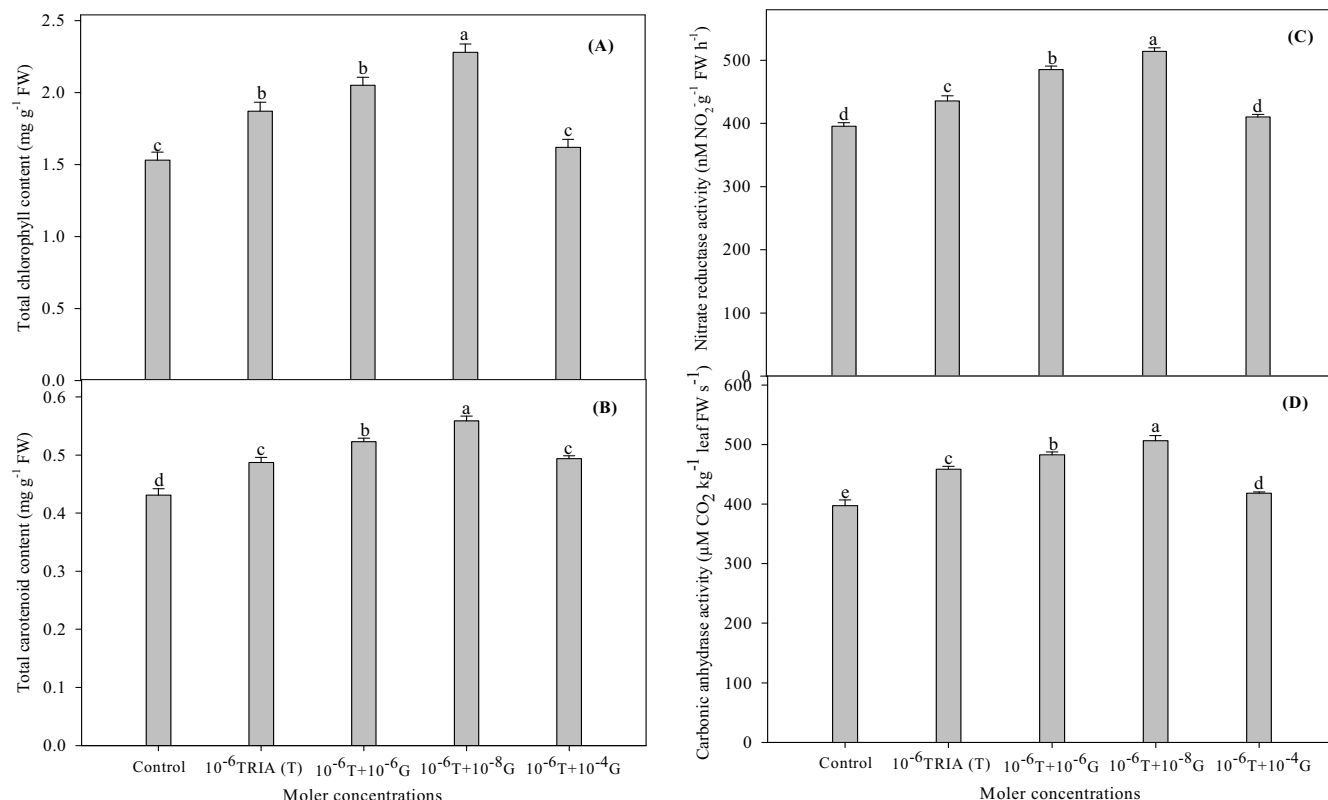
Number of umbels was maximum in plants treated with 10<sup>-6</sup> T + 10<sup>-6</sup> G followed by 10<sup>-6</sup> T + 10<sup>-8</sup> G by 58.90 and 31.50% than that of control (Table 2). Fruits per umbel were also found maximum in plants treated with 10<sup>-6</sup> T + 10<sup>-6</sup> G that produced 18.57% higher fruits per umbel exceeding the control (Table 2). However, the value given by treatment 10<sup>-6</sup> T + 10<sup>-6</sup> G equaled with that of treatments 10<sup>-6</sup> T + 10<sup>-8</sup> G and 10<sup>-6</sup> T in its effect. A combined application of 10<sup>-6</sup> T + 10<sup>-6</sup> G followed by 10<sup>-6</sup> T + 10<sup>-8</sup> G gave maximum 100-seed weight over the control (Table 2). However, treatment 10<sup>-6</sup> T showed poorest response among all treatments and the effect of treatment 10<sup>-6</sup> T + 10<sup>-8</sup> G was not significant on 100-seed weight (Table 2). As expected, seed yield was also found maximum in plants treated with 10<sup>-6</sup> T + 10<sup>-6</sup> G followed by treatment 10<sup>-6</sup> T + 10<sup>-8</sup> G and registered 109.5 and 57.7% higher values for seed yield respectively, as compared to control (Table 2). However, the effect of treatments 10<sup>-6</sup> T and 10<sup>-6</sup> T + 10<sup>-8</sup> G were found non significant for seed yield.

### DISCUSSION

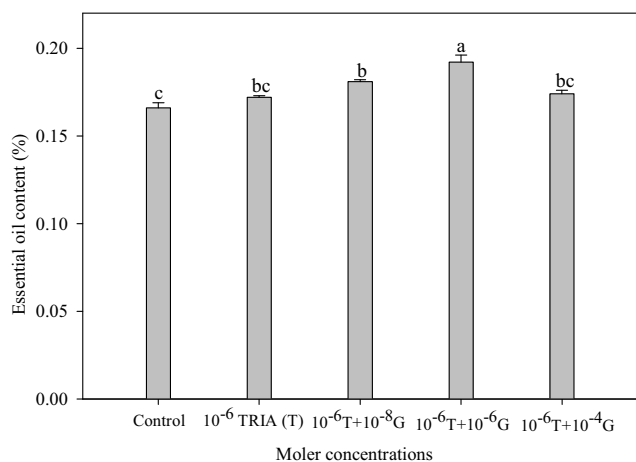
Compared with control, the observed increase in shoot and root lengths, fresh weight and dry weights in plants treated with TRIA and in combination with GA<sub>3</sub> could be ascribed by the pivotal roles of both in plants (Fig. 2A-D). GA<sub>3</sub> treatment promotes cell enlargement and cell division (Liu and Loy 1976; Moore 1989; Arteca 1996; Buchanan *et al.* 2000) while TRIA rapidly elicits a secondary messenger

**Table 2** Effect of spray of TRIA and the combined application of TRIA and GA<sub>3</sub> on yield parameters. Means within a column followed by the same letter are not significantly different ( $P \leq 0.05$ ). The data shown are means of four replicates  $\pm$  SE.

Treatments	Umbel number per plant	Fruits per umbel	100-seed weight	Seed yield
Control	7.3 $\pm$ 0.33 c	18.3 $\pm$ 0.33 d	0.698 $\pm$ 0.003 d	0.941 $\pm$ 0.08 c
10 <sup>-6</sup> TRIA (T)	7.6 $\pm$ 0.33 c	19.6 $\pm$ 0.33 bc	0.724 $\pm$ 0.005 c	1.078 $\pm$ 0.04 c
10 <sup>-6</sup> T+10 <sup>-8</sup> G	9.6 $\pm$ 0.33 b	20.3 $\pm$ 0.33 b	0.754 $\pm$ 0.006 b	1.484 $\pm$ 0.08 b
10 <sup>-6</sup> T+10 <sup>-6</sup> G	11.6 $\pm$ 0.33 a	21.7 $\pm$ 0.33 a	0.785 $\pm$ 0.006 a	1.971 $\pm$ 0.11 a
10 <sup>-6</sup> T+10 <sup>-4</sup> G	7.3 $\pm$ 1.00 c	18.6 $\pm$ 0.33 cd	0.714 $\pm$ 0.006 cd	0.991 $\pm$ 0.07 c



**Fig. 3** Effect of spray of combined application of TRIA and GA<sub>3</sub> on total chlorophyll content (A), total carotenoid content (B), nitrate reductase activity (C) and carbonic anhydrase (D).



**Fig. 4** Effect of spray of combined application of TRIA and GA<sub>3</sub> on essential oil content.

which moves rapidly throughout the plant resulting in stimulation of growth (dry weight increase) and water uptake (Ries and Wert 1988). GA<sub>3</sub> also stimulates the cells of germinating seeds to produce mRNA molecules that code for hydrolytic enzymes (Piskurewicz *et al.* 2008). GA<sub>3</sub> is a very potent hormone whose natural occurrence in plants controls their development.

Earlier studies have reported that GA<sub>3</sub> application as foliar spray on transplanted cuttings increased plant height (Sadowska *et al.* 1984; Dumivic *et al.* 2009). Srivastava and Srivastava (2007) reported that foliar spray of GA<sub>3</sub> application increased plant height and leaf length. An increase in growth parameters like shoot and root lengths, fresh and dry weights in plants treated with combined application of 10<sup>-6</sup> T + 10<sup>-6</sup> G is in accordance with the well known fact that exogenous application of plant growth regulators evokes the intrinsic genetic potential of the plant causing increase in elongation of internodes as a consequence of cell division and cell wall extensibility (Moore 1989; Taiz and Zeiger 2004; Khan *et al.* 2006). The growth promoting effects of TRIA on various attributes especially those on shoot and root lengths, fresh and dry weights has been reported by Misra and Srivastava (1991), Kumaravelu *et al.* (2000), Muthuchelian *et al.* (2003), Naeem and Khan (2005), Chaudhary *et al.* (2006), Khan *et al.* (2006), Sharma *et al.* (2006), Khan *et al.* (2007) Naeem *et al.* (2009), and Aftab *et al.* (2010) in various medicinal crops. Combined application of 10<sup>-6</sup> T + 10<sup>-6</sup> G also increased total chlorophyll and carotenoid content by 49 and 33.98%, respectively over their respective controls. In contrast, Srivastava and Shrivastava (2007) reported that GA<sub>3</sub> treatment significantly reduced total chlorophyll content. However, the present study reveals significant enhancement of chlorophyll and carotenoid content when both GA<sub>3</sub> and TRIA were applied together on the plant leaves. The earlier researchers suggested that TRIA directly activates the genes that control photosynthesis. These genes in turn activate the enzymes controlling the chemistry of photosynthesis (Houtz *et al.* 1985; Trewavas and Gilroy 1991). Similar effects of TRIA were obtained in several crops plants (Srivastava and Sharma 1990; Misra and Srivastava 1991; Ivanov and Angelov 1997; Kumaravelu *et al.* 2000; Chen *et al.* 2003; Muthuchelian *et al.* 2003). An increase in total chlorophyll and photosynthetic CO<sub>2</sub> assimilation and specific activity of Rubisco by TRIA (Houtz *et al.* 1985; Trewavas and Gilroy 1991; Khan *et al.* 2006, 2007; Naeem *et al.* 2009) and by GA<sub>3</sub> (Levy *et al.* 1986; Salisbury and Ross 1992; Taiz and Zeiger 2004) have also been recorded. A significant increase in carbonic anhydrase activity in plants treated with 10<sup>-6</sup> T + 10<sup>-6</sup> G and 10<sup>-6</sup> T + 10<sup>-8</sup> G were also observed. TRIA spray increased carbonic anhydrase (CA) activity, with 10<sup>-6</sup> T proving to be best. In the present study, TRIA-treated leaves showed a greater CA activity than the control (Fig. 3D). Such a res-

ponse of the plants to the applied TRIA is expected because TRIA increased the stomatal conductance that might have facilitated the diffusion of carbon dioxide into the stomata. In turn, the CO<sub>2</sub> might have been acted upon by CA. Finally, the CO<sub>2</sub> could be reduced by ribulose-1,5-bisphosphate carboxylase/oxygenase (RuBPCO) in the chloroplast stroma. A probable reason for the enhancement of CA activity due to TRIA application might be the de novo synthesis of CA, which involves translation/transcription of the genes associated (Okabe *et al.* 1980).

Enhancement of CA activity in treated plants might have been responsible for the enhanced rate of CO<sub>2</sub> fixation and hence have resulted in significant increase in fresh and dry weights of treated plants. Such a response at concentration of TRIA (10<sup>-6</sup> M) has also been reported by Ries and Houtz (1983), Srivastava and Sharma (1990), Misra and Srivastava (1991), Kumaravelu *et al.* (2000), Muthuchelian *et al.* (2003) and Naeem *et al.* (2009).

NR is the key enzyme in nitrogen metabolism and is responsible for the initiation of nitrate assimilation and hence protein synthesis. An increase in NR activity by combined application of TRIA and GA<sub>3</sub> may have exerted a pivotal role in enhancement of photosynthetic rate. The ultimate culmination of enhancement of CA and NR activity has increased overall growth and yield of treated plants as observed in Fig. 2A-D and Tables 1 and 2.

An adequate supply of mineral nutrients (N, P and K) in the initial stage of plant growth and development plays a pivotal role, since we have supplied uniform basal doses of nitrogen, phosphorus and potassium in the soil. Leaf-N and leaf-P contents were found significantly maximum in plant treated with 10<sup>-6</sup> T + 10<sup>-6</sup> G and 10<sup>-6</sup> T + 10<sup>-8</sup> G. GA<sub>3</sub> increased membrane permeability (Wood and Paleg 1972; Crozier and Turnbull 1984; Al-Wakeel *et al.* 1995). An increase in membrane permeability would facilitate absorption and utilization of mineral nutrient (Khan *et al.* 1998; Ansari 1996) and also transport of assimilate. This would also contribute towards enhancing the capacity of the treated plants for biomass production as reflected in the observed increase in fresh and dry weights of plant. The concentrations of leaf nutrients (N, P and K) were found significantly greater at 10<sup>-6</sup> M TRIA over the control. Enhancement in leaf-nutrients, particularly N, due to TRIA application could be attributed to the compositional or chemical change in plants leading to alterations in nitrogen concentration (Knowles and Ries 1981). Presumably, increased uptake of nutrients enhanced photosynthesis and improved translocation of photosynthates and other metabolites to the sinks that might have contributed to the improved yield of TRIA treated plants. These findings are in accordance with data on TRIA effects reported regarding plant nutrient elements (Ries and Houtz 1983; Kumaravelu *et al.* 2000; Chaudhary *et al.* 2006; Khan *et al.* 2006, 2007; Naeem *et al.* 2009).

This sustained increase in the above mentioned parameters of the treated plants which is expected to culminate in maximization of umbel number, number of fruits per umbel, 100 seed-weight and seed-yield would have a positive effect on the essential oil content.

Thus, it may be suggested that combined spray of GA<sub>3</sub> and TRIA (10<sup>-6</sup> T + 10<sup>-6</sup> G) on coriander plant would be highly effective for biomass production with increased essential oil content.

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