Role of IAA, GA_3 and riboflavin for crop improvement in fenugreek (*Trigonella foenum-graecum* L.)

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Abstract

Fenugreek (*Trigonella foenum-graecum* L.) is an important winter leafy vegetable with high nutritional values. Its seeds are used as spices and the whole plant is used in many home remedies and medicines throughout the world. Its foliage and seed productivity is much low in the region. Plant growth regulators (PGRs) and vitamin (riboflavin) are exogenously applied chemicals for enhancement of plant productivity. For this purpose, 50 mmol L^{-1} treatments of indole acetic acid, gibberellic acid, and riboflavin were applied on fenugreek variety Kasuri Methi. Results showed that GA_3 and indole-3-acetic acid significantly enhanced the root and shoot development, foliage growth and ions concentrations in shoot and root, and peroxidase dismutase (POD) and catalases (CAT) activities at seedling and vegetative stages. PGRs also increased the pod and seed yield of fenugreek. Riboflavin did not show any significant effect on fenugreek except that it increased the antioxidant activities at seedling stage. GA_3 was more effective as compared with IAA for the enhancement of biomass production, yield, and biochemical attributes of fenugreek. It was determined that indole acetic acid and gibberellic acid can be used to enhance the foliage biomass production and yield but GA_3 is superior over IAA in fenugreek. These outcomes can be useful for economic benefits with high production of this leafy vegetable.

Keywords: PGRs, vitamin, growth, yield, antioxidants

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Introduction

Fenugreek (*Trigonella foenum-graecum* L.) is an annual leafy vegetable and the oldest known cultivated medicinal plant that belongs to family Fabaceae (Khorshidian et al., 2016). High concentration of niacin, ascorbic acid, protein, and potassium in fenugreek make it a high nutritious vegetable (Mullaicharam et al., 2013). Fenugreek seeds and leaves are widely consumed in subcontinent of Indo-Pak as well as in many countries as a food flavoring and spices due to its strong flavor and aroma and as an ingredient in many traditional medicines (Bukhari et al., 2008). Fenugreek is grown in India, Pakistan, and Mediterranean countries. It is also known as a medicinal plant which has many pharmacological effects and is used as antioxidant, fungicide, hypoglycemic, stimulant of appetite, laxative, and hypercholesterolemia. Magnesium, sodium, and phosphorus are present in high amount in fenugreek seeds (Shaikh et al., 2013).

Plant growth regulators are known as phytohormones and are natural product when

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synthesized chemically and naturally in the plants (Javid et al., 2011). Plant growth regulators (PGRs) control cell differentiation, cell division, senescence of plants, and root and shoot growth (Al-Whaibi et al., 2012). Among PGRs, IAA plays a vital role in the growth and development of plant (Prusty et al., 2004). IAA also plays significant role in cell elongation, lessening wall pressure, increasing osmotic substance of cells, increasing permeability of water in cells, regulating protein, and cell wall synthesis. Indole-3-acetic acid also has a role in plant growth regulation (Wang et al., 2001). Indole acetic acid is called the key auxin which helps in controlling different physiological processes of plants for example cell division, cell enlargement, and differentiation of tissues that show various responses to light and gravity (Spaepen et al., 2007). Since the time of discovery of gibberellic acid, it has been considered as an important substance to regulate or control the height of the plant. When GA_3 is applied to the plants, it stimulates the elongation and growth of the shoot (Tanimoto, 2012). Gibberellic acid starts germination, breaks seed dormancy, promotes hypocotyl growth and cell division in cambial zone, intermodal length, and improves leaf size (Keykha et al., 2014). Gibberellic acid also promotes the growth parameters of plants such as number of leaves and leaf area, increases the production, and plays a part in improving the production of seed (Emongor and Ndambole, 2011).

Vitamins are called basic growth regulators because they are used in cellular metabolism of the plants. In numerous enzymatic reactions they play part as a cofactor (Amparo et al., 2010). Various vitamins have numerous biochemical and metabolic functions (Miret and Munne-Bosch, 2014). Different vitamins such as riboflavin indicate significant effects on physiological attributes, free amino acid, carbohydrates, and protein contents (Azooz et al., 2013). Riboflavin (vitamin B2) is essential for the metabolism of nutrient and protection of antioxidants (Ashoori and Saedisomeolia, 2014). In different physiological reactions of the plant it works as a coenzyme and is involved in anti-oxidation and peroxidation (Kennedy, 2016).

In Pakistan, PGRs are not used in vegetables for crop improvements. This is why less information is available on the enhancement of the growth, yield, and quality of fenugreek by IAA and GA_3. For this purpose, this study was aimed to find out the efficacy of IAA, GA_3, and riboflavin in fenugreek for better productivity of this vegetable.

Materials and Methods

A pot experiment was performed at University of Gujrat, Pakistan during 2018-2019. Fenugreek seeds variety Kasuri Methi were obtained from Pakistan Agriculture Research Council (PARC) Islamabad, Pakistan. Seeds were sown in 10 kg sandy clay soil in pots. The sandy loam soil was prepared by taking of about 60% sand, 10% clay, and 30% silt particles. The soil was mixed with organic manure with 1:1 ratio; no synthetic fertilizer was used and no disease was observed during the experiment. Treatments of Indole acetic acid (IAA), gibberellic acid (GA_3), and riboflavin were applied as foliar spray in one dose on fenugreek after 14 days of sowing. The treatments were:

T0=control (water)
T1= IAA (50 mmol L^{-1})
T2= GA_3 (50 mmol L^{-1})
T3= Riboflavin (50 mmol L^{-1})

The experiment was arranged in a completely randomized design (CRD) with six replicates. Four plants were taken for data collection at vegetative stage (60 days after sowing), maturity (120 days after sowing), and fruiting stage for biomass production (root length and shoot length, root dry biomass and shoot dry weights, numbers of branches and leaves, leaf area), photosynthetic pigments (chlorophyll “a” and “b” and carotenoids contents), and antioxidants pursuits, i.e. superoxide dismutase, peroxidase dismutase, and catalase. The number of pods and seed weight were determined at maturity.

Leaf area was calculated with a portable Handheld Leaf Area Meter (CI-203 Laser scanner). Chlorophyll ‘a’, ‘b’, and total chlorophyll, and carotenoid contents were estimated using the Arnon (1949) method. Total carbohydrates were estimated with the Anthrone method. Soluble protein was estimated following Bradford (1976).

Table 1
Means squares (MS) from the Analysis of Variance (ANOVA) for root development of fenugreek under IAA, GA_3, and riboflavin

Source of Variance	df	Root length		Root fresh weight		Root dry weight	
		Seedling stage	Vegetative stage	Seedling stage	Vegetative stage	Seedling stage	Vegetative stage
IAA	1	7.801***	98.350***	0.077**	0.051***	1.125ns	0.001ns
GA_3	1	7.22***	4.727**	0.021 **	0.001**	1.125ns	5.281ns
Riboflavin	1	4.651**	8.302 ns	2.812 **	0.016*	25.004ns	2.812ns
Interaction IAA× GA_3	1	0.101 ns	33.415**	0.018 ns	0.002ns	1.253ns	3.781ns
IAA× Riboflavin	1	0.72 ns	42.090**	0.077**	0.001ns	25.004ns	0.002*
GA_3×Riboflavin	1	0.101 ns	49.252***	0.018ns	0.002ns	53.005ns	3.781ns
IAA× GA_3×Riboflavin	1	7.22 ***	11.640 ns	0.021 ns	0.001ns	1.125ns	5.281ns
Error	16	0.438	3.030	0.008	0.002	1.687	4.489
Total	23						

ns: non-significant; *, **, and ***: significant at $p \leq 0.05$, 0.01, and 0.001, respectively

Estimation of CAT, POD, and SOD activities was determined by using Chance and Maehly (1955) procedure. Fresh leaves (5 g) were crushed in 5 ml of 50 mM cooled Phosphate buffer (pH 7.8), which was then put in an ice bath for extraction of enzymes activities. Afterwards, the crushed samples were centrifuged at 15,000 RPM for 20 minutes at 4 ℃ and supernatants were used for checking different enzymes activities. CAT solution (3 ml) was made to determine the catalase enzyme by adding 50 mM phosphate buffer (pH 7.0), 0.1 ml enzyme extract and 5.9 Mm H_2O_2. The reaction was started by addition the extract of enzyme in solution. Absorbance changes of CAT solution were determined at 240 nm. Changes in absorbance of 0.01 units per min. were utilized to characterize activity of CAT. Peroxidase dismutase solution (3ml) consisted of 50 mM phosphate buffer (pH 5.0), 40 Mm H_2O_2, 20 mM guaiacol, and 0.1 ml enzymes extract. At 470nm, absorbance changes of the solution were determined. A solitary POD action was characterized utilizing an assimilation move of 0.02 units every moment. Activity of superoxide dismutase was determined by calculating inhibition in photo-reduction of nitroblue tetrazolium by SOD enzyme. The SOD reaction was carried out by exposing the reaction mixture (containing 0.1 mM EDTA, 50 mM of phosphate buffer (pH 7.6), 10 µM riboflavin, 50 mM sodium carbonate, 0.1 mM EDTA, 50 µM NBT, 12 mM L-methionine, and 100 µL of crude extract in a 3.0 mL) to light for 15 minutes for incubation at standard/room temperature; then, absorbance was noted at 560 nm by a spectrophotometer. One unit of superoxide dismutase enzyme causes 50 percent inhibition of photochemical reduction of nitroblue tetrazolium. Root and shoot dry samples about 1g was digested with sulphuric acid and H_2O_2 by using the method of Wolf (1982) for determination of ions of potassium (K^+), sodium (Na^+), and calcium (Ca^{2+}). Concentrations of potassium, sodium, and calcium were measured by using a flame photometer (Model PFP7, Jenway Staffordshire, UK).

Minitab was used for analysis of data (Version: 19.2.0, Coventry, UK) and mean values were compared using Tukey's Test.

Results

Root development

Results indicated that IAA, gibberellic acid, and riboflavin had highly notable effect on root length and root fresh weight but it was non-significant for root dry weight in fenugreek (Table 1). The interaction of IAA× riboflavin, IAA × GA_3, gibberellic acid × riboflavin and IAA× GA_3 × riboflavin in most of the cases was non-significant except the interaction of GA_3, IAA, and riboflavin, which was remarkable at vegetative stage (Table 1). Higher root length was observed by the treatment of 50 mmol L^{-1} GA_3 at both growth stages. Higher root fresh biomass was noted by application of 50 mmol L^{-1} riboflavin at seedling stage and higher

root fresh biomass was noted with 50 mmol L^{-1} GA_3 at vegetative stage (Fig. I).

increased the number of leaves and 50 mmol L^{-1} GA_3 enhanced the growth of branches (Fig. III. C).

Table 2
Means squares (MS) from the Analysis of Variance (ANOVA) for shoot development of Fenugreek under IAA, GA_3 and riboflavin

Source of Variance	df	Shoot length		Shoot dry weight		Shoot fresh weight	
		Seedling stage	Vegetative stage	Seedling stage	Vegetative stage	Seedling stage	Vegetative stage
IAA	1	14.178***	283.815**	0.005**	1.711**	0.195**	13.521***
GA_3	1	22.278***	263.925**	0.001**	2.101***	0.061 ***	17.111***
Riboflavin	1	6.038**	13.132 ns	2E-04ns	0.101ns	0.007ns	5.121***
IAA× GA_3	1	0.428 ns	104.762 ns	85.584ns	0.245ns	0.031ns	0.501ns
IAA × Riboflavin	1	0.508***	906.315***	0.002ns	2.645***	0.112ns	6.661***
GA_3 × Riboflavin	1	0.428 ns	104.762 ns	85.584ns	0.245ns	0.031ns	0.501ns
IAA × GA_3 × Riboflavin	1	22.278***	263.925*	0.001ns	2.101***	0.061ns	17.111***
Error	16	0.535	39.734	9.501	0.132	0.036	0.136
Total	23						

ns: non-significant; *, **, and ***: significant at p≤ 0.05, 0.01, and 0.001, respectively

Shoot development

Indole acetic acid and gibberellic acid effects were highly significant on shoot length and shoot fresh and dry weights in fenugreek but riboflavin had highly significant results for shoot length at seedling stage and fresh weight of shoot at vegetative stage (Table 2). Interaction of GA_3 with riboflavin was also significant but the interaction of GA_3 with IAA and riboflavin was non-significant (Table 2). Higher shoot length and shoot fresh weight was observed with the treatment of 50 mmol L^{-1} GA_3 at both growth stages (Fig. II).

Foliage growth

Foliage growth of fenugreek, i.e. leaf area, number of branches and leaves, significantly improved with the treatment of GA_3, indole acetic acid, and riboflavin (Table 3). It was noted that the interactions of GA_3, IAA, and riboflavin with each other was non-significant in most cases. It was also noted that higher leaf area/plant was observed at growth (seedling stage) by 50 mmol L^{-1} GA_3 and at vegetative stage under the treatment of 50 mmol L^{-1} IAA (Fig. III. A-B). Maximum count of leaves and branches was noted by 50 mmol L^{-1} IAA at seedling stage. At vegetative stage, 50 mmol L^{-1} riboflavin

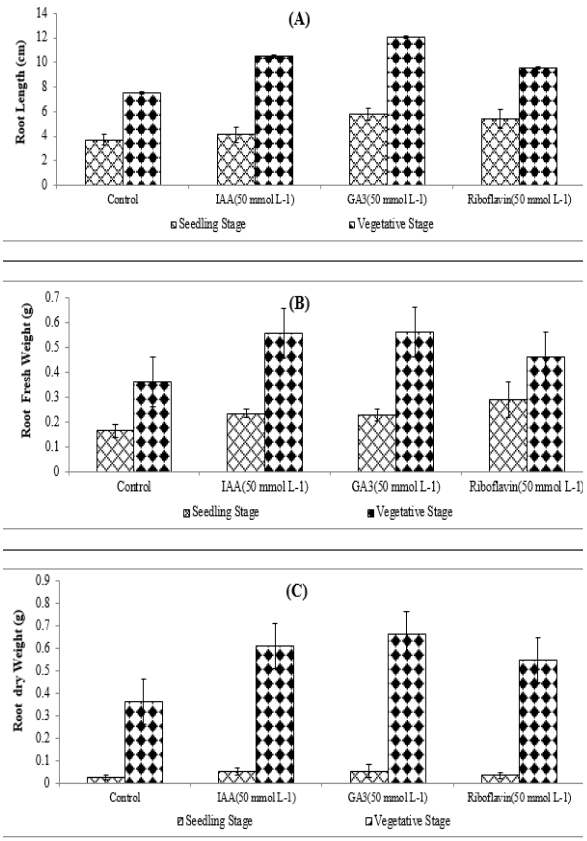

Fig. I. Development of roots in fenugreek under IAA, GA_3, and riboflavin at seedling and vegetative stages

Photosynthetic pigments

Table 3
Means squares (MS) from the Analysis of Variance (ANOVA) for foliage growth of fenugreek under IAA, GA_3, and riboflavin

Source of Variance	df	Leaf area/plant		Number of leaves/plant		Number of branches/plant	
		Seedling stage	Vegetative stage	Seedling stage	Vegetative stage	Seedling stage	Vegetative stage
IAA	1	0.071**	2.486**	10.125**	750.781***	9.031*	12.501*
GA_3	1	0.264**	2.726***	32.001**	87.781***	1.531***	24.512**
Riboflavin	1	0.062**	0.064**	28.125*	148.781ns	0.031ns	2.531*
IAA× GA_3	1	7.812*	0.324ns	18.101ns	57.781ns	2.531ns	3.125ns
IAA × Riboflavin	1	0.075ns	6.125***	15.125ns	140.281ns	9.031*	45.120***
GA_3 × Riboflavin	1	7.812ns	0.324ns	18.101ns	57.781ns	2.531ns	3.125ns
IAA × GA_3 × Riboflavin	1	0.264ns	2.726ns	32.001ns	87.781ns	1.531ns	24.512**
Error	16	0.081	0.379	15.145	43.968	1.156	2.583
Total	23						

ns: non-significant; *,**, and ***: significant at p≤ 0.05, 0.01, and 0.001, respectively

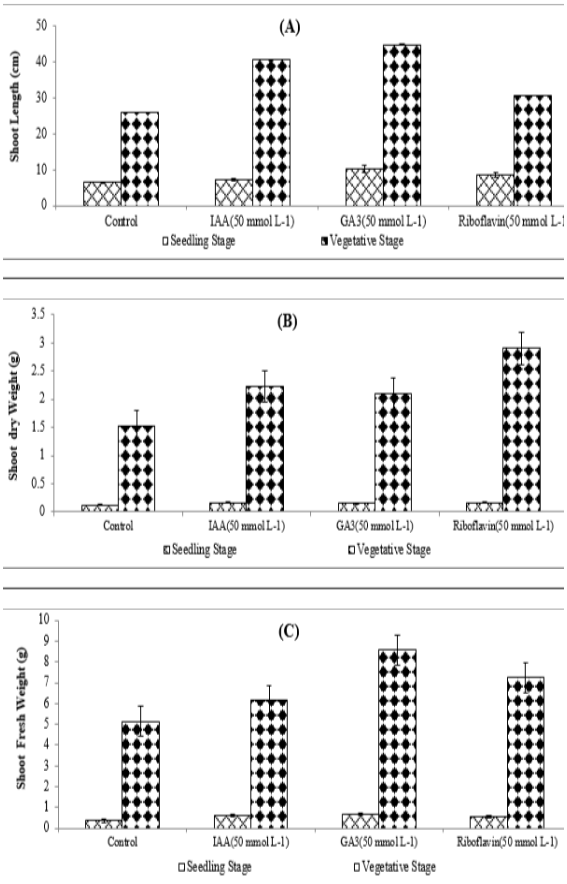

Fig. II. Development of shoots in fenugreek under IAA, GA_3, and riboflavin at seedling and vegetative stages

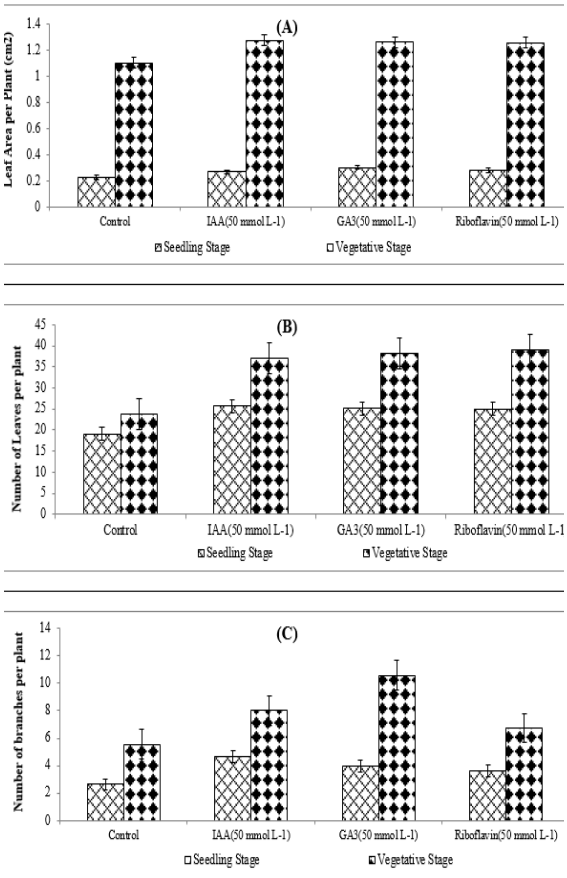

Fig. III. Foliage growth of fenugreek under IAA, GA_3, and riboflavin at seedling and vegetative stages

It was noted that the PGRs (IAA and GA_3) improved amount of photosynthetic pigments of fenugreek, i.e. chl. 'a', 'b', and carotenoids while the effects of riboflavin were non-significant on photosynthetic pigments (Table 4). In most of the cases, interactions between indole-3-acetic acid, gibberellic acid, and riboflavin were insignificant. At seedling stage, higher chl. a content observed with present at 50 mm mol L^{-1} of GA_3 . However,

at vegetative stage, higher contents of chl. a and carotenoid were calculated with 50 mmol L^{-1} GA_3. On the other hand, IAA increased chl. b contents (Fig. IV. A-B). Higher carotenoid and chl. b contents were noted with the applications of 50 mmol L^{-1} riboflavin (Fig. IV. C).

Ion contents

It was observed that IAA, GA_3, and riboflavin had highly significant increasing effects on ion accumulations of potassium (K^+) and calcium (Ca^{2+}) in roots and shoots of fenugreek except for sodium (Na^+) contents (Table 5). There were also significant interactions among IAA, GA_3, and riboflavin except in few cases. IAA applied in 50 mmol L^{-1} showed the highest accumulation of potassium (K^+) and calcium (Ca^{2+}) ions in roots and shoots of fenugreek at both growth stages except for Na^+ (Figs. V and VI). Accretion of Ca^{2+}, Na^+ and K^+ was higher in shoots as compared with roots in fenugreek (Figs. V and VI).

Antioxidant activities

Results for antioxidant activities showed that peroxidase dismutase (POD) and catalases (CAT) contents significantly increased with the treatment level of gibberellic acid and indole

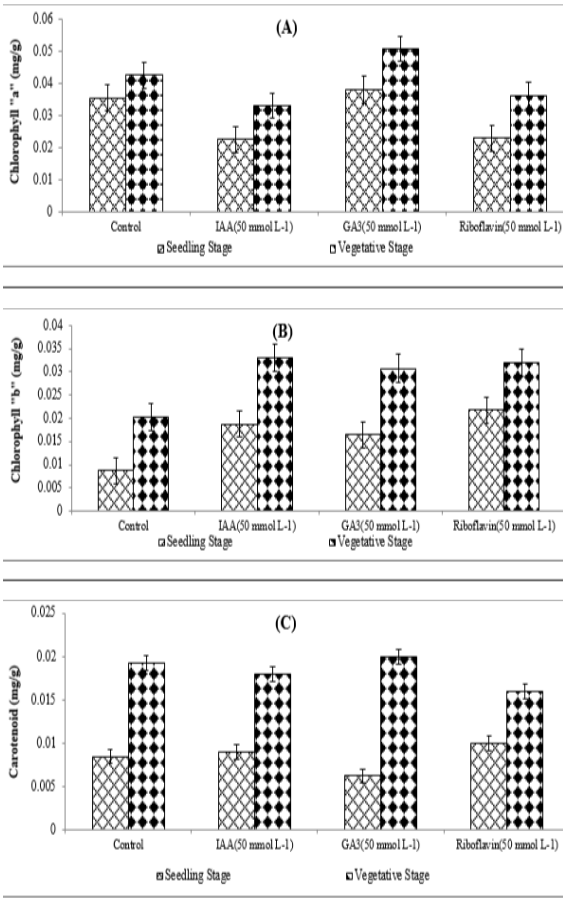

Fig. IV. Contents of photosynthetic pigments in fenugreek under IAA, GA_3, and riboflavin at seedling and vegetative stages

Table 4
Means squares (MS) from the Analysis of Variance (ANOVA) for photosynthetic pigments of fenugreek under IAA, GA_3, and riboflavin

Source of Variance	df	Chlorophyll-a		Chlorophyll-b		Carotenoid contents	
		Seedling stage	Vegetative stage	Seedling stage	Vegetative stage	Seedling stage	Vegetative stage
IAA	1	2.101**	0.023*	6.480**	1.757**	8.006*	3.003*
GA_3	1	3.781***	0.014**	1.512**	5.281*	3.125*	3.781*
Riboflavin	1	1.125ns	0.001ns	1.280**	2.257***	1.253ns	1.378ns
IAA × GA_3	1	6.050**	0.003ns	5.005ns	7.812ns	1.250ns	7.812ns
IAA × Riboflavin	1	8.041***	0.019ns	1.250ns	3.315***	3.231*	2.278ns
GA_3 × Riboflavin	1	6.055**	0.003ns	5.005ns	7.812ns	1.250ns	7.812ns
IAA × GA_3 × Riboflavin	1	3.781***	0.014ns	1.512ns	5.281ns	3.125ns	3.781ns
Error	16	6.791	0.047	1.402	1.207	6.770	6.760
Total	23						

ns: non-significant; *, **, and ***: significant at $p \leq 0.05$, 0.01, and 0.001, respectively

Table 5
Means squares (MS) from the Analysis of Variance (ANOVA) for ionic contents of fenugreek under IAA, GA_3, and riboflavin

Source of Variance	df	Sodium (Na^+) ions in root		Sodium (Na^+) ions in shoot		Potassium (K^+) ions in root		Potassium (K^+) ions in shoot		Calcium (Ca^{2+}) ions in root		Calcium (Ca^{2+}) ions in shoot	
		Seedling stage	Vegetative stage	Seedling stage	Vegetative stage	Seedling stage	Vegetative stage	Seedling stage	Vegetative stage	Seedling stage	Vegetative stage	Seedling stage	Vegetativ stage
IAA	1	1.579ns	3.465ns	9.735ns	0.017ns	6.133***	24.745***	0.023ns	13.158**	11.257***	15.521***	0.005***	0.087***
GA_3\	1	0.367ns	0.106ns	0.243ns	0.425ns	0.711***	4.651***	0.014 **	1.711*	1.960***	2.856***	0.001**	7.031*
Riboflavin	1	0.596*	0.487ns	1.106*	2.559*	0.717***	12.325***	0.001 ns	1.281 ns	3.962***	5.746***	0.002*	0.039**
IAA× GA_3	1	0.058*	0.338ns	1.069***	0.538**	0.664***	0.536***	0.003 *	1.557 **	0.465***	0.594**	0.003**	0.002*
IAA× Riboflavin	1	0.005ns	0.044ns	0.226ns	0.826*	0.658***	0.387***	0.019 *	1.030 ns	0.008***	0.004ns	0.005***	0.031***
GA_3 × Riboflavin	1	0.058*	0.338ns	1.069***	0.538**	0.664***	0.536***	0.003 ns	1.557 ns	0.465***	0.594**	0.003**	0.002*
IAA × GA_3× Riboflavin	1	0.367***	0.106*	0.243ns	0.425*	0.711***	4.651***	0.014 ns	1.711 ns	1.960***	2.856***	0.001ns	7.031ns
Error	16	0.008	0.021	0.064	0.040	0.024	3.187	0.047	1.520	4.020	0.051	3.625	4.718
Total	23												

ns: non-significant; *, **, and ***: significant at p≤ 0.05, 0.01, and 0.001, respectively

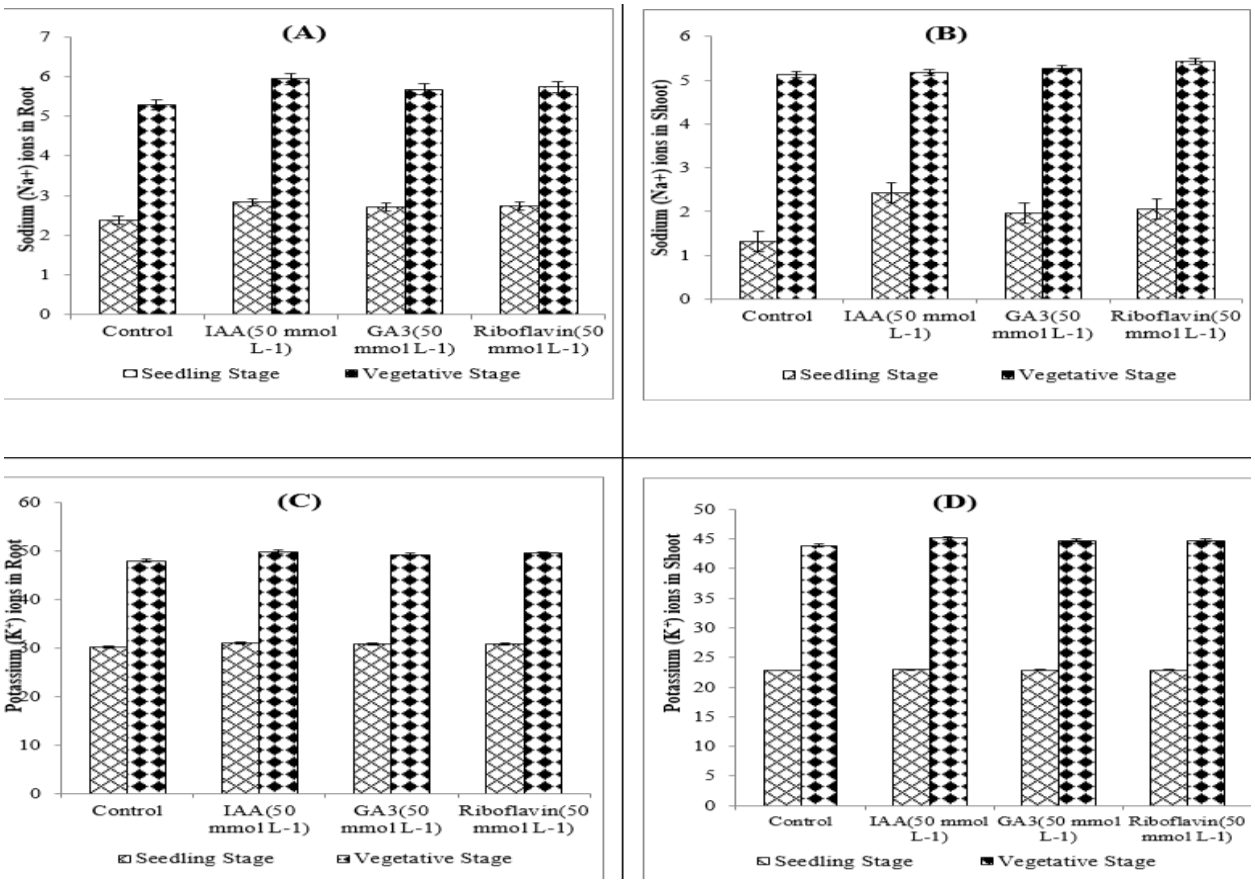

Fig. V. Ionic concentrations in fenugreek under IAA, GA_3, and riboflavin at seedling and vegetative stages

acetic acid. Riboflavin had significantly increasing results for superoxide dismutase (SOD), POD, and CAT at vegetative stage (Table 6). POD and catalases activities increased under the treatment with 50 mmol L-1 GA3 as compared with other treatments at seedling stage. SOD activity increased at vegetative stage under 50 mmol L-1 GA3 while POD and CAT activities improved under 50 mmol L-1 IAA at vegetative stage (Fig. VII).

Table 6
Means squares (MS) from the Analysis of Variance (ANOVA) for antioxidant activities and yield of fenugreek under IAA, GA_3, and riboflavin

Source of Variance	df	SOD at seedling stage	SOD at vegetative stage	POD at seedling stage	POD at Vegetative stage	CAT at seedling stage	CAT at Vegetative stage	Number of Pods/plant	Seed yield/plant
IAA	1	0.005ns	1.402ns	0.016 *	0.475*	0.017*	0.088*	0.0281*	0.781*
GA_3	1	5.821ns	0.036ns	9.112 **	0.060*	0.005*	0.022*	9.031*	3.781**
Riboflavin	1	0.002 ns	0.537*	5.951 ns	0.045*	0.002 ns	0.019*	11.281ns	0.031ns
IAA× GA_3	1	9.802 ns	0.050 ns	0.008 ns	0.056*	7.801 ns	0.006*	3.781*	0.281*
IAA × Riboflavin	1	9.921 ns	0.100 ns	0.011 ns	0.073*	0.003 ns	0.007*	69.031ns	7.031ns
GA_3 × Riboflavin	1	9.802 ns	0.050 ns	0.001 ns	0.056*	7.801 ns	0.006**	3.781ns	0.281ns
IAA× GA_3 × Riboflavin	1	5.281 ns	0.036 ns	0.012 ns	0.060*	0.005 ns	0.002**	9.031ns	3.781ns
Error	16	0.003	0.627	0.013	2.533	0.002	3.756	4.177	1.781ns
Total	23								

ns: non-significant; *, **, and ***: significant at p≤ 0.05, 0.01, and 0.001, respectively

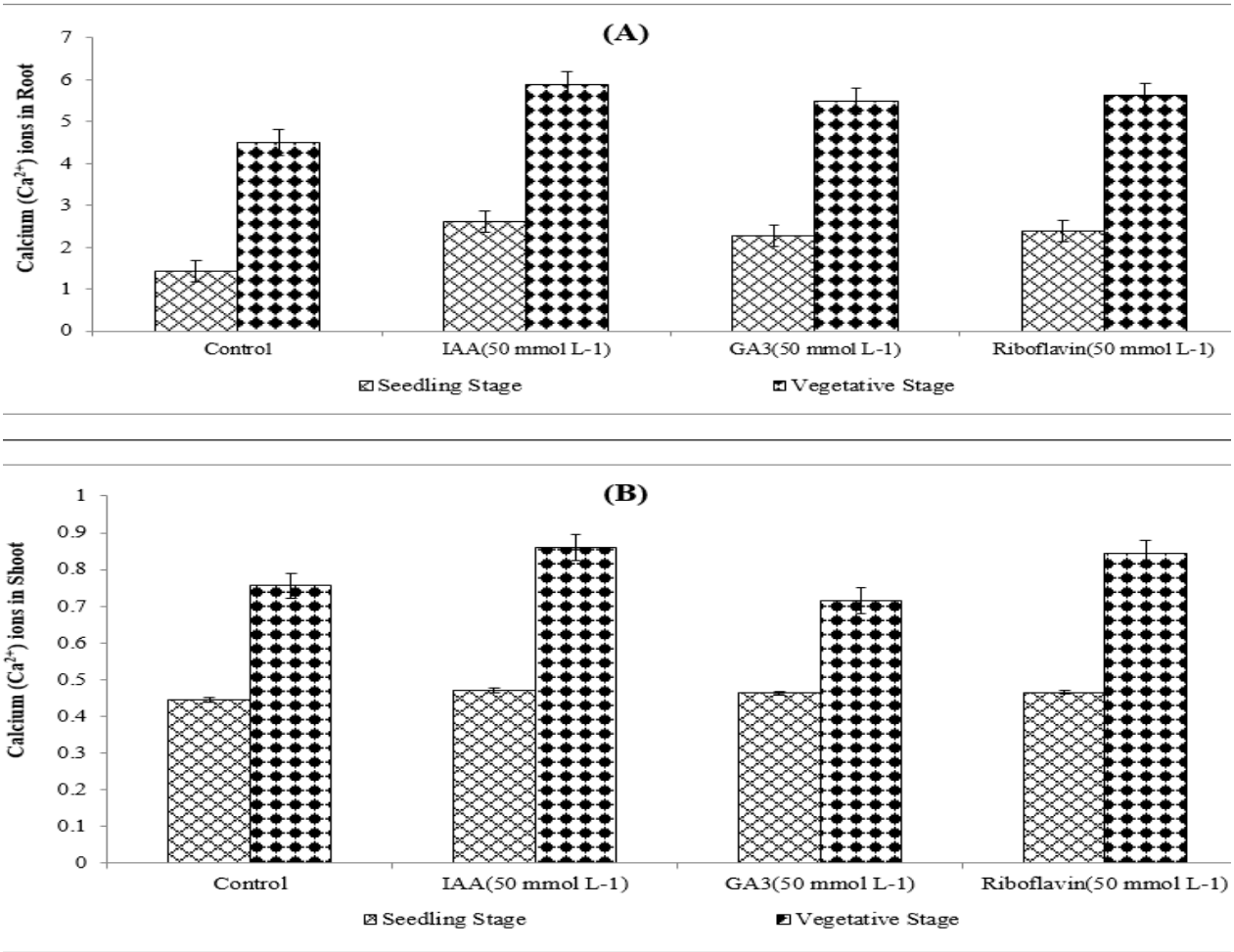

Fig. VI. Ionic concentrations in fenugreek under IAA, GA_3, and riboflavin at seedling and vegetative stages

Yield attributes

ANOVA revealed that the application of GA_3 and IAA had significant results for the number of pods and seed yield per plant of fenugreek while there was no significant effect by the application of riboflavin (Table 6). In most of the cases, interactions were also non-significant except

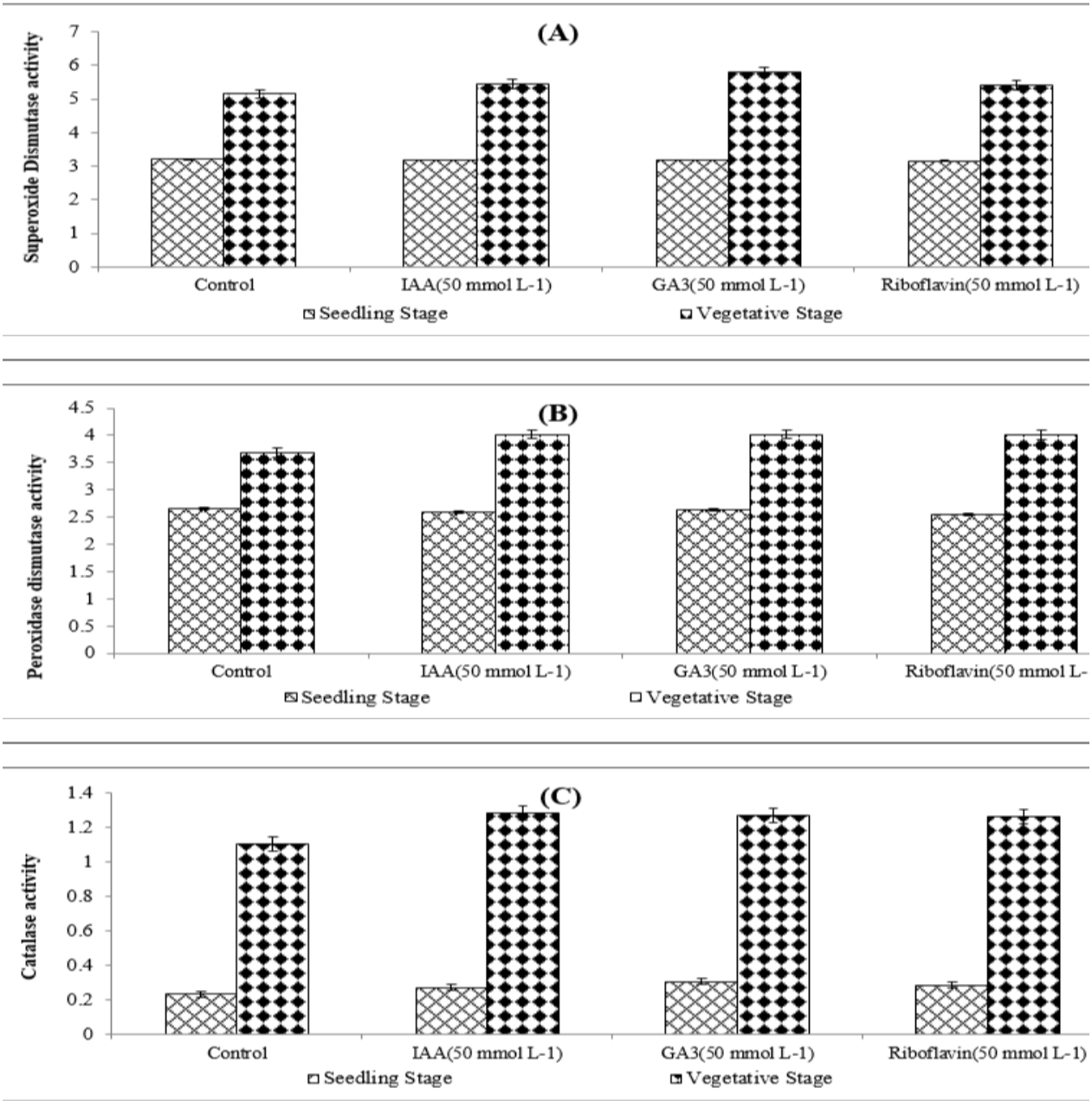

Fig. VII. Antioxidant activities of fenugreek in response to IAA, GA_3, and riboflavin at seedling and vegetative stages

under IAA × GA_3. Number of pods per plant improved by the application of 50 mmol L^{-1} IAA while higher seed yield was obtained by the application of 50 mmol L^{-1} GA_3 (Fig VIII).

Discussion

As results indicated, IAA and GA_3 enhanced the biomass production including root and shoot development and foliage growth. Because auxin regulated numerous physiological processes, e.g. tissue differentiation, cell division, and cell enlargement that show different response to light and gravity (Spaepen et al., 2007). Correspondingly, Sevik and Guney (2013) described that GA_3 and indole acetic acid applications improved the length of roots and shoots in *Melissa officinalis*. In many studies, it was noted that IAA and GA_3 are effective in increasing the plant productivity (root and shoot development) in various plants as in Arabidopsis (Zúñiga et al., 2013) and maize (Pilet and Saugy, 1987). In the present study, riboflavin also enhanced the root and shoot growth only for few

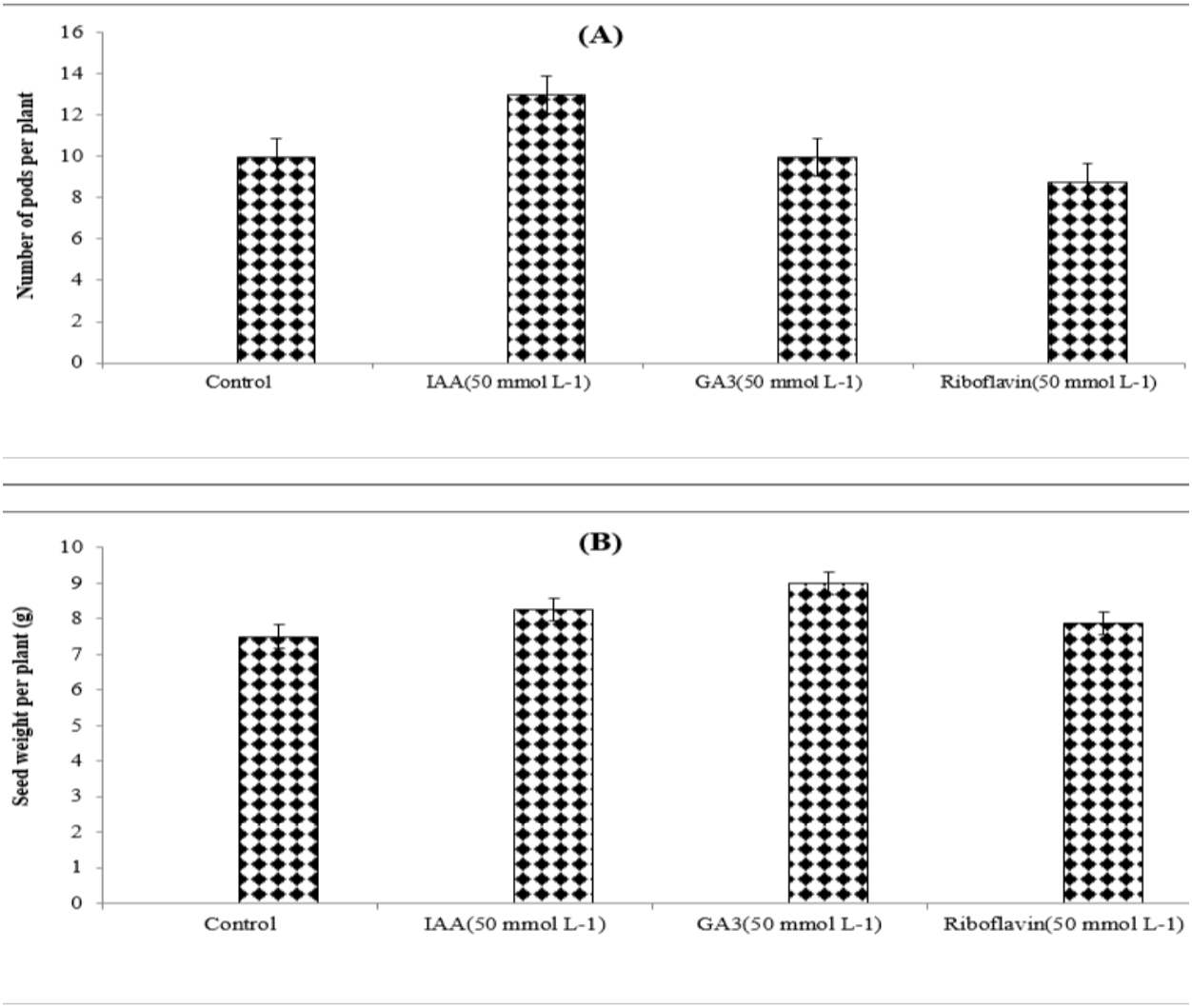

Fig. VIII. Yield attributes of fenugreek in response to IAA, GA_3, and riboflavin at seedling and vegetative stages

cases. Azooz (2009) and El-lethy et al. (2011) stated that riboflavin enhanced root and shoot fresh biomass by foliar application in *Pelargonium graveolens* and *Hibiscus sabdariffa*, respectively. It was also reported that IAA improved the leaf area in *Triticum aestivum* (El-metwally et al., 2015). On the other hand, Khan et al. (2006) stated that leaf area was enhanced with foliar treatment of gibberellic acid in tomato plants.

In fenugreek, it was noted that auxins increased photosynthetic pigments. These results are in accordance with many studies. For example, Chl. a content was enhanced by IAA treatment in date palm plant (Rasmia and Darwesh, 2014) and barley (Eleiwa et al., 2013). Similarly, Afroz et al. (2005) noted that GA_3 increased chlorophyll and carotenoid contents in mustard. Some researchers stated the riboflavin enhanced the contents of chlorophyll with foliar spray in *Pelargonium graveolens* (El-lethy et al., 2011).

Results showed that IAA and GA_3 exert positive effects in increasing the concentration of ion contents in roots and shoots of fenugreek. Concentration of ions such as Ca, K, Cl, and Na enhanced under the application of indole acetic acid in maize and date palm (Kaya et al., 2010: Rasmia and Darwesh, 2014). Change in antioxidant activities resulted in strong defense mechanism to cope with any stress conditions.

Ali et al. (2014) reported that enzyme activities such as SOD, POD, and CAT was improved by IAA in *Rosa indica.* Similarly, Mohammadi and Khavari-Nejad (2013) noted that IAA also enhanced the

antioxidant enzyme activities such as catalase and peroxidase in maize and *Glycine max* (Kaya et al., 2010). In the present study, riboflavin also had significant results for antioxidant activities at seedling stage of fenugreek. Similarly, riboflavin was reported to exert regulatory effects on antioxidant activities in *Pelargonium graveolens* (El-lethy et al., 2011).

Conclusion

It is concluded that gibberellic acid (GA_3) and indole-3-acetic acid (IAA) can be used to enhance the foliage biomass production and yield and GA_3 is superior over IAA in fenugreek. These outcomes can be useful for economic benefits with high production of this leafy vegetable.

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