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Evaluation of effect of selected Plant Growth Regulators on morphological traits and seed yield of *Fagopyrum esculentum* Moench of Himalayan Region

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ABSTRACT

Need for food and medicines is supposed to continue due to ever-growing world population. Investing in agriculture is the solution to reducing poverty and hunger in developing countries. Keeping that in mind present investigation was made to evaluate effect of plant growth regulators (PGRs) on morphology of Fagopyrum esculentum. Results revealed that IAA +BAP (100 mg l^{-1}) concentration was effective in enhancing number of leaves, stem diameter, number of inflorescence as well as seed yield. Number of leaves reduced in 100 mg l^{-1} of GAs, ABA, ABA+GAs and 25mg l^{-1} ABA treatments. Reduction in stem diameter (GAs 25 and 100 mg l^{-1}), number of inflorescence (ABA 25 mg l^{-1}) and seed yield (IAA+ABA 50 mg l^{-1}) was recorded. BAP alone as well as with ABA (ABA+BAP) and IAA (IAA+BAP) at 100 mg l^{-1} concentration showed significant increase in number of branches however lower concentration of ABA+GAs and BAP+GAs reduced it. Leaf surface area was enhanced with 100 mg l^{-1} concentration of BAP and BAP+GAs and declined with 25 mg l^{-1} concentration of ABA, GAs, ABA+GAs and higher concentration (50, 100 mg l^{-1}) of IAA+ABA.

***Keywords**: Fagopyrum esculentum,* plant growth regulators, Sustainable intensification, nutrition, antioxidant-rich, prebiotic, seed yield

Sustainable intensification is required to produce more food from the same area of land while reducing the environmental impacts (Royal Society of London, 2009). Many of the plant species that are cultivated for food are neglected and underutilized across the world, while they play a crucial role in the food security, nutrition and income generation of the rural poor (Magbagbeola *et al.,* 2010). One of such plant *Fagopyrum esculentum* Moench, commonly called buckwheat, is a potential functional food source (Ahmed *et al.,* 2014). It is an underutilized pseudo-cereal whose grains are used like cereals (Campbell, 1997). Entire buckwheat plant contains 2–5 times more phenolic compounds than oats and barley (Zdunczyk *et al.,* 2006). It has dark-hulled, triangular, starch-filled seeds, round and hollow lateral branches, heart shaped or somewhat arrow shaped leaves. It has compound raceme inflorescence that produces lateral 1-30 uniparous cymes (Quinet *et al.,* 2004). Flower of buckwheat is dimorphic i.e. the pin and the thrum type. The pin type has flowers with long pistils and short stamens whereas thrum type flower have short pistils and long stamens (Halbrecq et al., 2005). The seed is an achene, about 5 mm long with black soft hull, light green to white kernel (Krkoskova and Mrazova, 2005). It is also used in the form of flour-bread, rice, soup, cakes, pasta, crackers, cookies, pancakes and tortillas. Buckwheat grains and its by-products are a rich source of substance with valuable functional properties (Dziedzic *et al*., 2018).

Buckwheat grain is characterised by a high content of starch, a low content of á-gliadin and a high content of dietary fibre (Dziedzic *et al.,* 2012). Antioxidant-rich diets have been associated with a lower incidence of cancers, cardiovascular disease and age-related deteriorating processes (Kaliora and Dedoussis, 2007). Buckwheat is mainly used for the treatment of celiac disease as it is gluten free and eaten in place of wheat (Alvarez-Jubete *et al.,* 2010). Buckwheat tea is a popular health product in Asian and European countries (Qin *et al.,* 2013). Buckwheat could possible behaves as a prebiotic (Prestamo *et al.,* 2003). It has been reported that the ethyl acetate and ethanol extracts of the stem, seed and aerial parts of buckwheat show neuroprotective effect (Gulpinar *et al.,* 2012). The use of PGRs has gained importance for enhancing growth and efficient production. Phytohormones operate at the genetic level (Taiz and Zeiger, 2010). Foliar spray of plant growth regulators (PGRs) is significant in increasing morphological traits of plants. Number of studies is reported where PGRs enhanced stem diameter (Omar and Khudhur, 2015; Parvin and Haque, 2017), leaf area (Chetna *et al*., 2013; Ahmadi Lahijani *et al*., 2018); number of leaves (Taha *et al*., 2016; Garg et al., 2020), number of inflorescence (Nambiar *et al*., 2012; Sumathi

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et al., 2017), number of branches (Sumathi *et al*., 2017; Garg et al., 2020) and seed yield (Sumathi *et al*., 2017; Khan *et al*., 2020).

MATERIALS AND METHODS

The seeds of *Fagopyrum esculentum* Moench were obtained from Himachal Pradesh Agricultural University, Research Station, Sangla, Kinnaur (HP). Seeds of *Fagopyrum esculentum* selected for uniformity, damaged and insect infected seeds were discarded and the hollow ones were rejected by floating method in distilled water. Surface sterilization of seeds was done with 0.1% $HgCl_2$ prior to sowing, after which the seeds were rinsed three times with distilled water. Seeds were sown in the nursery beds, in the Herbal Garden of Shoolini University, Solan (Latitude 30°51'N, longitude 77°07'E and altitude 1195 m), where the average annual rainfall was 1315.6 mm. The average maximum and minimum temperatures were 32°C and 2°C, respectively. Nursery beds were watered regularly. When the first leaf appeared the seedlings were transferred to pots (20 cm diameter).The pots were filled with 3 kg uniform soil mixture containing soil: sand: farm yard manure (FYM) in 1:1:1 ratio. Three seedlings per pot in replicates of three were used for each treatment. No inorganic fertilizer and systemic pesticide were used during the experiment. Plant growth regulators spray was done after one week of transplanting the plants to pots. Four major hormones: indole acetic acid (IAA), benzylaminopurine (BAP), abscisic acid (ABA) and gibberellic acids (GAs), were used solely as well as in combinations i.e. IAA+BAP, IAA+ABA, IAA+GAs, ABA+BAP, BAP+GAs and ABA+GAs in concentration of 25, 50 and 100 mg l^{-1} through foliar spray. Sampling was done three times during life cycle of the plant i.e. after 30, 60 and 90 days. The following morphological and growth parameters were recorded at 30, 60 and 90 days of plant growth:

(i) Stem diameter (mm^2) (ii) Number of leaves per plant (iii) Leaf area (cm^2) (iv) Number of branches per plant (v) Number of inflorescence per plant (vi) Seed yield(g, per plant)

The data was analyzed statistically using Graph Pad Prism® 5.2. Mean values were calculated from measurements of three replicates and the standard error of means were determined. One way and two-way analysis of variance (ANOVA) was applied to determine the significance of results between different treatments and Bonferroni's post tests were performed at the significance level of $P<0.05$.

RESULTS AND DISCUSSION

Stem diameter

There was a regular increase in stem diameter with the advancement of plant age under all the treatments. Stem diameter increased at 30 days (2.90 mm, 31.8% increase from control), at 60 days (6 mm, 11.1% increase from control) and at 90 days (6.80 mm, 11.5% increase from control) in BAP 100 mg l^{-1} treated plants. Minimum stem diameter at 30 days was shown by GAs 25 mg l^{-1} (2 mm, 9.1% decrease from control). At 60 days (4.90 mm, 9.3% decrease from control) and 90 days (5.00 mm, 18% decrease from control) GAs 100 mg l^{-1} treated plants showed minimum stem diameter (Table 1). Maximum stem diameter at 30 days (3.10 mm, 40.9% increase from control), at 60 days (6.10 mm, 13% increase from control) and at 90 days (6.90 mm, 13.1% increase from control) was noted in IAA+BAP 100 mg l^{-1} treated plants. Minimum stem diameter (2.10 mm, 4.5% decrease from control) at 30 days was recorded in IAA + GAs 25 mg l^{-1}, IAA+ABA 25 mg l^{-1} and ABA+GAs 25 mg l^{-1} treated plants. Whereas, at 60 days (5 mm, 7.4% decrease from control) and 90 days (5.20 mm, 14.8% decrease from control) minimum stem diameter was observed in ABA+GAs 25 mg l^{-1} treated plants (Table no. 1).

It is clear that in general the stem diameter due to treatments for the specific period of growth didn't vary much from control. Cell proliferation in the cambium thickens plants stem and cytokinins are essential regulators of cambium growth (Matsumoto-Kitano *et al*., 2008). Present investigation revealed that BAP alone and in combination with IAA (IAA+BAP) at 100 mg l^{-1} concentration promoted stem thickness (Diameter). Increase in stem diameter may be due to cell expansion caused by cytokinins. Our results are in agreement with Naeem *et al*. (2004) who observed that kinetin which is a type of cytokinin when used with IAA (IAA+kinetin) was most effective in enhancing stem diameter in *Lens culinaris.* IAA alone has also shown an expansion in the stem diameter of *Dalbergia sissoo* (Omar and Khudhur, 2015). The increase in diameter of newly developed stem results from an increase in apical meristem activity. Decrease in stem diameter was noted in IAA+ GAs, IAA+ABA, ABA+ GAs treatments at low concentration (25 mg l^{-1}). Similar trend was seen in the shoot of lentil with the application of GA_3 (Naeem *et al*., 2004). As GA_3 increase shoot length of plant it cause decrease in concentration of available sugars, which results narrowing of shoot diameter (Naeem *et al*., 2004).

Number of leaves

Highest number of leaves was seen in BAP 100 mg l^{-1} treatment at both 30 days (10, 25% increase from control) and 60 days (16.30, 35.8% increase from control). Whereas, at 90 days BAP 50 mg l^{-1} treatment had maximum number of leaves (25.80, 17.3% increase from control). Maximum decrease in number of leaves

Table 1: Growth in *Fagopyrum esculentum* at different growth stages under various phytohormone treatments; values are mean±S.E. n=3, analysed by Two-way ANOVA followed by Bonferroni's multiple comparison test. Values followed by the same letter are not statistically different (P<0.05) compared with control

PGRs	Treatment	Number of leaves plant^{-1}			Stem diameter (mm^2)		
		30 Days	60 Days	90 days	30 Days	60 Days	90 days
IAA	25 mg l^{-1}	7.70±0.17a	12.20±0.18a	24.00±0.20b	2.30±0.05	5.60±0.13	6.30±0.07
	50 mg l^{-1}	8.10±0.22a	13.60±0.25b	24.40±0.24b	2.50±0.04	5.80±0.06	6.60±0.03
	100 mg l^{-1}	8.50±0.26a	13.80±0.23b	24.80±0.21b	2.80±0.06	5.90±0.11	6.70±0.08
BAP	25 mg l^{-1}	8.10±0.17a	14.70±0.15b	25.00±0.12b	2.50±0.11	5.70±0.10	6.30±0.06
	50 mg l^{-1}	9.10±0.26b	15.30±0.22b	25.80±0.18b	2.80±0.06	5.90±0.12	6.70±0.07
	100 mg l^{-1}	10.00±0.27b	16.30±0.19b	25.30±0.25b	2.90±0.09	6.00±0.04	6.80±0.04
ABA	25 mg l^{-1}	7.00±0.24b	11.70±0.21a	21.40±0.32a	2.20±0.04	5.60±0.06	6.20±0.20
	50 mg l^{-1}	7.80±0.21a	11.10±0.33b	20.30±0.24b	2.30±0.15	5.70±0.03	6.60±0.14
	100 mg l^{-1}	7.10±0.32b	10.34±0.31b	20.50±0.23b	2.40±0.10	5.80±0.12	6.60±0.08
GAs	25 mg l^{-1}	7.60±0.26a	12.90±0.22b	22.20±0.18a	2.00±0.07	5.10±0.15	5.50±0.06
	50 mg l^{-1}	7.10±0.38b	13.22±0.26b	22.70±0.24a	2.40±0.05	5.20±0.06	5.40±0.02
	100 mg l^{-1}	7.00±0.17b	11.30±0.25a	21.50±0.28a	2.30±0.03	4.90±0.08	5.00±0.12
IAA+BAP	25 mg l^{-1}	9.10±0.12b	15.00±0.25b	25.50±0.14b	2.60±0.12	5.70±0.10	6.50±0.05
	50 mg l^{-1}	9.82±0.28b	15.70±0.11b	26.00±0.27b	2.90±0.01	6.00±0.03	6.70±0.06
	100 mg l^{-1}	10.10±0.36b	16.10±0.23b	27.30±0.38b	3.10±0.05	6.10±0.07	6.90±0.14
IAA+ABA	25 mg l^{-1}	7.40±0.16a	12.00±0.13a	20.00±0.24b	2.10±0.10	5.60±0.03	6.10±0.06
	50 mg l^{-1}	7.20±0.34b	12.40±0.17a	20.70±0.15b	2.30±0.02	5.70±0.11	6.40±0.03
	100 mg l^{-1}	7.80±0.08a	12.30±0.15a	19.00±0.21b	2.50±0.04	5.90±0.06	6.60±0.10
IAA+ GAs	25 mg l^{-1}	9.10±0.37b	14.90±0.31b	23.30±0.22b	2.10±0.06	5.20±0.04	5.70±0.11
	50 mg l^{-1}	9.50±0.23b	15.20±0.28b	25.30±0.42b	2.30±0.08	5.30±0.03	5.90±0.06
	100 mg l^{-1}	9.70±0.34b	15.70±0.23b	24.00±0.35b	2.40±0.04	5.30±0.12	6.10±0.02
ABA+BAP	25 mg l^{-1}	8.20±0.25a	13.00±0.32b	22.10±0.18a	2.20±0.07	5.50±0.05	6.20±0.10
	50 mg l^{-1}	8.50±0.21a	13.50±0.22b	22.30±0.20a	2.40±0.04	5.70±0.12	6.30±0.09
	100 mg l^{-1}	8.90±0.31b	13.80±0.29b	22.70±0.23a	2.60±0.06	5.80±0.02	6.60±0.04
BAP+ GAs	25 mg l^{-1}	9.00±0.07b	14.00±0.25b	22.70±0.13b	2.20±0.10	5.30±0.11	5.80±0.03
	50 mg l^{-1}	9.30±0.12b	14.40±0.21b	23.30±0.09b	2.50±0.04	5.30±0.05	5.90±0.05
	100 mg l^{-1}	9.70±0.17b	14.80±0.23b	23.00±0.10b	2.60±0.07	5.40±0.09	5.30±0.04
ABA+GAs	25 mg l^{-1}	7.30±0.13a	11.60±0.33a	19.30±0.18b	2.10±0.06	5.00±0.11	5.20±0.05
	50 mg l^{-1}	7.10±0.12b	11.20±0.41b	18.90±0.16b	2.30±0.04	5.30±0.03	5.70±0.14
	100 mg l^{-1}	7.00±0.27b	11.00±0.14b	18.10±0.09b	2.30±0.06	5.50±0.04	5.50±0.07
CONTROL	D. W.*	8.00±0.140a	12.00±0.12a	22.00±0.15a	2.20±0.03a	5.40±0.10a	6.10±0.05a

*D. W. is distilled water

at 30 days was seen in ABA 25 mg l^{-1} and GAs 100 mg l^{-1} treated plants (7, 12.5% decrease from control), at 60 days in ABA 100 mg l^{-1} treated plants (10.34, 13.8% decrease from control) and at 90 days in ABA 50 mg l^{-1} treated plants (20.30, 7.7% decrease from control) (Table no. 1). Maximum increase in number of leaves was noted in IAA+BAP 100 mg l^{-1} treated plants at 30 days (10.10, 26.3% increase from control), 60 days (16.10, 34.2% increase from control) and 90 days (27.30, 24.1% increase from control) of plant growth. Whereas, minimum number of leaves at 30 days (7, 12.5% decrease from control), at 60 days (11, 8.3% decrease from control) and at 90 days (18.10 cm, 17.7% decrease from control) was seen in ABA+GAs 100 mg l^{-1} treated plants. It is evident from Table no. 1 that no variability due to different concentration of IAA in number of leaves was found in 30 days old plants. However all other treatments showed significant differences within 30 days of plant growth and with age this difference persisted. BAP and IAA+BAP had synergistic effect on number of leaves. ABA alone as well as with GAs and IAA reduced number of leaves.

In the present study, as the concentration of BAP and IAA+BAP increased the number of leaves increased and delayed the senescence. PGRs can delay or accelerate senescence. Delay in senescence resulted

Table 2: Leaf surface area in *Fagopyrum esculentum* at different growth stages under various phytohormone treatments; values are mean±S.E. n=3, analysed by Two-way ANOVA followed by Bonferroni's multiple comparison test. Values followed by the same letter are not statistically different (P<0.05) compared with control.

PGRs	Treatment	Leaf surface area (cm^2)		
		30 Days	60 Days	90 Days
IAA	25 mg l^{-1}	7.20±0.22a	15.20±0.19b	19.10±0.26a
	50 mg l^{-1}	7.60±0.18a	15.70±0.23b	20.00±0.13b
	100 mg l^{-1}	7.40±0.14a	16.00±0.25b	20.70±0.18b
BAP	25 mg l^{-1}	7.30±0.16a	15.10±0.10b	20.00±0.22b
	50 mg l^{-1}	7.50±0.12a	15.60±0.21b	20.60±0.15b
	100 mg l^{-1}	7.70±0.11b	16.30±0.12b	21.00±0.18b
ABA	25 mg l^{-1}	7.00±0.10a	13.10±0.22b	17.30±0.13b
	50 mg l^{-1}	7.30±0.09a	13.70±0.17b	17.70±0.14b
	100 mg l^{-1}	7.20±0.07a	13.50±0.15b	18.40±0.13a
GAs	25 mg l^{-1}	6.60±0.25a	13.30±0.17b	17.30±0.12b
	50 mg l^{-1}	6.70±0.16a	13.60±0.13b	17.70±0.20b
	100 mg l^{-1}	7.40±0.10a	14.20±0.11a	18.20±0.18a
IAA+BAP	25 mg l^{-1}	7.70±0.12b	14.70±0.13a	19.00±0.22a
	50 mg l^{-1}	8.10±0.16b	15.30±0.11b	19.70±0.12b
	100 mg l^{-1}	8.40±0.21b	15.70±0.14b	20.00±0.13b
IAA+ABA	25 mg l^{-1}	6.80±0.15a	13.30±0.24b	17.70±0.19b
	50 mg l^{-1}	6.30±0.13b	13.70±0.11a	17.30±0.19b
	100 mg l^{-1}	6.20±0.10b	13.00±0.13b	17.50±0.25b
IAA+GAs	25 mg l^{-1}	7.30±0.13a	15.00±0.17b	18.80±0.20a
	50 mg l^{-1}	7.70±0.18b	15.50±0.15b	19.00±0.11a
	100 mg l^{-1}	8.30±0.22b	15.80±0.16b	19.30±0.14b
ABA+BAP	25 mg l^{-1}	6.30±0.13b	13.70±0.12b	17.60±0.23b
	50 mg l^{-1}	6.40±0.12b	13.30±0.14b	17.90±0.26b
	100 mg l^{-1}	6.70±0.21a	14.20±0.11a	18.50±0.13a
BAP+GAs	25 mg l^{-1}	8.20±0.17b	15.30±0.12b	20.00±0.21b
	50 mg l^{-1}	8.30±0.12b	15.50±0.11b	20.50±0.14b
	100 mg l^{-1}	8.50±0.19b	16.00±0.15b	21.00±0.25b
ABA+GAs	25 mg l^{-1}	5.00±0.13b	13.70±0.28a	17.50±0.21b
	50 mg l^{-1}	5.30±0.12b	13.30±0.17b	17.70±0.15b
	100 mg l^{-1}	6.00±0.11b	14.00±0.13a	18.20±0.18a
CONTROL	D. W.*	7.10±0.11a	14.30±0.16a	18.60±0.22a

*D. W. is distilled water

increased number of leaves compared to other treatments, especially those plants treated with ABA. Hazarika *et al*. (2016) and Singh and Singh (2009) also reported increased number of leaves after BAP application. Increase in number of leaves on BA or BAP application may be due to increase in cell division caused by cytokinins and higher supply of assimilates mediated by application of BA (Dwivedi *et al.* 1999). IAA has been reported to increase number of leaves (Bhandari *et al*., 2009; Muthulakshmi and Pandiyarajan, 2015). Present study revealed that ABA+GAs (100 mg l^{-1}) had lowest number of leaves in *F. esculentum.* Similarly, GAs (150 ppm) decreased number of leaves in *Balanites aegyptica* during first season of growth (Mostafa and Abou Alhamd, 2011). Leite (2003) also noticed that foliar treatment with GA_3 had no effect on the number of leaves.

Leaf area

Leaf area showed an increasing trend with the advancement of plant age in all the treatments of phytohormones. BAP 100 mg l^{-1} treatment was most effective in eliciting maximal leaf area at 30 days (7.70 cm^2, 8.5% increase from control), at 60 days (16.30 cm^2, 14% increase from control) and at 90 days (21 cm^2, 12.9% increase from control) of growth in *Fagopyrum*

Table 4.3: Number of branches in *Fagopyrum esculentum* at different growth stages under various phytohormone treatments; values are mean±S.E. n=3, analysed by Two-way ANOVA followed by Bonferroni's multiple comparison test. Values followed by the same letter are not statistically different (P<0.05) compared with control.

PGRs	Treatment	No. of branches plant^{-1} 60 Days	No. of branches plant^{-1} 90 Days	PGRs	Treatment	No. of branches plant^{-1} 60 Days	No. of branches plant^{-1} 90 Days
IAA	25 mg l^{-1}	2.30±0.12b	3.40±0.15a	IAA+ABA	25 mg l^{-1}	3.00±0.05a	3.00±0.13b
	50 mg l^{-1}	2.70±0.13a	3.60±0.17a		50 mg l^{-1}	3.00±0.12a	3.30±0.10a
	100 mg l^{-1}	3.10±0.15a	4.30±0.14b		100 mg l^{-1}	3.30±0.09a	3.70±0.11a
BAP	25 mg l^{-1}	3.30±0.07a	3.90±0.12a	IAA+GAs	25 mg l^{-1}	2.60±0.02b	3.00±0.15b
	50 mg l^{-1}	3.50±0.10a	4.70±0.14b		50 mg l^{-1}	2.90±0.00a	3.00±0.12b
	100 mg l^{-1}	3.90±0.15b	5.00±0.19b		100 mg l^{-1}	3.00±0.04a	3.10±0.10b
ABA	25 mg l^{-1}	2.00±0.13b	3.00±0.07b	ABA+BAP	25 mg l^{-1}	3.70±0.00b	4.00±0.13b
	50 mg l^{-1}	3.00±0.05a	4.00±0.14b		50 mg l^{-1}	4.00±0.15b	4.70±0.13b
	100 mg l^{-1}	2.70±0.10b	3.60±0.08a		100 mg l^{-1}	4.30±0.11b	4.70±0.16b
GAs	25 mg l^{-1}	2.30±0.13b	2.70±0.22b	BAP+GAs	25 mg l^{-1}	2.00±0.08b	2.30±0.11b
	50 mg l^{-1}	2.80±0.08a	3.30±0.11a		50 mg l^{-1}	2.30±0.12b	2.50±0.14b
	100 mg l^{-1}	2.50±0.13b	3.40±0.04a		100 mg l^{-1}	2.40±0.13b	2.70±0.15b
IAA+BAP	25 mg l^{-1}	3.00±0.04a	4.20±0.12b	ABA+GAs	25 mg l^{-1}	1.20±0.05b	2.30±0.22b
	50 mg l^{-1}	3.20±0.11a	4.70±0.19b		50 mg l^{-1}	1.70±0.11b	2.70±0.14b
	100 mg l^{-1}	4.10±0.20b	5.00±0.24b		100 mg l^{-1}	2.00±0.20b	3.00±0.18b
CONTROL	D. W.*	3.10±0.09a	3.50±0.13a	CONTROL	D. W.*	3.10±0.09a	3.50±0.13a

*D. W. is distilled water

esculentum. GAs 25 mg l^{-1} treated plants at 30 days (6.60 cm^2, 7% decrease from control), ABA 25 mg l^{-1} treated plants at 60 days (13.10 cm^2, 8.4% decrease from control) and ABA 25 mg l^{-1} and GAs 25 mg l^{-1} treated plants at 90 days (17.30 cm^2, 7% decrease from control) was ineffective in enhancing leaf area. Maximal leaf area at 30 days (8.50 cm^2, 19.7% increase from control), at 60 days (16 cm^2, 11.9% increase from control) and at 90 days (21 cm^2, 12.9% increase from control) was noted in BAP+ GAs 100 mg l^{-1} treated plants. Whereas, at 30 days it receded in ABA+ GAs 25 mg l^{-1} treated plants (5 cm^2, 29.6% decrease from control), at 60 days in IAA+ABA 100 mg l^{-1} treated plants (13 cm^2, 9.1% decrease from control) and at 90 days in IAA+ABA 50 mg l^{-1} treated plants (17.30 cm^2, 7% decrease from control). It can be seen that the plant growth regulators IAA, BAP, ABA and GAs didn't show any variability at 30 days but thereafter IAA and BAP promoted leaf area, whereas ABA and GAs retarded leaf area. Combination of ABA with other PGRs retarded leaf area whereas GAs in combination with other PGRs promoted leaf area. Untreated (control) plants showed significant difference ($p<0.05$) with BAP (25, 50 and 100 mg l^{-1}), IAA+ABA (25, 50 and 100 mg l^{-1}), BAP+GAs (25, 50 and 100 mg l^{-1}) (Table 2).

Leaf area is generally considered as an index of plant growth. Therefore, increased leaf area is a confirmation that the plant growth regulators used are effective. In our results, it was observed that the leaf area increased by 100 mg l^{-1} concentration of BAP. Cytokinin is well known to stimulate leaf expansion (Sadak *et al*., 2013). Similar results of increased leaf area with BAP are reported by Ahmadi Lahijani *et al*. (2018). Significant increase in the leaf area by BAP together with GAs (BAP+GAs) at higher concentration might be attributed to the well recognized effects of GA_3 that it encourages cell extension with cell division (Taiz and Zeiger, 2010). Srivastava and Srivastava (2007) and Ahmad Dar *et al*. (2015) reported that application of GA_3 could increase leaf length in *Catharanthus roseus* and fenugreek. In present study, leaf area decreased in ABA+GAs (25 mg l^{-1}) and IAA+ABA (50, 100 mg l^{-1}) treatments. This is similar to the results of Nair et al. (2009) and Naeem et al. (2004). ABA inhibits leaf expansion (Alves and Setter, 2000).

Number of branches

Number of branches was measured at a regular interval of 60 and 90 days. Both at 60 days (3.90, 25.8% increase from control) and 90 days (5, 42.9% increase from control) duration, BAP 100 mg l^{-1} treated plants revealed highest number of branches than other applied phytohormones. Lowest number of branches at 60 and 90 days was observed in ABA 25 mg l^{-1} (2, 35.5% increase from control) and GAs 25 mg l^{-1} treated plants (2.70, 22.9% increase from control). ABA+BAP 100 mg l^{-1} treated plants at 60 days (4.30, 38.7% increase from

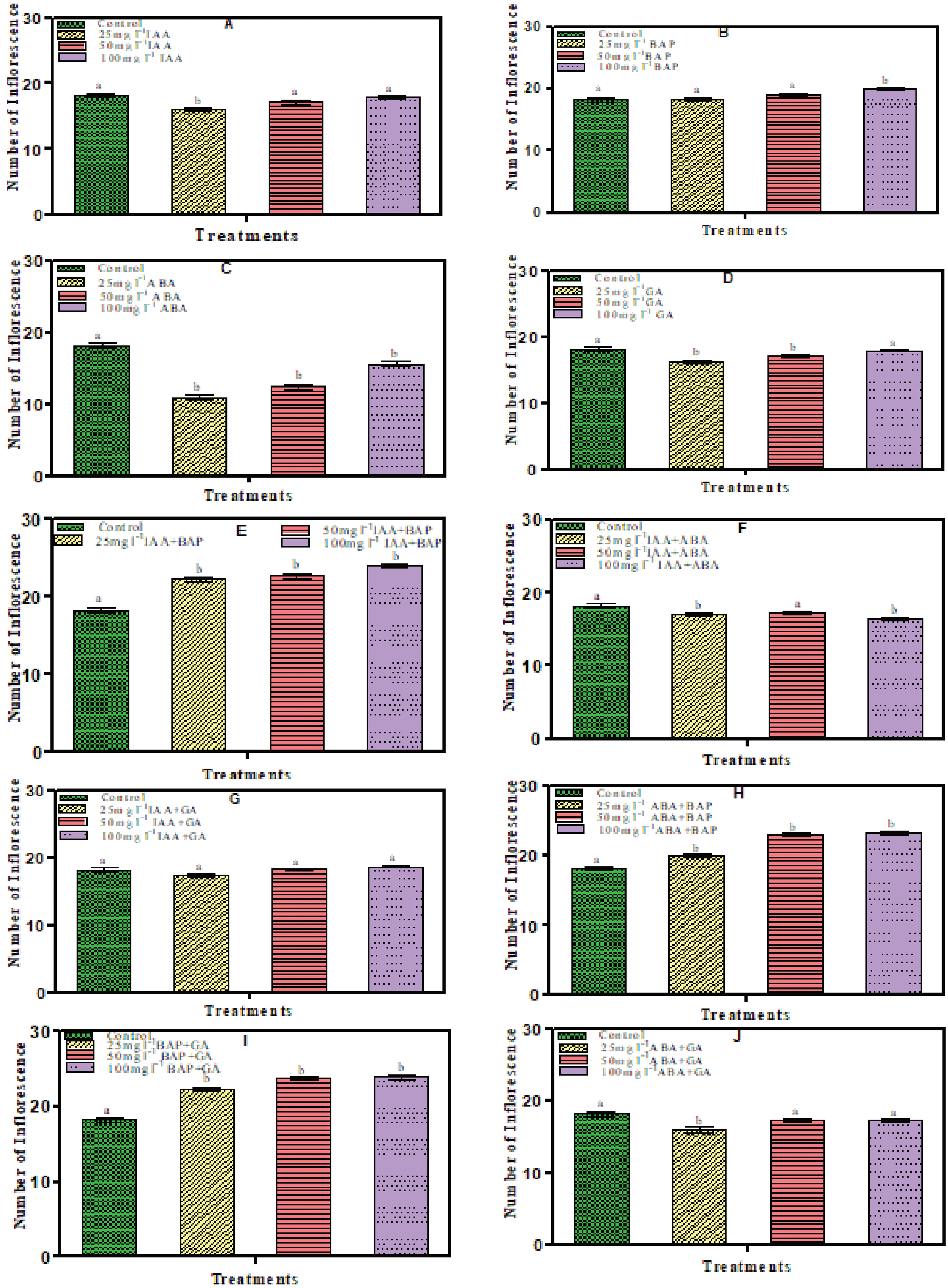

Fig. 1: Number of inflorescence in *Fagopyrum esculentum* on 60 days of plant growth treated with IAA (A), BAP (B), ABA (C), GAs (D), IAA+BAP (E), IAA+ABA (F), IAA+GAs (G), ABA+BAP (H), BAP+GAs (I) and ABA+GAs (J). Values are mean±SE; n=3, analysed by one-way ANOVA followed by Tukey's multiple comparison test. Values followed by the same letter are not statistically different (P<0.05) compared with control.

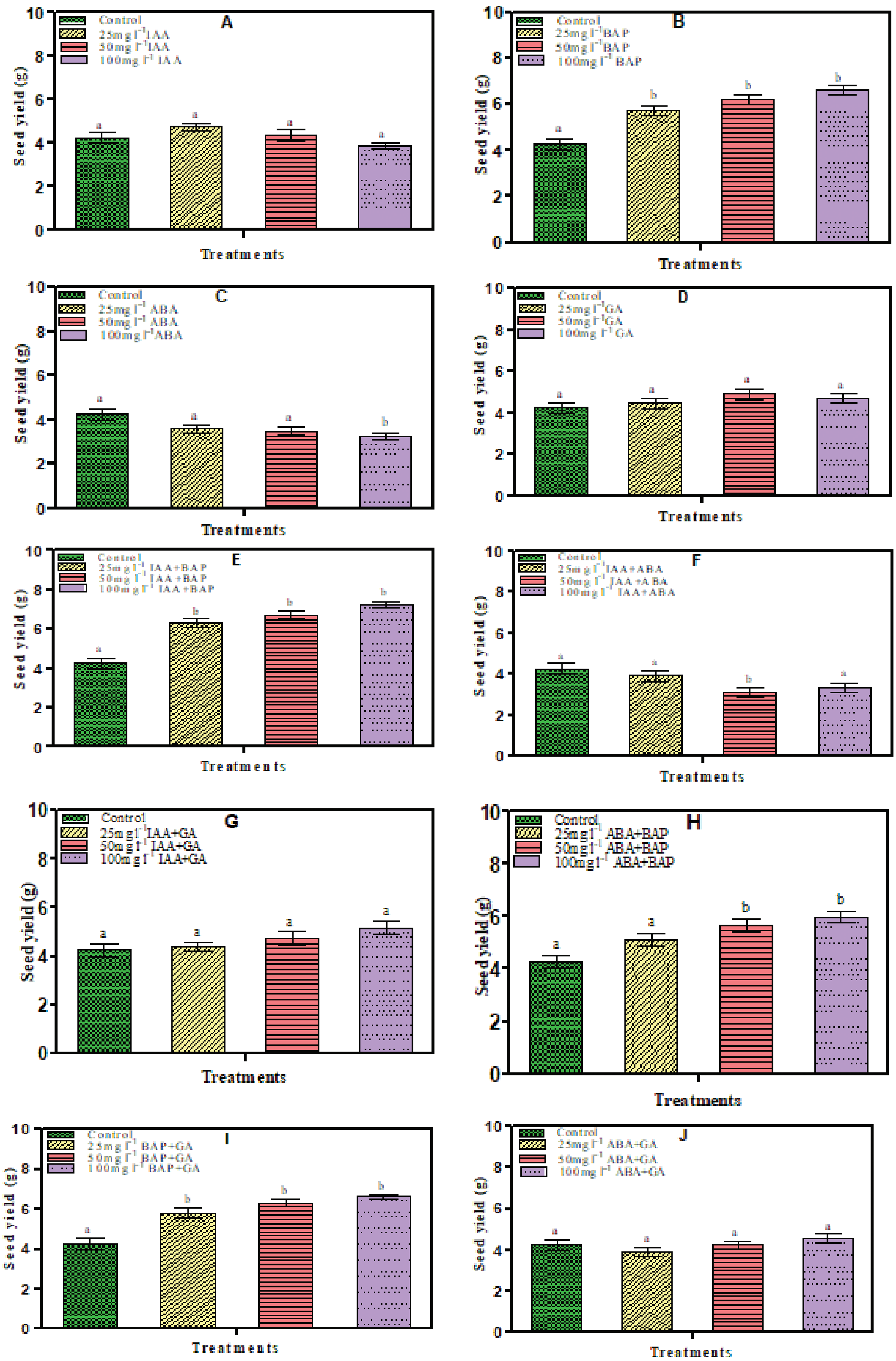

Fig. 2: Seed yield in *Fagopyrum esculentum* on 90 days treated with IAA (A), BAP (B), ABA (C), GAs (D), IAA+BAP (E), IAA+ABA (F), IAA+GAs (G), ABA+BAP (H), BAP+GAs (I) and ABA+GAs (J). Values are mean±SE; n=3, analysed by one-way ANOVA followed by Tukey's multiple comparison test. Values followed by the same letter are not statistically different (P<0.05) compared with control.

control) and IAA+BAP 100 mg l^{-1} treated plants at 90 days showed maximum number of branches (5, 42.9% increase from control). Minimum number of branches at 60 days (1.20, 61.3% decrease from control) was recorded in ABA+GAs 25 mg l^{-1} treated plants. Whereas, at 90 days BAP+GAs and ABA+GAs at 25 mg l^{-1} showed lowest number of branches (2.30, 34.3% decrease from control) (Table no 3). It is clear that in general, BAP alone and with IAA (IAA+BAP) promoted number of branches, especially at 90 days. ABA+BAP treatment had synergistic effect on number of branches. Treatment with ABA+ GAs, BAP+GAs and IAA+GAs retarded number of branches.

Number of branches per plant was significantly influenced by the BAP treatment. These results are in agreement with Britto *et al*. (2003) and Abdelgadir *et al*. (2009). It is known fact that cytokinin enhance axillary bud outgrowth (Shimizu-sato *et al*., 2009). King and Van Staden (1988) reported that application of cytokinin is effective in inducing the axillary buds at pea node. Phytohormones combination of ABA+BAP and IAA+BAP showed maximum number of branches at higher concentration. This coincides with the findings of Naeem *et al*. (2004) who reported increased number of branches in *Lens culinaris* on application of IAA alone and in combination with kinetin (IAA+kinetin). ABA is dormancy hormone and reported to hinder the growth of active buds (Yaish *et al*., 2010), but in the present studies, when ABA in combination with BAP was applied to plants the number of branches increased. It is a known fact that abscisic acid has multiple functions in the developmental processes of plants (sah *et al*., 2016). Lower concentration (25 mg l^{-1}) of ABA+GAs and BAP+ GAs decreased number of branches in present investigation. Similar findings were observed by Naeem *et al*. (2004) in lentil plant where GA_3+kinetin decreased number of branches as compared to control. Leite (2003) also noticed that foliar treatment with GA_3 had no effect on the number of branches.

Number of inflorescence

Flower initiation started within 30 days of plant growth and by 60 days prominent inflorescence was observed on the plant. Highest number of inflorescence at 60 days was achieved by BAP 100 mg l^{-1} treated plants (20, 9.9% increase from control). Whereas, minimum number in ABA 25 mg l^{-1} treated plants (11, 39.6% increase from control) (Fig. 1). In combined treatment of PGRs, maximum number of inflorescence at 60 days was marked in IAA+BAP 100 mg l^{-1} treated plants (24, 31.9% increase from control) and lowest number in ABA+GAs 25 mg l^{-1} treated plants (16, 12.1% decrease from control) (Fig.1). It can be concluded that IAA (Fig. 1 A), ABA (Fig. 1C) and GAs (Fig. 1D) alone as well as in combination with each other retarded inflorescence number. BAP alone (Fig. 1B) had not much variation from control but in combination with all other hormones (Fig. 1E, H and I) it promoted inflorescence number.

In the present study, the number of inflorescence per plant increased with the application of BAP alone or in combination with auxins. Several studies have shown that number of inflorescence increase with exogenous application of cytokinins such as in *Dendrobium* (Wang *et al*., 2009), *Jatropha curcas* (Pan, 2011) and *Dendrobium* (Nambiar *et al*., 2012). Increased number of inflorescence due to cytokinins may be due to the fact that it promotes flower bud formation (Bernier and Périlleux, 2005). BAP with GAs at 100 mg l^{-1} enhanced inflorescence number significantly. Similar findings were reported with BA+ GAs on flowering of *Picea sitchensis* (Tompseet, 1977). Our results are consistent with effect of GAs on flowering by Wahyuni *et al*. (2011) on *Brunonia australis* and Chetna *et al*. (2013) on *Withania somnifera*. Number of flowers has been shown to increase under IAA effect in Lentil (Khalil *et al*., 2006) and *Verbascum thapsus* (Bhandari *et al*., 2009). In this study, number of inflorescence reduced in plants given treatment of ABA+GAs (25 mg l^{-1}). Prat *et al*. (2008) had similar findings with treatment of gibberellins. Function of gibberellins in flowering is difficult because different species react differently to them (Prat *et al*., 2008). The inhibitory effect of gibberellins on flowering is well recognized (Bradley and Crane, 1960; Retamales *et al*., 2000). ABA application decreased inflorescence number in grapes (Palma and Jackson, 1989).

Seed yield

Maximum seed yield at 90 days was shown by BAP 100 mg l^{-1} treated plants (6.63 g, 55.6% increase from control), while minimum in ABA 100 mg l^{-1} (3.27 g, 23.2% decrease from control) treated plants. In combination of PGRs treatment, maximum seed yield after 90 days was seen in IAA+BAP 100 mg l^{-1} (7.26 g, 70.4% increase from control) treated plants and minimum in IAA+ABA 50 mg l^{-1} (3.15 g, 26.1% decrease from control) treated plants (Fig. 2). It is clear from Fig 2 that BAP (Fig. 2B), IAA+BAP (Fig. 2E), ABA+BAP (Fig. 2H) and BAP+GAs (Fig. 2I) enhanced seed yield, whereas ABA (Fig. 2C) and IAA+ABA (Fig. 2F) reduced it. Other applied hormonal treatments showed no variations in seed yield.

Seed yield was highest in BAP and IAA+BAP treatments at 100 mg l^{-1} concentration. Flower development is vital for enhancing seed yields. BAP treatment significantly increased seed yield by increasing the total number of flowers. Increase in the number of flowers in BAP treated plants may be due to the role of cytokinin in the regulation of inflorescence meristem activity and size (Kiba and Sakakibara, 2010). Published researches has also reported enhanced seed yield with BAP in *Jatropha curcas (*Pan and Xu*, 2011) and Triticum aestivum* (Bagdi *et al*., 2011). Similar to our results, Shawkat (2005) also reported that by spraying plants of summer squash (*Cucurbita pepo*) with IAA (100 and 200 mg/1) plant yield improved, in comparison to control plants. Also, Bhandari *et al*. (2009) while working on *Verbascum thapsus* revealed that IAA (200 ppm) was best treatment for number of fruits. Decrease in seed yield was revealed by plants given treatment of IAA+ABA (50 mg l^{-1}). Perez-Jimenez *et al*. (2015) also observed reduced seed yield in *Capsicum annuum* by ABA treatments. IAA (200 ppm) produced lowest number of seeds in soybean plant (sarkar *et al*., 2002).

Exploitation of 'underutilized' crops can contribute effectively to promote nourishment and biological sustainability. *Fagopyrum esculentum* Moench is one of the essential neglected crops having high nutritive and medicinal value. The productivity of crop is quite less. From the present study it can be concluded that PGRs effectively increased morphology and productivity of *Fagopyrum esculentum*. Combination of PGRs were more effective than solely applied PGRs. Higher concentration was effective than lower concentration of different treatments. IAA+BAP at 50 and 100 mg l^{-1} gave best results in terms of morphology, and productivity. The results of the present study call for further research on mechanism of PGRs action by using molecular approaches.

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