OnLine Journal of Biological Sciences 1 (9): 801-804, 2001
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Effect of Growth Hormones *i.e.*, GA_3, IAA and Kinetin on the External Morphology and Flowering of *Phaseolus aureus* L.

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Abstract: Effect of the growth hormones *i.e.*, gibberellic acid (GA_3), indole-3-acetic acid (IAA) and Kinetin were studied on the shoot of *Phaseolus aureus* L., after 45 and 60 days of treatment and was compared with control. Hormones were used individually *i.e.*, 100 ppm GA_3, 100 ppm IAA and 20 ppm Kinetin and in combinations *i.e.*, 100 ppm GA_3 + 100 ppm IAA, 100 ppm GA_3 + 20 ppm Kinetin, 100 ppm IAA + 20 ppm Kinetin and 100 ppm GA_3 + 100 ppm IAA + 20 ppm Kinetin. Applied GA_3 proved to be very effective individually as well as in combinations thus increasing the number of internodes and leaves as compared to control as well as IAA and Kinetin treatments. Likewise early initiation of flowering was observed in the GA_3 treatments. The maximum number of flowers were borne on the plants treated with GA_3. The IAA and Kinetin treatments did not show any significant increase/decrease in the number of internodes and leaves when compared with control. Moreover, IAA and Kinetin did not show any significant effect on the floral initiation and on the number of flowers per plant.

Key words: Shoot, leaves, flowering, hormones, GA_3, IAA, kinetin, *Phaseolus aureus* L.

Introduction

The simultaneous interaction of growth hormones *i.e.*, Auxins, Gibberellins and Cytokinins in regulating various aspects of growth and differentiation appears to be a general feature of higher plants. GA_3 increased the length of stem internodes and accelerated the onset of floral development (Hernandez, 1997). In *Pisum sativum* L., GA_3 was responsible for the internode growth (Ingram *et al.*, 1986). Chaudhry and Zahur (1992) have reported that IAA augments expansion in the cells while GA_3 shows inhibition in the diameter. In the internal morphology elongation has been observed in the cells by these workers. Applied IAA inhibits the length of shoot (Adams and Ross, 1983). Cytokinins ordinarily inhibit elongation of stem (Fox, 1964) and stimulate growth by swelling rather than elongation (Katsumi, 1962). GA_3 plays important role in physiological processes of leaf growth (Harada and Vergara, 1972). Kumar *et al.* (1981) reported that potato plants produced fleshy and dark green leaves with the application of IAA. Similarly, Kinetin treated plants also have lushy leaves thus showing chloroplast development. Moreover, cultures of tobacco callus become green in light if a cytokinin is supplied. Stetler and Laetsch (1965) have found that the conversion of proplastids into chloroplasts with grana is specifically stimulated by cytokinins.

Auxins can either promote or inhibit floral initiation in plants. Inhibitions have been reported for a wide diversity of species *i.e.*, Xanthium, Kalanchoe, Soybean, Barley, Sunflower, Calendula and others (Lang, 1952 and Livermann, 1955). IAA has been shown to induce flowering in pine apple (Van Overbeck, 1946). More recently Ritenowi *et al.* (1996) have reported that IAA plays an important role in axillary bud development. Nakayama *et al.* (1962) have reported that flowering can be enhanced by cytokinins. GA_3 treated plants of *Stellaria longipes* golidedid not show flowering (MacDonald *et al.*, 1986). Contrarily Bonnet *et al.* (1987) reported that GA_3 induced flowering in Western hemlock. Gibberellins are involved in flowering growth in some plants (Rao *et al.*, 1980).

Materials and Methods

Phaseolus aureus forms the basic subject of the present work. Effects of growth hormones *i.e.*, Gibberellic acid (GA_3), Indole-3-acetic acid (IAA) and Kinetin have been observed on the shoot of the plant. The seeds were sown in earthenware pots and were watered at regular intervals.

In order to study the effects of growth hormones on external morphology, the plants were ear-marked according to their treatments. When the cotyledons were fully opened and oriented horizontally with respect to the hypocotyledonary axis 27 μl of every hormonal treatment was applied on the apical meristem of the plant. This treatment was repeated after every 24 h. The effect of hormones was studied on the number of internodes per plant and number of leaves as well as flowering *i.e.*, early or late flowering following treatments and thus compared with control. The date of appearance of 1st flower bud was recorded. The total number of flowers per plant was also counted and compared with control. The mean of 10 readings was taken and calculated as final (Steel and Torrie, 1981).

Results

Number of internodes per plant: The number of internodes per plant was recorded in treated as well as in control plants. The 100 ppm GA_3 treated plants showed a significant increase as the number of internodes after 45 as well as 60 days in compare to control. In IAA treatments the number of internodes showed no increase in comparison to control after 45 days. However, after 60 days there was an insignificant increase in number of above mentioned treatments. Applied Kinetin revealed an increase in number when compared with control after 45 as well as 60 days.

In the mixed dose of 100 ppm GA_3 + 100 ppm IAA, 100 ppm GA_3 + 20 ppm Kinetin and 100 ppm GA_3 + 100 ppm IAA + 20 ppm Kinetin registered a remarkable increase in the number as compare to control. The mixed dose of IAA and Kinetin showed no response however an insignificant increase in the number was recorded (Table 1).

Number of leaves per plant: The number of leaves in control were 6 after 45 days and 6.2 after 60 days. The 100 ppm GA_3 treated plants showed an increase which being 7.5 after 45 days and 9 after 60 days. The extraneous GA_3 proved to be more effective than applied IAA and Kinetin. In IAA the increase in number was 7.2 and 7.5 after 45 and 60 days while in Kinetin treatments the number was 6.2 and 7.3 after 45 and 60 days.

The combined doses of GA_3 with IAA and Kinetin showed a marked increase in number when compared with control. The mixed dose of IAA and Kinetin registered increase over the control but it was less than that with mixed GA_3 doses. All the

Table 1: Effect of growth hormones on external morphology of shoot of *P. aureus* L.

Treatments	45 days		60 days	
	No. of internodes per plant	No. of leaves per plant	No. of internodes per plant	No. of leaves per plant
Control	3.0 ±0.00	6.0 ±0.00	3.2 ±0.319	6.2 ±0.197
100 ppm GA_3	4.7 ±0.288	7.5 ±0.526	5.3 ±0.281	9.0 ±0.00
100 ppm IAA	3.0 ±0.00	7.2 ±0.439	4.0 ±0.00	7.5 ±0.517
20 ppm Kinetin	3.5 ±0.511	6.2 ±0.821	4.2 ±0.021	7.3 ±0.293
100 ppm GA_3 + 100 ppm IAA	4.2 ±0.325	6.6 ±0.742	5.1 ±0.011	9.0 ±0.00
100 ppm GA_3 + 20 ppm Kinetin	4.4 ±0.163	7.3 ±0.873	5.2 ±0.150	9.0 ±0.00
100 ppm IAA + 20 ppm Kinetin	3.5 ±0.635	6.4 ±0.619	3.7 ±0.531	7.6 ±0.520
100 ppm GA_3+ 100 ppm IAA + 20 ppm Kinetin	4.7 ±0.819	7.8 ±1.965	5.0 ±0.00	9.0 ±0.00

Table 2: Effect of growth hormones on flowering time and number of flowers per plant after 60 days.

Treatments	Appearance of 1st flower bud (days)	No. of flower per plant
Control	55	12.5 ±1.344
100 ppm GA_3	51	24.5 ±1.106
100 ppm IAA	56	10.5 ±0.764
20 ppm Kinetin	54	13.7 ±1.529
100 ppm GA_3 + 100 ppm IAA	53	15.5 ±2.329
100 ppm GA_3 + 20 ppm Kinetin	53	17.0 ±1.362
100 ppm IAA + 20 ppm Kinetin	56	11.4 ±0.556
100 ppm GA_3+ 100 ppm IAA + 20 ppm Kinetin	53	19.0 ±1.528

Fig. 1: Effect of 100 ppm GA_3 on extension growth compared with control

Fig. 3: Effect of 20 ppm Kinetin

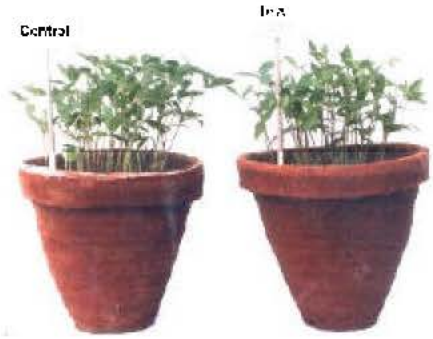

Fig. 2: Effect of 100 ppm IAA compared with control

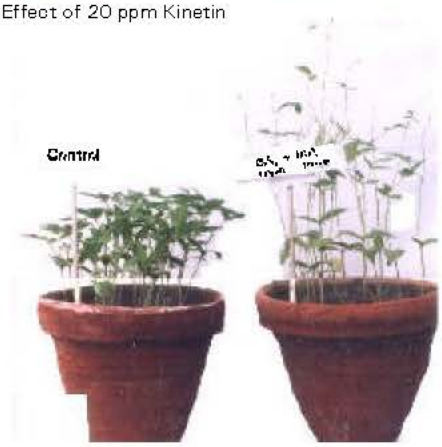

Fig. 4: Effect of 100 ppm GA_3 + 100 ppm IAA showing remarkable increased length

Fig. 5: Effect of 100 ppm GA_3+20 ppm Kinetin showing increased length

Fig. 6: Effect of 100 ppm IAA + 20 ppm Kinetin showing lush green leaves

Fig. 7: Effect of 100 ppm GA_3+100 ppm IAA + 20 ppm Kinetin showing more number of internodes and leaves with increased shoot length

3 hormones when applied simultaneously proved to be more effective than all the other doses thus increasing the number up to 7.8 and 9 after 45 and 60 days, respectively (Table 1).

Time of flowering: Hormones play an important role on the early initiation of flowering. Therefore, the date of appearance of 1st flower bud was noted in treated as well as in control plants. The 1st flower bud appeared after 55 days in control. In 100 ppm GA_3 treated plants it appeared 4 days earlier *i.e.*, early flowering was observed in comparison to control. On the other hand, the flowering in IAA treatments was delayed only for one day when compared with control. In the Kinetin treatments it appeared after 54 days. In the mixed doses of 100 ppm GA_3 + 100 ppm IAA, 100 ppm GA_3+ 20 ppm Kinetin and 100 ppm GA_3 + 100 ppm IAA + 20 ppm Kinetin the 1st flower bud was observed after 53 days. In the combination of 100 ppm IAA+20 ppm Kinetin the flowering was delayed only for 2 days when compared to control (Table 2).

Number of flowers per plant : The number of flowers per plant in treated as well as in control were counted after 60 days. In control the number of flowers were 12.5. The 100 ppm GA_3 proved to be most effective thus increasing the number to 24.5 when compared to control. Extraneous IAA applications did not reveal any significant effect when compared with control. The number being 10.5. The Kinetin treatments were less effective than the GA_3 dose, the increase in number was 13.7 in comparison to control.

In the mixed dose of 100 ppm GA_3 + 100 ppm IAA an increase of 15.5 was observed over the control. The 100 ppm GA_3 + 20 ppm Kinetin showed an increase in comparison to control which was 17. The 100 ppm IAA + 20 ppm Kinetin treatment proved to be ineffective. When all the three hormones were applied simultaneously, the number of flowers were 19 in comparison to 12.5 in control (Table 2).

Discussion

The marvelous extension growth with applied GA_3 is well known (Zahur, 1974 and Awan *et al.*, 1999). There was also a significant increase in the length of shoot and consequently in the number of internodes as well as number of leaves (Table 1; Fig. 1). However, the diameter showed a decrease. Although the leaves increased in number but they were light green in colour, this may be due to rapid consumption of nutrients during accelerated increase in length (Allsopp, 1965). In IAA treated plants inhibition in total shoot length was observed which was accompanied by the narrowing of internodes (Kumar *et al.*, 1981). There was a negligible increase in the number of leaves with the application of IAA, however, leaves were dark green and healthy when compared with control as well as with GA_3 (Fig. 2). Chaudhry (1997) working on *Abelmoschus esculentus* reported that IAA treated plants had dark green leaves. This may be due to enhanced chloroplast development. The Kinetin treated plants showed an increase in the number of internodes and number of leaves. However, no effect on the elongation of shoot was exhibited (Fig. 3). Cytokinins ordinarily inhibit elongation of stem (Fox, 1964) and stimulate growth by swelling rather than elongation (Katsumi, 1962). The leaves of Kinetin treated plants were lush green and more healthy than control, this may be due to the cause mentioned above.

The mixed doses of GA_3 + IAA and GA_3 + Kinetin revealed an increase in the number of internodes and leaves per plant after 45 days as well as after 60 days in comparison to control (Fig. 4 and 5). Gorden and Buess (1973) reported that GA_3 + IAA

treatments increased the number of leaves. However, Kabar (1997) reported that GA_3 alone and GA_3 + Kinetin are equally effective but in this experiment GA_3 showed rapid elongation and an increase in the number of internodes and number of leaves but in combination with Kinetin it became less effective. The combined dose of IAA + Kinetin showed an insignificant increase in the number of internodes and number of leaves (Table 1). Similar effects have been reported by Vanderhoef and Key (1968) working on soybean. In this treatment the leaves were lush green in colour with expanded leaf surfaces (Fig. 6). This may be due to the chloroplast development with grana and accelerated rate of photosynthesis (Stetler and Lactsch, 1965). When all the three growth hormones were applied simultaneously, they caused an increase in the number of internodes and leaves which was accompanied by increased shoot length (Fig.7). Similar reports have been given by Iqbal and Mahmood (1980) and Ingram *et al.* (1986).

The reports of Hernandez (1997) and Awan *et al.* (1999) are on record pertaining to GA_3 application on flowering, similarly in the present work this hormone induced early flowering. Moreover, increased number of flowers were seen when compared with control. The IAA treatments showed no significant delay in flowering and likewise no effect on the number of flowers was observed when compared with control. According to Lang (1952) and Liverman (1955) auxins inhibit floral initiation in some plants. Moreover, low concentrations of auxins are ineffective (Chaudhry, 1997). However, no significant delay was registered in this experiment. Nakayama *et al.* (1962) have reported that flowering can be enhanced by cytokinins. Similarly, the application of Kinetin showed early flowering and an increase in the number of flowers in comparison to control (Table 2).

The combined doses of GA_3 + IAA and GA_3 + Kinetin showed early flowering which was accompanied by more number of flowers in comparison to control. This may be attributed to the applied mixed dose of GA_3 because IAA as mentioned above has no positive effect. This is further proved by the reports of Thompson and Guttridge (1959). The combined dose of IAA and Kinetin showed late flowering in comparison to control, and proved to be ineffective in increasing the number of flowers. This delay in flowering may be attributed to the mixed dose of IAA (Chaudhry, 1997). When all the three hormones *i.e.*, GA_3 + IAA + Kinetin were applied simultaneously they caused early initiation of flowers. This treatment proved to be the most effective in increasing the number of flowers per plant (Tables 2). This clearly showed the effect of GA_3 and Kinetin on the initiation of flowering. Sachs *et al.* (1959) have reported that each of the hormones can regulate flowering in certain plants and that GA_3 was invoked as endogenous control of flowering. Furthermore, the threshold required for flowering may have been acquired earlier with the combination of these three hormones.

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