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Effect of Indole Butyric Acid (IBA) on Early Root Formation(Tomato " Sahil" Hybrid) Cuttings

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Abstract
Present study was designed to explore a suitable technique for early root development in tomato cuttings, minimizing the cost incurred on the purchase of hybrid seeds for each and every time. Production of new crop through hybrids seedlings/cuttings and to get one or two extra crops while perennials crop introduced through cuttings which can survive for 2-3 years if there is no insect, pest & disease attack. Bio-regulators affect fundamental processes of plant growth and development. Experiment was conducted at NTHRI Shinkiari Mansehra, to investigate the growth and survival rate of tomato hybrid "Sahil" to response of varying concentration of ***Indole 3-Butyric Acid***. IBA was applied @ (0.2 ppm, 0.4 ppm, and 0.6 ppm) on seedlings of tomato (*Solanum Lycopersicon* L.) to observe the effect on survival rate and root formation. It observed that the stem thickness, number of leaves, number of roots, root length and fresh weight of root were maximum at T_3 (0.6 ppm). It was also found that the increasing concentration of ***Indole 3-Butyric Acid*** up to 0.6 ppm enhanced the all agronomic traits /growth of tomato hybrids seedlings under high shade. Moreover study was initiated as offseason vegetables during winter seasons both shade in walk-in tunnel and IBA had affected significantly.

Keywords: *Solanum Lycopersicon* L, IBA, shade, roots, cuttings, seedlings.

Introduction
Tomato (*Solanum Lycopersicon L.*) is native to South America and belongs to the family S*olanaceae*. [1] . It is one of the most widely cultivated economically high value vegetable in the world. Botanically, tomato is fruits (berry), but they are commonly referred to as vegetable. It is perennial in nature but is grown as an annual crop in Pakistan. In many countries it is considered as "poor men's Orange" because of its attractive appearance & nutritive value. Today, it is recognized as one of the important commercial and dietary vegetables crops. Apart from the commercial value of tomato, it has also served as a suitable well-established reproducible model organism for the study of many biological process. A well ripe tomato (100 g of edible portion) contains water (94.1%), energy (23 calories), calcium (1.0 g), magnesium (7.0 mg), vitamin A (1000 IU), ascorbic acid (22 mg), thiamine (0.09 mg), riboflavin (0.03 mg) and niacin (0.8 mg). The average height of the plant is 100-300 cm, with a weak and woody stem that often vines over other plants. The leaves are 10-25 cm long, odd pinnate, with 5-9 leaflets on petioles, each leaflet up to 8 cm long, with a serrated margin [2] .Tomato cultivated on an area of 47100 ha with an annual production of 502300 tons in Pakistan [2] . Province wise contribution is 25% by Khyber Pakhtunkhwa, 38%, Baluchistan, 16%, Punjab and 21% of Sindh share as well. Seventy two percent of the total tomato production is contributed from spring crop and 28% from summer season crop [3]. In district Mansehra, tomato comes under very high value crop grown in the local area of Srin/ Pakhal valley but frost / chilled is major limiting factor in its production. Moreover, tomato fruit is perishable in nature. There are no cold storage/ house facilities accessible for the farmers to store it in this very district. Mostly farmer grow tomato in this area because of its high price but there is no idea for hybrid cultivation. At present, the tomato growers are diverting to the hybrids cultivation from OPV var: in some of the area due to its high production but unfortunately they could not found the true hybrid due to adoptability problem. The determinate tomatoes are

preferred by commercial growers as they wish to harvest the whole field at once. Indeterminate tomatoes are preferred by home growers and small stake holders /farmers who like to sell ripened fruit in the market throughout the season [2]. Among commercially tomato grown Cv/Vr: in the country is low yielder as compared to hybrids. The advantages of hybrid tomato cultivars are uniformity in shape and size, increased vigour, early maturity, high yield and resistance to specific pests and pathogens [4].

The Chemical/ growth promoting hormones (GPH) is used in commercial horticulture to improve plant growth and yield. Can used safely on fruits, vegetables and leafy crops. Products produce under hormonal treatment are safe to eat, like wise naturally organic product producer. Indole 3-Butyric Acid is a plant bio-regulator belonging to the auxin group referred as organic compounds either natural or synthetic that modifies or controls one or more specific physiological processes within a plant. IBA that regulate growth and influence various developmental process, including stem elongation, early root formation, callus formation, enhance flowering, enzyme induction and leaf & fruits senescence. They can accelerate or retard the growth and maturation rate [5]. Indole 3-Butyric Acid (IBA) is the leading plant hormone used to promote the formation of roots and to generate new roots in the cloning of tomato plants through cuttings. However, IAA promotes the shoot growth. It had a very positive effect on the number of adventitious roots formation in tomato cuttings. [6] .Treated seed of tomato with IAA, IBA and NAA with concentration of 25 mg/L, 50mg/L, 75mg/L, 100mg/L,125mg/L and 150mg/L apply on six genotypes in CRD with three replications showed that seedlings emergence was enhanced by all bio-regulator treatments at 100mg/L relative to control, the highest values being 92.1, 88.4 and 89.4% for the IAA, IBA and NAA treatments, respectively, marked reduction was recorded at higher concentrations of 125 and 150 mg/L for all test genotypes. [5]

Bio-regulators affect fundamental processes of plant growth and development. Indole-3-acetic acid (IAA), Indole-3-butyric acid (IBA) and Naphthalene acetic acid (NAA) are plant bio-regulators belonging to the auxin group. Plant bio regulators are organic compounds, that modify or controls one or more specific physiological processes within a plant. They can accelerate or retard the growth or maturation rate or otherwise alter the behaviour of plants or their products. [7]. Growing tomato cuttings in an ordinary soil and well decomposed FYM is much cheaper and easily adoptable for the tomato growers compared to growing of tomato cuttings with the use of growth hormones. It proved that tomato lateral shoots can be grown successfully in a media prepared with soil and well decomposed FYM in 1:1 ratio. [2] Half strength of MS medium with different concentrations of auxins, such as, IAA and IBA, were used for in vitro root formation in five varieties of tomato. All cultured shoot showed rooting response and showed positive response towards healthy root development. In case of IAA thin long roots were initiated from the cut ends at the base of shoots. While in case of IBA, though similar type of roots formed in all concentrations but the root growth in IBA supplemented media was slow. Among the four combinations tested, 0.2 mg/I IAA, showed best result with increased number of average root for all varieties. All the varieties showed good number of roots with 0.2 mg/I IAA. [8].

Peat, per litre and sand are more suitable than soil-based compost for rooting side shoots. Adding fertilizers onto the media increased the transplanting growth [9]. Tomato cuttings required plant growth regulators such that IBA, IAA and NAA to promote rooting. Singh, 1999 [10], reported that use of IAA and IBA concentrations (500 and 1000 ppm) on tomato side shoots along with control showed enhancement in rooting and growth of side shoots. Chao *et al* , 2001 [11], revealed that raising of tomato seedlings had some difficulties especially in early spring and winter because of lack of appropriate method, there should be an appropriate methods to enhance raising of tomato seedlings. [12]. IndoleAcetic Acid (IAA) at the lowest concentration stimulated stem elongation as well as root numbers and roots length. Effect of different concentrations (1500, 3000, 45000 and 10,000 ppm) of IBA on ten tomato cultivars. Naphthalene Acetic Acid (NAA) significantly increased the number of root and root length [13]. Adventitious root formation in tomato cuttings was totally suppressed with the application of IAA and IBA combination. They further observed the best root formation in tomato cuttings in 1.00 mg NAA/L. [14]. There is need of exploring an efficient method of vegetative propagation of tomato plant for its cheaper multiplication and cultivation throughout the year. Growing hybrid tomato through seed is very much expensive due to its high price. The price of single seed of hybrid is 0.13 $ at present .On the other hand poor farmer cannot afford it such a high price. [2,18].

The aim of present study was to explore suitable hybrids which can grow as perennial with short periods of dormancy. The treatments of growth promoting hormones which influenced the agronomic traits and early root formation which not only reduce the mortality but also increase fruiting habits in tomato plants , while the farmers can also utilized this vary technology to save their precious money to spent on its purchase again & again.

2. Materials and methods

2.1. Site location

National Tea High Value Crops Research Institute (PARC) Shinkiari, District (Mansehra) Khyber Pakhtunkhwa (Pakistan) was selected to conduct the investigation under their established infrastructure of high shade during February to July 2014 with the objectives to determine the seedlings response which influenced by various concentrations of IBA growth regulators on Tomato (*Solanum lycopersicum* L.).

2.2. Germplasm collection

For the purpose of research Tomato "Sahil" (hybrids cuttings) were collected from Gene bank of NTHRI and was used in the experiment. All the cuttings were very succulent at the time of treatments

2.3. Experimental design

The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replicates. A plot size of 2×3 f^2 was maintained for each treatment with 27 cuttings /treatments. The high shade nursery tunnel with 70-30 % light intensity green net as MS was used to cover the cuttings from direct sunlight and enhance the temperature inside the tunnel.

2.4. IBA treatment (ppm)

Prior to the treatment of succulent cuttings the edge of lateral portion of each has prepared with sharp knife to avoided the damage of cuttings and any fungus attack before insertion in the medium for rooting . The treatments concentration of IBA were as under. Formula used to make a solution, a quantity of solute is added to a known volume of solvent, adding 2-6 mg of the rooting hormone IBA (the solute) to 1liter of ethanol [15].

T1 0.2 ppm(IBA)
T2 0.4 ppm(IBA)
T3 0.6 ppm(IBA)

2.4. Parameters observed

After the treatments of all cuttings as per experimental design the following parameters were recorded

2.4.1. Number of roots

The number of roots after 15 days interval were recorded randomly form randomly selected plants and counted mean data for analysis.

2.4.2 Number of leaves

Average number of leaves counted for growth performance from randomly selected plants

2.4.3 Thickness of stem

Thickness of stem measured by Vernier caliper and counted on average basis

2.4.4 Number of flower

Average number of flowers counted for growth performance from randomly selected plants

2.4.5 Length of roots

Randomly selected plants were uprooted from the media and data been collected by simple measuring tap.

2.4.6 Fresh weight of roots

Fresh weight were recorded immediately after collection of the roots and preserved.

2.5 Statistical analysis

The data obtained were subjected to the analysis of variance using MSTATC [16]. The statistical differences between treatments were detected using LSD Test at 0.05% probability.

3. Results and discussion

3.1. Experimental results and discussion on roots, leaves, stem thickness, flower, root length, fresh root weight.

Experiment on Indole Butyric Acid (IBA) response to early root formation & growth in tomato hybrid cuttings under high shade nursery was conducted with the aim to explore early, more root formation with less mortality %age and diseased free plants production. Results are analysed and discussed in accordance with the procedure of [16]. The statistical differences between treatments were detected by using LSD Test at 0.05% probability and presented in tables 1-6 while ANOVA tables are presented in table (1a- 6a) respectively.

3.2. Roots length (cm)

The significant differences were observed among the different concentrations of IBA treated seedlings for root length table 1-a. IBA concentration of T3 (0.6 ppm) produced maximum roots length of seedlings with 4.32 cm against the IBA concentration of T1 (0.2 ppm) with 2.51 cm, while T2 (0.4 ppm) remained at par with T3 (0.6 ppm) table 1. Root length contributed towards all the entire morphological and physiological component. IBA clearly enhance the root length before its normal time.

Table 1: Effect of different concentrations of IBA on tomato hybrid "Sahil" cuttings for root length

Treatments IBA(ppm)	**Roots length (cm)**
T3 (0.6)	4.31 a
T2 (0.4)	3.10 b
T1 (0.2)	2.51 c
LSD (% 5)	0.33
CV (%)	4.47

Table 1-a: ANOVA for roots length.

Source	**Df**	**Sum of Square**	**Mean Square**	**F-Value**	**Prob.**
Replication	2	0.64889	0.32444		
Treatments	2	5.01449	2.50724	112.97	0.0003
Error	4	0.08878	0.02219		
Total	8	5.75216			

P value = 0.0003

However it was also observed that if the concentration increased may be the root length also increased as earlier. Similar results were reported by [8].

Figure 1: IBA experiment conducted under high shade tunnel at NTHRI.

3.2. Stem thickness (mm)

There was a significant variation among the stem thickness of different concentrations of IBA treated seedlings (2 a). Concentration of IBA T3 (0.6 ppm) produced maximum stem thickness of seedlings with 0.53 mm against the IBA concentration of T1 (0.2 ppm) with 0.35 mm while T2 (0.4 ppm) remained at par with T1 (0.2 ppm) (table 2). The stem thickness of seedlings plays a vital role in enhancing the water and nutrients transportation. May be IBA affected this hybrids efficiently not only promote growth but also increased diameter of stem which will be support fruits at time of fruit initiation and share the load because healthy plants is a sign of healthy fruits . These results supported the findings of [17,11].

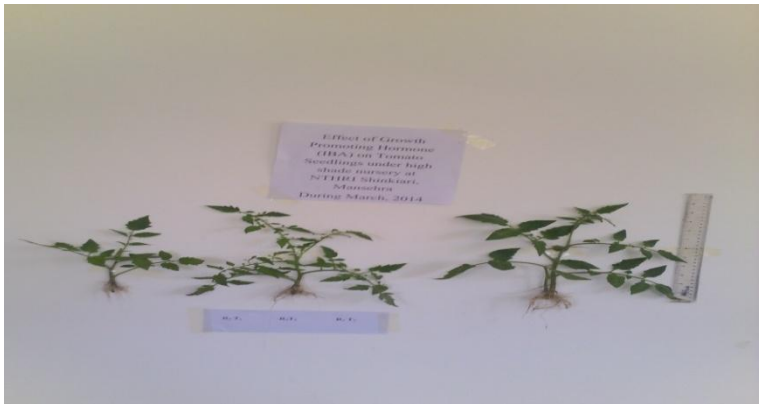

Figure 2: Different concentration of IBA showing root formation during study period.

Figure 3: Single Tomato plant of Sahil Hybrid showing response for root formation.

Table 2. Effect of different concentrations of IBA on tomato hybrid "Sahil" cuttings for stem thickness.

Treatments IBA(ppm)	**Stem thickness (mm)**
T3 (0.6)	0.53 a
T2 (0.4)	0.38 b
T1 (0.2)	0.35 b
LSD (% 5)	0.11
CV (%)	12.14

Table 2 a: ANOVA for Stem thickness.

Source	**Df**	**Sum of Square**	**Mean Square**	**F-Value**	**Prob.**
Replication	2	0.00676	0.00338		
Treatments	2	0.05309	0.02654	10.10	0.0273
Error	4	0.01051	0.00263		
Total	8	0.07036			

P value = 0.0273

3.3. Number of leaves

Statistically non-significant differences were observed among the different concentrations of IBA treated seedlings for number of leaves (table 3a). T3 (0.6 ppm) produced maximum number of leaves of seedlings with 32.00 against the IBA concentration of T1 (0.2 ppm) with 22.83 while T2 (0.4 ppm) remained at par (table 3). With T1 (0.2 ppm) and T2 (0.4 ppm) (table 3). It was observed that IBA concentration applied to the cuttings affected physiologically less as comparatively on leaves , may be IBA play a vital role for roots instated of axillaries and flowers buds . The number of leaves per seedling plays a vital role in enhancing the photosynthesis. There are identical results that were reported by [11].

Table 3: Effect of different concentrations of IBA on tomato hybrid "Sahil" cuttings for leaves.

Treatments IBA(ppm)	No of leaves
T3 (0.6)	32.00 a
T2 (0.4)	24.44 ab
T1 (0.2)	22.83 b
LSD (% 5)	7.96
CV (%)	13.30

Table 3 a: .ANOVA for Number of leaves .

Source	Df	Sum of Square	Mean Square	F-Value	Prob.
Replication	2	27.534	13.7671		
Treatments	2	143.683	71.8417	5.82	0.0654
Error	4	49.377	12.3443		
Total	8	220.595			

3.4. Number of Roots

Roots play an important role in plants and the basic source of foods supplies, it effect all parts of plants therefore IBA not only initiate the roots but also produce more number of roots for effective survival of plants. All the treated seedlings differed statistically significantly for number of roots in this study (table 4a).

Table 4. Effect of different concentrations of IBA on tomato hybrid "Sahil" cuttings for roots

Treatments IBA(ppm)	No of roots
T3 (0.6)	24.89 a
T2 (0.4)	19.11 b
T1 (0.2)	16.39 b
LSD (% 5)	3.35
CV (%)	7.35

Table 4a ANOVA for Number of roots

Source	Df	Sum of Square	Mean Square	F-Value	Prob.
Replication	2	60.323	30.1617		
Treatments	2	113.057	56.5284	25.79	0.0052
Error	4	8.768	2.1919		
Total	8	182.148			

P value = 0.0052

Concentration of IBA T3 (0.6 ppm) produced the maximum number of roots of seedlings with 24.89 against the IBA concentration of T1 (0.2 ppm) with 16.39 while T2 (0.4 ppm) remained at par with T1 (0.2 ppm) (table 4). Number of roots contributed towards all the morphological and physiological component and also give sport to plant against uprooting. There were comparable results that were reported by [5] .

3.5. Number of flowers

Data presented the non-significant differences were observed among the different concentrations of IBA treated seedlings for number of flowers (table 5a). IBA concentration of T3 (0.6 ppm) obtained maximum number of flowers with 0.77 against the IBA concentration of T1 (0.2 ppm) with 0.55 while T2 (0.4 ppm) remained at par with T1 (0.2 ppm) and T3 (0.6 ppm) after (table 5). Poor formation of flowers may be due to

highly chilly weather, same results reported by [18]. In local climatic condition the weather remains unfavourable till May, but in this case the experiment was initiated during February due to treatment plan of IBA. It has also been concluded from the results that immediately after the imitation of roots plants must be shifted to the normal media or under tunnel for more flowers formation and fruiting later on. The number of flower per seedling plays a vital role in enhancing the yield. These results supported the findings of [12].

Table 5**.** Effect of different concentrations of IBA on tomato hybrid "Sahil" cuttings for flowers

Treatments	Number of flowers
T3 (0.6)	0.77 a
T2 (0.4)	0.66 ab
T1 (0.2)	0.55 b
LSD (% 5)	0.21
CV (%)	14.30

Table 5**a:**ANOVA for Number of flowers

Source	Df	Sum of Square	Mean Square	F-Value	Prob.
Replication	2	0.05447	0.02723		
Treatments	2	0.07260	0.03630	4.00	0.1112
Error	4	0.03633	0.00908		
Total	8	0.16340			

P value = 0.1112

3.6. Fresh weight of roots (mg)

All the treated seedlings statistically differed significantly in case of fresh weight of roots in this study (table 6a). Concentration of IBA T3 (0.6 ppm) revealed maximum fresh weight of roots 0.29 mg of seedlings with against the IBA concentration of T1 (0.2 ppm) with 0.15 mg while T2 (0.4 ppm) remained at par with T1 (0.6 ppm) (table 6). Fresh root weight showed the presence of carbohydrates and starch contents presuming the strong plant base. Fresh weight of roots contributed towards all the morphological and physiological component. These results confirmed the findings of [6, 18]. Figures 1 showed the physical practice on the ground under tunnel, while (figure 2-3) reflected the root formation, growth and numbers of roots per plant. It was also observed that some roots become yellowish after it uprooted from the media, may be due to over watering in cold weather. However there were no symptoms of any disease at all thought the study period.

Table 6**:** Effect of different concentrations of IBA on tomato hybrid "Sahil" cuttings for fresh root weight

Treatments	Fresh weight of roots (mg)
T3 (0.6)	0.29 a
T2 (0.4)	0.19 b
T1 (0.2)	0.15 b
LSD (% 5)	0.09
CV (%)	4.47

Table 6a**:** ANOVA for Fresh weight of roots

Source	Df	Sum of Square	Mean Square	F-Value	Prob.
Replication	2	0.00887	0.00443		
Treatments	2	0.03120	0.01560	8.07	0.0395
Error	4	0.00773	0.00193		
Total	8	0.04780			

P value= 0.0395

Conclusion

It was concluded from this study that ***Indole Butyric Acid*** (IBA) has no significant effect on tomato hybrid seedling for number of flower and number of leaves, but have a signification effect on tomato hybrid seedlings for stem thickness, number of roots, root length and fresh weight of roots and also no mortality found in all treatments. It was also found that the increasing concentrations of IBA enhance the growth of tomato hybrids seedlings. IBA concentration at the rate of 0.6 ppm was found best in sand, therefore, recommended for further study to produce quality production of tomato hybrid seedlings.

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