EFFECT OF VARIOUS CONCENTRATIONS OF IN-DOLE BUTYRIC ACID (IBA) ON OLIVE CUTTINGS

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The research study was conducted at Pakistan Oilseed Development Board Tarnab Research Center Peshawar to evaluate the effect of different concentrations of indole butyric acid (IBA) at 0, 100, 200 and 300 ppm on hardwood and semi-hardwood cuttings of olive. Cuttings were taken in the month of October from an eight-year-old mother plant and dipped in the required IBA solutions for five seconds. The highest survival percentage, shoot length, shoot diameter, number of leaves per shoot, number of branches per shoot, number of roots per plant, root length and root diameter were recorded in both types of cuttings treated with IBA 300 ppm. The maximum number of branches per shoot was observed in hardwood cuttings as compared to semi-hardwood cuttings. Also hardwood cuttings treated with IBA (100 ppm) gave the highest number of branches per shoot. On the basis of overall performance either type of olive cuttings treated with IBA solution (300 ppm) showed the best results to propagate olive. **Keywords:** olive, Olea europea, IBA concentrations, plant survival, root initiation, cuttings

Einfluss verschiedener Konzentrationen von Indolbuttersäure (IBA) auf Olivenstecklinge. Die Studie wurde am Pakistan Oilseed Development Board Tarnab Research Center Peshawar durchgeführt, um die Wirkung verschiedener Konzentrationen von Indolbuttersäure (IBA) bei 0, 100, 200 und 300 ppm auf verholzte und halbverholzte Stecklinge bei Olive zu untersuchen. Stecklinge wurden im Monat Oktober von einer acht Jahre alten Mutterpflanze genommen und fünf Sekunden lang in die entsprechenden IBA-Lösungen getaucht. Maximale Überlebensrate, Trieblänge, Triebdurchmesser, Anzahl der Blätter pro Trieb, Anzahl der Austriebe pro Trieb, Anzahl der Wurzeln pro Pflanze, Wurzellänge und Wurzeldurchmesser wurden bei beiden Arten von mit 300 ppm IBA behandelten Stecklingen aufgezeichnet. Die maximale Anzahl der Austriebe pro Trieb bei verholzten Stecklingen wurde mit der bei halbverholzten Stecklingen verglichen. Verholzte Stecklinge, die mit IBA (100 ppm) behandelt wurden, ergaben die höchste Anzahl an Austrieben pro Trieb. Hinsichtlich der Gesamtleistung zeigten beide Arten von Stecklingen die besten Ergebnisse für die Vermehrung von Olive bei einer IBA-Behandlung mit 300 ppm.

Schlagwörter: Olive, Olea europea, IBA-Konzentration, Überlebensrate, Wurzelbildung, Stecklinge

Olive (Olea europea), locally called Zaitoon or Khuna, belongs to the Oleaceae family. The olive can be consumed as a fruit, or oil extracted from it can be used for various purposes. Olive oil accounts for 20 to 30 percent of the fresh weight of the fruit and is well known throughout the world for its nutritive and medicinal properties. The oil is nutritious because of the higher contents of oleic acid with therapeutic effect (ASHRAFI et al., 2008). In human history, the domestication of olives started approximately 6500 years ago (LOUKAS and KRIMBAS, 1983). Olives originated from Palestine, Lebanon, Syria and Cyprus and later cultivation was spread to Italy, Spain and North Africa (SIMMONDS, 1976).

Olive can be grown from seed with high success rate of germination; however, growing from seed is a very slow process and it takes time to become a plant that is fit for budding, grafting or fruiting. Therefore BALOCH (1994) suggested that other methods of propagation are to be tried to get fruits as quickly as possible. Cuttings is one of the simplest and most rapid ways of propagation, using hardwood and semi-hardwood cuttings (SIDDI-QUI and HUSSAIN, 2007). Plants can synthesize 3-types of endogenous auxins for root initiation at early stages of development (AINI et al., 2009; RASHOTTE et al., 2003; SIMON and PETRASEK, 2011). For the last few decades many researchers have been involved in determining the role of IAA and IBA in root initiation and development. While the development of primary root received a lot of attention, the formation of lateral and adventitious roots is not well understood (CASSON and LINDSEY, 2003). However, it is known that IBA effectively induces lateral rooting, adventitious rooting and acrobasal type of the rooting system in other species (CHHUN et al., 2003; Chhun et al., 2004; Ludwig-Muller et al., 2005; Ste-FANCIC et al., 2005; WANG et al., 2003).

In Pakistan, millions of wild olive trees (Olea cuspidata and Olea ferrugenea) are found in Federally Administrated Tribal Area, Khyber Pakhtunkhwa, Potohar region and Northern Baluchistan. The presence of such large numbers of wild olive trees indicates that the agro-climatic conditions of these areas are favourable for olive cultivation. Additionally, a small number of different olive varieties belonging to Olea europea that yields a high number of fruits were introduced into various parts of the country. Currently, there is a demand for the establishment of a large number of olive varieties in these regions in the shortest possible time. The present study is therefore planned to evaluate the performance of different types of olive cuttings treated with different concentrations of indole butyric acid (IBA).

MATERIALS AND METHODS

All the experiments were conducted at Pakistan Oilseed Development Board Tarnab, Peshawar. A Randomized Complete Block Design (RCBD) with the split plot arrangement was used for experimentation, having three replications. Types of cuttings were allotted to main plots while IBA concentration to subplots. Two types of cuttings were used; hardwood cuttings were taken from dormant, mature stems which were fully dormant with no obvious signs of active growth while semi-hardwood cuttings were prepared from partially mature wood of the current season's growth. The cuttings were taken from an eight year old olive tree of the 'Leccino' cultivar. The experiment was performed in the month of October and twenty cuttings of uniform length (5 to 6 inches) and leaf number were used for each treatment. Cuttings were dipped in different IBA concentrations of 0 ppm = control, 100, 200 and 300 ppm for 5 seconds and then planted in polyethylene bags with media of sand, silt and farmyard manure in combination of 1:1:1 for callus formation up to the end of April. The cuttings were planted in nursery beds in the first week of May. A distance of 5 cm between cuttings was maintained while rows were kept 45 cm apart with a two-third portion buried in soil. For partial shade, cuttings were planted in plastic tunnels with the help of dipper and covered with nylon shade cloth. All the cultural practices, irrigation, fertilizers, weed control and intercultural practices were applied uniformly during the experiment. The data regarding plant survival percentage (%), number of days to bud sprout, number of shoots per plant, shoot length (cm), shoot diameter (cm), number of leaves per shoot, number of branches per shoot, number of roots per plant, root length (cm) and root diameter (cm) were recorded at the end of November.

The data were analyzed by using Randomized Complete Block Design (RCBD) with eight treatment combinations, replicated three times and means were further assessed for differences through Least Significant Difference (LSD) test. Statistical computer software, MSTATC (Michigan State University, East Lansing/Michigan, USA), was applied for computing both the ANOVA and LSD (STEEL et al., 1997).

RESULTS AND DISCUSSION

PLANT SURVIVAL PERCENTAGE (%)

The data (Table 1) regarding plant survival percentage showed that there is a significant effect of different IBA concentrations on the plant survival percentage, whereas the effect of cutting type and the interaction between IBA concentration and cutting type did not significantly differ. The highest plant survival (69.17 %) was found in plots having cuttings treated with 300 ppm of IBA, followed by cuttings treated with 200 ppm of IBA, whereas the minimum number of plants survived in control. These results are in accordance with those of MARSHALL and WARRING (1985) and SIDDIQUI and HUSSAIN (2007).

The survival of the cuttings treated with high concentrations of IBA may be directly linked with the capacity of the growth regulator to stimulate the generation of adventitious roots. The adventitious roots absorb mineral nutrients from the soil, which helps in the survival of the cuttings. Evidence for the involvement of IBA, but not IAA, in lateral root or adventitious root development was reported in rice by WANG et al. (2003). CHHUN et al. (2003 and 2004) observed that IBA was also able to induce adventitious roots or lateral roots, while IAA gave the same response at 20-fold higher concentrations.

NUMBER OF DAYS TO BUD SPROUT

It is indicated from the results that IBA concentration applied to different type of olive cuttings did not affect the number of days to bud sprout significantly (Table 1). The reason for this phenomenon is that the IBA or auxin application does not have a direct effect on bud sprout and shoot development, but is mainly responsible for inducing lateral roots or adventitious roots. WAHAB et al. (2001) and MITRA and BOSE (1990) also reported that there is no effect of IBA application on number of days to bud sprout. Bud sprouting may be due to the stored

Table 1: Effect of IBA concentrations and type of cuttings on plant survival percentage, number of days to bud sprout, number of shoots per plant, shoot length (cm) and shoot diameter (cm) of olive

Type of cutting	Plant survival percentage	Number of days to bud sprout	Number of shoots per plant	Shoot length (cm)	Shoot diameter (cm)
Hardwood ¹	46.67	31.33	2.67	16.28	0.26
Semi-hardwood ¹	36.25	31.17	1.92	13.10	0.23
Significance level	n.s.	n.s.	n.s.	n.s.	n.s.
IBA conc. (ppm) ²	······································				
0	5.00 c	30.33	1.83	4.96 d	0.14 b
100	42.50 b	31.33	2.17	13.42 c	0.26 a
200	49.17 b	32.00	2.50	18.84 b	0.29 a
300	69.17 a	31.33	2.67	21.55 a	0.29 a
LSD ($\alpha = 0.05$)	5.40	n.s.	n.s.	1.74	0.04
		Level of sigr	nificance		
Interaction $(C \times I)$	n.s.	n.s.	n.s.	n.s.	n.s.

¹The results were obtained after treating hardwood and semi-hardwood with 100 ppm IBA

The least person a mixture of both hardwood and semi-hardwood treated with various concentrations of IBA Mean with identical letters in column do not differ significantly from each another; n.s. = non significant at 5 % level of probability

C × I = interaction of types of cutting and IBA

food materials (carbohydrates) in the cuttings.

NUMBER OF SHOOTS PER CUT-TING

The statistical analysis of the data showed that there is no significant effect of IBA concentration, cutting type and the interaction between IBA concentration and cutting type on number of shoots per cutting (Table 1). SIDDI-QUI and HUSSAIN (2007) observed the same effect of IBA on the number of shoots per cutting in Ficus hawaii. The reason for this may be that there was an equal length of the cutting above the soil surface and hence equal number of nodes.

SHOOT LENGTH (CM)

The data regarding shoot length showed that there is a significant effect of IBA concentration on shoot length, whereas the effect of cutting type and the interaction between IBA concentration and cutting type did not significantly differ. The highest shoot length (21.55 cm) was recorded in plots treated with 300 ppm of IBA while minimum shoot length (4.96 cm) was recorded in control (Table 1). This significant effect of IBA concentration on shoot length is in accordance with that of NAGHMOUCHI et al. (2008). As a growth promoter IBA promotes cell division, which results in early rooting leading towards efficient absorption of mineral nutrient and hence maximizes shoot length.

SHOOT DIAMETER (CM)

The data showed that there is a significant effect of IBA concentration on shoot diameter, whereas the effect of cutting type and the interaction between IBA concentration and cutting type did not significantly differ. The maximum shoot diameter (0.29 cm) was recorded in plots treated with 300 ppm of IBA concentration, followed by 0.26 cm where 200 ppm of IBA was used, whereas the minimum shoot diameter (0.14 cm) was recorded in control (Table 1). The significant effect of IBA on shoot diameter might be due to high number of roots and lea-

ves. These results are in accordance with those of SIDDI-QUI and HUSSAIN (2007).

NUMBER OF LEAVES PER SHOOT

There was a significant effect of IBA concentration on number of leaves per shoot, whereas the effect of cutting type and the interaction between IBA concentration and cutting type did not significantly differ. The maximum leaves per shoot (49.17) were recorded in cuttings where IBA concentration was used at the rate of 300 ppm, whereas the minimum leaves per shoot (9.33) were recorded in control (Table 2). IBA plays a direct role in enhancing root development, which, in turn increases shoot length and hence number of leaves, the result is similar to that of WAHAB et al. (2001). An increase in number of roots due to IBA application may necessitate the increased activity of photosynthesis, transpiration and respiration in leaves and thus may result in the increase in number of leaves per cutting (MUHAMMAD, 1987).

NUMBER OF BRANCHES PER SHOOT

The statistical analysis of the data showed that there is a significant effect of IBA concentrations, cutting type and the interaction between IBA concentration and cutting type on the number of branches per shoot (Table 2). The highest number of branches per shoot (5.33) was observed in response to 300 ppm of IBA, followed by 5.17 in response to 100 ppm of IBA, while the minimum number of branches per shoot (1.00) was recorded in control. The highest number of branches per shoot (4.58) was recorded in hardwood cuttings, while the minimum number of branches per shoot (3.58) was recorded in semi-hardwood cuttings. STEFANCIC et al. (2005) reported that IBA has significantly influenced the number of branches per shoot. It is a fact that IBA directly affects the number of roots and root growth and indirectly affects shoot length, which may result in a high number of branches per shoot. The type of cutting has also shown a significant effect on number of branches per shoot. It may be because the hardwood cuttings of olive contains

Type of cutting	Number of leaves per shoot	Number of branches per shoot	Number of roots per plant	Root length (cm)	Root diameter (cm)
Hardwood ¹	36.58	4.58 a	14.42	8.15	0.12
Semi-hardwood ¹	31.17	3.58 b	10.17	7.36	0.11
Significance level	n.s.	*	n.s.	n.s.	n.s.
IBA conc. $(ppm)^2$					
0	9.33 c	1.00 b	3.33 d	3.09 d	0.07 d
100	32.00 b	5.17 a	8.17 c	7.04 c	0.10 c
200	45.00 a	4.83 a	17.17 b	9.80 b	0.13 b
300	49.17 a	5.33 a	20.50 a	11.09 a	0.16 a
LSD ($\alpha = 0.05$)	4.59	0.51	1.43	0.79	0.02
		Level of sig	nificance		
Interaction ($C \times I$)	n.s.	*	n.s.	n.s.	n.s.

Table 2: Effect of IBA concentrations and type of cuttings on number of leaves per shoot, number of branches per shoot, number of roots per plant, root length (cm) and root diameter (cm) of olive

¹The results were obtained after treating hardwood and semi-hardwood with 100 ppm IBA

²The data represent a mixture of both hardwood and semi-hardwood treated with various concentrations of IBA

Mean with identical letters in column do not differ significantly from each another; n.s. = non significant and * = significant at at 5 % level of probability

 $C \times I$ = interaction of types of cutting and IBA

more food material than the semi-hardwood cuttings.

NUMBER OF ROOTS PER PLANT

Significant variation in number of roots per plant was observed in relation to different IBA concentrations, whereas the effect of cutting type and the interaction between IBA concentration and cutting type did not significantly differ. The maximum roots per plant (20.50) were recorded in cuttings where IBA concentration was used at the rate of 300 ppm, followed by 17.17 where IBA concentration was used at the rate of 200 ppm, whereas the fewest roots per plant (3.33) were recorded in control (Table 2). The induction of maximum number of roots per plant in the cuttings treated with high concentration of IBA was also reported by QADDOURY and AMSSA (2004) and AINI et al. (2009). This effect of IBA may be due to the fact that cambial activities involved in root induction are stimulated by increasing IBA concentration DAOUD et al. (1989).

ROOT LENGTH (CM)

The statistical analysis of the data showed that there is

a significant effect of IBA concentration on root length, whereas the effect of cutting type and the interaction between IBA concentration and cutting type did not significantly differ. The highest root length (11.09 cm) was recorded in plots where IBA concentration was used at the rate of 300 ppm, followed by 9.80 cm where IBA concentration was used at the rate of 200 ppm, whereas the minimum root length (3.09 cm) was recorded in control (Table 2). The increase in root length may be due to the maximum number of branches per shoot whose tips produces more auxin which results in root elongation, and the effect of metabolites translocation and carbohydrates metabolism. BHATT and TOMAR (2010) also found increase in root length in Citrus auriantifolia with increasing IBA concentration.

ROOT DIAMETER (CM)

IBA concentration has significantly affected the root diameter, whereas the effect of cutting type and the interaction between IBA concentration and cutting type did not significantly differ (Table 2). The maximum root diameter (0.16 cm) was recorded in plots treated with 300 ppm of IBA, followed by plots treated with 200 ppm of IBA, whereas the minimum root diameter (0.07 cm)

was recorded in control. Olive cuttings showed a positive response towards the increasing concentration of IBA regarding root diameter. The present findings can be confirmed by SINGH and SINGH (2005), who also reported that IBA has a significant effect on the root diameter compared to all other growth regulators like indole acetic acid and naphthalene acetic acid.

CONCLUSION AND RECOMMENDA-TIONS

Based upon the findings of this study, it is recommended that the cuttings of olives should be treated with IBA at 300 ppm or higher till maximum plant survival percentage, shoot length, shoot diameter, number of leaves per shoot, number of branches per shoot, number of roots per plant, root length and root diameter are achieved. There was no significant difference between using hardwood cuttings and semi-hardwood cuttings except number of branches per shoot.

REFERENCES

- AINI, A.S.N., GOH, B.L. AND RIDZUAN, R. 2009: The effects of different indole-3-butyric acid (IBA) concentrations, two light regimes of in vitro rooting and acclimatization of in vitro teak (Tectona grandis) plantlets. Afr. J. Biotechnol. 8: 6151-6161
- ASHRAFI, Z.Y., SADEGHI, S., MASHHADI, R.H. AND HAS-SAN, A.M. 2008: Allelopathic effects of sunflower (Helianthus annuus) on germination and growth of wild barley (Hordeum spontaneum). J. Agric. Tech 4: 219-229
- BALOCH, A. 1994: Phases of plant growth. Islamabad National Book Foundation, 1994
- Внатт, В.В. AND TOMAR, Y.K. 2010: Effects of IBA on rooting performance of Citrus auriantifolia Swingle (Kagzi-lime) in different growing conditions. Nature Sci. 8(7): 8-11

- CASSON, S.A. AND LINDSEY, K. 2003: Genes and signalling in root development. New Phytologist 158: 11-38
- Снним, Т., Такета, S., Tsurumi, S. and Ichii, M. 2003: The effects of auxin on lateral root initiation and root gravitropism in a lateral rootless mutant Lrt1 of rice (Oryza sativa L.). Plant growth regulation 39: 161-170
- CHHUN, T., TAKETA, S., TSURUMI, S. AND ICHII, M. 2004: Different behaviour of indole-3-acetic acid and indole-3-butyric acid in stimulating lateral root development in rice (Oryza sativa L.). Plant growth regulation 43: 135-143
- DAOUD, D.A., AGHA, K.H., ABU-LEBDA, M.S. AND AL-KHAIAT 1989: Influence of IBA on rooting of leafy olive cutting. Olive 6: 28-30
- LOUKAS, M. AND KRIMBAS, C.B. 1983: History of olive cultivars based on their genetic distances. J. Hortic. Sci. 58: 121-127
- LUDWIG-MULLER, J., VERTOCNIK, A. AND TOWN, C.D. 2005: Analysis of indole-3-butyric acid-induced adventitious root formation on Arabidopsis stem segments. J. exper. bot. 56: 2095-2105
- MARSHALL, K.F. AND WARRING, G. 1985: Effect of number of roots on survival percentage and shoot development in cuttings. Plant Cell Physiol. Culture 16: 563-569
- MITRA, S.K. AND BOSE, T.K. 1990: Fruits, tropical and sub-tropical. - Calcutta: Naya Prokash, 1990
- Muнаммаd, H. 1987: Rooting response of olive cuttings to various concentrations of Indole Butyric Acid. Asian Journal of Agriculture 9: 129-132
- NAGHMOUCHI, S., KHOUJA, M.L., REJEB, M.N. AND BOUSSAID, M. 2008: Effect of growth regulators and explant origin on in vitro propagation of Ceratonia siliqua L. via cuttings. Biotechnol. Agron. Soc. Environ. 12(3): 251-258

- QADDOURY, A. AND AMSSA, M. 2004: Effect of exogenous indole butyric acid on root formation and peroxidase and indole-3-acetic acid oxidase activities and phenolic contents in date Palm off shoots. Bot. Bull. Acad. Sinica 45: 127-131
- RASHOTTE, A.M., POUPART, J., WADDELL, C.S. AND MU-DAY, G.K. 2003: Transport of the two natural auxins, indole-3-butyric acid and indole-3-acetic acid, in Arabidopsis. Plant Physiol. 133: 761-772
- SIDDIQUI, M.I. AND HUSSAIN, S.A. 2007: Effect of indole butyric acid and types of cuttings on root initiation of Ficus hawaii. Sarhad J. Agric. 23(4): 919-925
- SIMMONDS, N.W. 1976: Evolution of crop plants. London: Longman Group Ltd., 1976
- SIMON, S. AND PETRASEK, J. 2011: Why plants need more than one type of auxin. Plant Sci. 180: 454-460

- STEEL, R.G.D., TORRIE, J.H. AND DICKEY, D.A. 1997: Principles and procedures of statistics: A biological approach. New York: McGraw-Hill, 1997
- STEFANCIC, M., STAMPAR, F. AND OSTERC, G. 2005: Influence of IAA and IBA on root development and quality of Prunus 'GiSelA 5' leafy cuttings. HortScience 40: 2052-2055
- WAHAB, F., NABI, G., NAWAB, A.M. AND SHAH, M. 2001: Rooting response of semi-hardwood cuttings of guava (Psidium guajava L.) to various concentrations of different auxins. OnLine J. Biol. Sci. 1: 184-187
- WANG, S., TAKETA, S., ICHII, M., XU, L., XIA, K. AND ZHOU, X. 2003: Lateral root formation in rice (Oryza sativa L.): differential effects of indole-3-acetic acid and indole-3-butyric acid. Plant growth regulation 41: 41-47

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