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EFFECT OF WOUNDING, AUXIN TYPE AND CONCENTRATION ON ROOTING OF LEMON VERBENA (*Aloysia triphylla* (L'HER.) BRITTON) PLANT

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ABSTRACT

Aloysia triphylla (L'Her.) Britton (Verbenaceae) is a deciduous shrub known as lemon verbena and is an important medicinal and aromatic plant. The experiments were carried out at the Farm of the Medicinal and Aromatic Plants, and Laboratory of Agric. Botany Dept., Fac. Agric., Mansoura Univ. during the seasons of 2006, 2007 and 2008. The experiments aimed to study the effect of wounding and auxin type and concentration on rooting of *Aloysia triphylla* plant. Anatomical changes during the formation and emergence of adventitious roots were performed in addition to determination of endogenous IAA. The results showed that both IBA and NAA increased rooting percentage and number and length of roots of the cutting. The effects of both growth regulators depended on the concentration used. Best results for number and length of roots per cutting were achieved using 2000 ppm IBA and 1000 ppm NAA. Wounding of the cuttings in addition to the use of auxins improved rooting of the cuttings, and the best rooting resulted from wounding the cutting plus treating them with 2000 ppm IBA and 1000 ppm NAA. Anatomical studies showed that narrow phloem tissue, cambial zone and vascular rays beside badly and slowly formation of the vascular connection between the new roots and the vascular tissues of the cuttings seemed to be mostly responsible for difficult to root cuttings. Results showed also that number of roots per cutting, and root length were increased with increasing endogenous IAA content. This increase correlated with auxin treatments, auxin type and their concentrations.

INTRODUCTION

Aloysia triphylla (L'Her.) Britton belongs to Family Verbenaceae is an aromatic shrub formerly classified as *Aloysia citriodora* (Ort.), *Lippia citriodora* (Ort.) HBK., *Verbena citriodora* and *Verbena triphylla* L'Her. Leaves collected before flowering contain high quality essential oil of lemon scent, and its main constituents are limonene, spathulenol, geranial, citral, carvone, nerol, geraniol, borneol, terpineol and linalool, (Figueiredo *et al*., 2004). The essential oil of lemon verbena is used in perfumery, in medicine as an antifungal, antipyretic, sedative, antineuralgic, and antispasmodic, diuretic, diarrhea, stomachic, asthma and fever, and the leaves are also used in culinary to add flavor to food and beverages, (Keville, 1999).

Rooting of stem cuttings is an important horticultural tool for the propagation of ornamental shrubs. Factors affecting the rooting of cuttings can include application of auxins, seasonal timing at which cuttings are collected, type of cuttings, wounding, physiological and histological factors as well as an imbalance of endogenous phytohormones. Synthetic auxins are known to stimulate adventitious root formation on cuttings. The cuttings response to auxins treatments depends on the type of cutting, the auxin type and its concentration used as well as its formulations (Hartmann *et al*., 2007). IBA is the most widely used among the root promoting auxinic compounds because it is non-toxic and effective over a wide range of species (Couvillon, 1988). In spite, with few deciduous ornamental shrubs, hardwood cuttings which are prepared during the winter or early spring rooted best than other cuttings type, (Sari and Qrunfleh 1995). Al-Salem and Karam (2001) on *Arbutus andrachin* cutting found that wounding by making two opposite longitudinal incisions at the base of the cutting increased rooting percentage and root number, length, and fresh and dry weights in basal cuttings treated with IBA. Ibrahim (1989) found that application of IBA at 150 and 250 ppm significantly increased the rooting percentage of *Aloysia triphylla (*L'Herit) Britton, and the middle cuttings treated with IBA at 250 ppm in talc powder gave the best results.

Stefanini *et al.* (2004) reported that treatment with 250 mg IBA / l + boric acid of stem cuttings of *Aloysia triphylla (*L'Herit) gave the highest number of roots, longest roots, highest rooting percentage, heaviest fresh and dry weights of roots.

Hartmann *et al.* (2007) concluded that the origin of adventitious roots from callus tissue and out cut surface has been associated with difficult –to-root species. Other research workers reported that the stimulatory effect of synthetic auxins compounds on rooting ability was correlated with increasing endogenous IAA content (Pluss and Meier, 1989).

The aim of the search was to study the effect of wounding, auxin type and concentration on rooting of *Aloysia triphylla* (L'Her.) Britton hardwood stem cutting.

MATERIALS AND METHODS

Rooting experiments were carried out at the Farm of the Experimental Station of Medicinal and Aromatic Plants, as well as Laboratory of Agric. Botany Dept., Fac. Agric., Mansoura Univ. during the growing seasons of 2006, 2007 and 2008.

The experiments were carried out as follows :

The first growing season (2006):

Cuttings (15 cm long) were prepared from the inter medial part of 2 years old hardwood branches in February. The cutting base was dipped into a distilled water as control and 250, 500, 1000, 2000 and 4000 ppm IBA alcoholic solution (50 % ethanol) for 5 seconds.

The second growing season (2007):

Hardwood cuttings (15 cm long) at the end of February, considering the results of the first growing season, were prepared to study the effect of two types of auxins either IBA or NAA at 1000 and 2000 ppm as well as wounding on rooting of lemon verbena stem cuttings. Concerning the wounding treatment, the basal cuttings were wounded before dipping in an alcoholic solution of either IBA or NAA by using one longitudinal deep incision

1 cm long penetrating the wood. The bases of cuttings were dipped into an alcoholic solution of both auxins for 5 seconds.

The third growing seasons (2008):

Rooting experiments were carried out at the end of February by using hardwood cuttings treated with the same treatments in the second growing season.

All experiments in the growing seasons were laid out in a complete randomized design with three replicates, using 20 cuttings for each replicate. At the end of the experiments (two months after planting), the percentage of rooted cuttings, average length of roots (cm) were measured and the average roots and shoots per rooted cutting were counted .

Anatomical studies :

In the third season (2008), the anatomical studies were carried out , one cm long , samples were excised from the base of cuttings 2 and 4 weeks after planting and at the end of the experiments. The samples were immediately fixed in FAA, 70 % alcohol solution, dehydrated in alcohol series followed by xiline for clearing and embedded in a paraffin wax (52-54 ̊C m.p.). Cross and longitudinal sections 20µm were prepared by rotary microne stained in saffranin –light-green combination and mounted in Canada balsam (Geriach, 1977). The sections were photographed.

Determination of endogenous auxins :

Determination of endogenous auxins was carried out during only the last growing season at the Principal Central Lab., Fac. Agric., Cairo University. Fresh samples from the rooted cuttings were taken to determine indole- acetic acid (IAA) at the end of the experiment. In this respect, 10 g of fresh rooted cuttings were weighed and ground in a cold mixture of chlorophorm and methanol 60 : 140 (v:v). The combined acidic ethyl acetate phase was reduced in volume to be used for Gas liquid chromatography determination of acidic hormones, (Addicott *et al*., 1964). Gas liquid chromatography (GLC) - Mass Spectrometry (MS) has been used for further identification of total indoles (Gaskin *et al.*, 1973).

RESULTS AND DISCUSSION

The effects of different concentrations of IBA on rooting of *Aloysia triphylla* L. two years old stem cuttings are shown in Table (1). Treating hardwood cuttings with IBA at different concentrations increased the percentage of rooting, and number and length of roots (cm) per rooted cutting, as well as average number of shoots per rooted cutting. The effects of IBA on rooting depended on the concentrations used; the best rooting (50 %) was achieved when hardwood cuttings collected during February were treated with IBA at 2000 ppm. In spite of some success of rooting obtained, lemon verbena plant was still difficult to root.

The use of higher concentration of IBA (4000 ppm) decreased rooting, which could be attributed to the toxicity of the high IBA concentration that inhibits growth (Pierick, 1986) and causes reduction in number of root primordia (Carpenter and Cornell, 1992).

Accumulation of reserve food material especially carbohydrate is necessary for root formation and its growth (Hartmann *et al.,* 2007). The production of auxins with break of bud dormancy is correlated with an increase of the starch mobilization to the base of cuttings caused by enhanced activity of hydrolytic enzymes (Nanda and Anand, 1970). Successful rooting with hardwood cuttings collected in February could be attributed to an increase of endogenous auxins content, decreased ABA due to break of bud dormancy (Kweon and Sun, 1996) and renovation of vegetative growth.

Table (1): Effect of different concentrations of IBA on rooting of *Aloysia triphylla* (L'Her.) Britton cuttings during the growing season of 2006.

IBA treatments (ppm)	Rooting (%)	Average root No.	Average root length (cm)	Average shoots No.
Control	10	3	0.5	1
250	15	5	2	1
500	28	7	2.5	2
1000	36	10	4.2	2
2000	50	15	8.2	3
4000	28	8	3.2	2
L.S.D at 5 %	**8.31**	**2.82**	**1.02**	**1.05**

Results in Table (2) indicated that cuttings treated with either IBA or NAA at all concentrations (with or without wounding) increased the percentage of rooting, the average number and length (cm) of roots per rooted cutting. Generally, IBA treatment was more effective on rooting than NAA treatment in all cases.

The effects of either IBA or NAA on rooting depended on the concentration used. IBA at 2000 ppm was more effective on promoting root formation than other concentrations used. In addition, best rooting was achieved with wounded cuttings treated with IBA at 2000 ppm.

The stimulatory effect of auxins on adventitious root formation may be attributed to the auxin, which causes cell elongation, swelling of tissues, cell division, development of root primordia and formation of adventitious roots (Pierick, 1986). Mato and Vietez (1986) pointed out, that IBA treatments may control endogenous auxin levels of cutting either through direct regulation of IAA oxidase system or indirectly through the transport of auxin protectors.

In addition, the effect of IBA may be due largely to its accumulation at the base of cuttings to a suitable level for initiation and development of roots (Thomas, 1982). The promotive effect of IBA on rooting could be partially due to the reduction of ABA (absicic acid) content especially soon after treatment (Sawky *et al.,* 1988).

Mackenzie *et al.* (1988) pointed out, that wounded tissues may stimulated cell division and production of root primordia. This is due to a natural accumulation of auxins and carbohydrates in wounded area (Hartmann *et al.,* 2007). The effect of wounding to promote rooting could also be due to the fact that wounding increases the penetration of IBA into the deep-seated tissue of phloem parenchyma and so facilitates root formation (Howard, 1986).

Table (2) : Effect of wounding and different concentrations of IBA and NAA on rooting of *Aloysia triphylla*(L'Her.) Britton hardwood cuttings during the growing seasons of 2007 and 2008.

Treatments	Season 2007			Season 2008		
	Rooting (%)	Average root No./ Cutting	Average root length/ cutting (cm)	Rooting (%)	Average root No./ cutting	Average root length/ cutting (cm)
Control	11.0	03.0	0.5	10.00	04.0	0.6
1000 ppm IBA	50.0	11.0	5.0	52.00	13.0	7.0
W+1000 ppm IBA	61.0	14.0	7.0	58.00	16.0	9.0
2000 ppm IBA	65.0	17.0	9.0	63.00	19.0	13.3
W+ 2000 ppm IBA	81.0	24.0	12.5	80.00	23.0	17.0
1000 ppm NAA	45.0	10.0	4.0	46.00	12.0	5.0
W+1000 ppm NAA	48.0	12.0	4.5	50.00	15.0	6.0
2000 ppm NAA	30.0	08.0	3.3	33.00	10.0	4.0
W+2000 ppm NAA	15.0	05.0	2.0	20.00	06.0	2.0
L.S.D at 5 %	**6.3**	**2.0**	**3.2**	**6.8**	**2.3**	**2.5**

W=wounding

1- Anatomical changes in the cuttings during the rooting process :

The stem structure of *Aloysia triphylla* resembled the description written by Gattuso *et al.* (2008).

The outer most cell layer constitutes the epidermis, then follows the cortex tissue, its outer most region at the ribs consists of 2-4 cell layers of angular collenchyma and inner region contains more parenchyma cell layers. Vascular bundles are open collaterally and arranged in a complete cylinder; it reinforced by a discontinuous ring of phloem fibers.

Figure (1) shows a cross section of stem cutting taken from two-year-old shoots at the starting of rooting experiments. The most remarkable anatomical feature in the basal portion of cuttings is the early formation of both periderm and secondary vascular tissues. The early secondary growth takes place in the stem due to the activity of vascular cambium.

The stem has narrow phloem tissue, cambial zone, phloem and xylem rays, so that; the vascular bundles became compactly arranged in the vascular cylinder.

Two weeks after planting, no visible changes were found in the examined section of un-treated cuttings (Fig., 2).

Fig. (1) : Cross section of two years old stem cutting of *Aloysia triphylla* at the start of the experiment.
Note : early formation of secondary growth and perideried tissue (Per)
(obj. x10 oc. x10).

Per : Periderm
Co : Cortix
Ph : Phloem
CZ : Cambial zone
SX : Secondary xylem
PF : Phloem fibers

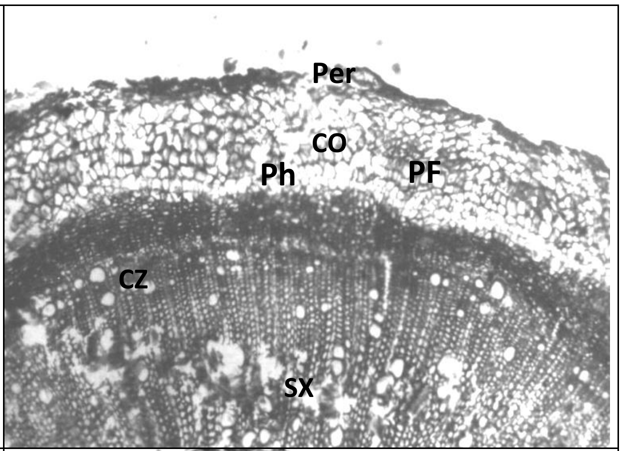
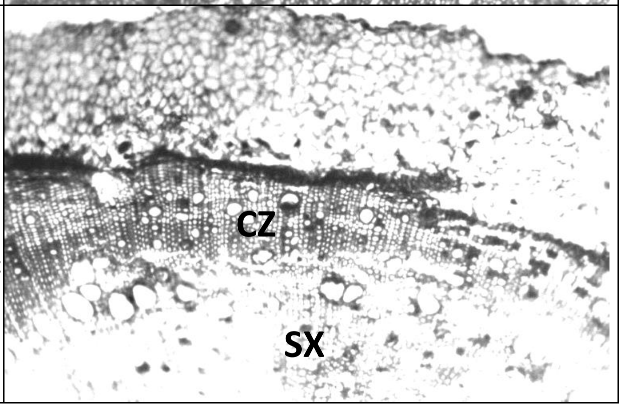


Fig. (2) : Cross section through the base of untreated cutting, 15 days after planting.

Note : no visible anatomical changes compared with the section taken at the start of the experiment (obj. x10 oc. x10).

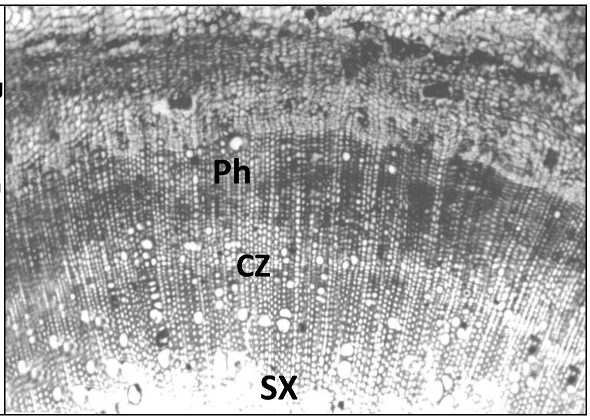
SX : Secondary xylem
CZ : Cambial zone



However, sections taken from the basal portion of treated cuttings with IBA two weeks after planting showed that there was a great proliferation and extension of phloem tissue, cambial zone and vascular rays (Fig., 3) In addition, formation of some meristimatic cells with dense cytoplasm was observed as well as initiation of some root primordia from cambial zone.

Fig. (3) : Cross section through the base of cutting affected by IBA at 2000 ppm, 15 days after planting showing great proliferation and extension of phloem tissue and cambial zone.
(obj. x10 oc. x10).

Ph : Phloem
CZ : Cambial zone
SX : Secondary xylem



It also shows crushed cells of the epidermis, cortex and phloem tissues due to increasing the pressure of secondary tissue. Growth and passage of the root primordia (Fig., 4) seemed to include an increase in the activity of hydrolytic enzymes (White and Lovell 1984). The cambial zone (Fig., 5) and xylem rays (Fig., 6) were the main tissues which developed the adventitious roots.

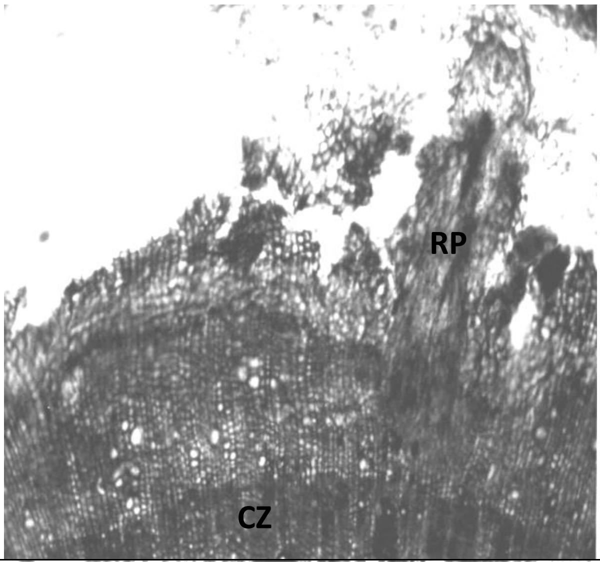


fig. (4) : Cross section through the base of cutting affected by IBA at 2000 ppm, 15 days after planting showing initiation of root primordia from cambial zone (obj. x10 oc. x10).

RP : Root primordia
CZ : Cambial zone

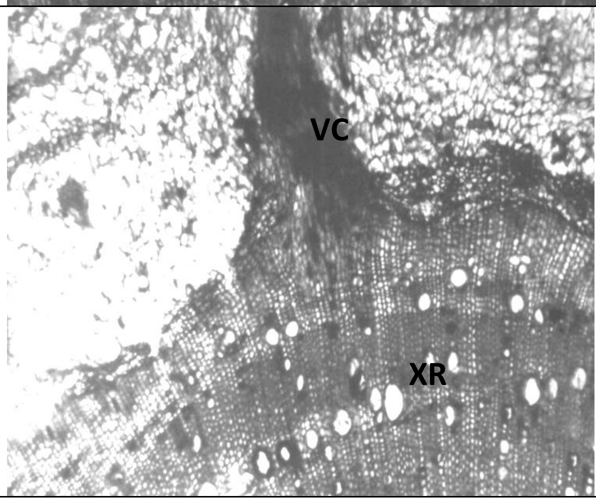


Fig. (5) : Cross section through the base of treated cutting with IBA at 2000 ppm showing development of root primordia from xylem rays. (obj. x10 oc. x10).

VC : Vascular connection

XR : Xylem rays

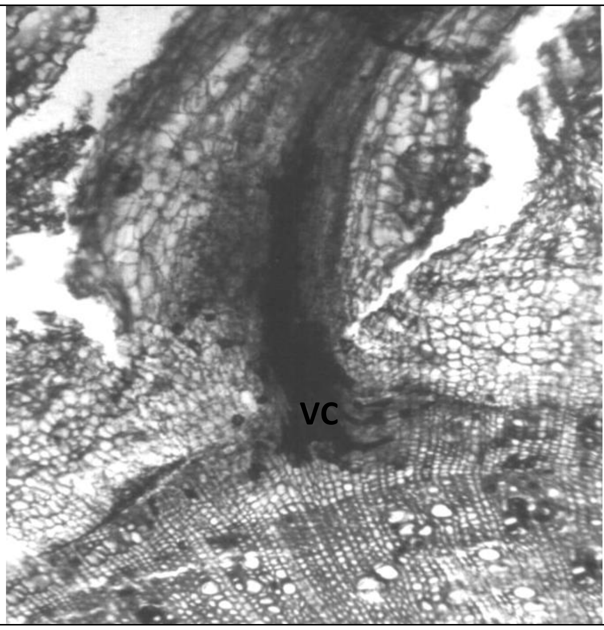


Fig. (6) : Cross section of untreated cutting, 4 weeks after planting.

Note : bad development of the vascular connection between the cutting and emerging root (obj. x10 oc. x10).

VC : Vascular connection

Four weeks after planting both untreated (Fig., 6) and treated cuttings with IBA (Fig., 7) showed some rooting. The vascular connection in untreated cuttings was not as good compared with treated ones. This phenomena may be attributed to formation of tylosis in the xylem vessel that blocked the translocation through the xylem vessels. Moreover, vascular transition tissues were also clearly developed in the growing roots of treated cuttings with IBA at 2000 ppm (Fig., 7). In addition, some roots were developed near the cut surface of cuttings (Fig., 8).

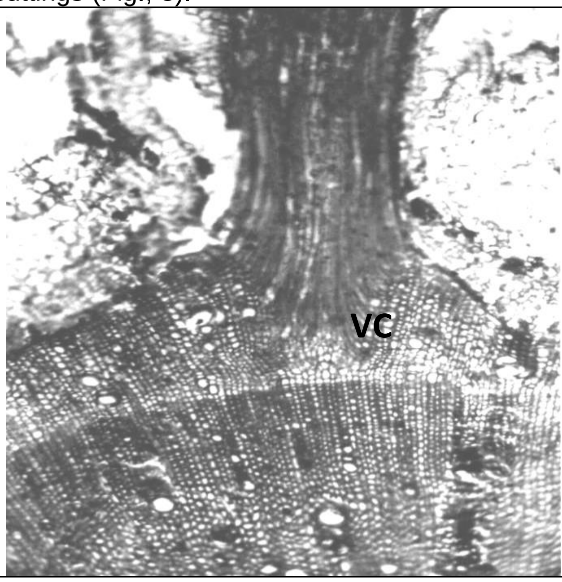


Fig. (7) : Cross section of treated cutting with IBA at 2000 ppm, 4 weeks after planting showing a good developed vascular connection between the cutting and emerging root (obj. x10 oc. x10).

VC : Vascular connection

Fig. (8) : Longitudinal section through the basal portion of rooted cutting, 4 weeks after planting showing some of the adventitious roots developed near the cut surface of cuttings and some root primordia initiated from cambial zone (obj. x10 oc. x10).

RP : Root primordia

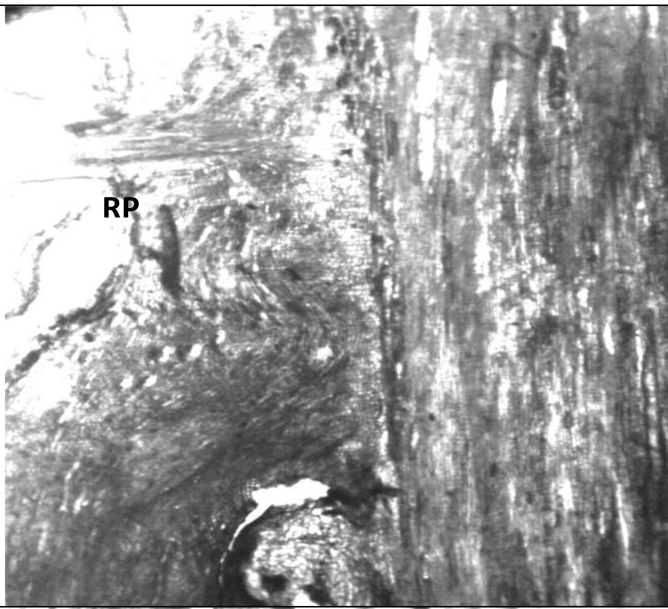
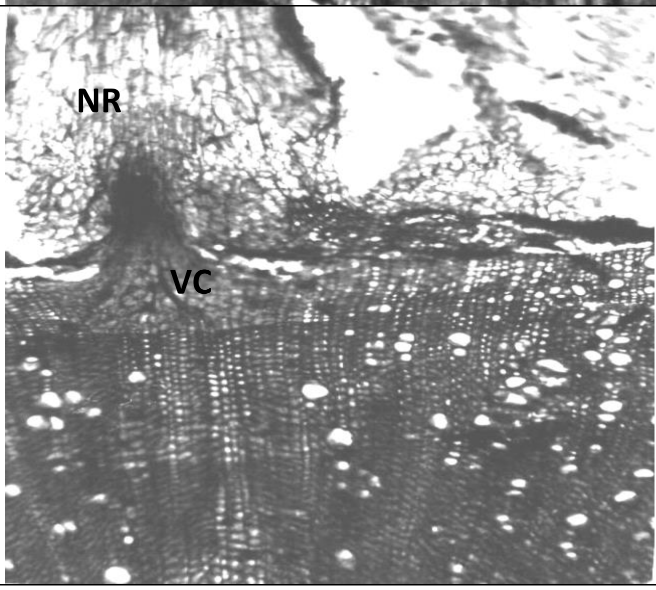


Fig. (9) : Cross section of untreated cuttings, 4 weeks after planting.

Note : weak and bad developed of vascular connection between the cuttings and emerging roots (obj. x10 oc.x10).

NR : New root
VC : Vascular connection



These observations indicate that *Aloysia triphylla* cuttings are difficult-to-root. In this respect, Hartmann *et al.* (2007) noted that, origin of adventitious roots from callus tissue or at the cut surface of cuttings has been associated with difficult-to-root species.

Formation of vascular connections plays an important role in the successful rooting of lemon verbena. The results indicated that the new adventitious roots were formed in cuttings then died. This phenomenon may be due to weak vascular connection between the roots and vascular tissues of the cuttings as shown in Fig., (9) compared with Fig., (7).

The stimulatory effect of IBA on the adventitious root formation may be attributed to its effect on cell elongation, swelling of tissues, cell division, development of root primordia and formation of adventitious roots (Pierick, 1986 and Hussein, 2008). Moreover, IBA treatment increased vascular cambial activity which produced histological events leading to root initiation and root primordia formation (Davies *et al*., 1982).

Mato and Vietez (1986) pointed out, that IBA treatments may control endogenous auxin levels of cutting, and Hartmann and Kester (1968) reported that IBA is the best rooting promoter due to its fast auxin activity and slow destruction by enzymatic system.

2- Anatomical changes after wounding and treating cuttings with IBA :

In the basal and sub-basal regions of cuttings other anatomical changes took place in response to wounding.

Sections taken from the basal and sub-basal regions of cuttings showed significant anatomical differences compared with those at the beginning of the experiment (Fig., 1).

Most of the anatomical changes occurred in treated cuttings on the wound area and on the opposite side of wound area. The deep incision penetrated to the wood and callus formation was started (Fig., 10), and heavy callus at the wound area on either side of the split was developed.

Callus is produced by cortex tissue and cambial zone. Later, callus expanded in two opposing directions and gradually occupied the gap between the separate halves of the woody cylinder (Fig., 11) as well as proliferation of phloem tissue and cambial zone (Fig., 12).

Meantime, some root primordia developed on the opposite site of the wound area (Fig., 13). Moreover, the root primordia emergence and growth outward were observed through other stem tissue. The emergence and growth of root primordia caused crush of the outer tissues (Figs., 13 & 14).

Four weeks after planting, the adventitious roots emerging from the cutting as well as vascular connection between the main stem and the emerging roots were observed in the growing roots (Fig., 15). This connection ensured the translocation of nutrients and hormonal factors to the growing roots.

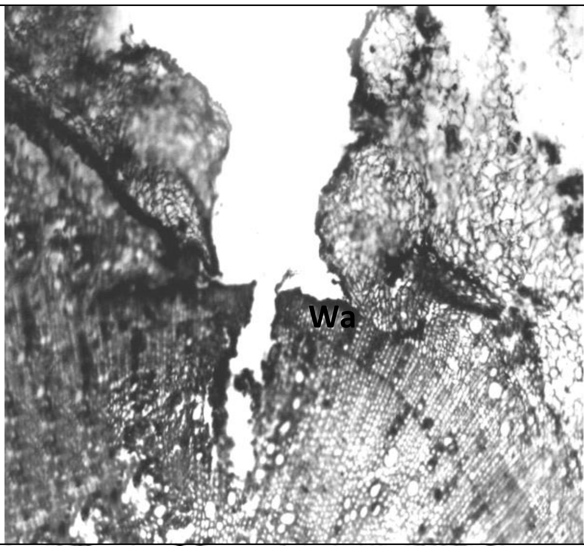


Fig. (10) : Cross section through the basal portion of wounded cutting at the start of the experiment showing the deep incision penetrated to the wood (obj. x10 oc. x10).

W : Wound area

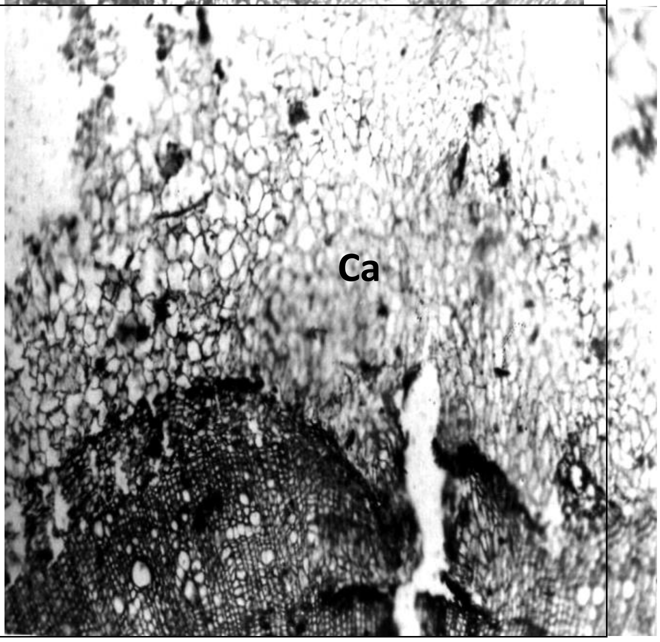


Fig. (11) : Cross section at the wound area of treated cutting two weeks after planting.

Note : developed heavy callus tissue which expanded in two opposing direction and gradually occupies the wound area (obj. x10 oc. x10).

Ca : Callus

Fig. (12) : Cross section of the opposite site of the wound area showing great proliferation of phloem tissue and cambial zone (obj. x10 oc. x10).

Ph : Phloem
CZ : Cambial zone

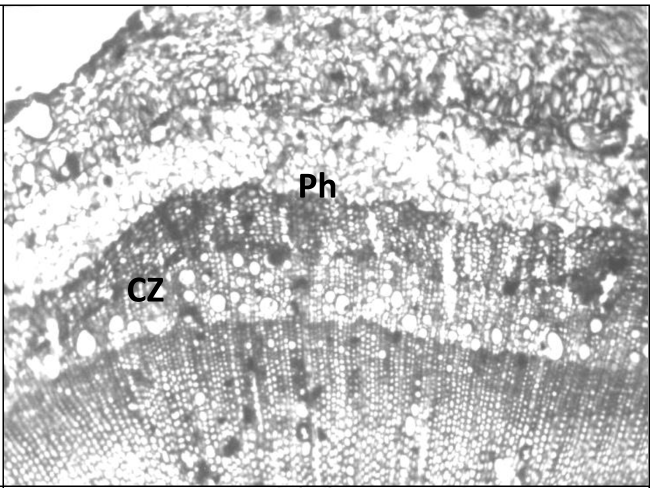


Fig. (13) : Cross section in the basal portion of wounded cutting showing the deep incision and the opposite site

(obj. x10 oc. x10).

WA : Wound area
RP : Root primordial

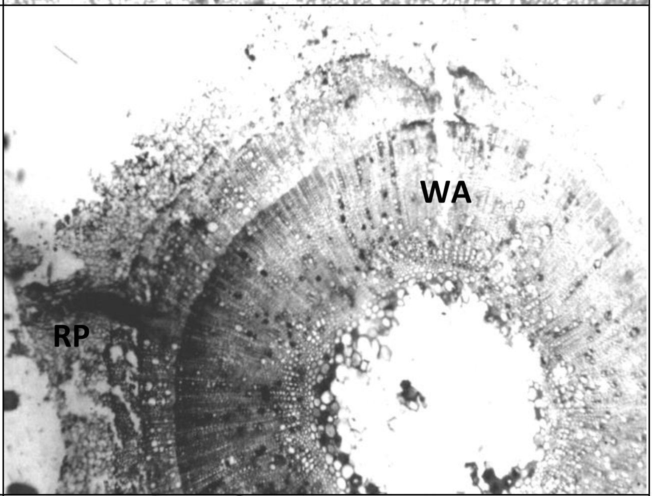
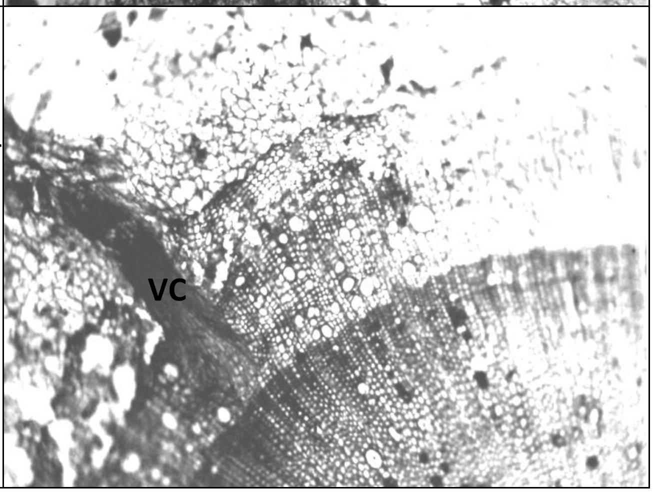


Fig. (14) : Cross section at the opposite site of wound area, 2 weeks after planting.

Note : development of root promerdia and vascular connection is started in the initiated root primordia (obj. x10 oc. x10).

VC : Vascular connection



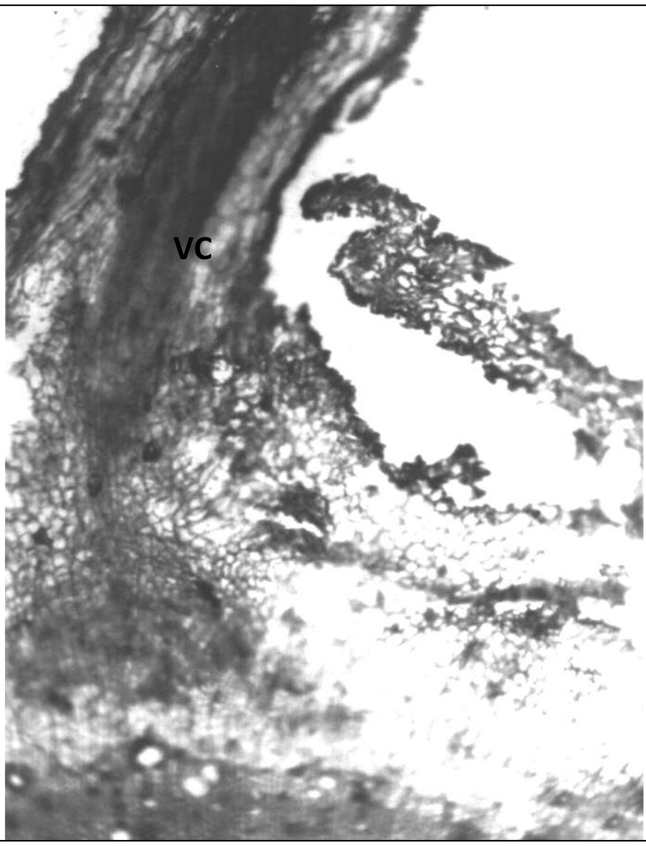


Fig. (15): Cross section of wounded stem cutting treated with IBA at 2000 ppm, 4 weeks after planting showing developed formation of the vascular connection between the cutting and emerging root (obj. x10 oc. x10).

VC : Vascular connection

Generally, the wounded cuttings treated with IBA showed early formation of both root primordia and vascular connection between the cutting tissue and the root primordia. Moreover, the roots developed faster and more abundantly than either treated with IBA alone or control ones.

The promoting effects of wounding and its interaction with IBA on rooting may be attributed to stimulation of cell division, production of root primordia (Hartmann and Kester, 1975). This is due to greater absorption of water and applied growth regulators by tissues at the base of the cuttings as well as natural accumulation of auxins and carbohydrates in the wounded area (Hartmann *et al*., 2007). Raviv and Reuveni (1984) reported that starch accumulation at the base of avocado cuttings was highly correlated with the number of rooted cuttings. Moreover, in some plant species, wounding seems to stimulate root development and penetration through a sclerenchymatic rings which form a mechanical barrier against the developing root primordia (Shawky *et al,*.1988).

It seems that *Aloysia triphylla* cuttings were difficult-to-root due to narrow cambial zone, xylem and phloem rays. These anatomical parameters showed a very strong influence on the process of root formation. In addition, wounding was very useful in promoting the rooting process of *Aloysia triphylla* stem cuttings especially when followed by IBA treatments since rooting developed faster, and the roots were more abundant and longer than either untreated treatments (control) or those treated with IBA alone.

Relationship between rooting ability and endogenous indole-3-acetic acid (IAA) :

The relationship between rooting ability and root characters of lemon verbena and endogenous IAA is shown in Table (3).

It is clear that rooting ability, number of roots per rooted cutting, and root length were increased with increasing endogenous IAA content. This increase correlated with auxin treatments, auxin type and their concentrations.

The wounded cuttings treated with IBA at 2000 ppm gave the best rooting and highest level of endogenous IAA content. A positive correlation was reported between endogenous IAA content and rooting ability of cuttings (Shin *et al*., 1988 and Sagee *et al*., 1992). Kweon and Sun (1996), reported also that endogenous IAA promoted both root initiation during the early stage and root primordia as well as increased number of cells per root primordium. The auxins can come from buds or young leaves or may be applied exogenously (Pluss and Meier, 1989).

Table (3) : Effect of wounding and different concentrations of IBA and NAA on rooting of *Aloysia triphylla* hardwood cuttings during the growing season of 2008.

Treatments	Rooting (%)	Avg. root No/cutting	Avg. root length/cutting (cm)	*IAA (mg/1oo g fresh weight)
Control	10.00	4.00	0.60	0.20
1000 ppm IBA	52.00	13.00	7.00	0.50
W+1000 ppm IBA	58.00	16.00	9.00	0.55
2000 ppm IBA	63.00	19.00	13.30	1.25
W+ 2000 ppm IBA	80.00	23.00	17.00	2.75
1000 ppm NAA	46.00	12.00	5.00	1.10
W+1000 ppm NAA	50.00	15.00	6.00	1.25
2000 ppm NAA	33.00	10.00	4.00	0.31
W +2000 ppm NAA	20.00	6.00	2.00	0.25
L.S.D. (5%)	6.82	2.27	2.47	

*** IAA determination was not replicated.**

Webster and Van Hof (1970) mentioned that the promotive effect of auxin on initiation and development of adventitious roots may be attributed to that auxin can enhance DNA as well as RNA and protein synthesis which are prerequisites for cell division. In addition, an increase in endogenous auxin content due to both IBA and NAA treatments may be attributed to their effects on the control of endogenous auxin production through, either direct regulation of IAA oxidase system or the transport of auxin protectors (Mato and Vietiez, 1986).

Although, these results support the suggestion that high levels of endogenous auxins are associated with promotion of root formation on cuttings, adventitious root formation is regulated by complex interactions between endogenous factors which affect the various developmental stages of root formation, among of them endogenous auxin content in cuttings.

It could be concluded that **l**emon verbena hardwood cuttings should be collected during February and treated with wounding before dipping in either IBA 2000 or NAA 1000 ppm in order to produce highest rooting parameters. Two anatomical factors seemed to be the most responsible for difficult to root cuttings ; narrow phloem tissue, cambial zone and vascular rays in addition to badly and slowly formation of the vascular connection between the new roots and the vascular tissues of the cutting.

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تأثير التجريح ونوع وتركيز الأوكسين على تجذير نبات اللويزا

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اللويزا شجيرة متساقطة الأوراق تعرف باسم الفربينا الليمونية تنتمى الى العائلة الفربينية ، وهي نبات طبي وعطري هام . أجريت هذه التجارب في مزرعة النباتات الطبية والعطرية ومعمل قسم النبات الزراعي بكلية الزراعة جامعة المنصورة خلال أعوام ٢٠٠٦ ، ٢٠٠٧ ، ٢٠٠٨ ، بهدف دراسة تأثير التجريح ونوع وتركيز الأكسين على تجذير العقل الخشبية لنبات اللويزا. وقد تم دراسة التغيرات التشريحية خلال تكوين وخروج الجذور العرضية بالإضافة إلى تقدير المحتوى الداخلي من اندول حمض الخليك. وأظهرت النتائج أن المعاملة بكل من اندول حمض البيوتريك و نفثالين حمض الخليك بجميع التركيزات المستخدمة أدت الى زيادة النسبة المئوية للتجذيروعدد الجذوروطولها للعقلة واعتمد تأثير كلا منظمي النمو على التركيز المستخدم . و تم الحصول على أفضل النتائج لكل من النسبة المئوية للتجذيروعدد الجذوروطولها للعقلة عند استخدام اندول حمض البيوتريك بتركيز ٢٠٠٠ جزء فى المليون و نفثالين حمض الخليك بتركيز ١٠٠٠ جزء فى المليون . كما أدى تجريح العقل بالإضافة إلي إستخدام الاكسينات الى تحسين تجذير العقل ، ونتج أفضل تجذير من تجريح العقل بالإضافة إلى معاملة العقل باندول حمض البيوتريك تركيز ٢٠٠٠ جزء في المليون أو نفثالين حمض الخليك بتركيز ١٠٠٠ جزء في المليون. وأظهرت الدراسات التشريحية أن ضيق خلايا اللحاء ومنطقة الكمبيوم والأشعة الوعائية بالإضافة إلى سوء وبطء تكوين التوصيل الوعائي بين الجذور الحديثة والأنسجة الوعائية غالبا ما تكون مسئولة عن صعوبة تجذير العقل ، وأظهرت النتائج أيضا أن عدد الجذور وطولها يزيد بزيادة المحتوى الداخلي من أندول حمض الخليك ، وأن هذه الزيادة ترتبط بالمعاملة بالأكسين ونوعه وتركيزه.

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