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Plant Growth Regulators and Rooting Media: A Viable Approach for Growth and Performance of Citrus

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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Review Article

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ABSTRACT

Plant growth regulators accompanied with appropriate growth medium can boost up the growth and performance of plants to a greater extent. PGRs such as auxin, gibberellin and cytokine play an exaggerative role in growth and development of the citrus. Moreover, the study reflects a clear overview of the impact of PGRs and different growth media to encourage rooting percentage, number of leaves, survival percentage, sprouting time, and root length of citrus cuttings. Citrus is a versatile fruit crop having a huge number of varieties, cultivars, and species; comes in different colours and flavours. Although, there are different methods used in the multiplication of citrus plant which includes budding, grafting, cutting, air layering, etc. However, propagation by cutting technique is the most commercial and farmer's friendly approach. Different elements such as concentration of PGRs, age of plants and environmental factors are responsible for the successful growth of the citrus cuttings. Naphthalene acetic acid (NAA) and indole-3-butyric acid (IBA) are the two types of auxin, which are most commonly utilized in citrus propagation. Rooting medium plays

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a crucial role in the supply the minerals and availability of water in citrus. An absolute rooting medium must have more enough nutrients and water holding capacity with better drainage as well as sufficient air spores for excellent gaseous exchange that fruitfully influence the rooting ability and also have a profound effect on growth and development in citrus cutting. The best time for cutting of citrus stems is in between the July and August.

Keywords: Plant growth regulators; growing medium; citrus cutting.

1. INTRODUCTION

The fundamental elements such as sunlight, water, nutrients, O_2, and CO_2 are necessary for the growth and development for the plants. However, some chemical substances significantly promote the growth of the plant to a great extent, which known as the plant growth regulators often called as phytohormones [16]. Plant growth regulators are further divided into two major groups which including growth promoters and growth inhibitors. Some common plant growth promoters include auxin, gibberellin, and cytokine [57]. These substances are mainly responsible for the growth of plant shoots, leaves, roots, etc. On the other hand, the growth inhibitors hinder the growth of the plants along with encouraging dormancy in seeds. Some common plant growth inhibitors are abscisic acid and ethylene. In the case of the Citrus plant, it consists of a wide range of varieties, species, and hybrids. The best method of propagating these plants is vegetative or asexual propagation methods, especially the cutting method [44]. However, plant growth regulators in case of citrus cutting play a great role which introducing faster growth of the plant such as roots and shoots [47]. Besides this, the rooting media also has a great influence on the growth of citrus cuttings. However, the study demonstrates the concept of plant growth regulators especially auxin and rooting media along with their significant effects in the growth of citrus cuttings. Further, the discussion portion includes the role of auxins in citrus plant growth, the impact of growth medium to enhance the performance of citrus cuttings, and the evaluation of different physiological parameters in citrus cutting and the importance of plant growth regulators (PGRs) and rooting media to increase of the citrus cutting growth and development.

2. CRITICAL ANALYSIS OF THE EFFECTS OF AUXIN ON THE GROWTH AND PERFORMANCE OF CITRUS CUTTING

There are different types of plant growth regulators used for growing citrus cuttings but among all, auxin has been proven as an effective growth regulator to encourage the growth of different plant parts including roots, shoots, leaves, etc [10]. According to different studies, Indole-3-butyric acid (IBA) and Naphthalene acetic Acid (NAA) are the most common forms of auxins used for plant growth for stem cuttings [39]. Both forms of auxins in different concentrations can show different impacts on plant growth. Therefore, the application of appropriate concentration is essential to get desirable growth of different parts of the plant.

2.1 Effects on Rooting Performance

According to Ruchitha and Poojashree, (2021) the exogenous application of auxin can enhance the vegetative growth of different fruit crops to a great extent. The study also includes the importance of propagating citrus by cutting, air layering, or micropropagation application of such PGRs can create a great effect. Especially in the cutting method of plant propagation Indole-3-butyric acid proved as an effective solution. However, some cultivators also apply Naphthalene acetic Acid as PGR which also shows effective results [49]. Accordingly, Harshita and Singh, (2022) stated that the hormones are the key factors that ensure the ultimate growth of the plants, especially the IBA. In the case of root formation of citrus cuttings, it plays a huge role. However, the study illustrates different varieties and cultivars of citrus and their growth response in different recommended doses of IBA and NAA. IBA was recommended for most of the citrus varieties for observing maximum root formation [25]. Budiarto *et al.,* (2019) stated a experiment, which is about the root of citrus plants and the particular growth pattern of the plant. Different factors are responsible for the growth of citrus plants. From the study, individuals can understand about different aspects that influence the root and shoot growth of citrus. However, plant growth regulators play a significant role in the growth of different parts of citrus plants. The effects of root growth patterns can further encourage the application of appropriate doses of such plant

growth regulators to citrus cuttings [9]. Al-Jubori *et al.,* (2022) stated the application of IAA and IBA auxin and cytokinin to encourage the vegetative multiplication of *Citrus aurantifolia.* It also includes the growth of average shoot number and rooting percentage of citrus cuttings. However, different concentrations of these PGRs (IAA and IBA) are essential for the successful growth of the plant parts [3].

2.2 Effect of Auxin on the Number of Leaves

According to the study of Patel *et al.,* (2021), the influence of IBA and NAA at various doses to encourage the growth of plant leaves especially focusing on cuttings of Kagzi lime. Based on the experiment result, cuttings show more effective growth in the combination of IBA and NAA plant growth regulators rather than when applied individually. However, PGR @ 3000 ppm of IBA + 3000 ppm of NAA proved to be effective for growing citrus leaves [43]. On the other hand, Das *et al.,* (2021) demonstrated the application of gibberellin and NAA for the vegetative growth of *Citrus limon.* They conducted a total of eight experiments by applying different doses of gibberellin and NAA. However, the citrus plants in which NAA was applied at a rate of 50 ppm showed the most effective vegetative growth. Therefore, it can be said that NAA, another form of auxin, has a great role in growing leaves of a plant along with growing shoots [12].

Gautam *et al.,* (2022) illustrated that the different concentrations of PGRs has impart the direct impact on the shoot growth and number of leaves of *Citrus limon* Burm and it was observed that the shoot growth and number of leaves was maximum at the combined dose of 2000 ppm IBA and 1000 ppm NAA [21].

2.3 Effect on the Survival Percentage

According to the study by Mariano, (2022), the survival percentage of citrus cuttings, especially focusing on the species of Calamansi, Calamandarin, and Dalandan. The study illustrates the application of different hormones such as IBA, IAA, NAA, hormex, and Saredex in different doses. However, most of the citrus cuttings of three species show maximum survival rate to the application of NAA at 400 and 500 ppm. In Calamansi, Calamandarin, and Dalandan the survival rate was 100%, 80%, and 100% respectively [41].

On the other hand, Kumar and Kumar, (2022) stated about the impact of IBA to analyze the survival rate of citrus cuttings, especially focusing on the *Citrus Limon* Burm. According to the experiment between which were compared between ten sets of experiments done by applying various concentrations of IBA in different growth media. However, plants treated with 800 ppm IBA concentration showed the maximum survival percentage [34]. Accordingly, the study of Kumar and Singh, (2020) the role of Indole-3-butyric acid (IBA) and Polyhydroxybutyrate (PHB) on *Citrus aurantifolia* from cuttings. According to the study the maximum survival rate between the total of nine treatments which were applied in various doses of IBA and PHB. The result of the experiment saws that maximum citrus cuttings were survived in the concentration of 2500 ppm IBA and 1000 ppm PHB [35].

2.4 Effect on the Day Required for Sprouting

The study of Gnawali *et al.,* (2022) demonstrates the vegetative growth of *Citrus limon* in the treatment of IBA in various doses. An experiment was designed to observe sprouting percentage, day of sprouting, length of leaves and shoots, etc. Different concentrations of IBA including 0 ppm, 500 ppm, 100 ppm, 2000 ppm, 3000 ppm, 4000 ppm, and 5000 ppm were used in the experiment. According to the result, the maximum sprouting percentage was observed at 4000 ppm IBA concentration. Along with that, the days for sprouting were about 33.16 [23]. According to the study of Kour *et al.,* (2022), individuals can gain knowledge about the growth response of *Citrus* × *Gambhir* Lush (Rough lemon) with the application of IBA plant growth regulator in various doses including 500 ppm, 1000 ppm, and 1500 ppm. In his experiment all the lemon cuttings were treated with the selected concemtration of IBA solution. However, based on the result it can be said that the maximum sprouting percentage in the cuttings was found in the IBA solution @ 500 ppm. The total time required for sprouting was 130 days [31]. On the other hand, Sharma, (2022) stated the growth behavior of C*itrus aurantifolia* in different concentrations of auxin, especially in the form of IBA. Along with that, it was also found that the minimum time required for citrus sprouting was about 16.83 days @ 0 ppm IBA concentration. However, IBA @ 500 ppm showed the best response of the citrus cuttings in terms of sprouting percentage [52].

Table 1. Different concentrations of auxin to enhance the number of leaves of Citrus

SL. NO.	Citrus varieties	Concentration of Auxin	Number of leaves (%)	References
1.	*Citrus aurantifolia* (Acid lime)	@ 3000 ppm IBA + 3000 ppm NAA	20.97	[43]
2.	*Citrus limon (*L.*)* (Lemon)	@ 50 ppm NAA	66.33	[12]
3.	*Citrus limon* Burm.	@ 2000 ppm IBA + 1000 ppm NAA	14.28	[21]

Table 2. Different concentrations of auxin influence the survival percentage of citrus

SL. NO.	Citrus varieties	Concentration of Auxin	Survival percentage	References
1.	*Citrus Sp*. (Calamansi, Calamandarin, Dalandan)	@ 400 and 500 ppm NAA	80-100%	[41]
2.	*Citrus Limon* Burm.	@ 800 ppm IBA	89.90 %	[34]
3.	*Citrus aurantifolia*	@ 2500 ppm IBA + 1000 ppm PHB	82.66 %	[35]

2.5 Effect on the Root Length

According to the study of Rajangam *et al.,* (2022), the effect of the IBA hormone in the growth of acid lime which was recorded by comparing the root growth of acid lime seedlings. A total of five concentrations of IBA were applied including 500 ppm, 1000 ppm, 1500 ppm, 2000 ppm, and 2500 ppm. However, based on findings maximum root growth was found in the solution is IBA @ 2000 ppm. In this treatment, the root length of the citrus cuttings was about 26.15 cm. Along with that, the fresh and dry weight of the root was also high in this IBA concentration [46]. Khan and Nabi, (2023) stated in their study that the significance of auxin hormone to enhance root and shoot growth of plants. However, they described those different forms of auxin, especially IBA, IAA, and NAA play a great role in the root induction of fruit crops. IBA is essential for growing adventitious roots of the plants whereas NAA is a great solution for inducing root growth of citrus cuttings [29]. On the other hand, the study of Kumar *et al.,* (2023) demonstrates the effect of auxin in the growth of different parts in *Citrus sinensis* L. Osbeck or sweet orange. Three sets of treatments were done by using the concentrate of IBA @ 1000 ppm, 2000 ppm, and 3000 ppm to examine the best result. Most of the hardwood cuttings of sweet orange showed maximum root growth in the IBA solution of 3000 ppm. However, the study also includes rooting percentage, number of roots, and root weight of citrus cuttings [37].

3. EVALUATION OF THE EFFECT OF GROWTH MEDIUM ON GROWTH AND PERFORMANCE OF CITRUS CUTTINGS CRITICALLY

3.1 Effect on the Rooting Percentage

According to Ferlito *et al.,* (2020), the medium that was used for the growth of *Carrizo citrange* was conifer wood biochar, one of the most significant substitutes for the nursery of citrus fruits, giving a fruitful result for the growth of the roots. That indicates that the replacement of the peat medium with the conifer wood biochar by 25%, which occurred an increase in the rooting percentage by more over 50%. Malakar *et al.,* (2019) highlighted that different growing media such as sand (M1), soil rite (M2; Mixture of 75 percent Irish peat moss and 25 percent horticulture grade perlite), soil mixture (M3; mixture of fine garden soil, sand and farmyard manure at a ratio of 2:1:1) and cocopeat (M4) were used for the species *Citrus aurantiifolia;* by the application of such medium showed a positive result for the rooting percentage as well. Those types of rooting medium increased the rooting percentage by 20-25% for that citrus species [19]. Meanwhile working with *Citrus sinensis* Albrecht *et al.,* (2020) mainly emphasized Murashige and Skoog agar medium which is a very famous is mainly differentiated with macronutrients of Calcium, Magnesium, and Nitrogen as well as micronutrients like zinc, copper, and iron which helps in the growth and

development of roots in the cuttings of citrus spp [1]. According to Vibhute *et al*., (2022), all the organic compounds provided a wonderful position for the Plant growth hormones to act upon and gave the rooting percentage almost a hike of 50%. For those kinds of nutrient factors Murashige Skoog agar mediumwais a very benevolent type of medium in terms of root and shoot growth which enables the hormones to act accordingly [60]. The studies done by Shanker *et al.,* (2019) on the *Citrus aurantifolia,* stated the fact that using of a mixture of peat soil with FYM with or without sand in a ratio of 1:1 showed somewhat considerable growth in the root length and number of roots. But the medium which is used without the sand did not show much of a positive result, on the other side the medium consisting of the sand showed somewhat increasing results in the root growth, by 5- 10%. Here, only the application of the FYM is not good enough for the growth and development of roots on the citrus cutting also Plant Growth Regulators has not enough significant effect on the rooting of the cutting of citrus species [51].

3.2 Effect on the Number of Leaves

In the research work shown by Kumar and Kumar (2022), the growth media used for the growth of *Citrus aurantifolia* considerably had a lot of things like slit soil, sand, FYM, vermicompost as well as cocopeat. All those things were extremely rich in almost all kinds of macro as well as micronutrients and were thought to be very much nourishing for the increase in the number of leaves on the cuttings of citrus, that was also demonstrated by Deshlehra *et al.,* (2022). However, after the results, the number of leaves did not increase as much as assumed before the culture.

The percentage increased only by 16.7 which was considerably lesser than what was thought and hence, over-nourishment can hinder the development of the leaves in some cases [34][13]. In the last case scenario, as portrayed by Lal (2023), the most traditional and convenient method of sand and soil plant growth media was being used for the citrus species of *Citrus jambhiri*, which showed a spectacular result for the increase in several leaves. It resulted in an increase of 17.3 which was quite high for such a simple type of plant growth medium and was surprisingly good [38]. Choudhary *et al.* (2022) mentioned that the traditional growth medium escalated the actions of plant growth regulators and gave a positive result. Hence the second plant growth medium gave the maximum healthy hike in the number of leaves as it has the composition of the maximum number of organic compounds [11].

3.3 Effect on Survival Percentage

In the studies shown by Rehman *et al.,* (2020), the growth media of *Citrus aurantifolia* was somewhat given a different result where along with the traditional method of sand and soil, a mixture of compost or vermicompost was added with the growth medium in a ratio of 1:1:1 that contained peat mass, sawdust as well as leaf parts [48]. Anitha *et al*., (2023), also stated that the whole mixture was very nourishing and was full of essential micro as well as macronutrients that were very significant for the citrus species' plant growth and survival and gave the plant some better scopes to survive. That species of Citrus survived even more than that which was only grown in sandy and slit soil and showed healthy side effects. It concludes the fact that the growth media provided a space for the plant growth regulators to fully act over the species and give them healthy growth along with higher survival chances which states that the higher the percentage of nourishing organic compounds in the growth media, the higher the chances of survival [5]. In the second scenario shown by Zulia and Gustianty (2023), the species of *Citrus limon L.* was tried growing in a completely different growth media which was uncommon and new for a citrus species and that was the ZPT extracts of green beans [61]. Al-Zuhairi *et al*., (2021), considered that green beans were full of phosphorus and potassium and could easily serve as the bed for the growth of a citrus species and it came up with better survival results than lemons that had previously been grown in some kinds of orchards with basic plant growth medium [4].

3.4 Effect on the Day Required for Sprouting

According to the studies of Kangabam *et al.,* (2020), when the growth media consisted of Garden soil, sand, and vermiculite along vermicompost in the ratio of 1:1:1:1 for the culture of *Citrus jambhiri Lush*., there was a lumpsome decrease in the required day length for the sprouting of the seedlings. the previous day's length which was approximately 8-10 days was reduced to 6 days and sprouting started after that [27].

Table 3. Different growth mediums to impact rooting percentage of citrus cutting

SL. NO.	Citrus Species	Growth medium	References
1	*Carrizo citrange*	Conifer wood biochar	[19]
2	*Citrus aurantiifolia*	Sand, soil rite, Mixture of 75 percent Irish peat moss and 25 percent horticulture grade perlite, soil mixture, and cocopeat	[40]
3	*Citrus sinensis* (L.) Osbeck.	Murashige and Skoog (MS) agar-nutrient medium	[1]
4	*Citrus aurantiifolia*	a mixture of peat soil + FYM with or without sand in1:1:1 ratio	[51]

Table 4. Different growth mediums affect the number of leaves of citrus cutting

SL. NO.	Citrus Species	Growth medium	Number of leaves (%)	References
1	*Citrus aurantifolia*	slit soil, sand, FYM, vermicompost, cocopeat.	16.7	[34]
2	*Citrus Gambhir*	sand and soil	17.3	[38]

Table 5. Various growth mediums to nurture survival percentage of citrus cutting

SL. NO.	Citrus Species	Growth medium	Survival (%)	References
1	*Citrus aurantifolia*	slit soil, sand, vermicompost	20	[48]
2	*Citrus limon L.*	ZPT extracts of green beans.	45	[61]

Table 6. Several growth mediums to enhance sprouting days of citrus cutting

SL. NO.	Citrus Species	Growth medium	No. of required days	References
1	*Citrus Gambhir Lush*	Garden soil, sand, vermiculite, vermicompost	6	[27]
2	*Daisy Mandarin*	Murashige and Skoog agar media	7	[7]

Qadri *et al*., (2021) elaborated that the growth medium effectively decreased the number of days by being a pool of both macro and micronutrients and serving as a faster nutrition tub for the citrus species [45]. According to Brar *et al.,* (2020), the growth media of *Daisy Mandarin* when substituted with MS media instead of sands and soils, the day length for sprouting was reduced to 7 from 11 despite being not grown in a traditional growth media. They were treated with Nitrogen, zinc as well and Indole acetic Acid. To some extent, they were also exposed to Gibberellic Acid and Ethylene for the early respiration surge or the climacteric of their fruits and as a result their root lengths also showed a considerable amount of growth before the actual day length. They were also provided with photo selective covering during their growth period. Hence, Murashige and Skoog agar media, if backed up by proper supporting conditions can provide better results for reducing days for sprouting for the citrus species even in the absence of traditional growth media [45].

3.5 Effect on the Root Length

Firstly, the study shown by Kumar *et al.,* (2022) depicting the root length of Citrus sinensis showed a great positive variation in the fact that consisted of the growth media with sand, soil, and FYM and peat in the ratio of 1:1:1:1 which is about 5 cm approx. The length variation is quite high for the citrus species and gives a particular base for the action of the plant growth regulators especially auxin.

Whereas when the same species was being used under a plant growth media of cocopeat along with Trichoderma sp. In a ratio of 1:1, it failed to show the same results as it has the absence of basic plant growth media sand and soil and failed to trigger the work action mode of

the plant growth regulators. Hence, it is clear from the above studies that despite using the same plant growth regulators, there has been a major shift in the results of the same citrus species and that is only because of the variation in the plant growth media. Cocopeat despite being a good grower medium, alone fails to increase the growth rate of the root length [36]. From the studies of Arunadevi *et al.,* (2019), Citrus aurantifolia showed their best result of root length growth when they were allowed to grow on a salty media and were pushed with NAA, their root length almost increased to 69.7% and hence it showed a positive result [6]. So, it can be concluded from another study written by Samaradiwakara *et al.,* (2023) that even with the barest minimum growth media components, citrus fruits can show amazing growth in their root lengths only if they are provided with the right amount of needful plant growth regulators such IBA and NAA [6].

3.6 Critical Illustration of Number of Leaves Per Cuttings

According to the study of Kumar *et al.,* (2023), the effect of different PGRs especially auxin on the growth of shoots, roots, and leaves of fruit crops. IAA, IBA, and NAA help in the expansion of leaves per cutting whereas gibberellin helps to enhance the shoot growth. Along with that, it can influence the fruit quality of citrus [32]. On the other hand, Maurya *et al.,* (2022) represent the importance of auxin in different forms and concentrations to initiate the formation of roots in citrus plants. However, experiment on *Citrus limon* Burm. by applying IBA and NAA in different concentrations such as 1000 ppm, 1500 ppm, and 2000 ppm. In this expriment the main factor which was recorded that the effect of seasonal factor on the growth and development of the cutting, for this they had planted the cuttings on the two different seasons, one is Rainy and on the other hand thay selected Spring. However, based on the result it can be seen that the citrus cuttings have shown a great response in the application of IBA at 2000 ppm concentration. A maximum number of leaves in citrus cuttings were found in the treatment of IBA solution at a rate of 2000 ppm. In the Spring season, the number of leaves in citrus cuttings was 12.44 which was the highest among all. On the other hand, in the Rainy season, the number of leaves in cuttings was 14.06 [42]. In an experiment by Rathour *et al*., (2023), the effects of various PGR (IAA, IBA and NAA) rates on the development of shoots and growth of cuttings from the stem in Lemon (Citrus limon Burm.) cv. Pant lemon-1 was examined. The conditions were westerners UP, and the treatments were T 15 (3000 ppm IBA + 1000 ppm NAA) and ppm IBA + 750 ppm NAA) [47]. It was also observed in the paper of Dhakad *et al.* (2020) that the cuttings immersed in (2000 ppm IBA + 1000 ppm NAA) concentration had significantly higher days to first buds originate sprout (8.53 days), emerging (83.33%), plant height (15.55cm), number of shoots per cutting at 30, 60, and 90 days after planting (2.98, 3.88, and 4.88 respectively), number of leaves per cutting (14.28), shoot diameter (3.41mm), and final survival (83.33%) of cuttings [14]. According to Sharma (2022), for Citrus aurantifolia there was plant growth medium like garden soil, silt soil, FYM, and vermicompost along with cocopeat in the ratio of 1:1:1:1:1 where there was the application of Indole acetic Acid, Gibberellic acid, and the gaseous hormone Ethylene as well in the concentrations of 1000, 300 and 460 ppm respectively [52]. As stated by Kavinprashanth *et al.* (2021) spray of NAA was also done to restore the cutting like a new plant or the parent plant. The results were more or less leaves were found per cutting which were approximately 19.2 much greater than the original one or the parent one. Hence, the combination of the concentrations of the Plant growth regulators (IBA and NAA) as well as the growth media components brought about a positive change in the citrus species cutting [28].

4. ANALYSIS OF THE EFFECT OF PGRS AND ROOTING MEDIA IN COMBINATION ON THE NUMBER OF LEAVES IN A CRITICAL MANNER

According to Verma *et al.,* (2022), PGR and rooting media play a significant role in the development process of plants in their versatile aspects of growth. However, PGRs are considered the naturally occurring or synthetic compounds that play a crucial role in regulating the plants in their different phases [59]. On the other hand, rooting media refers to the substrates utilized for propagating plants through cutting as mentioned by Singh *et al.,* (2019). From this perspective, the number of leaves is considered a fundamental parameter in assessing plants as it reflects the overall productivity and health of the plant. As citrus propagation is a common practice for commercial and home citrus cultivation and at the same time, the success of this propagation highly depends on the proper selection of the PGRs such as IAA,

IBA, NAA, etc plant growth and the rooting media as expressed by Fruit (2023) [53]. However, the number of leaves on the cutting is of particular interest among the numerous aspects that impact the performance of citrus cuttings. Particularly, Auxins are the commonly used PGRs in the rooting cuttings. Additionally, IBA is a widely employed synthetic auxin known for its efficiency in the context of root initiation [53]. Kumar *et al.,* (2022) highlighted the influence of IBA concentrations on the number of leaves in the case of citrus cuttings. Along with this, different studies have evaluated the relationship between the concentration of IBA and the total number of leaves within citrus cuttings which sheds light on the critical role of this hormone in the propagation of citrus plants [36]. According to Al-Jabbari *et al.* (2020), increasing the IBA concentration from 1000 ppm to 6000 ppm led to a statistically significant increase in the number of leaves. This result indicates a substantial effect of IBA concentration within leaf production. Overall, there was a significant rise in the number of leaves with increasing the IBA concentration [2].

Here, Zivzic and Salakhani are the two types of IBA hormones that directly affect the total number of leaves. On the other hand, there is a requirement to investigate the potential negative impacts of high IBA concentration on other aspects of citrus cutting development as evaluated by Upadhyay, *et al.,* (2020). Here, it is important to ensure that root development and overall plant health are not compromised while increased leaf production is valuable. Moreover, the balance between root and shoot growth is crucial for the plant as stated by Vega, *et al.,* (2019) [58]. Additionally, the excessive focus on leaf production might compromise the overall health of the cutting. Cytokynins on the other hand are another group of PGRs that can affect the number of leaves in the context of the citrus cutting as highlighted by Eisa *et al*., (2022) [15]. From this perspective, the opinion of Estrella-Maldonado *et al.,* (2022) highlights that the application of cytokinins like Kinetin can increase the number of leaves in the citrus cutting, particularly during the early stage of growth. During the consideration of the utilization of PGRs (IAA, IBA or NAA), the choice of the particular chemical as well as its concentration is significant [18]. According to Khandaker *et al.,* (2022), cytokinins and IBA can positively impact leaf production where the optimal concentration may differ. In the case of rooting, it is important to choose the effective media for the successful propagation of the citrus cuttings [30]. Accordingly, Ullah, (2020) stated that numerous types of media including soil, peat, vermiculite, and perlite have varying properties that can impact the development of the roots as well as the leaves in cuttings [55]. Hayat *et al.,* (2023) stated that different growth media like rice husk, wheat straw, bagasse, etc have been also utilized in citrus propagation. The traditional soil delivers essential nutrients but may suffer from poor aeration that potentially hampers the root growth of the plants. In this aspect, the quality as well as the composition of soil matters a lot and efficiently impacts the number of leaves [25]. Loam-based soil mixtures result in a higher leaf count than a sandy soil mixture as approached by Brischke and Wegener (2019) [8]. On the other hand, Suthar *et al.,* (2021) stated soil-less media including vermiculite, peat, and perlite offer moisture retention and developed aeration in Kagzi lime. They play a crucial role in generating a controlled environment for the development of roots effectively [54]. The research addresses that a 1:1 peat-perlite mixture can result in a higher number of leaves as indicated by ESRİNGÜ *et al.,* (2022) [19]. On the other hand, pure peat or perlite does not have the significant potential for providing the higher number of leaves that its mixture contains. In the context of citrus cutting propagation, the interaction or coordination between PGRs especially auxin and rooting media is a vital aspect. Additionally, it must be followed by the individuals to increase the total leaf numbers in which case, the choice of the rooting medium and PGR should be complementary to optimize the number of leaves and overall growth. The research in this aspect highlights that the combination of 1000 ppm IBA with a perlite vermiculite mixture can result in a high leaf count within various treatment combinations as delivered by Idol *et al.,* (2019) [26]. However, a nutrient-rich medium and a well-draining medium possess a significant role in encouraging the development of a larger amount of the leaves. Overall, it is important to mention that while a large number of leaves may indicate better initiation growth, the crucial factor is that it does not provide a guarantee for long-term success. Additionally, it should not be overlooked here to focus on the overall health and the adaptability of the cutting and at the same time, its ability to withstand transplantation into the field. Moreover, the utilization of effective rooting media and PGEs can have a crucial influence on the number of leaves in citrus cuttings as mentioned by Ghimire *et al.,* (2022) [22].

Table 7. Various growth mediums to advance root growth percentage of citrus cutting

SL. NO.	Citrus Species	Growth medium	Root Growth (%)	References
1	Citrus sinensis	soil, sand, FYM, peat.	5	[36]
2	Citrus aurantifolia	saltwater media	69.7	[6]

Table 8. The effect of the interaction among IBA doses and vegetative growth of the cuttings [2]

Cultivars	Hormone doses	Rooting rate (%)	Roots number	Roots length (mm)	Survival rate (%)	Shoot length (mm)	Shoot number	Leaves number
Zivzik	0	0.67 BC	13.59 B	89.08 D	0.67 BC	110.47 G	3.13 CD	23.03 G
	1000	0.73 B	27.08 A	84.79 D	0.77 AB	128.69 FG	3.17 CD	39.37 FG
	2000	0.67 BC	12.74 BC	98.91 D	0.70 B	148.51 EF	2.58 D	49.76 EFG
	4000	0.63 BC	10.42 CD	100.31 D	0.63 BC	162.83 DE	3.17 CD	71.73 DE
	6000	0.93 A	9.68 D	76.50 D	0.93 A	177.48 D	2.70 D	56.38 EF
Salakhani	0	0.67 BC	7.05 EF	153.98 C	0.67 BC	273.01 AB	3.68 BC	93.94 D
	1000	0.50 C	7.03 EF	155.84 C	0.50 CD	241.50 C	4.31 B	100.96 CD
	2000	0.53 C	7.16 EF	186.98 B	0.40 DE	262.62 BC	4.28 B	137.70 B
	4000	0.23 D	5.44 F	227.69 A	0.23 E	285.55 AB	5.44 A	177.44A
	6000	0.50 C	8.47 DE	230.76 A	0.50 CD	300.23 A	3.65 C	130.19 B

5. CONCLUSION

The study centralises the concept of using plant growth regulators and root growth mediums in terms of enhancing the formation of spontaneous roots facilitating the rooting process of cuttings in different citrus species. The function and contribution of auxins in plant growth of citrus species, the effect of growth medium on the performance in citrus cuttings, an assessment focused on the number of leaves that citrus cuttings have, and the significance of PGRs especially auxin as well as rooting media for increasing the leaves numbers in citrus species are all covered in the discussion segment of this review paper. Along with that, the study includes the role of different growing medium intended for sprouting new plants from cuttings and supports the root growth of the cuttings and help them to grow into plants. However, the study also emphasizes the application of auxin and growth mediums in different concentrations for better nourishment of the citrus cuttings. For the vegetative development of new citrus cuttings, various rooting media such as sand, dirt, humus, peat moss, and vermiculite are utilized. Various studies directed to analysis of the usefulness of numerous rooting media as growth mediums to properly develop citrus cuttings among different species of citrus family have been highlighted to spot their essentiality in plant growth and how their performance affects the entire growth additionally showcased in the assignment.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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