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Effect of chemicals on growth and buddability in rough lemon (*Citrus jambhiri* Lush) rootstock seedlings under semi arid irrigated ecosystem of Haryana

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Abstract
An experiment was conducted with the objective to advance and enhance the buddabiltiy in rough lemon (*Citrus jambhiri* Lush) seedlings comprising foliar application of T_1 (GA_3 50 ppm + Urea 0.5%), T_2 (GA_3 50 ppm + Urea 1%), T_3 (GA_3 100 ppm + Urea 0.5%), T_4 (GA_3 100 ppm + Urea 1%), T_5 (NAA 50 ppm + Urea 0.5%), T_6 (NAA 50 ppm + Urea 1%), T_7 (NAA 100 ppm + Urea 0.5%), T_8 (NAA 100 ppm + Urea 1%), T_9 (ABA 50 ppm + Urea 0.5%), T_{10} (ABA 50 ppm + Urea 1%), T_{11} (ABA 100 ppm + Urea 0.5%), T_{12} (ABA 100 ppm + Urea 1%) and T_{13} (Control) twice after 30 and 60 days after transplanting. Maximum seedling height 95.83cm and numbers of leaves 98.17/plant was measured in treatment T_4 (GA_3 100 ppm + Urea 1%) at 180 days after transplanting. Maximum stem diameter 7.16mm was measured in treatment T_{11} (ABA 100 ppm + Urea 0.5%). The highest per cent seedlings 57.14, 76.19 and 95.24% attained buddable stage in treatment T_{11} at 150 DAT, 165 DAT and 180 DAT respectively. Overall, foliar spray of ABA (100 ppm + Urea 0.5%) twice at 30 and 60 DAT resulted in 57% buddable seedling over control (14%) in rough lemon rootstock 150 DAT.

Keywords: Plant growth regulator, urea, foliar spray, rough lemon, buddability

Introduction
In India, the area under citrus is 1.03 million ha with production of 13.20 million tonnes and average productivity of 9.78 tones/ha. (NHB data base). Citrus occupy third rank after mango and banana. Among the citrus cultivars, sweet orange, mandarin, limes, lemon and grapefruit are important and occupy a major acreage in the state. They are propagated by budding mainly on rough lemon *(Citrus jambhiri,* Lush) rootstock. Rough lemon originated in the Himalayan foothills of India and is the most widely used rootstock for citrus. Availability of sufficient, superior quality planting material and its effective distribution to the orchardists is likely to improve orchard efficiency. Under sub-tropical conditions of Haryana and surrounding states, the time required to grow citrus rootstock seedlings to a size suitable for budding may be as long as one and half to two years. The entire process is time consuming and labour intensive as, irrigations, fertilization and plant protection measures are to be taken up at regular intervals at all nursery stages thereby increasing cost of production. Shortening this time would benefit to the nurserymen by reducing cost of production. After budding operation, although the citrus plant becomes ready for plantation in about six to eight months, it can only be planted in the orchard, in spring or autumn. Hence, the plant can only be transplanted around one year after budding. So, it takes about total three years from seed sowing to reach to farmers' field. Use of plant growth regulators and chemicals is a boon to horticulture and is being used for various purposes with an ultimate aim to increase production. Gibberellic acid takes part in rapid cell division, which mainly helps to increase the thickness of seedling girth (Hoda *et al.* 2010) [6] in citrus. Naphthalene Acetic Acid (NAA) belongs to synthetic forms of auxins play key role in cell elongation, cell division, vascular tissue, differentiation, root initiation, apical dominance and leaf senescence (Davies, 1987) [5]. Foliar application of NAA has also been found to increase plant height, number of leaves per plant in different crops (Lee, 1990) [10]. Plant growth retardants are applied in agronomic and horticultural crops to reduce unwanted longitudinal shoot growth without lowering plant productivity (Rademacher, 2000) [12].

So, keeping in view the above facts the present investigation was carried out with the objective to reduce the time and cost of production of healthy Kinnow plants on rough lemon rootstocks for the benefit of entrepreneurs and farmers with the intervention of growth regulators and urea under semi arid irrigated ecosystem.

Materials and Methods

The present investigation was carried out at department of Horticulture, CCS Haryana Agricultural University, Hisar during 2016-17 in Randomized Block Design (RBD) with three replication. Fruits of rough lemon were harvested directly from the single plant during the month of September. Seeds were extracted manually and sown in the primary nursery. In the mid of March, healthy and uniform seedlings were transplanted in the secondary nursery. These seedlings were used for the experiment, comprising of foliar application of thirteen treatments viz. T_1 (GA_3 50 ppm + Urea 0.5%), T_2 (GA_3 50 ppm + Urea 1%), T_3 (GA_3 100 ppm + Urea 0.5%), T_4 (GA_3 100 ppm + Urea 1%), T_5 (NAA 50 ppm + Urea 0.5%), T_6 (NAA 50 ppm + Urea 1%), T_7 (NAA 100 ppm + Urea 0.5%), T_8 (NAA 100 ppm + Urea 1%), T_9 (ABA 50 ppm + Urea 0.5%), T_{10} (ABA 50 ppm + Urea 1%), T_{11} (ABA 100 ppm + Urea 0.5%), T_{12} (ABA 100 ppm + Urea 1%) and T_{13} (Control, water spray) twice after 30 and 60 days after transplanting (DAT) on the same seedlings**.** Solution of growth regulators were prepared by dissolving the required quantity in small amount of absolute alcohol and then final volume was made with distilled water. Height of the seedlings was measured from soil surface to the plant tip in centimeters with a meter scale, while diameter was measured with Vernier calipers at 15 cm above ground level. Leaves of each seedling was counted and averaged per replication as well as treatment. The seedling that attained stem diameter of 6 mm at 15 cm above ground level was considered ready for budding and hence, observations on buddable percentage was determined on the basis of stem thicken. The mean values of different treatments were analysed with the help of a windows based computer package OPSTAT along with corresponding standard error of mean (S.E.±). The critical difference at 5 per cent level of significance was computed.

Results and Discussion

Seedling height: Mean seedling height differed significantly among treatments (table-1). Mean seedling height increased significantly with treatments T1 to T8 and decreased with treatments T9 to T12 over control. Maximum seedling height (73.52 cm) was measured in treatment T_4 (GA_3 100 ppm + Urea 1%) followed by (70.77 cm) in treatment T_2 (GA_3 50 ppm + Urea 1%), whereas, minimum seedling height (46.31cm) was measured in treatment T_{11} (ABA 100 ppm + Urea 0.5%) significantly followed by (47.98 cm) in T_9 (ABA 50 ppm + Urea 0.5%). Similarly, seedling height increased significantly with every increase in the number of days after transplanting (DAT) irrespective of treatment. Maximum seedling height (74.78 cm) was found at 180 DAT and minimum (43.41 cm) at 30 DAT.

The interaction of days after transplanting and treatment was also significant. Maximum seedling height (95.83 cm) was measured at 180 DAT in treatment T_4 (GA_3 100 ppm + Urea 1%) and minimum (41.28 cm) was observed with the treatment T_6 (NAA 50 ppm + urea 1%) at 30 DAT. In the present study, the higher seedling height with GA_3 and urea spray may be attributed to the cell multiplication and elongation in the cambium tissue of the internodal region, because gibberellic acid apparently activates the metabolic processes or nullifies the effect of growth inhibitor. NAA has been shown to greatly increase cellulose fiber formation in plants (Suman *et al.* 2017) [14]. Arne and Mitchell (1969) [1] also reported that ABA inhibits stem elongation probably by its inhibitory effect on gibberellic acid. These results are in accordance with the findings of Dalal *et al.* (2004) [4] and Jadhav *et al*. (2007) [7] in rangpur lime.

Table 1: Effect of different chemicals on the seedling height (cm) of rough lemon

Treatment	Days after transplanting (DAT)						
	30	60	90	120	150	180	Mean
T_1: GA_3 50 ppm +Urea 0.5%	44.18	48.50	66.73	71.20	74.10	79.00	63.95
T_2: GA_3 50 ppm +Urea 1%	44.17	52.67	71.20	80.31	85.47	90.80	70.77
T_3: GA_3 100 ppm +Urea 0.5%	43.70	49.83	70.81	73.40	75.82	82.85	66.07
T_4: GA_3 100 ppm +Urea 1%	43.67	54.33	75.78	82.94	88.56	95.83	73.52
T_5: NAA 50 ppm +Urea 0.5%	44.43	47.33	61.70	64.50	68.54	75.58	60.35
T_6: NAA 50 ppm +Urea 1%	41.28	49.17	62.50	67.28	72.94	79.11	62.13
T_7: NAA 100 ppm +Urea 0.5%	43.33	49.67	68.03	73.38	76.12	81.33	65.31
T_8: NAA 100 ppm +Urea 1%	43.83	53.17	70.38	76.15	81.74	87.95	68.87
T_9: ABA 50 ppm +Urea 0.5%	42.37	44.10	44.48	47.49	53.26	56.00	47.98
T_{10}: ABA 50 ppm +Urea 1%	42.83	43.17	46.52	50.17	57.45	60.08	50.04
T_{11}: ABA 100 ppm +Urea 0.5%	41.77	42.53	43.72	45.66	49.50	55.17	46.31
T_{12}: ABA 100 ppm +Urea 1%	42.10	43.13	44.16	48.73	53.20	58.96	48.27
T_{13}: Control	42.70	45.17	52.00	57.62	63.23	69.50	55.04
Mean	43.41	47.59	59.82	64.53	69.23	74.78	
C.D. at 5%	Treatment=1.32 Days=1.02 Treatment × Days=2.91						

Stem diameters: Stem diameter of the seedling influenced significantly by various treatments, days after transplanting and their interaction. Treatments T9 to T12 and T5 significantly increased stem diameter over control, whereas, all remaining treatments were at par with control. Maximum stem diameter (5.54 mm) was measured in treatment T_{11} (ABA 100 ppm + Urea 0.5%) closely followed by (5.41 mm) in T_{12} (ABA 100 ppm + Urea 1%). Treatment T_9 (5.30 mm) and T_{10} (5.28 mm) were found at par with each other. The minimum stem diameter (4.64 mm) was observed in treatment T_4 (GA_3 100 ppm + Urea 1%) and it was at par with T_1, T_2 and T_3 with values 4.80, 4.67 and 4.71 mm, respectively. Maximum stem diameter (6.20 mm) was observed at 180 DAT and minimum (3.71 mm) at 30 DAT. While comparing their interaction, maximum stem diameter (7.16 mm) was observed at 180 DAT with treatment T_{11} (ABA 100 ppm + Urea 0.5%) and it was at par with treatment T_{10} (6.75 mm) and T_{12} (6.88 mm) at 180 DAT and minimum stem diameter

(3.59 mm) was observed in control at 30 DAT. Nitrogen shows the positive correlation with GA_3 which resulted into maximum stem diameter as GA_3 plays role of cell elongation. ABA possesses the capacity of inhibiting elongation of normal light-grown internodes, and of preventing the outgrowth of lateral shoots, hence more stem diameter. Increase in the stem diameter with the application of GA_3 were earlier reported by Modesto *et al.*(1996) & Kadam *et al.* (2011) in rangpur lime and Kadam *et al.*(2010) in Kagzi Lime.

Table 2: Effect of different chemicals on stem diameter (mm) of rough lemon seedlings

Treatment	Days after transplanting (DAT)						
	30	60	90	120	150	180	Mean
T_1: GA_3 50 ppm +Urea 0.5%	3.80	3.97	4.61	4.89	5.51	6.04	4.80
T_2: GA_3 50 ppm +Urea 1%	3.65	3.78	4.50	4.83	5.42	5.83	4.67
T_3: GA_3 100 ppm +Urea 0.5%	3.72	3.84	4.47	4.85	5.45	5.93	4.71
T_4: GA_3 100 ppm +Urea 1%	3.71	3.96	4.36	4.75	5.39	5.68	4.64
T_5: NAA 50 ppm +Urea 0.5%	3.80	4.04	4.80	5.03	5.66	6.23	4.93
T_6: NAA 50 ppm +Urea 1%	3.69	4.07	4.68	4.90	5.58	6.02	4.82
T_7: NAA 100 ppm +Urea 0.5%	3.75	4.10	4.71	4.99	5.51	5.96	4.84
T_8: NAA 100 ppm +Urea 1%	3.85	4.02	4.61	4.93	5.63	5.94	4.83
T_9: ABA 50 ppm +Urea 0.5%	3.68	4.20	5.32	5.66	6.28	6.67	5.30
T_{10}: ABA 50 ppm +Urea 1%	3.72	4.21	5.16	5.57	6.28	6.75	5.28
T_{11}: ABA 100 ppm +Urea 0.5%	3.62	4.25	5.57	5.94	6.67	7.16	5.54
T_{12}: ABA 100 ppm +Urea 1%	3.67	4.24	5.40	5.91	6.37	6.88	5.41
T_{13}: Control	3.59	3.96	4.83	4.99	5.19	5.56	4.69
Mean	3.71	4.05	4.85	5.17	5.76	6.20	
C.D. at 5%	Treatment=0.16 Days=0.10 Treatment×Days=0.41						

Number of leaves: All the treatments were found significantly effective in increasing the number of leaves per plant over control (Table-3). However, maximum numbers of leaves 82.19 per seedling were counted with the application of treatment T_4 (GA_3 100 ppm + Urea 1%) and minimum 67.40 leaves per seedling was counted in treatment T_{13} (control). It was also observed that the number of leaves per seedling increased significantly with gradual increase in the number of days after transplanting (DAT) irrespective of the treatments. However, the maximum number of leaves 86.41 per seedling was observed at 180 DAT and minimum 44.69 at 30 DAT. The interaction between days after transplanting and treatment was also found significant. Maximum number of leaves 98.17 per seedling was observed in treatment T_4 (GA_3 100 ppm + Urea 1%) at 180 DAT and minimum number of leaves 43.17 in control at 30 DAT. This increase in number of leaves per seedling increased may be due to increase in seedling height as evident from the investigation. Increase in number of leaves with GA_3 and urea treatment may be due to the enhanced physiological processes to form new leaves at faster rate and promotion of linear growth which creates an ideal condition to develop new leaf primordial. Similar results reported by Patil *et al.*(1996) in khirni rootstock, Singh and Sheo-Govind (2000) in *Citrus reticulata*.

Table 3: Effect of different chemicals on number of leaves per seedling of rough lemon

Treatment	Days after transplanting (DAT)						
	30	60	90	120	150	180	Mean
T_1: GA_3 50 ppm +Urea 0.5%	47.33	68.67	78.83	83.83	89.83	93.50	77.00
T_2: GA_3 50 ppm +Urea 1%	44.33	66.50	82.90	89.17	84.50	88.17	75.93
T_3: GA_3 100 ppm +Urea 0.5%	43.17	73.83	80.50	85.50	88.50	94.17	77.61
T_4: GA_3 100 ppm +Urea 1%	44.02	78.50	87.83	90.17	94.47	98.17	82.19
T_5: NAA 50 ppm +Urea 0.5%	43.83	63.17	68.17	81.83	76.33	80.17	68.92
T_6: NAA 50 ppm +Urea 1%	43.67	61.67	66.67	72.83	75.17	78.83	66.47
T_7: NAA 100 ppm +Urea 0.5%	44.67	61.00	77.50	80.97	85.17	88.50	72.97
T_8: NAA 100 ppm +Urea 1%	43.83	67.33	82.63	87.83	91.47	94.83	77.99
T_9: ABA 50 ppm +Urea 0.5%	45.50	68.50	71.33	75.50	77.07	80.10	69.67
T_{10}: ABA 50 ppm +Urea 1%	46.83	64.83	76.50	79.17	82.83	84.67	72.47
T_{11}: ABA 100 ppm +Urea 0.5%	45.17	59.67	72.50	75.50	79.83	82.50	69.20
T_{12}: ABA 100 ppm +Urea 1%	45.67	65.27	72.65	74.13	76.80	80.50	69.27
T_{13}: Control	43.00	61.73	70.83	72.83	76.83	79.17	67.40
Mean	44.69	66.21	76.06	80.71	82.98	86.41	
C.D. at 5%	Treatment=1.29 Days=1.35 Treatment × Days=4.60						

Buddability

All the treatments were found significantly effective in increasing the buddability of the seedlings over control from 150 DAT onwards (Table-4). Highest percentage of buddable seedlings 57.14, 76.19 and 95.24% was observed in T_{11} (ABA 100 ppm + Urea 0.5%) and lowest (14.28, 33.33 and 71.43%) in T13 (control) at 150 DAT, 165 DAT and 180 DAT, respectively. This shows that foliar application of ABA along with Urea resulted into more than 50% buddable seedling one month advance as compared to control. ABA being growth inhibitor might have not allowed the sprouting of the buds and thus these buds might have delayed growth and increases the diameter of the stem, hence advanced the buddability as evident from the present investigation. Whereas, role of auxins and GA3 in increasing buddabilty is mediated through increase in overall growth i.e. Height and number of leaves which results into increased photosynthetic activity and accumulation of more metabolites which increases the stem

diameter. These results are in conformity with the earlier findings of Dalal *et al.* (2002) [3] and Dalal *et al.* (2004) [4] in rangpur lime.

Table 4: Effect of different chemicals on buddable percentage of rough lemon seedling

Treatment	Days after transplanting			
	135	150	165	180
T_1: GA_3 50 ppm +Urea 0.5%	1.00	23.80	47.62	76.19
T_2: GA_3 50 ppm +Urea 1%	1.00	23.80	47.62	71.43
T_3: GA_3 100 ppm +Urea 0.5%	1.00	19.05	47.62	76.19
T_4: GA_3 100 ppm +Urea 1%	1.00	19.05	42.85	71.43
T_5: NAA 50 ppm +Urea 0.5%	1.00	39.00	61.90	85.71
T_6: NAA 50 ppm +Urea 1%	1.00	28.57	52.38	76.19
T_7: NAA 100 ppm +Urea 0.5%	1.00	28.57	61.90	76.19
T_8: NAA 100 ppm +Urea 1%	1.00	35.14	57.14	85.71
T_9: ABA 50 ppm +Urea 0.5%	1.00	52.38	66.67	90.48
T_{10}: ABA 50 ppm +Urea 1%	1.00	52.38	66.67	90.48
T_{11}: ABA 100 ppm +Urea 0.5%	1.00	57.14	76.19	95.24
T_{12}: ABA 100 ppm +Urea 1%	1.00	52.14	71.43	95.24
T_{13}: Control	1.00	14.28	33.33	71.43
C.D. at 5%	NS	3.32	6.45	5.47

It may be concluded from the study that foliar application of NAA and ABA @50-100ppm along with urea 0.5-1.0% at 30 and 60 days after transplanting advanced the buddabilty and more percentage of buddable seedlings in rough lemon rootstock of citrus. The effect of ABA was more pronounced.

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