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Effect of growth regulators, chemical and organic wastes on the seed germination and seedling diameter of Rangpur lime

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Abstract
The present investigation entitled "Effect of growth regulators, chemical & organic wastes on germination & seedling diameter of Rangpur lime'', was conducted at All India Co-ordinated Research Project on Citrus, Department of Horticulture, Mahatma Phule Krishi Vidyapeeth, Rahuri during 2013-14. The field experiment was laid out in Randomized Block Design (RBD) with three replications and ten treatments whereas laboratory experiment was laid out in Completely Randomized Design (CRD) with three replications and ten treatments, comprising pre-soaking treatment of Gibberellic acid (GA_3) at 50 and 100 ppm, Naphthalene acetic acid (NAA) at 50 and 100 ppm, KNO_3 at 1% and 2%, cow urine 50% and 100% and cow dung paste and control. The observation on germination percentage, seedlings diameter were recorded. Among all the treatments, T_2 and GA_3 100 ppm was the best treatment for germination under laboratory (88.86%) as well as field conditions (69.88%) whereas, the highest seedling diameter (1.67 cm) was recorded in treatment T4, NAA 100 ppm.

Keywords: Germination, Rangpur lime, seedlings, treatment, Gibberellic acid, diameter

Introduction
Citrus fruits have a prominent place among people and extensively grown in tropical and subtropical regions. Citrus is often regarded as a 'queen of fruits'. It is one of the choicest fruits having high consumer's preference both as fresh fruit as well as for its refreshing processed juice. Genus citrus is a unique in its diversity of forms and no other fruit can parallel it. Citrus fruit possess greater adaptability to different climatic conditions. Internationally, citrus plantation in the world is confined to 0-40º latitude from north to south of the equator covering different soil and climatic conditions (Ghosh, 2000) [6]. The plants of this rootstock are vigorous, precocious and prolific with produce. It makes a good union with a number of citrus species like sweet orange, mandarin and pummelo. This rootstock is raised through seed only. In seed propagated plants better and quicker germination of seeds and production of maximum number of nucellar seedlings is highly essential to meet the increasing demands of cultivators in shortest possible time. The problem in Rangpur lime propagation is heavy mortality of the seedlings in the primary nursery stage. Gupta, (1989) [7] reported that seed coat of lime acts as a barrier because it interfere with early germination of seed due to presence of certain inhibitory substances that delay the germination. The experimental evidences under Citrus Fruit Research Scheme, Nagpur (1944-48) indicates that the rate and extent of seed germination and seedling growth in Rangpur lime is not satisfactory.
Germination of Rangpur lime seeds sown without any pre-treatment is between 27-30% (Singh *et al.,* 1970) [18]. The seeds take about 20-40 days to germinate and the seedling growth in the nursery stage is also very slow, and hence it takes longer time, nearly about 15-18 months to attain buddable size. Slow growth at early period of Rangpur lime seedlings has been reported by Kawathalakar and Kunte (1974) [12] under Nagpur condition. In order to make the nursery practices efficient, the rootstock seedlings must attain good health, vigour and size for budding. It is, therefore, highly essential to accelerate the germination and growth rate of Rangpur lime seedlings to attain buddable size earlier.

Material and Methods
The research carried out at All India Co-ordinated Research Project on Citrus, Department of Horticulture, Mahatma Phule Krishi Vidyapeeth, Rahuri during 2013-14. Geographically, the central campus is situated in between 19^0 47′ to 19^0 57′ North latitude and 74^0 19′ to 74^0 32′ East longitude. It is situated at about 525 metre above the mean sea level.
Climatically, this area falls in semi-arid and sub-tropical zone with an annual rainfall varying from 307 to 619 mm. Average annual rainfall being 475 mm.

Most of the rainfall is received through South-West monsoon. The annual mean maximum and minimum temperatures are 34.5 ^{0}C and 18.3 ^{0}C, respectively.

The seeds of Rangpur lime were treated as follows

T_1: Soaking seeds in 50 ppm GA_3
T_2: Soaking seeds in 100 ppm GA_3
T_3: Soaking seeds in 50 ppm NAA
T_4: Soaking seeds in 100 ppm NAA
T_5: Soaking seeds in 1.0% KNO_3
T_6: Soaking seeds in 2.0% KNO_3
T_7: Soaking seeds in cow urine 50%
T_8: Soaking seeds in cow urine 100%
T_9: Soaking seeds in cow dung paste
T_{10}: Soaking seeds in distilled water (control)

(Note- All the seeds were soaked for 24 hours in growth regulators, chemical and organic wastes.)

After imposing the treatment, seeds were sown for germination. The experiment was carried out adopting Completely Randomized Design (CRD) with 10 treatments and 3 replication in field condition whereas under laboratory conditions, treated seeds of Rangpur lime were placed according to between paper method. The rolled towel papers with seeds were kept in growth chambers, at 30±2 ºC temperature. The light was provided for 8 hours a day by using 2 numbers of white long fluorescent tube lights. As and when required, water was sprinkled on paper to maintain sufficient quantity of moisture. The data recorded were statistically analyzed by using the technique of analysis of variance as suggested by Panse and Sukhatme (1987) [15].

The observation, germination percent under laboratory condition was recorded by counting the germinated seeds 28 days after sowing and expressed in percentage. Emergence of plumule was taken as a criterion for germination (Jun inouye *et al*., 1966) [10]

Germination percent under field condition

Germination counts were made at 30 days after sowing and expressed in percentage.

Seedling diameter

The seedling diameter was recorded with help of digital vernier caliper at an interval of 60, 120, 180, 240, 300, 360 and 420 days after sowing and expressed in cm.

Results and Discussion

Germination percent under laboratory condition

The observation recorded indicated that there were significant variations among different treatments. The treatment of GA_3 100 ppm (T_2) recorded highest germination (88.86%) as well as maximum percentage increase over control (21.32%) and it was significantly superior over rest of the treatments followed by NAA 100 ppm i.e. T_4 (85.62%) and KNO_3 1.0% i.e. T_5 (83.15%) whereas lowest germination (73.24%) was recorded in control. Table (1)

Table 1: Effect of growth regulators, chemical and organic wastes on seed germination under laboratory condition

Treatment	Germinaiton (%)	Increase over control (%)
T_1	81.97	11.91
T_2	88.86	21.32
T_3	79.64	8.73
T_4	85.62	16.90
T_5	83.15	13.53
T_6	82.17	12.19
T_7	80.79	10.30
T_8	80.11	9.30
T_9	75.02	2.43
T_{10} (control)	73.24	-
$SE_{m\pm}$	2.322	-
CD @1%	6.984	-

Germination percentage under field condition

The highest germination (69.88%) as well as maximum percentage increase over control (29.40%) was observed in seeds treated with GA_3 100 ppm i.e. T_2 and it was significantly superior over rest of the treatments followed by i.e. T_4 NAA 100 ppm with respect to germination (64.67%) as well as percentage increase over control (19.75%), i.e. T_1 GA_3 50 ppm (61.33%, 13.57%) and i.e. T_6 KNO_3 2.0% (61.33%, 13.57%). The Lowest germination (54.00%) was recorded in control.

Table 2: Effect of growth regulators, chemical and organic wastes on seed germination under field condition

Treatment	Germination (%)	Increase over control (%)
T_1	61.33	13.57
T_2	69.88	29.40
T_3	58.00	7.40
T_4	64.67	19.75
T_5	60.00	11.13
T_6	61.33	13.57
T_7	58.00	7.40
T_8	61.00	12.96
T_9	54.67	1.24
T_{10} (control)	54.00	-
$SE_{m\pm}$	1.682	-
CD @5%	5.052	-

There are variations among different treatments. i.e. T_2 GA_3 100 ppm recorded highest germination (88.86%) as well as maximum percentage increase over control (21.32%) and it was significantly superior over rest of the treatments followed by i.e. T_4 NAA 100 ppm (85.62%) as well as percentage increase over control (16.90%). Table (2)

The promotive effect of GA_3 on seed germination might be due to increased alpha amylase activity which catalyses the starch conversion into simple carbohydrates and chemical energy is liberated which is used up in the activation of embryo (Shepley *et al.,* 1973) [16]. The results are in conformity with the findings of Shant and Rao (1973), Abohassan *et al.,* (1979) [1] in Kagzi lime, Choudhary and

Chakrawar (1980 and 1981) [4, 5] in Kagzi lime and Rangpur lime, Gupta (1989) [7] in Kagzi lime, Kalalbandi (2002) in Rangpur lime and Sulabha Kherdekar (2003) [17] in Kagzi lime. In the present experiment under laboratory condition, the germination percentage was higher when compared to the corresponding treatments under field condition. This may be attributed to the unfavorable soil conditions and soil fauna which could adversely affect the germination of Rangpur lime seed under field conditions. Further laboratory condition, temperature was constantly maintained (around 30 ºC) with the maximum fluctuation of ± 2 ºC, whereas, the same was not controlled in field conditions. Hence, these two reasons might have contributed to the differences in the values observed in laboratory and field condition experiment. Similar observation was also made by Kamel and Nanda (1986) [11] in *Cassia fistula* seeds. Higher germination in case of GA_3 pre-soaking Rangpur lime seed in this study may be due to the fact that the exogenous application of GA_3 antagonizes application the effect of inhibitors (Brian and Hemming, 1958 and Wareing *et al.,* 1968) [19]. GA_3 helps in synthesis of enzymes. One of these enzymes α-amylase converts the starch into simple sugars. These sugars provide energy that is required for various metabolic and physiological process of germination. Gibberellin also enhances cell elongation, so the radical can push through the endosperm, seed coat that restricts its growth (Hartmann and Kester, 1972) [8]

Seedling diameter

At 60 days, highest stem seedling diameter of Rangpur lime was obtained in i.e. T_4 NAA 100 ppm (0.24 cm) which was highly significant over rest of treatments. This treatment was at par with i.e. T_2 GA_3 100 ppm (0.23 cm), i.e. T_6 KNO_3 2.0% (0.23 cm), i.e. T_1 GA_3 50 ppm (0.22 cm), i.e. T_5 KNO_3 1.0% (0.22 cm) and T_8 Cow urine 100% (0.21 cm). However all the treatments were significantly superior over the control (0.16 cm). After 120 days, i.e. T_4 NAA 100 ppm noticed high value for stem diameter (0.28 cm) followed by T_1 GA_3 50 ppm (0.27 cm) and T_2 GA_3 100 ppm (0.27 cm). The minimum stem diameter (0.22 cm) was recorded in control. All the treatments were significantly superior over the control.

At 180 days, maximum stem diameter (0.33 cm) was recorded in i.e. T_4 NAA 100 ppm which is closely followed by i.e. T_6 KNO_3 2.0% (0.32 cm), i.e. T_1 GA_3 50 ppm (0.31 cm) and i.e. T_2 GA_3 100 ppm (0.31 cm), i.e. T_3 NAA 50 ppm (0.30 cm) and T_7 Cow urine 50% (0.29 cm). All the above treatments were significantly superior over the control (0.25 cm). Data on 240 days, indicated that, i.e. T_4 NAA 100 ppm recorded that maximum stem diameter (0.50 cm) followed by T_2 GA_3 100 ppm (0.42 cm), i.e. T_6 KNO3 2.0% (0.41 cm) and T_1 GA_3 50 ppm (0.39 cm). Minimum stem diameter was observed in control (0.29 cm). At 300 days, maximum stem diameter obtained in T_4 NAA 100 ppm (0.53 cm) and lowest diameter recorded (0.41 cm) in control. After 360 days, highest stem diameter was recorded in T_4 NAA 100 ppm (1.15 cm) and the lowest diameter in control (0.92 cm). All the treatments were significant and were at par except T_9 i.e. soaking of seed in cow dung paste (0.95 gm). It was seen that, at 420 DAS, the highest stem diameter was obtained in T_4 NAA 100 ppm (1.67 cm) followed by T_6 KNO_3 2.0% (1.57 cm). The lowest stem diameter was obtained in control (1.23 cm). All the treatments was significant over control except T_9 soaking of seed in cow dung paste (1.37). table (3)

Table 3: Effect of growth regulator, chemical and organic wastes on seedling diameter (cm)

Treatment	Seedling diameter (cm)						
	60 DAS	120 DAS	180 DAS	240 DAS	300 DAS	360 DAS	420 DAS
T_1	0.22	0.27	0.31	0.39	0.48	1.05	1.47
T_2	0.23	0.27	0.31	0.42	0.51	1.12	1.60
T_3	0.21	0.26	0.30	0.37	0.43	1.02	1.43
T_4	0.24	0.28	0.33	0.50	0.53	1.15	1.67
T_5	0.22	0.22	0.28	0.35	0.44	1.03	1.40
T_6	0.23	0.24	0.32	0.41	0.52	1.07	1.57
T_7	0.20	0.22	0.29	0.35	0.43	1.00	1.43
T_8	0.21	0.23	0.28	0.37	0.51	1.03	1.50
T_9	0.20	0.22	0.27	0.31	0.43	0.95	1.37
T_{10}	0.16	0.22	0.25	0.29	0.41	0.92	1.23
$SE_{m\pm}$	0.010	0.012	0.016	0.019	0.028	0.044	0.059
CD @5%	0.030	0.036	0.046	0.056	0.082	0.129	0.176

The increase in stem diameter with NAA, GA_3, KNO_3 and cow urine may be due to cell expansion rather than cell division and supply of nutrient elements. The results are in accrodance with Choudhary and Chakrawar (1980) [4] in Kagzi lime and Agha *et al.,* (1990) [2] in orange and citrange. These findings are also in conformity with the findings of Kumar and Prasad (1997) [13] in papaya, Nema *et al.,* (1996) [14] in khirni and Jadhav (2000) [9] in Jambhiri and Rangpur lime rootstock.

Conclusions

From the experimental findings, following conclusions may be drawn in respect of germination percentage and seedling diameter. Under field condition, seeds treated with GA_3 100 ppm and NAA 100 ppm exhibited superior result in germination percentage, number of seedlings per seed and overall growth of Rangpur lime seedlings. Under laboratory condition, seeds soaked in GA_3 100 ppm and NAA 100 ppm solutions exhibited encouraging results in seed germination. GA_3 100 ppm, NAA 100 ppm, KNO_3 2.0 per cent recorded maximum seed germination in very short period of time under laboratory conditions rather than field conditions. Highest seedling diameter was exhibited in NAA 100 ppm.

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