

GERMINATION STUDIES OF THE SEEDS OF CITRUS SINENSIS (L.) OSBECK

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ABSTRACT

Germination studies of the seeds of *Citrus sinensis* (L.) Osbeck were carried out in the laboratory. The seeds were subjected to chemical and physical treatments namely; soaking in hydrogen peroxide, soaking in concentrated tetraoxosulphate (vi) acid for different time interval and also treatment with Potassium nitrate, removal of seed coat and puncturing of seed coat. The result indicated that light promoted seed germination over dark germinated seeds. Treatment of punctured seeds with Potassium nitrate enhanced germination over the control and other treatments. The result indicated that the seed coat of *Citrus sinensis* are hard and prevent germination.

Keywords: Germination, Seeds, *Citrus sinensis*, treatments, Soaking

1. INTRODUCTION

Citrus sinensis (L.) Osbeck which is commonly called sweet orange and it was the most commonly grown fruit in the world. (Morton, 1987). Orange is an evergreen flowering tree reaching 25 ft(7.5m) in height, and has a rounded crown of slender branches. The twigs are twisted and angled, young ones may bear slender semi-flexible, blunted spines in the leaf axils. The fruit is globose, oblate or oval 6.5-9.5 cm wide, containing an essential oil. The outer rind which is the epicarp is orange or yellow in colour when ripe, the inner rind which is the mesocarp is white, spongy and non-aromatic. The pulp which is the endocarp is yellow, orange or more or less red in colour, consists of tightly packed membranous juice sacs enclosed in 10 to 14 wedge-shaped compartments which are readily separated as individual segments.

Citrus sinensis has several uses which includes edible uses, medical uses and industrial uses. In the home, oranges are commonly peeled, segmented and utilized in fruits cups, salads, gelatins and numerous other deserts, and as such garnish on cakes, meats and poultry dishes. They were also squeezed daily in the kitchen for juice (Wilton et al., 1945).

Seed germination is a crucial stage in the life history of plants. Germination is a non-reversible process. The result of germination is clearly visible. Germination includes a series of events which transform a dehydrated resting embryo with a barely detectable metabolism into one vigorous metabolism cumulating in growth and end with the elongation of the embryonic axis which results in the penetration of the structure surrounding the embryo by the radicle. (Bewley and black 1985; Bewley, 1997). Various plants require different condition for successful seeds germination. Seed formation is affected by environmental condition, however the most important external factors that determine germination of seed are temperature, water, oxygen and light (Siegal, 1990).

Seed germination is the return of metabolic activities and growth by the seed tissue to give rise to a new plant by the development of the embryo. Some seeds do not germinate immediately after dispersal even if suitable conditions of growth are provided. In this period growth of the seeds remains suspended and it is said to be in the rest or dormant stage. This phenomenon is called dormancy of seeds. It may occur due to immature embryo, hard or impermeable seed coat, and presence of inhibitors like abscissic acid.

2. MATERIALS AND METHODS

2.1 Source of Plant Materials- Matured orange (*Citrus sinensis*) were sourced from choba in Obio/Akpor Local Government area of Rivers State and taken to the laboratory. The orange were ruptured and the seed were collected. Dried and kept in a bottle and stored in a refrigerator.

2.1.1 Viability Test By Floatation Method- The seeds were Poured into a container water to test for viability. The once that floated were removed and are regarded as non-viable seeds, while the once that sank were used to carry out this experiment.

2.2 GERMINATION TEST- For each germination studies, ten (10) seeds of *Citrus sinensis* were put inside 9cm Petri dish lined with whatman's filter paper and replicated 3 times per treatment. Each Petri dish was moistened with 10ml water (depending on the treatment being carried out), placed in light or dark condition and incubated at 25°C.

2.3 SEED DORMANCY BREAKING TREATMENT- The seeds were subjected to various seed dormancy breaking treatment as outlined below;

2.3.1 Mechanical treatment by puncturing- Seed were punctured with needle and germinated in the light and dark condition and percentage germination were recorded after 7days.

2.3.2 Removal of Seed coat Seed coats were removed with blade and germinated in light condition and percentage germination were recorded after 7days.

2.3.3 Treatment with hydrogen peroxide (H_2O_2)- Seeds were soaked in hydrogen peroxide for different time intervals ranging from 5, 10, 20, 40, 60, 80, 120 minutes after which the seeds were washed in running tap water and germinated in light condition.

2.3.4 Treatments with concentrated tetraoxosulphate (vi) acid (H_2SO_4)- Seeds were soaked in tetraoxosulphate (vi) acid at various time intervals ranging from 2,4,6,60,80 and 120 minutes, after which the seeds were washed in running tap water and germinated in light condition.

2.3.5 Preparation of Potassium Nitrate (KNO_3)- Different condition of potassium Nitrate were prepared namely; 1mM, 10mM and 100mM concentrations were prepared by dissolving 0.101g, 1.01g and 10.1g of KNO_3 respectively into a liter of deionized water. The seeds were incubated in these KNO_3 concentration and germinated.

2.3.6 Treatment of Intact Seeds, Punctured Seeds and Seeds with Seed coat Removed in Potassium Nitrate- Intact seeds, punctured seeds and seeds with seed coat removed were treated with different concentrations of potassium Nitrate namely: 1mM, 10mM and 100mM and incubated in light condition.

3. RESULTS AND DISCUSSION

3.1 GERMINATION OF INTACT SEEDS

The results of the germination of intact seeds of *Citrus sinensis* are shown in figure 1.0.

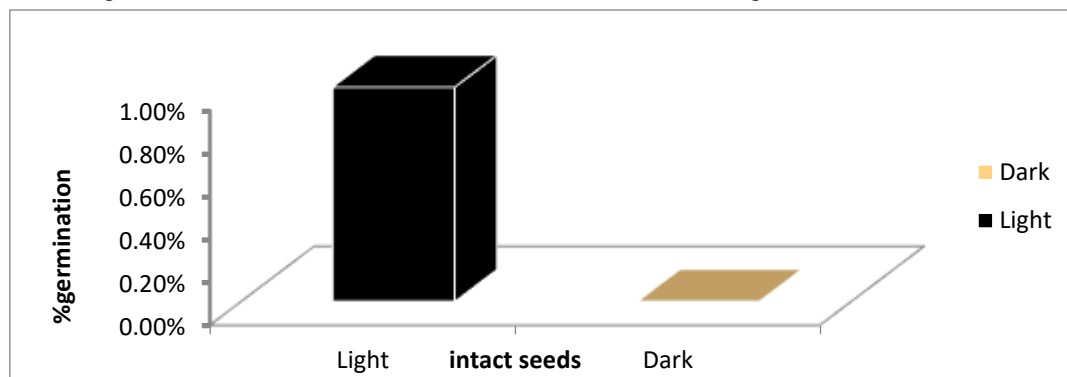


Figure 1.0 percentage Germination of intact seeds incubated in the light and dark condition at 25°C for 7 days.

The result showed that the germination of intact seeds incubated in the light was (2%) those germinated in the dark gave (0%), indicating that germination of intact seeds were poor in both light and dark conditions.

3.2 MECHANICAL TREATMENT BY PUNCTURING

Figure 2.0. showed the results of the germination of punctured seed of *Citrus sinensis*

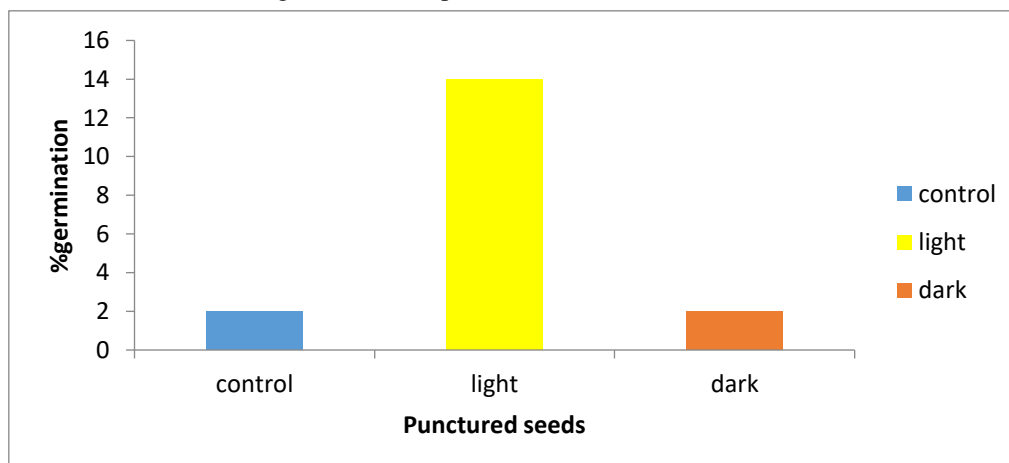


Figure 2 Percentage Germination of punctured seeds incubated in light and dark condition at 25°C for 7 days.

The results showed that there were improvement in the seeds punctured and exposed to light (14%) when compared to intact and punctured seeds germinated in the dark. Light enhance germination over intact and punctured seeds germinated in the dark

3.3 REMOVAL OF SEED COATS

The results of the germination of removal of seed coats of *Citrus sinensis* is shown in Figure 3.0.

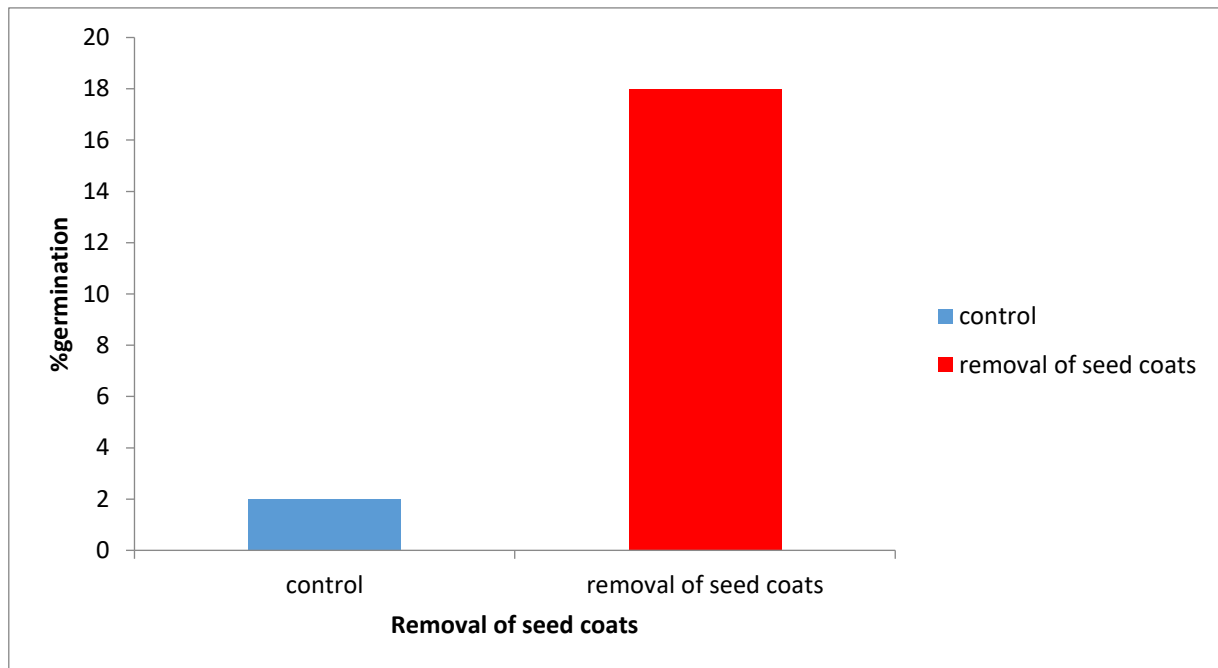


Figure 3.0 Percentage Germination of removal of seed coats incubated in light condition at 25°C for 7 days.

When the seed coats were mechanically removed, percentage germination were enhanced for seeds exposed in light when compared to intact seeds germination in light. Again, seed coat removal improves the germination of *Citrus sinensis* seeds.

3.4 TREATMENT WITH HYDROGEN PEROXIDE(H_2O_2)

The result of the germination of seeds soaked in hydrogen peroxide for various time interval is shown in Figure 4.0.

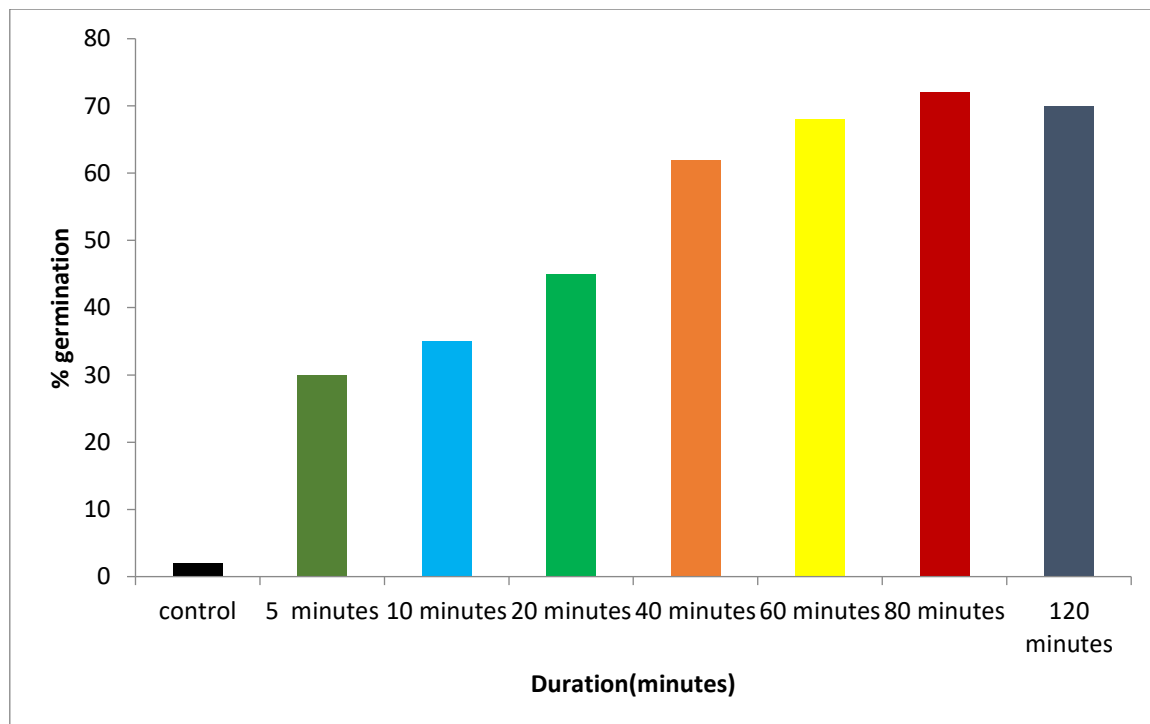


Figure 4.0 Percentage germination of seed soaked in hydrogen peroxide for various time interval and germinated in light condition at 25% for 7 days.

The results showed that there were an increase in germination with time of soaking except for 120 minutes treatment. 80 minutes treatment gave the highest percentage germination of 72% H_2O_2 is an oxidizing agent and may have acted on the seed coats to reduce its impermeability, allowing the entry of water and gaseous exchange.

3.5 TREATMENT WITH CONCENTRATED TETRAOXOSULPHATE (VI) ACIDS(H_2SO_4)

The result of the germination of seeds soaked in tetraoxosulphate(vi) acids (H_2SO_4) for various time interval is shown in figure 5.0

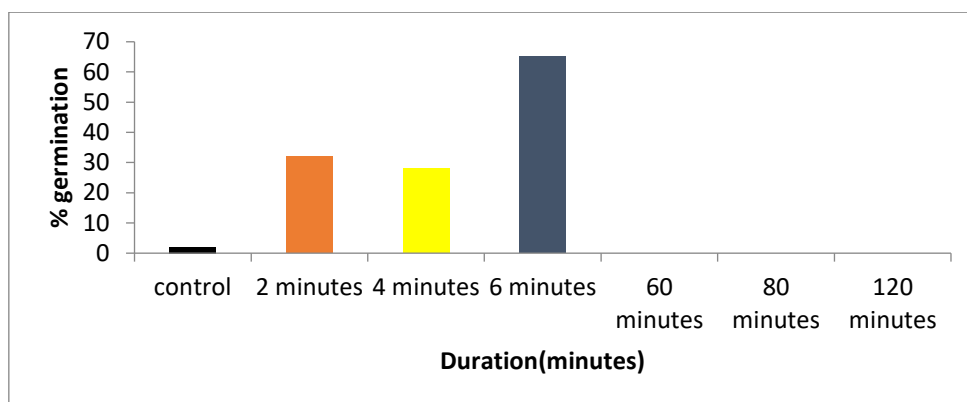


Figure 5.0 the Percentage Germination of seed soaked in tetraoxosulphate(vi) acid for various time interval and incubated in light condition at 25°C for 7 days.

The results showed that there were an increase in germination. 6 minutes treatment gave the highest percentage germination of 64%. At 60, 80, 120 minutes, germination were inhibited.

3.6 GERMINATION OF INTACT SEEDS IN POTASSIUM NITRATE

The result of the germination of intact seed incubated in different concentration of KNO_3 is shown in Figure 6.0.

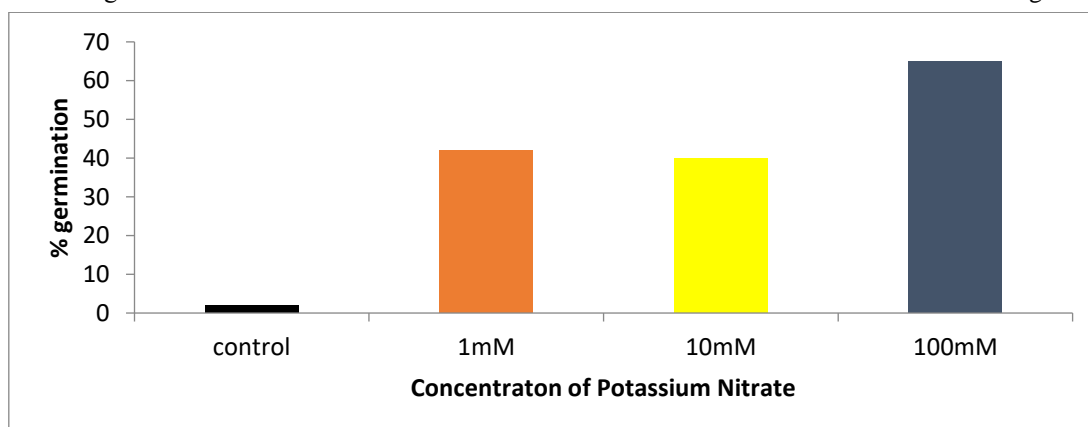


Figure 6.0: percentage Germination of intact seed germinated in KNO_3 and incubated in light condition at 25°C for 7 days.

The germination of intact seeds in Potassium Nitrate showed that Potassium Nitrate (KNO_3) enhance germination of intact seeds. 100mM treatment gave the highest germination (66%), 1mM gave 42% and 100mM gave the least germination (40%). Percentage germination were significantly increased over the control intact seeds.

3.7 GERMINATION OF PUNCTURED SEEDS IN POTASSIUM NITRATE

The result of the germination of punctured seeds incubated in different concentrations of KNO_3 is shown in figure 7.0.

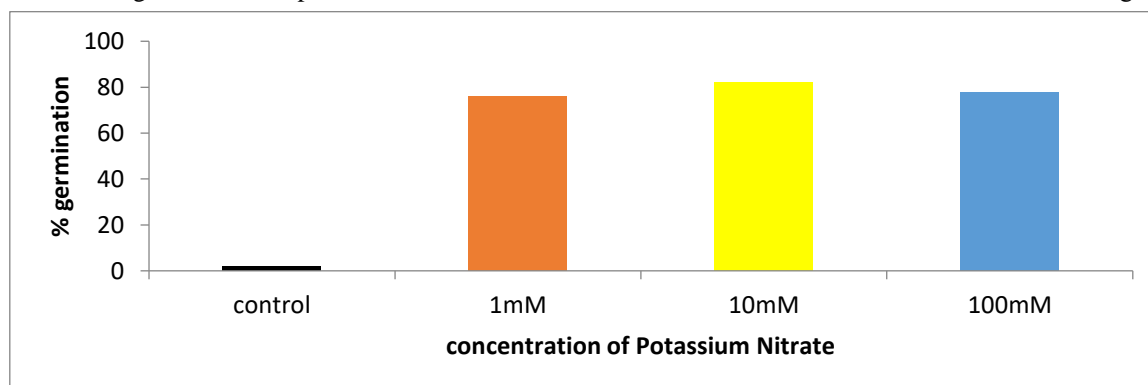


Figure 7.0 the percentage Germination of punctured seed germinated in KNO_3 and incubated in light condition at 25°C for 7 days.

The germination of punctured seeds incubated in Potassium Nitrate indicated that Potassium Nitrate promoted the germination of punctured seeds. 10mM treatment gave the highest percentage germination of (82%), following by 1mM with 76% and 100mM with 78%.

3.8 GERMINATION OF SEED WITH SEED COAT REMOVED IN KNO_3

The result of the germination of seeds with seed coat removed incubated in different concentrations of KNO_3 is shown in figure 8.0

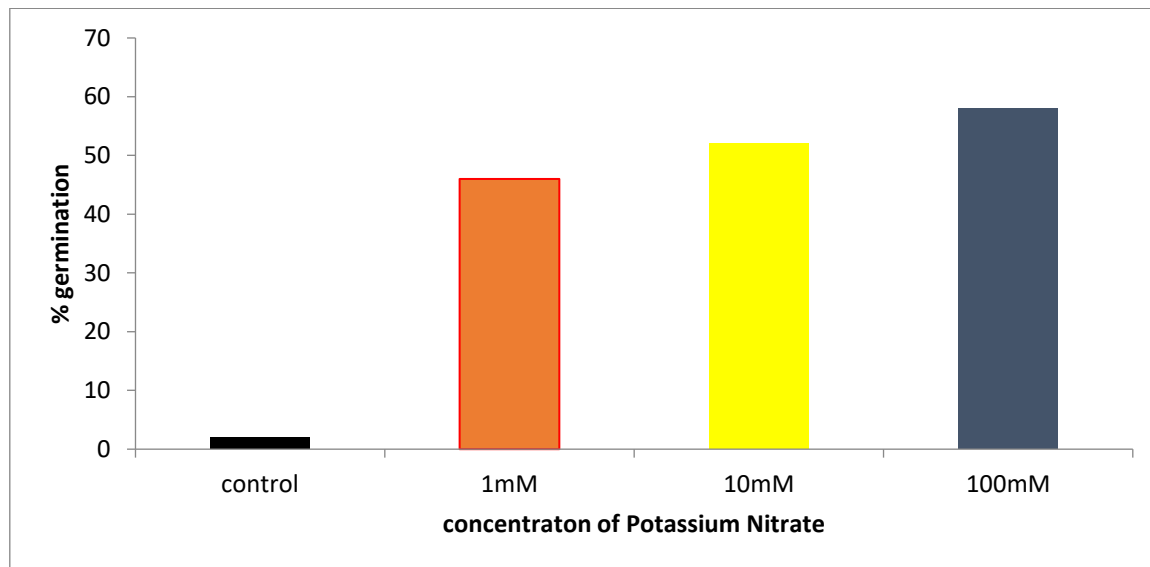


Figure 8.0: Percentage Germination of seeds with seed coat removed, incubated in KNO_3 and germinated in light condition at 25°C for 7 days.

The percentage germination of seed with seed coat removed showed gradual increase in germination with increase in concentration of potassium nitrate (KNO_3). 100mM had the highest germination (58%), followed by 10mM (52%), and then 1mM (46%).

4. DISCUSSION

Percentage germination of intact seeds of *Citrus sinensis* showed poor germination when germinated in the light (2%), and dark condition (0%). Intact seeds exhibit poor germination in both dark and light.

Puncturing of seeds and removal of seeds coat promoted germination (14%) and (18%) respectively when exposed to light conditions over intact seeds (2%) and punctured germination in that (2%). Seed coat of *Citrus sinensis* can be said to be impermeable to uptake of water and gaseous exchange. Germination in this case, is prevented by hard seed coat. Tungate et al., (2002) suggested that seed coat may restrict entrance of water and respiratory gases during imbibition and this may be the major cause of poor germination.

Hydrogen peroxide is an oxidizing agent. It oxidizes the seed coat of *Citrus sinensis* by making the seed coat permeable for water and oxygen and also breaking physical dormancy caused by the seed coat. Dormancy breaking often involves change in membranes.

This generally occurs only in within hydrated seeds (Derek, Bewley et al, 2006). Seeds of *Citrus sinensis* soaked in hydrogen peroxide (H_2O_2) germinated and showed progressive increase in germination with increase in time of soaking (figure 4.0).

Hydrogen peroxide greatly improve germination over the intact and other treatment (puncturing and seed coat removal) and this indicated that hydrogen peroxide can be used to improve the poor germination of seeds. 80minutes soaking in hydrogen peroxide gave the highest germination in the light condition (72%). Agrawal and Dadlani., (1995) reported that hydrogen peroxide improved the germination of *Poa pratensis*.

The soaking of *C. sinensis* in concentrated tetraoxosulphate(VI) acid showed that H_2SO_4 is an oxidizing agent that improve germination of seed in the light condition. 6 minutes acid treatment gave the highest germination (64%). Treatment beyond 6 minutes inhibited germination.

The germination of intact seeds, punctured seeds and seeds with seed coat removed (figure 6.0,70 and 8.0) indicated that potassium nitrate (KNO_3) improved germination over the control. Finch, Clay and Dent (2002) reported that potassium nitrate broke dormancy and improved germination in seed of *Prunus avium*.

5. CONCLUSION

Citrus sinensis seeds are dormant and showed poor germination due to hard seed coat which makes it impermeable to water and gases required for germination.

Treatment such as removal of seed coats, soaking of seeds in concentrated tetraoxosulphate (vi) acid, treatment with hydrogen peroxide and treatment with potassium nitrate promoted germination of Citrus sinensis by the process of softening and oxidizing the seed coat. In nature, soil contains potassium nitrate and this may enhance germination of Citrus sinensis.

Treatment of punctured seeds with potassium nitrate was the most effective followed by the soaking of seeds in hydrogen peroxide and treatment of intact seeds with potassium nitrate.

6. REFERENCES

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