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Germination Biology of Bell (*Passifloria burifolia* L.) and Sugar Apple Seeds (*Annona squamosal* L.) in Southern Nigeria

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ABSTRACT: Germination biology of two species of the apple – Bell apple (*Passifloria burifolia* L.) and Sugar apple (*Annona squamosal* L.) was undertaking to determine the possibility of growing apple in Southern Nigeria. Some seeds were sown directly in a garden soil in polythene bags, while other seeds were subjected to different treatments like scarification with sand paper, in boiling water for 1 minute, placing in 10% H_2SO_4, puncturing seeds with needle and soaking seeds in tap water for 24hours. Seedlings of Bell apple and Sugar apple were planted directly in garden soil using the square system method of planting. Effects of different treatments on seed germination and growing of seeds and seedlings were investigated. Seedling growth parameters such as number of leaves, measurement of stem girth, leaf area and plant height for both seedlings grown from seeds and seedlings planted directly into the garden soil were studied at a 7day (weekly) interval. The water holding capacity of soil, nutrients availability, soil conductivity and pH were also investigated. Observations revealed that seeds soaking enhanced the rate of germination of the seeds of sugar apple, and the garden soil enhanced the growth rate of seedlings of Bell apple.

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The most important aspect of plant development is germination. It describes the protrusion of the radical from a seed coat. Germination also refers to the formation of a sporeling from a spore, such as hyphae spores from fungal spores. Though, in a broad sense, germination can be described as the development of a small life or germ into a larger being (Raven *et al.*, 2005). Fully developed seeds contain an embryo and a store of food supplies, which are covered in a seed coat in most plant species. Some plants have a variable number of seeds that lack embryos, which do not germinate and are referred to as dormant seeds. Dormancy is a measure for plant survival or perennation. Dormant seeds are ripe seeds that do not germinate because external environmental factors inhibit metabolic processes and cell growth from getting started. Under ideal conditions, seeds germinate, and embryonic tissues resume growth, eventually forming seedlings. Physical dormancy is caused by impermeable seed coats that prevent oxygen from entering the seed. When the seed coat wears away enough to allow gas exchange and water absorption from the atmosphere, this is broken. The metabolic and growth rates of cells are affected by temperature. Different types of seeds germinate at different temperatures, as do seeds from the same plant. Seeds germinate within a temperature range, and they can germinate above or below that range. Many seeds germinate at temperatures 16 - 24 ^{o}C. As a result, certain seeds need to be exposed to cold temperatures (Vernalization) in order to break their dormancy. Physiological dormancy is a form of dormancy in which seeds depend on temperature to break their dormancy. Some seeds can only germinate if they are exposed to high temperatures during a forest fire, causing their seed coats to crack. This is a type of physical inactivity (Baskin and Baskin, 2014). Crop germination is influenced by both internal and external influences. The most important external variables are the correct temperature, water, oxygen or air, and sometimes light or darkness (Ravan *et al.*, 2005). Different plants need different variables for successful seed germination. This is also determined by the seed variety and is closely linked to the plant's natural environment's ecological conditions. The possible germination response of some seeds is influenced by environmental factors during seed formation; the majority of these responses are seed dormancy (Ravan *et al.*, 2005). Germination can be triggered by light or darkness, which is a form of physiological dormancy.

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Despite the fact that most seeds are unaffected by light or darkness, many seeds, especially those found in forests, will not germinate until an opening in the canopy allows enough light for seedling growth (Raven *et al.*, 2005). The development of certain plants is hampered by delayed germination. Examples of such are the bell and sugar apple seeds (*Passifloria burifolia* L. and *Annona squamosal* L.). Apple trees can be found all over the world, with the *Malus* genus being the most widely planted. The tree's wild ancestor, *Malus sieversii*, can still be found in Central Asia, where it is believed to have originated. Apples have been grown in Asia and Europe for thousands of years, and European colonists brought them to North America. China produced half of the world's apple production in 2017, which totaled 83.1 million tonnes (FAOSTAT 2017). Many people plant apple seeds in their gardens in the hopes of growing apples that are similar to store-bought apples, or at the very least to feed their families, only to discover after a lot of time and effort that the apples they harvest are nothing like the original. Aside from the fact that the new tree would bear little or no resemblance to the parent apple from which the seed was derived, successfully and timely growing apple seedlings can be difficult. Since apples are sold along the road, on the street, and all over Nigeria, the current project (research), which centres on investigating the possible best method of growing these species of apple (Bell and Sugar apple) in the southern part of Nigeria with reference to Edo State, is expected to have a high economic value.

MATERIALS AND METHODS

Collection of Seeds and Seedlings Material: Seeds of apple and seedlings of two species of apple, bell apple (*Passiflora burifolia*) and sugar apple (*Annona squamosa*) were however, graciously provided by National Institute of Horticultural Research and Training (NIHORT) Ibadan, Oyo State, Nigeria. A total of eighty seeds of apple and eighteen seedlings of the two species of the apple were obtained.

Seed viability test: The seeds were put in a bowl of water and left for 15 mins. Submerged seeds were collected and used, while the ones that remained afloat were discarded. Seventy five seeds submerged and were used for the research work.

Garden soil: Twenty polythene bags, each containing 10kg of garden soil were prepared and three seeds were sown in each bag of garden soil at 3cm depth. A total of sixty seeds were sown. The garden soil was obtained from the staff residential quarters in the University of Benin environment. Germination records were taken on a seven day interval up to 42 days. Seeds that failed to sprout after 42 days were regarded as not germinated.

Soil physicochemical parameters: Soil physicochemical analyses of soil used in the experiment was conducted as provided in Bray and Kurtz (1945a,b); APHA (1985)

Seed treatment: Fifteen seeds were separately subjected to different pre-sowing treatment as follows; a set of seeds were scarified using sand paper; while another were punctured using needle. The third set of seeds were soaked in hot water for 2 mins, and the fourth soaked in tap water (pH 6.7 – 7.2) for 24 hrs. The fifth group was soaked in 10% H_2SO_4 for 1 min.

Germination: All the treated seeds were placed on moist filter paper and put in Petri dishes. Appearance of the radicle at the micropyle region of the seed occurred seven days after sowing, in case of the seeds soaked in tap water for 24hours. Germination did not occur with seeds given other treatments.

Planting of seedlings: Seedlings of the eight bell apple (*Passiflora burifolia*) and eight sugar apple (*Annona squamosa*) were planted in garden soil prepared measuring 60 x 60 x 60cm dimension at a distance of 5m adopting square planting system. Watering of the soil was done after planting and later done every three days.

Seedlings measurement: Measurement of height, stem girth, leaf area and number of leaves of seedlings were observed periodically and values recorded.

Statistical analysis: The data shown in the tables were mean ($\overline{X}$) values taken in triplicate from each sample investigated. The statistical method employed was the analysis of variance according to Sokal and Rohlf (1973).

RESULTS AND DISCUSSION

Table 1 shows soil physicochemical paranmeters prior to sowing. Soil was slightly acidic and had a nitrogen content of 0.53% and an available phosphorus level of 4.15 mg/kg respectively.

Table 1: Chemical Composition of Garden Soil Used for Planting Seedlings and Seeds of Apple

Nutrients parameter	Garden soil value
Ph	6.2
Organic carbon (%)	1.64
Total organic matter (%)	2.19
N (%)	0.53
Avail. P (mg/kg)	4.15
K (Cmole/kg)	0.41
Ca (Cmole/kg)	4.01
Mg (Cmole/kg)	2.26
Na (Cmole/kg)	0.44

Table 2: Germination of the Apple Seeds Treated/Scarified Before Sowing On Filter Paper.

	Germination					
Time (days)	7	14	21	28	35	42
Treated seeds						
(a) *Annona squamosa*						
Control	0	0	0	0	0	0
Seeds scarified with sand paper	0	0	0	0	0	0
Seeds punctured with needle	0	0	0	0	0	0
Seeds soaked in hot water for 2 min	0	0	0	0	0	0
Seeds soaked in tap water for 24hrs	0	67.9	67.9	67.9	67.9	67.9
Seeds soaked in 10% dilution of H_2SO_4 for 1 min	0	0	0	0	0	0
(b) *Passiforia burifolia*						
Control	0	0	0	0	0	0
Seeds scarified with sand paper	0	0	0	0	0	0
Seeds punctured with needle	0	0	0	0	0	0
Seeds soaked in hot water for 2 min	0	0	0	0	0	0
Seeds soaked in tap water for 24hrs	0	0	33.3	67.9	100.0	100.0
Seeds soaked in 10% dilution of H_2SO_4 for 1 min	0	0	0	0	0	0

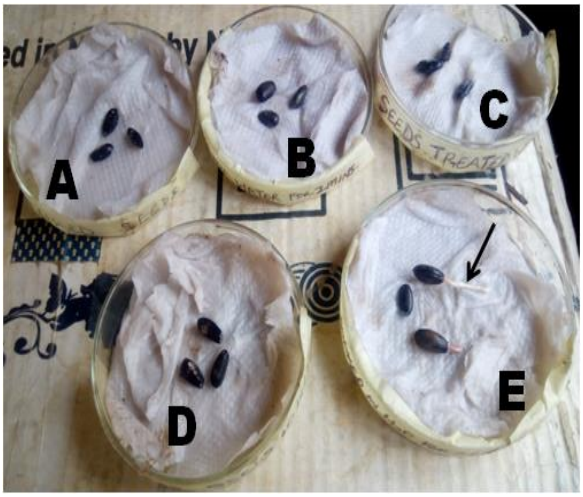

Plate 1: Seeds of the apple seed subjected to different treatment and placed on moist filter paper in Petri dishes. A -Seeds scarified with sand paper, B -Seeds punctured with needle, C- Seeds soaked in hot water for 2 min, D - Seeds soaked in 10% dilution of H2SO4 for 1 min, E - Seeds soaked in tap water for 24hrs

Plate 2: Seedling grown from seeds soaked in tap water for 24 hours.

Plate 3. (a) *Annona squamosa* and (b) *Passiforia burifolia*) seedling obtained as seedling at 35 days after planting.

Plate 4: (a) Sugar apple and (b) Bell apple plants at age 350 days after planting in garden soil, showing a newly developed fruit (white arrow).

The control seeds did not germinate within 42 days after germination (Table 2, Plate 1). Similarly, those

seeds treated with hot water, acid or scarification did not germinate as well. However, presoaking the seeds in tap water for 24 hours resulted to seed germination after one week. For *P. burifolia*, the earlier seed germinated at the third week, but only when seeds were presoaked in tap water for 24 hours.

Results showed selected plant growth parameters after 350 days of sowing. No significant differences in leaf area ($p>0.05$) were reported; leaf area in the bell apple plant was 89.21cm^2, compared to 90.61 cm^{2}in the sugar apple plant (Table 3, Plates 2 – 4). There were significantly more leaves in the bell apple plant (126) than in the sugar apple (91) respectively. Whereas the sugar apple had had a fruit, the Bell apple had none.

Table 3: Plant Parameters at 350 Days after Sowing

Parameters	Bell apple (*Passifloriaburifolia*)	Sugar apple (*Annonasquamosa*)	p-value
Leaf area (cm^2)	89.21±12.33	90.61±4.32	0.374
Stem girth (mm)	7.62±2.11	7.82±0.62	0.621
Plant height (cm)	121.3±21.34	129.4±11.32	0.118
*No. of primary branches	13±2	10±4	0.301
*No. of secondary branches	2±1	4±2	0.089
*No. of leaves/plant	126±11	91±9	***0.028***
5 leaf weight (g DM)	4.6±1.3	3.8±1.6	0.119
*No. of fruits per plant	0	1	NA

**Mean presented to the nearest integer*

For many years now, apple fruits are purchased along the road and the streets of most parts of Nigeria. Apples are grown in some parts of northern Nigeria and presently propagated by the Research Institute in the south west of the country. The fruit is not grown by farmers, by choice in the southern part of the country; hence the quest to undertake this research on how it can be grown in the southern part of the country. In order to actualize this objective, seeds of apple fruits were got from the roadside vendors examined with a view to germinating them. It was thus observed that the seeds of the apple fruit sold along roadside and on the street could not germinate, even when scarified. This is because these seeds from such fruits are polyploidy in nature. The search for apple seeds within Nigeria that can germinate was obtained successfully with seeds and seedlings of sugar apple and bell apple from National Institute of Horticultural Research and Training (NIHORT) Ibadan, Oyo State, Nigeria. These seedlings were successfully grown. The two set of seedlings showed different growth rates. The bell apple seedlings showed rapid growth in height, leaf number and leaf length. However, the sugar apple seedlings showed stunted growth in height, reduced leaf number and leaf length when compared to the bell apple seedlings. The leaves of both seedlings showed peculiar morphological features of bell apple and sugar apple respectively. The bell apple showed opposite arrangement of leaves with long sharp and pointed leaf tips; while the sugar apple seedlings showed alternate arrangement of leaves with blunt tips. The germination of the seeds was found to be epigeal. It was observed that thirty eight out of the sixty seeds sown, germinated. This amounted to 63.3% germination of the total seeds sown in garden soil in polythene bags. The seed that was treated by soaking in tap water for 24hours had 66.7% germination. At day 7, there was no germination of any of the seeds sown in garden soil in polythene bags. However, few germination of seeds (nine seeds) was observed in day 14 and maximum germination of twenty seven (27) seeds was observed in day 21. This is an indication that the *Annona squamosa* seed require more time (twenty one (21) days) for germination. For the germination of *Annona squamosa* using different treatments, mechanical scarification using sand paper resulted in 0% germination. Also, seed puncture using needle resulted in 0% germination. Likewise seeds soaked in hot water for 2minutes and seeds placed in 10% H_2SO_4 for 1 minute showed no germination. However, seeds soaked in tap water for 24 hours showed 66.7% germination in less than 21 days. Heydecker and Coolbear (1977) reported that pre-soaking of seeds in water increased percentage of germination in many species. It was observed that the number of days for germination was shortened from 21 days to 7 days. The failure of the seeds of *Annona squamosa* to respond to boiling water treatment, scarification with sand paper, soaking in boiling water and placing in 10% H_2SO_4 treatments may be due to loss of viability. The seedlings from the seeds soaked in tap water for 24 hours showed more relative growth rate and development of more leaves initially as compared to the seedlings that were sown directly in the garden soil in polythene bags. The value of soil nutrients analysis to the critical values of soil nutrients required for the germination and relative growth of *Annona squamosa* indicates that nitrogen, phosphorus and potassium in the garden soil are within the range of the critical values of the recommended standard values of humus soil needed for apple germination and growth (Muhr *et al.*, 1965).

Conclusion: The present study has shown that apple seeds can be grown by farmers in the southern part of Nigeria. There is however, to be a major component of input from research, for seeds that would germinate.The present study may not have been conclusive in its findings, because of time constraint, since the results were not followed up to fruiting of both plants. The present study however, has given a pointer to debunk the notion that apple cannot be grown in southern part of Nigeria.

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