EVALUATION OF TREE SEED GERMINATION CAPACITY: *EUCALYPTUS MICROCORYS* IN SOUTHERN PROVINCE OF RWANDA

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Abstract

In Rwanda, most trees and shrubs are propagated using seeds but seed germination information is limited due to inadequate research. Most of people claim that after acquiring tree seeds they meet problems comprising low germination rates of seeds, abnormality of seedlings, impurity and others constraints. The aim of this study was to evaluate the germination capacity of tree seeds. *E. microcorys* was used in the experiments which consisted of three treatments (T1: Rwasave nursery soils, T2: Ruhande Arboretum soils and T3: Sterilized sandy soils) with three replications. The temperature has been controlled at 25-35^{0}C at laboratory level. All tests were examined daily to ensure that the moisture content of the substrate is near optimum. The laboratory germination began after three days while the nursery started in nine days after sowing. It was recommended to all foresters and farmers that to ensure the germination of seeds, they must consider the type of seeds, the limiting environmental factors distinguished as edaphic (soil depth, fertility, texture and structure, presence of excessive assimilable carbonates or chlorides); climatic (temperature, aridity and humidity) and biotic (pests, diseases and competition from vegetation) and reflect on all silvicultural management practices.

Keywords: *Silviculture, Eucalyptus microcorys seeds, Germination capacity, Tree nursery.*

1. INTRODUCTION

The Forestry sector is playing key roles in supporting the livelihood of all Rwandans especially by providing most of the energy consumed by the bulk population. The roles include controlling soil erosion, protecting water catchments and supplying other goods and ecological services (MINIFOM, 2010). The forest cover now is low as 9% of total area of Rwanda (MINAGRI, 2012). The recent inventory of forestry resources in Rwanda, however, revealed that forestry and woodlands cover an estimated area of 240,746 ha per 10% of the total land (Rutabingwa, 2008). Forests contributed up to 80% of total energy needed in 2007 (BEST, 2009).

The amount and quality of the gained yield in the form of seeds depend on many factors i.e. genotype, environmental conditions, seed storage, natural or unnatural selection as well as conscious seeds ennoblement (Sekutowski *et al.*, 2015).

Broadly, most of trees (eucalyptus, grevillea, cyprus, alnus sp. etc) are spread using seeds but the germination capacity is low due to inappropriate tree nursery practices, management by foresters and inadequate research (Mng'omba *et al.*, 2007).

Furthermore, *E. microcorys* is an exotic tree comes from New South Wales and Queensland Australia. The first introduced in Ruhande Arboretum was coming from Pretoria in Republic of South Africa (Kalinganire A., 1988*).* It grows in forests near the coast on moderate to well fertile drained soils in a protected sunny position. Tallow wood is drought and frost tender. The precipitation varies from 900 to 1500 mm/year and the temperature of average 30^0C. The dormancy of the semen of Eucalyptus has a satisfactory germination at the optimal temperature; in condition that they are viable and mature (Turnbull, 1977). The high evapotranspiration of the forest prevented rapid leaching of salts from the soil complex, in which salts were present but not in dangerous amounts (FAO, 1979).

The Eucalyptus forest soils have a fairly low pH; they are likely to be deficient in phosphorus and also nitrogen, in spite of numerous tree species, shrubs and herbs of legumes under the dominant eucalypts.

In East African countries particularly in Uganda, *E. microcorys* has proved a preferred species because lower branches of edge trees are persistent and downward sweeping (Kingston, 1974).

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In addition, the germination test determines the maximum germination potential, or viability, of the seed. The germination rate of a particular seed lot is a key indicator as to how that seed will perform at the field (20/20 Seeds Labs Inc., 2010).

We emphasized on increases in the effectiveness of these appreciated exotic tree species propagation, as the plantation of such species can significantly contribute not only to protect watersheds and environment but also to hasten the process of creating the vegetation cover on deforested areas. Besides, we suggest that the production of exotic adaptive tree species, which can be planted during restoration of deforested areas, is an issue worth responsibility to use in generating direct monetary income (revenues) for households, public entities and increasing the biodiversity.

The aim of the conducted research was to evaluate the germination capacity of tree seeds using *E. microcorys* in Southern region of Rwanda.

2. MATERIALS AND METHODS

2.1 Description of the Study area

Ruhande Arboretum is located (altitude: 1737m; latitude 2°36´S and longitude 29°44´E) in Southern Province of Rwanda, Huye District, Ngoma Sector. The average annual rainfall is 1,232 mm and the average annual temperature (T^0):19.6^{0}C. The botanical garden is a plantation forest of 200 hectares organized into 529 plots intercalated by alleys of about 6 m wide (Burren, 1995). This forest was created in 1934 under the request of the former resident of the colonial territory of Rwanda-Urundi (Musafiri, 2014; Ndayishimiye and Nyirabuhoro, 2015).

According to Rwanda Agricultural Board (RAB), Arboretum of Ruhande is the largest arboretum in Africa and unique forestry resource internationally appreciated for its fine collection of Eucalypts (Burren, 1995). It is composed of over 207 native and exotic species, including 143 hardwoods with 69 Eucalyptus species, 57 softwood and 3 bamboo species (Nsabimana , 2013 and Varhammar *et al*. 2015).

The soil in arboretum is classified as a Ferralsols according to FAO (1998), formed from the parent material of schists and granites mixed with mica schist and quartzite (Steiner, 1998; Verdoodt and Van Ranst, 2003).

2.2 Seed Sample Collection

A sample of seeds of *E. microcorys* stored in Ruhande Tree Seeds Center (CGF) was randomly collected. The seed identity was defined by its N° of seed lot: 574/012, date of collection: August, 2012; local name:

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inturusu and provenance: Ruhande-UR. The sample was weighted using the electrical balance.

2.3 Methodology

Experimental design

a. In laboratory

The seed sample was 10gm which was divided into three equal parts. Each sample had 3.33grs. We made three replications and two treatments (T1: Soil from Rwasave tree nursery, T2: Soil from Ruhande Arboretum and T3: Sterilized sandy soil [Control]).

The following formula was used for to evaluate the seed purity of *E. microcorys*:

$$Purity\ (\%) = \frac{Weight\ of\ pure\ seed\ (gr)}{Total\ weight\ of\ working\ samples\ (gr)}\ x\ 100$$

(ISTA, 1996)

The amount of water to be added to the sand was calculated as follows (*Anonymous, 1952).*

$$X\ ml\ of\ water\ to\ be\ added\ to\ each\ 100grs\ of\ sand = \frac{118.3ml}{Weight\ of\ 118.3\ of\ sand\ in\ gr}\ x\ (20.2\text{-}8.0)$$

The Laboratory Germination Capacity (LGC) is an estimate of the maximum number of seeds likely to germinate under optimum conditions. The total time required for the test depends on species where 4 weeks were enough for Eucalypts *(*Palzer, 2002*)*. For Eucalyptus species, it is usually determined as number of viable seed per gram. However, it is calculated by a simple formula below:

$$LGC\ (\%) = \frac{Number\ of\ germinated\ seeds}{Total\ number\ of\ seeds\ in\ dish}\ x\ 100$$

(ISTA, 1996)

After determining the LGC, we can calculate the number of viable seed per kg as follows:

$$Number\ of\ viable\ seed\ per\ kg = \frac{N\ X\ LGC}{100}$$

(ISTA, 1996); where *N: number of seed in 1kg.*

b. At nursery level

The seed sample was 10grs which was divided into three parts as the treatments (T1: 3.0grs [Control], T2: 3.4grs and T3: 3.6grs). The treatments differed from each other by their quantity. Three replications were used.

Based on routine nursery operations, the likely percentage germination was the same as FGC. Therefore,

$$Number\ of\ germinated\ seeds = \frac{N\ X\ FGC}{100}$$

(*ISTA, 1996*)

Likewise, the total number of seedlings in any sample *E. microcorys* can be calculated by the portioned method after determining the number of seedlings that raised in the testing sample.

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The percentage of germinated seed that grow to produce a live seedling at the end of the period in the nursery can be called ‘‘*S*’’. Thus, *the number of germinated seedlings from 1 kg of seeds* = $\frac{N\ X\ S}{100}$ *(ISTA, 1996)*

Not all the live plants in the nursery at the time of planting will be of suitable quality to plant because some do not meet the basic criteria of ‘target seedlings’. The poor quality seedlings must be culled and discarded from the planting programme. The percentage of seedlings that are of acceptable quality for planting can be called ‘‘*P*’’.

The number of quality seedlings from 1 kg of seeds = $\frac{N\ X\ P}{100}$ *(ISTA, 1996)*

The number of quality seedlings per kilo of seeds at the end of the growing period in the nursery is estimated as: $\frac{N\ X\ FGCXSXP}{1000000}$ *(ISTA, 1996)*

Once the total seedling requirement for a species is known, it is possible to calculate the total seeds requirement by the following formula:

Total weight of seed required (kg) = $\frac{N\ X\ 1{,}000{,}000}{N\ X\ FGCXSXP}$ *(ISTA, 1996)*

3. OBSERVATIONS AND RESULTS

a. Laboratory analysis

Seed purity test

The purity analysis of a seed sample in the seed testing laboratory refers to the determination of the different components of the purity *viz.*, pure seeds, other crop seeds, weed seeds and inert matter (TNAU, 2014). The purity of *E. microcorys* was 98% where at the impurity counted 2%. We found that in 10 gr of *E. microcorys*, there is 9.8gr of pure seeds and 0.2 gr of the inert matters (Table 1).

Table 1. Estimation of seeds purity of *Eucalyptus microcorys*.

	Quantity of sample (gr)	Percentage (%)
Pure seeds	9.8	98%
Inert matter	0.2	2%
Total	10	

Laboratory germination capacity

Seeds were planted in plates containing washed sand (particle size diameter 0.6–0.8 mm), moistened with distilled water. Each petri dish was covered with paper filter on which treated seeds of eucalyptus were sown

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3.33grs per petri dish. Three replications were used in this experiment. The seeds sowed in the sterilized sandy soil (control), Rwasave tree nursery and Ruhande soils showed a high number of germinated seeds of 95000, 68100 and 67300 seedlings respectively. The LGC is 40.9%, 29.1% and 29.1% with respect to the treatments. The total of number of seeds germinated in 1 kg was estimated at about 230.400 seedlings, which resulted at 99.3% of sown seed (Table 2). The environmental conditions such as climatic (temperature, aridity and humidity) and biotic (pests, diseases and competition from vegetation) factors were controlled after setting up the experiments. The temperature have been maintained at 25-35^{o}C

Table 2. Estimation of germination rate and number of germinated seeds

Treatments (T)	Average number of seed germinated in 1kg	LGC (%)
1	68,100	29.3
2	67,300	29.1
3	95,000	40.9
Total	**230,400**	**99.3**

Where: **T1**: Soil from Rwasave tree nursery, **T2**: Soil from Ruhande Arboretum and **T3**: Sterilized sandy soil [Control].

Germination capacity with time

The seed germination period of *E. microcorys* ranged between 3-8 days from sowing date. The number of germinated seeds vary with the time (Figure 1). The day one showed a high number of seed compared to the last day (day 5).

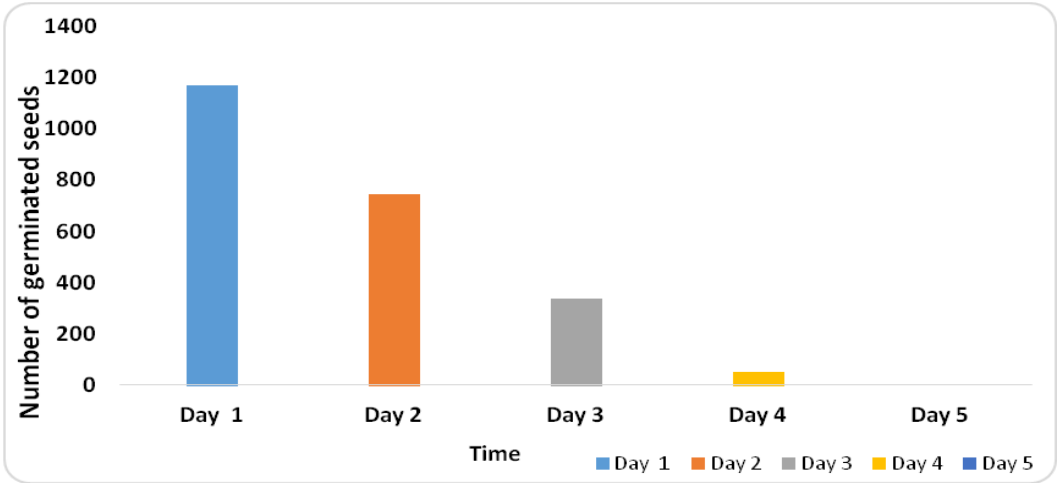

Figure 1. *E. microcorys* seed germination dynamics

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Laboratory viable seedlings

A viable seed is one which is capable of germination under suitable conditions (Bradbeer, 1988). For Eucalyptus species, it is usually determined as number of viable seed per gram. The number of viable seed per 1kg is 228,787 seedlings (Fig. 2).

The maximum number of seeds likely to germinate under optimum conditions was 99.3 % of the total number of the seeds in 10 gr (Table 2). The germination rate of each petri dish marked a high germination capacity of 40.9% and more viable seeds (95,000 seedlings).

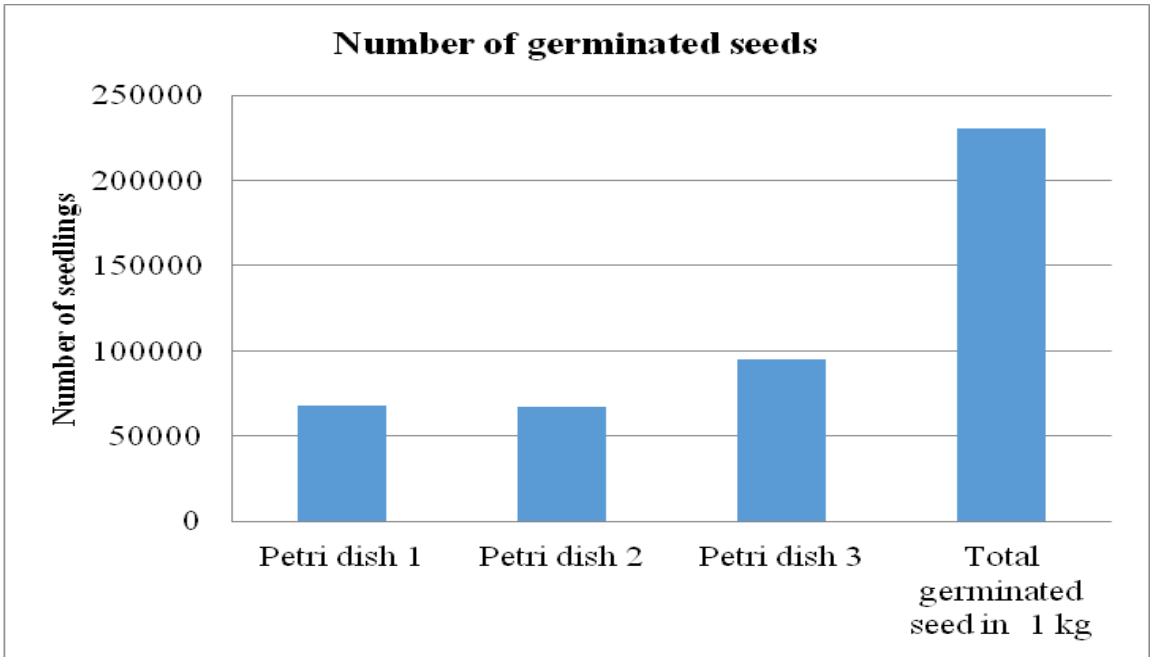

Figure 2. Number of germinated seeds

b. Nursery analysis

Field Germination Capacity (FGC) and number of seedlings

The seeds started to germinate after 9 days from the sowing date. The germination process took about 30 days. The germinated seeds were 64.200 seedlings which was equivalent to FGC of 28.86%.

Quality seedlings at the end of nursery

The results in table 3 showed that the number of seedlings pricked out per kg are 49.600 seedlings which are equivalent to 77.2%; the germinated seed that survived to produce quality seedlings 49.562 seedlings which are equivalent to 99.9% while quality seedlings at the end of the nursery activities

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were 45.462 seedlings which was equivalent to 91.7%.

Table 3. Illustration of total quality seedlings at the end of nursery

Items	QSS	Percentage (%)
GS	64,200	28.86
SP	49,600	77.2
GSSQ	49,562	99.9
QSENA	45,462	91.7

Where QSS: Quantity of survived seedlings; GS: Total number of germinated seeds per one kg; GSSQ: Germinated seed that survived to produce quality seedlings; QSENA: Quality seedlings at the end of the nursery activities and SP: Number of seedlings pricked out per one kg.

c. Statistical analysis

The ANOVA (table 4) confirmed that the difference are highly *significant* at a level of 5% probability ($p > 0.05$) depends upon the quantity sown. The coefficient of variation is 0.0012%. The standard deviation is 137.2438. The correlation coefficient is $R^2 = 0.9865$; this means that the correlation is moderately perfect.

Table 4. Analysis of variance (ANOVA)

SV	SS	df	MS	F	P-value
Between Groups	150677.89	6	25113	5.58E+03	.000**
Within Groups	9	2	4.5		
Total	150686.89	8			

**= highly significant; CV% = 0.0012; STDEV = 137.2438

4. DISCUSSIONS

Ranal and Santana (2006) reported that it is clear that researchers, gardeners, seed sellers and foresters must choose the measurements, environmental factors (light intensity, day length, night length, light color, water, water quality, gravity, crowding, temperature, nearby plants (by chemical agents), genetics, oxygen availability, seed condition, seed age, seed coat condition, seed size and other environmental conditions can have measurable effects on seed germination), and interval of evaluation according to their objectives.

Gardeners, worldwide, have a number of ideas of other environmental factors that may influence germination such as the phases of the moon, tidal effects, and planting with companion seeds. In order to germinate and break dormancy a seed has to

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absorb quite a bit of water (Ranal and Santana, 2006).

Germination is normally carried out in germination cabinets under controlled environment (Lars, 2000). Besides, the laboratory and nursery results indicated that the germination rate in laboratory is higher than the germination rate in the nursery due to the controlled conditions affecting the germination such as the weather (high moisture content), the temperature and the types of soils (moist sand) used to test. Also, we observed that the number of germinated seed per kg in laboratory is 230,400 seedlings whereas in nursery is 64,200 seedlings. Soil temperature plays a key role in promoting seed germination (Schonbeck and Egley 1980). The temperature has been maintained at 25^0C where the germination marked a high level (99.3%).

Germination is defined as 'the emergence and development of the seedling to a stage where the aspects of its essential structures indicate whether or not it is able to develop further into a plant under favourable conditions in the soil' (ISTA 1996 and ISTA, 1998). The exact criteria of evaluation vary slightly between species, e.g. in eucalypts a seed is considered to have germinated when the radicle has developed normally and the cotyledons have emerged from the seed coat and have unfolded (Boland et al. 1980).

On the other hand, the timing of germination or the breaking of the dormancy is important to the success of the young seedling and the seed has to somehow respond to signals in its environment in order to germinate at appropriate times (Brad W., 2003 and Donohue K., 2002). The reason why in laboratory the germination begun after 3 days and took 5days (Fig. 1) whereas in nursery begun after 9 days and took about 30days as the temperature and moisture were maintained in laboratory.

The results (table 2 and 3) showed that the LGC is 99.3% whereas FGC is 28.86%. With reference to Lars (2000), the germination or viability test should indicate the potential germinability which, with proper handling, should reflect expected germination in the nursery. Germination potential is most directly determined in a germination test: under the appropriate conditions everything that can germinate should germinate.

The results of this experiment showed a high correlation ($R^2 = 0.9865$) and the results are highly significant at a level of 5% of probability due to the quantity of seed sown (Table 4).

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In the experiments carried out in laboratory and nursery level, the results showed that *E. microcorys* seeds are viable and have a high germination rate when they are treated in good conditions (ISTA, 1996) by considering all factors affecting germination of seeds and storage behaviors.

Watering, weeding around the out-planted seedling and the protection against big herbivores enhances seedling growth. The separated effects of these factors could be helpful for the out-planted seedling management.

Forest trees of Rwanda are under enormous deforestation pressure and many other catastrophes such as diseases, damages by insects, pests and hence more research work is warranted to enhance tree seeds propagation and domestication programs. Furthermore, more research work is needed to determine proper handling, nursery care practices and storage of tree seeds, particularly for exotic tree seeds that are not well known in the region. There is no conclusive evidence that seed provenance influences seed germination capacity.

Finally, this study evaluated the seeds germination capacity and seedling production, we recommend all foresters and researchers to consider all silvicultural management practices when propagating and producing seeds.

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