

EFFECTS OF GERMINATION ECOLOGY ON IN VITRO GERMINATION PERFORMANCE OF HIGHLAND BAMBOO (*Yushania alpina*) SEED COLLECTED FROM KEFA, SOUTH WEST ETHIOPIA

Belete G. Tesfaye^{1*}, Yigardu M. Mengesha², Smegnew M. Birlie^{2*}, and Marshet N. Gebeyehu³

^{1,2} Central Ethiopia Environment and Forest Research Center, Addis Ababa Ethiopia

² Ethiopian Environment and Forest Research Institute (HQ), Addis Ababa, Ethiopia

³ Jimma Environment and Forest Research Center, Jimma Ethiopia

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EFFECTS OF GERMINATION ECOLOGY ON IN VITRO GERMINATION PERFORMANCE OF HIGHLAND BAMBOO (*Yushania alpina*) SEED COLLECTED FROM KEFA, SOUTH WEST ETHIOPIA. *Yushania alpina* is an African endemic bamboo species, and it is a valuable resource in ecological and socioeconomically value in Ethiopia. However, low germination is a challenge for seedlings production besides its seed availability is scarce. To improve the seed germination capacity, using different germination ecology treatments are needed. Hence, this study was initiated to investigate the effects of different germination ecology, and to determine the qualities, size, and yield of *Y. alpina* seed. The collected matured fruits were processed, and then the cleaned (pure) and not cleaned (impure) seed were used for this study. This study had two phases; the first was to measure the purity, moisture content, seed character, seed yield; and the second was to investigate the effects of different germination ecology for in vitro seed germination of *Y. alpina* using pure and impure seeds. In this result, the purity, moisture content, seed size, seed weight, and grain yield were determined for cleaned *Y. alpina* seed. The interaction effects of seed type and germination ecology were highly significant on all germination parameters. The highest germination capacity (55%) of pure seed was recorded on T2 (paper + ambient temperature), followed by 38% on T1 (sand + ambient temperature), and 31% on T3 (paper + incubator (25°C)); however, these treatments produced lower results in the impure seed. In addition, the highest (23.5 days) mean germination time was recorded on T3, followed by 13.06 on T2 using impure seed, but the lowest (2.5 and 2.01) value was recorded on T1 and T2 of pure seeds. Hence, this result concluded that using sand media at ambient temperature as germination ecology is preferred to enhance the germination capacity of *Y. alpina* seed. Also, seed surface disinfection using antifungals is recommended to reduce seed contamination.

Keywords: Germination ecology, germination, *Y. alpina*, pure and impure seed

PENGARUH EKOLOGI PERKECAMBAHAN TERHADAP KINERJA PERKECAMBAHAN IN VITRO BENIH BAMBU DATARAN TINGGI (*Yushania alpina*) DARI KEFA BARAT DAYA ETHIOPIA. *Yushania alpina* adalah spesies bambu endemik Afrika, dan merupakan sumber daya yang berharga dalam nilai ekologi dan sosial ekonomi di Ethiopia. Namun demikian, daya berkecambah yang rendah merupakan tantangan dalam produksi bibit selain ketersediaan benih yang langka. Untuk meningkatkan daya kecambah benih diperlukan perlakuan ekologi perkecambahan yang berbeda. Oleh karena itu, penelitian ini dilakukan untuk menyelidiki pengaruh ekologi perkecambahan yang berbeda, dan untuk menentukan kualitas, ukuran, dan hasil benih *Y. alpina*. Buah matang yang dikumpulkan diproses, dan kemudian biji yang dibersihkan (murni) dan tidak dibersihkan (tidak murni) digunakan untuk penelitian ini. Penelitian ini memiliki dua fase; yang pertama adalah mengukur kemurnian, kadar air, karakter benih, hasil benih; dan yang kedua adalah menyelidiki pengaruh ekologi perkecambahan yang berbeda untuk perkecambahan benih in vitro *Y. alpina* menggunakan benih murni dan tidak murni. Hasil penelitian menunjukkan kemurnian, kadar air, ukuran benih, berat benih, dan hasil biji ditentukan untuk benih *Y. alpina* yang dibersihkan. Pengaruh interaksi jenis benih dan ekologi perkecambahan sangat nyata pada semua parameter perkecambahan. Kapasitas perkecambahan tertinggi (55%) benih murni tercatat pada T2 (kertas + suhu lingkungan), diikuti oleh 38% pada T1 (pasir + suhu lingkungan), dan

*Corresponding author: bgetnet704@gmail.com

31% pada T3 (kertas + inkubator (25°C). ; namun, perlakuan ini menunjukkan hasil yang lebih rendah pada benih yang tidak murni. Selain itu, waktu perkecambahan rata-rata tertinggi (23,5 hari) tercatat pada T3, diikuti oleh 13,06 pada T2 menggunakan benih tidak murni, tetapi nilai terendah (2,5 dan 2,01) tercatat pada T1 dan T2 benih murni. Oleh karena itu, hasil ini menyimpulkan bahwa penggunaan media pasir pada suhu kamar sebagai ekologi perkecambahan lebih disukai untuk meningkatkan kapasitas perkecambahan benih *Y. alpina*. Selain itu, desinfeksi permukaan benih menggunakan antijamur dianjurkan untuk mengurangi kontaminasi benih.

Kata kunci: Ekologi perkecambahan, perkecambahan, *Y. alpina*, benih murni dan benih tidak murni

I. INTRODUCTION

Yushania alpina (K. Schum) is a monocarpic plant that generates flowers and gives seeds once in its lifetime, and its growth is restricted to highland areas at altitudes varying from 2400 to 3500 masl (Ramanayake & Yakandawala, 1998). It propagates sexually and asexually from seed and vegetative parts, respectively. The vegetative propagation (through stump, rhizome, culm cutting, and offset) may be disastrous if the age of the source plant is too high besides its high labor cost requirement, difficult for transportation, and also it is not much liked for large scale propagation because of explants shortage could occur (Ayana, Gure, & Embaye, 2014; Embaye, 2003; Mulatu & Fetene, 2014). Hence, propagation through seed ought to be the priority if there is no germination challenge because of physical and biological behaviors. However, propagation of *Y. alpina* through seed remains a challenge to establish and develop small and large scale plantation forests due to long flowering cycles, poor storage characteristics, and short viability of seeds as a result of its recalcitrant, presence of disease and pests (Ayana et al., 2012, 2014; Bahru et al., 2015).

Moreover, seeds are a living biological end product of genetic and environmental interactions, and their behavior can't be predicted with certainty (Loha, Tigabu, Teketay, Lundkvist, & Fries, 2006; Mamo, Mihretu, Fekadu, Tigabu, & Teketay, 2006). Hence, seed germinations explain high variability between species and seed lots, and even within seed lots. In addition, effects of provenances on germination of different species' seeds and

seedlings related traits quality exhibited high significant differences besides the magnitude of genetic variation that is considerably more than the environmental variation (Bahru et al., 2015; Derero et al., 2012; Fredrick, Muthuri, Ngamau, & Sinclair, 2015; Loha et al., 2006, 2008; Mamo, Mihretu, Fekadu, Tigabu, & Teketay, 2006). Carles, Lamhamedi, Beaulieu, Stowel, Colas, & Margolis (2009) also reported the strong influence of genotype and growing environment of the mother tree on the weight and size of the seed; besides to this the required average time for germination can also be correlated positively with seed size and weight (Norden, Daws, Antoine, Gonzalez, Garwood, Chave, 2008).

In addition, a procedure for collection and storage of seed, pre-sowing treatment, and sowing media with the environment have determined the physical quality (purity), moisture content, and germination of the seed, which the latter depends on the condition of the first two parameters (Loha et al., 2006, 2008; Wang, Lu, & Zhao, 2009). So far, the storage period at which the seed is able to germinate and also can be used for planting and production depends on its genetic makeup and storage condition (Mrda, Crnobarac, Dušanić, Jocić, & Miklič, 2011). Hence, seed longevity is especially important for low-cost propagation for plantation forest development and to maintain the quality of seedlings. However, the *Yushania alpina* seed longevity is seriously very short and shows low germination performance. Hence, finding different seed germination enhancement techniques using growing media with environments, and different cleaning stage

seed is an urgent issue to foster and support the current interest of bamboo forest development endeavors. Therefore, the objective of the study was to enhance the germination potential by using different growing media with the environment in addition to determining seed size and yield, and seed quality of *Yushania alpina* seed for releasing alternative propagation technology to produce in mass and quality seedlings.

II. MATERIAL AND METHOD

A. Study Area Description

Mature fruits of *Y. alpina* were collected in August 2020 from Desta site, Buta Kebele of Adeyo District in the Kefa Zone of South Nation, Nationalities and Peoples Region, Ethiopia. It is far distance, about 530 km from Addis Ababa and lies between 7°8' to 7°26'N latitude and 36°15' to 36°50'E longitude. The elevation ranges from 500 to 3000m above sea level.

This study was conducted in the Tree Seed Quality Control laboratory of Central Ethiopia Environment and Forest Research Center, Addis Ababa. This laboratory had been established in 1975 in the forest sector, which is now well designed and organized in all aspects of tree seed quality test, and it is the only place for tree seed quality test in the country.

B. Seed Collection, Handling, and Processing

To ensure maximum genetic diversity, mature fruits were collected from ten selected clumps of bamboo stands at a distance of at least 100 m apart between them (FAO, 1975). The collected fruits stayed in dispersing and open for overnight to avoid mold and then put in perforated plastic bags to transport safely to the Central Ethiopia Environment and Forest Research Center (CE-EFRC). In the seed processing section, the fresh mature fruits were subjected to open-air drying on the net bed covered by nylon cloth for one week. After drying, it was rubbed to extract naked and

covered seeds, and then also further cleaned by using mortar and pestle for onward laboratory test, storage, and distribution. Finally, the cleaned seeds were placed in perforated plastic bags and stored at room temperature for experiment and distribution. Then, seed yield, seed size, seed quality test, and other different experimental tests were started and continued step by step within a month after the collection of the seeds.

C. Determination of Purity Analysis and Moisture Content of Seeds

After the collected seeds were processed, purity analysis and moisture content of *Y. alpina* seeds were determined in the CEE-FRC Tree Seed Quality Control laboratory following the methods used by FAO (1985). For purity analysis, two samples of 30 gm each were taken from the total seed lots of *Y. alpina* containing all the impurities and weighed. Following this, only the pure seeds were selected and reweighed separately. The percentage of pure seed was calculated as follows:

$$\text{Purity\%} = \frac{\text{Weight of Pure seed}}{\text{Total Weight of Original Sample}} \times 100 \quad \text{..(1)}$$

For moisture content analysis, two samples of 2 mg each were taken from the total seed lot of *Y. alpina*, and then the weighted sample was subjected to drying and the moisture content was determined using an automatic analyzer machine (Keren DBS -Kern & Sohn Company, Germany). This advanced machine worked both in the weighing and drying process; finally, it gave the moisture content result automatically within 15 to 30 minutes.

D. Determination of Grain Yield, Seed Size and Weight of *Y. alpina* Seed

The seed weight of *Y. alpina* was determined by following the FAO guide for forest seed handling with special reference (FAO, 1985). Accordingly, eight replicates of 100 pure seeds were randomly taken from the total seed lot and then the weight of thousand seeds was calculated to put as a report. Hence, the 1000

pure seed weight was converted to the number of pure seeds per kg:

$$\text{Number of seed/kg} = 1000\text{gm} \times \text{TSW}(\text{gm}) \dots\dots(2)$$

To measure the size of *Y. alpina* seed, a total of sixty seeds in 3 replicates of 20 seeds per unit were randomly taken from the total seed lot of *Yushania alpina*. Following this, the seed length and seed width/girth were measured in centimeters (cm) using a seed caliper (Mamo et al., 2006).

E. The Effect of Growing Media and Environment on *Y. alpina* Germination Capacity

As growing media with the environment is the most important factor for germination, different growing media such as moisture blotting paper and pure sand were prepared by using 11 cm and 16.5 cm diameter petri dish, respectively before the moisture content was done. The treatments were set from their growing media and environment, i.e.; T1= sand + ambient temperature, T2 = paper + ambient temperature, and T3= paper + incubator (25°C) were tested for pure and impure seed. A total of hundred seeds with four replications were used for each treatment, and it was laid out in a completely randomized design (CRD) in the laboratory bench and germination cabinet. To maintain the optimum moisture, the experiment was sprayed equally using distilled water as required through the observation. Finally, data on the germinated seed was recorded in five-day intervals until 30 days of sowing. Also, the number of seedlings raised per kg of seeds was also calculated after the germination percentage was obtained using the following formula:

$$\frac{\text{No of seedlings raised}}{\text{kg of seeds}} = \frac{\text{No. of seeds}}{\text{Kg}} \times \text{germination}(\%) \times \text{purity}(\%) \dots\dots(3)$$

F. Germination Percentage and Mean Germination Time

1. Germination Percentage

Pure seed samples were taken from the stock stored at room temperature, and hence a total

of sixty seeds were used to conduct germination test. Seeds were sown uniformly and watered as needed to keep moist but not wet. Then after continues attendant germination data record took place. Accordingly, germination percentage was the dividend of germinated and total sowed seed, and its value was expressed in percent as follows:

$$\text{Germination Capacity}(\%) = \frac{\text{Total Germinated Seed}}{\text{Total Sowed Seed}} \times 100 \dots\dots(4)$$

The emergence or development of a radicle from the seed embryo was considered as germinated seed to evaluate germination capacity (FAO, 1985). The number of germinated seeds was recorded through seven days intervals, and also abnormal seeds, seeds infected with fungus, and not geminated seeds were considered non-viable.

2. Mean germination Time (MGT)

MGT was also calculated by adopting Eq.5 from Ellis and Roberts (1980 as cited in Mavi et al. (2010).

$$\text{MGT} = \frac{\sum Dn}{\sum n} \dots\dots\dots(5)$$

Where :
n is the final germinated seeds on day D, and D is the total days of the germination time.
Germination speed/Peak Value – which is adopted from Ayana, Tadesse, and Kebede (2012)

$$\text{PV} = \frac{\text{GP}}{\sum Dn} \dots\dots\dots(6)$$

Where:
PV= peak value, GP= germination percentage,
Dn= total germination days

G. Data Analysis

Germinated seed, seed yield and quality, seed size, and others were collected as required. Germination percentage, mean germination time, peak value, seedlings produced from 1 kg seed (SPs/Kg), purity%, moisture%, and seed size and yield were calculated from the collected raw data using simple statistics in MS excel 2016. In addition, germination percent, peak value, mean germination time, and seedlings production per 1 kg seed (Sps/

Kg) data were subjected for analysis of variance using SAS 9.4 version (SAS, 2008). Also, the data were evaluated using tables and graphs. Then, Duncan multiple range test (DMRT) at 5% confidence interval was used for mean separation.

III. RESULT AND DISCUSSION

A. Analysis of Variance

The analysis of variance revealed that the interaction effect of seed cleaning stage (SCS) and treatment is very highly significant ($P \leq 0.001$) on germination percentage (GP), mean germination time (MGT), and seedlings production per 1kg seed, but only highly significant on Peak value (PV), i.e., germination speed (Table 1). In addition, both factors alone are also very highly significant on GP, PV, MGT, and SpKg, however, the treatment is highly significant at PV ($P \leq 0.01$). Accordingly, Bahru, Mulatu, and Kidane (2015) and Bahru et al. (2012) also reported the significant effect of different seed germination ecology (presowing treatment) on the germination performance of Ethiopian indigenous bamboo species. However, the germination performance is also highly influenced by the growing ecology of the seed sources. Due to this, the significance of germination treatments in this study was agreed with the previously mentioned reports, but the magnitude of germination percentage obtained from *Yushania alpina* seed was completely different. This is due to variations from genetics through segregation and the mother plant's growing agroecology condition.

B. Determination of Purity Analysis and Moisture Content of Seeds

According to the FAO guideline, (1985), the purity and moisture content of *Y. alpina* seed were found at 88.4% and 3–5.4%, respectively (Table 1). Hence, the seeds are said to be pure seeds, and also the sowing material moisture content has a vital conditioning effect for storage, germination energy, and capacity (Domin et al., 2020). Purity analysis of a given seed sample is the first test to identify mature, pure, and germinating seeds for storage and further tests such as the determination of moisture content, seed weight, and seed length and width (FAO, 1985). Also, seed moisture has an influence on seed longevity in the storage, and hence checking the moisture level of the seed in the given sample is very crucial before storage (FAO, 1985). This seed purity (88.4%) is slightly better, however, its moisture content range (3–5.4%) is highly lower than that of compared with Bahru et al. (2015) who reported 86% and 6.9–8% for *Arundinaria Alpina* seed purity and moisture content percent, respectively; this might be due to the difference of seed source location and its time of collection.

The analysis of the seed moisture content gave 5.4% when tested within ten days after collection, but it was gradually declining to 3% after a month which is very low according to the seed moisture standard range (ISTA, 1993; Hartmann & Kester, 1983) and it leads to low germination capacity. This indicates that the germination capacity depends on the moisture content of the seed which is supported by the

Table 1: ANOVA summary of Seed Germination Parameters of *Yushania alpina* L

Variation Source	DF	GP MS	PV MS	MGT MS	SpKg MS
SCS	1	6016.67***	0.60***	758.93***	8642.35***
Treatment	2	482.00***	0.03**	161.02***	692.31***
SCS*Treatment	2	232.67***	0.02**	136.48***	334.03***
CV		10.29	25.73	28.86	10.29

Remarks: *** = very highly significant ($p \leq 0.001$), ** = highly significant ($p \leq 0.01$), SCS= Seed Cleaning Stage, MS = Mean Square, SpKg = Seedlings production from 1 kg seed

reports of Loha, Tigabu, Teketay, Lundkvist, and Fries (2006), and Loha, Tigabu, and Teketay (2008) stated the complex adaptive traits of seed germination which depends on the condition of purity and moisture content parameters in addition to its genes makeup and environmental factors. Wang, Lu, and Zhao (2009) also stated the influence of temperature, storage time length, and light on seed germination, which are great determinants to maintain seed moisture content and viability.

In addition to low germination capacity, the low moisture content could also affect the growth of some of the radicals into shoots which means it becomes dead rather than developing shoots and roots for release into the nursery as a seedling. Because *Y. alpina* seed is a recalcitrant seed and has short longevity which are the major causes that the radicals could die rather than grow to complete seedlings as reported by Embaye (2003) but not only this, it might be due to loss or absence of enough endosperms as a source of food for the seed embryo to stay alive and to have enough energy for germination. Furthermore, the germination capacity has also been determined by the size and weight of the seed besides the influence of moisture and purity parameters.

C. Determination of Grain Yield, Seed Size and Weight of *Y. alpina* Seed

Grain yield is an important parameter to determine the extracted seed output from the collected fresh fruit biomass, and know how to have enough amount of seed for planned bamboo species propagation through seed besides being cost-effective by minimizing collection time and cost. According to this, about 201.7 gm of extracted seed yield was obtained from 1500 gm of collected fresh fruit, which is 13.5% of the total collected biomass (Table 2). This indicates that the grain yield of *Y. alpina* is very low, and needs to collect a huge amount of fresh fruit biomass because above 86% of biomass yield is changed to trash or straw when it was processed and cleaned to produce pure seed. However, the seed size and its weight are key determinant factors for grain yield and germination capacity or percentage.

In this study, the mean value of *Y. alpina* seed length and width were 0.47 and 0.25 cm, respectively, and also the weight of thousand seeds was recorded at 7.38 gm which is highly against the result of Bahru et al. (2015) who reported 17 gm of thousand seed weight from the *Y. alpina* sample collected from the Dawa district Guji zone, Oromia Regional State, Ethiopia. This might be due to the difference of seed size (length and width) difference that is influenced by environmental factors.

Table 2: Determination of seed size and quality parameters

Species	Purity (%)	MC (%)	TSW (gm)	No of Seed /kg	Seedlings /kg	SL (cm)	SW (cm)
<i>Yushania alpina</i>	88.4	3 -5.4	7.38	135567.945	65,913.14	0.47	0.25

Remarks: MC – moisture content, TSW – thousand seed weight, SL – seed length, SW – seed weight

Table 3: Determination of grain yield and tree diameter

Species	Min DBH (mm)	Max DBH (cm)	FFW (gm)	ESY (gm)	ESY (%)
<i>Yushania alpina</i>	1.3	6cm - 19cm	1500	201.7	13.45

Remarks: DBH – diameter at breast height, FFW – fresh fruit weight, ESY – extracted seed yield

Moreover, Carles et al. (2009) also reported the strong influence of genotype and growing environment of the mother tree on the weight and size of the seed; besides the required average time for germination can also be correlated to the seed size and weight (Norden et al., 2008). The lowest and largest seed length was also 0.4 and 0.5 cm whereas the lowest and largest seed width was also 0.2 and 0.3 cm, in respective order.

According to the mean seed weight determination, the computed number of seeds was 135, 567.95 in one kilogram of *Y. alpina* pure seed (Table1). Therefore, 65, 913.14 seedlings were raised from one kilogram of *Y. alpina* pure seeds using the first 5.4% moisture content with (55%) of seed germination capacity that was sowed on a petri dish with moist blotting Whatman paper, however, the germination capacity differed based on the growing media and the environment for the studied seed (Figure 1). This indicates that it has more than 43.41% seedlings advantages when compared with the reports of Bahru et al. (2015) who obtained 37,301 seedlings from 17 gm of thousand seed weight having 6.9–8% moisture content and 73% germination capacity of *Y. alpina* pure seeds. Moreover, there were 1.3cm and 6–19cm lowest and largest diameter at breast height of *A. alpina* natural forest trees, respectively, which seems to have corresponded with the maximum diameter (up to 20 cm) of indigenous bamboo species of Ethiopia (Embaye, 2003).

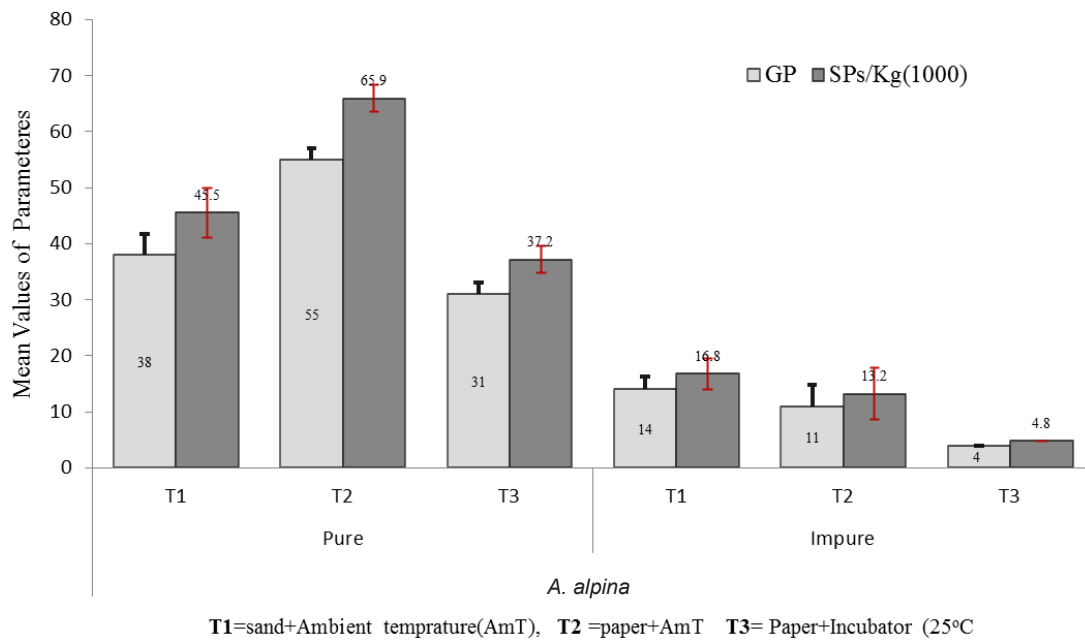
D. The Effect of Growing Media and Environment on Seed Germination Capacity

According to the ANOVA result in Table 1, the interaction of treatments and seed cleaning stage has shown significant influence on studied parameters in the germination of *Yushania alpina* seed. In this result, the combinations of two different factors; seed cleaning stage and seed germination ecology treatments showed different germination percentages. Pure seeds having 5.4% moisture content gave the highest germination percentage (55%) on T2 and also followed by 38% on T1 and 31% on T3 (Table 4; Figure 1). Such a wide range of germination percentage differences might be due to a very crucial factor of seed germination such as temperature and moisture variation of growing media, and the environment in addition to favorable internal conditions of the seed as reported by Hartmann and Kester (1983). Also, Yerima, Tiamgne, Fokou, Tziemi, and Van-Ranst (2015) reported that growing substrates had a significant effect on germination and seedling emergence. In addition, the highest germination obtained on T2 was recorded after the shoot emerged above the sand; hence there might be a probability that not all seeds that can radicle emerge as a shoot. Therefore, comparing the germination percentage of seeds sowed on Whatman paper, and sand growing media may not be always true, but the seed sowed on sand media is cost-effective, and applicable everywhere either at farmer level or nursery site of governmental and private institutions.

Table 4: Means of seed germination parameters of *Yushania alpina* L.

Genotype	SCS	Treatment	GP (mean±SD)	PV (mean±SD)	MGT (mean±SD)	Sps/Kg (mean±SD)
<i>Yushania alpina</i>	Cleaned seed	T1= Sand + AmT	38.00±3.65b	0.41±0.04ab	2.49±0.24c	45.54±4.37b
		T2= Paper + AmT	55.00±2.00a	0.52±0.14a	2.03±0.55c	65.91±2.40a
		T3= Paper + Incubator (25°C)	31.00±2.00c	0.33±0.06b	3.14±0.65c	37.15±2.40c
	Not cleaned seed	T1= Sand + AmT	14.00±2.31d	0.18±0.00c	5.59±0.10c	16.78±2.77d
		T2= Paper + AmT	11.00±3.83d	0.09±0.03c	13.06±5.07b	13.18±4.59d
		T3= Paper + Incubator (25°C)	4.00±0.00e	0.05±0.01c	22.75±2.63a	4.79±0.00e
CV			10.29	25.73	28.86	10.29

Remarks: the values having the same letter are not significantly different, AmT = Ambient Temperature



T1=sand+Ambient temprature(AmT), T2 =paper+AmT T3= Paper+Incubator (25°C)
 Figure 1. In vitro germination capacity of *Y. alpina* seed cleaning stage at different germination ecology treatments

On the other hand, the impure seeds having 5.4–6% moisture content gave the highest (14%) germination percentage on sand media at room temperature (T1) which was better than that of those sowed on T2 (11%) and (4%) on T3 (Table 4; Figure 1). The lowest germination capacity (4%) was recorded from the impure seed sowed on Whatman paper in a 25°C incubator (T3). As shown in Figure 1, the germination capacity of the impure seed was highly lower than that of the pure seed. This indicates that impure seeds should not be preferred to use as a valuable source of propagation for *Y. alpina* neither in small nor large-scale forest establishment and development purposes.

As compared to Bahru et al. (2015) who reported 73% germination of *A. alpina* pure seed having 0.59 cm length and 0.17 cm width, this highest recorded germination percentage (55%) of pure seed is very low. This might be due to the difference in seed size and agroecology of germplasm sources. Moreover, this might also be due to seeds living in biological end products of genetic and environmental interaction, in which the magnitude of genetic variation is considerably

more than the environmental variation (Bahru et al., 2015; Derero et al., 2012; Fredrick et al., 2015). Hence, the seed conditions explain high variability between species and seed lots, even between seed lots, across years and provenances due to environmental factors besides genetic differences (Loha et al., 2006, 2008; Mamo et al., 2006). Because of this, the seeds' behavior cannot be predicted certainly (Mamo et al., 2006). FAO (1985), and Ghosh and Singh (2011) also reported heavy and larger seeds had weighed more per seed, and hence they contained more food reserves which are more likely to have higher germination percentages by providing more energy to produce initially vigorous seedlings. Therefore, seed size and weight are important characters in the selection of well-adapted and highly productive seed sources.

In addition to the germination capacity of seeds, the influence of seed germination ecology treatments also determined the pattern and mean germination time. Accordingly, the lowest (2.01) mean germination time was recorded from pure seed sowed on T2 and followed by 2.5 on T1 (Table 4, Figure 2). However, the

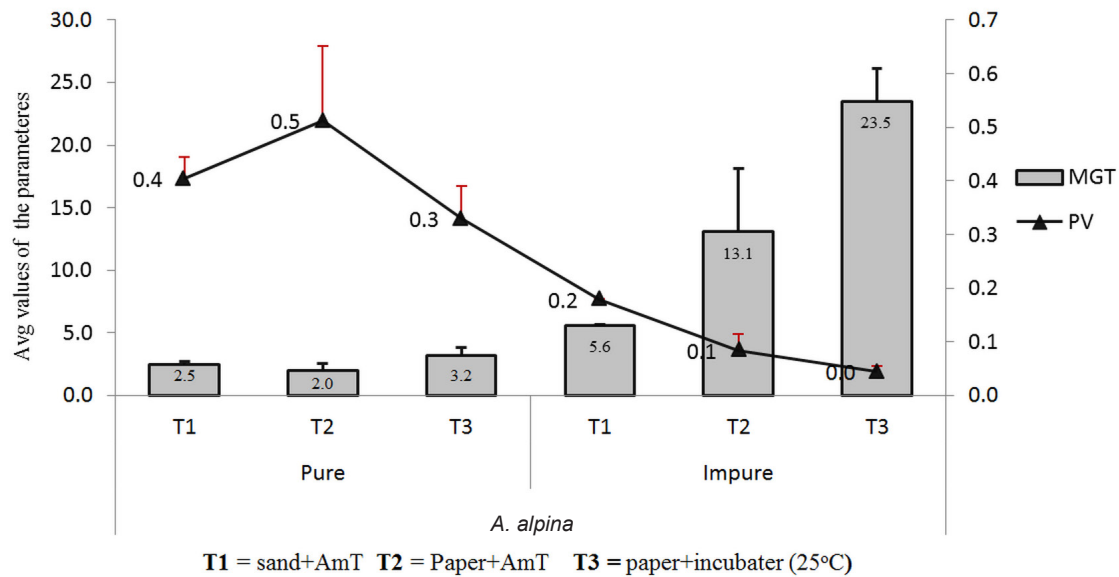


Figure 2. The effect of in vitro seed germination ecology treatments on MGT and PV

highest (23.5 days) mean germination time was obtained from impure seed sowed on T3, followed by 13.06 days on T2. As a result, using pure seed sowed on Whatman paper in ambient temperature (T2) had vigorous and fast germination; hence it seems to have better germination speed than germination on T1 and T3. This difference might be due to the presence of direct light in addition to sustaining conducive moisture and optimum temperature to the seed for reducing the periods or duration of mean germination time. This also indicates that the seed cleaning stage has a significant detrimental effect on germination. This agrees with the statement of Yerima et al. (2015), the short duration to germinate is highly important for the vigourity of seedling emergence whereas the long duration becomes fatal.

According to Figure 2, the recorded values of mean germination time were significantly different regarding the seed cleaning stage and growing medium with the environment. However, it is better to use pure seed sowed on sand media at ambient temperature (T1) as a germination ecology treatment, considering the cost and applicability of the technology at community level for bamboo establishment and development without any advanced and

cost incurring materials requirements besides the mandated institution. As an economically and environmentally valuable plant, the development and adaptation of easy to apply and effective propagation method of highland bamboo in every aspect is an urgent affair for sustainable use and socioeconomic value. Hence, the output of this research could be helpful and used as an input to propagate *Y. alpina* species for the developmental endeavors and income source of the community.

IV. CONCLUSION

Germination improving technology is an urgent affair for successful forest development of *Yushania alpina* plantations. However, it has unsatisfactory seed germination due to its recalcitrant seed behavior, and hence its seed longevity is seriously very short and becomes a low germination performer. To confront the low germination challenge, different germination ecology treatments using seed cleaning stages were tested to promote germination potential. Hence, the pure seed had a significant influence to increase germination percentage using seed germination ecology treatments. The highest germination percentage and short mean germination time were obtained on T2 (paper +

ambient temperature), followed by germinations on T1 (sand + ambient temperature), and T3 (paper + incubator (25°C)), respectively. However, the impure seed revealed the least and most unsatisfactory germination percentage in all treatments. Hence, this concluded that using pure seed on sand media with ambient temperature is more advisable and preferred to adopt cost-wise and easy propagation of *Yushania alpina* everywhere and by whomever.

Based on the current research result the following recommendations can be given as follows:

- The germination study on different germination ecology treatments is a one season seed collection so that it should be conducted using across season collection.
- Also, the germination study on different germination ecology treatments used seeds at 10 days after collection, hence it should be designed and conducted using seeds having different storage period.

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