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Seed Germination Performance of Nine Arabica Coffee (*Coffea arabica* L.) Varieties Under the Laboratory Condition After Six Months of Storage Period

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Abstract

Coffee seeds are classified into intermediate seeds which require high moisture content during the storage and possess a short shelf life. Seed germination testing aims to determine the potential of a seed lot, evaluate seed quality during the storage and also to estimate the success of seedling in the nursery. This experiment aimed to determine the germination performance of several Arabica coffee varieties after being stored for six months at controlled temperature. Nine varieties of Arabica coffee, namely Abesinia-3, Andungsari-1, BLP, Gayo-2, Goiaba, Kartika-1, Kayumas, MP-3, and S-795 were used in this study. The seeds were harvested in the 2019 harvest season (September) and the germination was tested in April 2020. The germination test was carried out in laboratory by using petridish. The result showed that the tested Arabica coffee seeds still had a high germination percentage which was more than 85%. Only three varieties namely S-795, Abesinia-3, and Goiaba had low germination growth (<80%). Meanwhile, Kayumas, BLP, and Kartika-1 coffee varieties had the best germination growth with an average length of 34.19 mm; 39.79 mm and 39.41 mm, respectively. Kartika-1 variety needs 6–7 days to germinate; Kayumas, BLP, and Goiaba varieties took 7–8 days; S-795 and MP-3 took 10 days; as well as Andungsari-1, Abesinia-3, and Gayo-2 which took 10–12 days. Arabica coffee seeds that had a short germination time will had a high germination rate and otherwise. Each variety displayed a different germination performance. BLP, Kartika-1, Gayo-2 and MP-3 varieties had high potential germination, 86.67%; 89.17%; 86.67%, and 93.33%, respectively. Kayumas, BLP, and Kartika-1 varieties showed significantly longer germination, namely 34.19 mm; 39.79 mm and 39.41 mm, respectively. The fastest of average germination time was possessed by Kartika-1 variety (6.54 days) and the highest seed germination rate was also possessed by Kartika-1 variety (0.153 per day).

Keyword: Arabica coffee, seed germination, viability

INTRODUCTION

Arabica coffee is an essential commodity which possesses high economic value. In 2018/2019, Indonesia ranked the fourth as the world coffee producer and in the same time the consumption rate of coffee in Indonesia showed positive trends (Wibowo, 2019). The majority of cultivated Arabica coffee is predominantly self-pollinating and cultivated by seeds. The self-pollination characteristic of Arabica coffee causes high genetic uniformity in the coffee plant population. The variations of Arabica

coffee population are caused by the spontaneous mutation of genes that control the character of the fruits and seeds as well as the natural crossover that occurs between two types of Arabica coffee (Clifford & Willson, 1985; Gebreselassie *et al.*, 2010). Planting material in the form of seeds in Arabica coffee is very popular (Rosa *et al.*, 2011; Nasiro *et al.*, 2017) because in addition to having genetic variation in a low population, it is also easier to distribute and produce large numbers of plants in one harvest period. The main obstacle to planting material of seeds on coffee plants is the short shelf life period. Coffee seeds are able to germinate shortly after harvest and seed viability can decline rapidly. Coffee seeds are classified as intermediate seeds (Ellis *et al.*, 1990; Hong & Ellis, 1996; Selmar *et al.*, 2006) because they require high moisture content during the storage and the shelf life of coffee seeds is relatively short (Dussert *et al.*, 1997; Rahardjo, 2012). The shelf life of coffee seeds will be longer if stored at low temperatures. Nasiro *et al.* (2017) revealed that coffee seeds stored at 15ºC were still able to maintain their germination of 71–78% while being stored at room temperature (± 28ºC) was only able to maintain seed germination of 43–55%. Coffee seeds still germinate after being stored for 10 months at 15°C and the moisture content of 10–11% (Ellis *et al.*, 1990). The storage using low moisture content is contrary to the provisions of the Regulation of Ministry of Agriculture No. 89 (2013) which stated that the moisture content of coffee seed stored is 30–40%. Nasiro *et al*. (2017) showed that coffee seeds stored at 15°C were able to maintain seed viability more than 80% for five months of storage while coffee seeds stored at room temperature only lasted for three months with a germination percentage of more than 80%. In contrast, Rosa *et al.* (2011) found that coffee seeds stored at 10–15°C and 10–11% moisture content will produce poor quality seeds while if at 20°C still produce good quality seeds. Apart from storage temperature, the germination rate is also influenced by the level of fruit maturity at harvest. Coffee seeds harvested from fully ripe fruit (physiologically ripe) have the most optimal germination compared to seeds harvested when still green or yellowish towards red (Rosa *et al.*, 2011; Baliza *et al.*, 2012).

The germination testing aims to determine the potential of germination in the seeds lots and can be used to compare the quality among the seeds lots. It is also used to predict the success of seedling in the nursery (Sudrajat *et al.*, 2017). The use of seeds which has high physiology potential is urgently needed for the success of production and it is also significantly contributing in the early process of seedling (Rahimi, 2013; Castan *et al.*, 2018). Therefore, the evaluations of germination potential in the seeds which have faster seed deterioration period and have been stored for a certain time are urgently needed. Seeds able to be germinated when the root bulges or shoots appear. According to Silva (2002), the germination process of seeds starts when water was absorbed through an inhibition process and it lasts when the embryo axis begins to elongate, usually a potential root. Seeds can germinate when placed in appropriate condition of environment, adequate water supply, and proper light so the testing in laboratory can be the basic to measure the viability potential of the seeds (Kader, 2005; Eira *et al.*, 2006; Sudrajat *et al.*, 2017). This research aimed to investigate the germination characteristics of some varieties of Arabica coffee seeds after being kept in the room with controlled temperature.

MATERIALS AND METHODS

This research utilized nine varieties of Arabica coffee seeds, those are: Abesinia-3, Andungsari-1, BLP, Gayo-2, Goiaba, Kartika-1, Kayumas, MP-3, and S-795 which have been kept storage in the room temperature 20–22°C for 6 months. Those seeds were harvested in 2019 (July–September) and were germinated in April 2020. The seeds were evaluated and germinated in petridish.

The research was conducted at Plant Breeding Laboratory of Indonesian Coffee and Cocoa Research Institute. The temperature was ±28ºC in the morning and afternoon and ±30ºC in the noon with relative humidity ranging 55–70%.

The Arabica coffee seeds from each variety was peeled and soaked in fungicide mankozeb solution 0.2% formulation (w/v) for one night in the room temperature. The seeds are then germinated in petridish coated by wet wipes and paper wrap. Each petridish contains 30 coffee seeds and was repeated for four times then germinated in room temperature. The observation of seed germination was every day until 21 day after germination (Rahardjo, 1988). The seeds can be observed if the root bulged or shoot appeared. The variables of seed germination as follow: Moisture content (WC) =

$$\frac{m_1 - m_2}{m_1 - m_0} \times 100$$

m_0 is the weight of cup and the cover; m_1 is the weight of cup, cover, and coffee seeds before the drying; m_2 is the weight of cup, cover, and coffee seeds after the drying. The drying process is conducted in the 105ºC oven temperature for 16 hours (SNI (2008)). Germination percentage (GP) =

$$\frac{n\,KN}{N} \times 100\%$$

n KN is the number of normal sprouts; N is the number of seed to germinate (Adapted from Sudrajat *et al*. (2017); Baliza *et al.* (2012)). Abnormal germination (AG) =

$$\frac{n\,KA}{N} \times 100\%$$

n KA is the number of abnormal sprouts; N is the number of seeds to germinate (Adapted from Sudrajat *et al.* (2017)). Potential seed germination (PSG) = $GP + AG$, GP is germination percentage; AG is the percentage of abnormal germination. Mean of total length (SL) =

$$\frac{\sum_{i=1}^{k} SL}{n}$$

SL is the total length of each seedling; n is the total number of seedlings evaluated (Adapted from Silva *et al*. (2019)). Mean of shoot length (HL) =

$$\frac{\sum_{i=1}^{k} HL}{n}$$

HL is the length of the shoot of each seedling; n is the total number seedlings evaluated (Adapted from Silva *et al.* (2019)). Mean of root length (RL) =

$$\frac{\sum_{i=1}^{k} RL}{n}$$

RL is the length of the root of each seedling; n is the total number seedlings evaluated (Adapted from Silva *et al*. (2019)). Mean of the root/shoot ratio (RRA) =

$$\frac{\sum_{i=1}^{k} RRA}{n}$$

RRA is the ratio between the root and the shoot of each seedling; n is the total number of seedlings evaluated (Adapted from Silva *et al.* (2019)). Mean germination time (MT) =

$$\sum_{i=1}^{k} \text{ni. ti} \Big/ \sum_{i=1}^{k} \text{ni}$$

t_i = time from the start of the experiment to the i^{th} observation; n_i = the number of seeds germinated in the time i; k = the last time of germination (Ranal *et al*., 2009). Coefficient of variation of germination time (CVt) =

$$\frac{S_t}{MT} \times 100$$

S_t is standard deviation of germination time; *MT* is mean germination time of seeds (Ranal

et al., 2009). Mean germination rate (MR) = $\frac{1}{MT}$

MT is mean germination time of seeds (Ranal & Santana, 2006). Uncertainty of the germination (U) = $-\sum_{i=1}^{k} f_i \, Log_2 f_i \quad ; \quad f_i = \frac{n_i}{\sum_{i=1}^{k} n_i}$

f_i is relative frequency of germination; n_i is number of seeds that germinate on the day i; k = last day of observation (Ranal *et al.*, 2009). This study implements Completely Randomized Design using four replications. The data was analysis by using SAS 9.0 software and continued by Duncan test if there is significant difference.

RESULTS AND DISCUSSION

The result of ANOVA test showed that various accession of Arabica coffee had different outputs in the parameter of germination percentage, potential of seed germination, shoot length, root length, total length of seedlings, mean germination time, coefficient of variation of germination time, mean of germination rate, and uncertainty (data was not presented). Each accession of Arabica coffee had each qualified seed. The result of moisture content test after storage period showed that the moisture content of seeds was still about 21.11–39.20%. Based on the Regulation of Agriculture Ministry of Republic of Indonesia Number 89 (2013), the moisture content of coffee seeds ranged from 30% to 40% so that several varieties observed were maintained according to these conditions. The Arabica coffee of Kayumas variety (28.43%), BLP (25.65%), Kartika-1 (26.85%), Gayo-2 (21.11%) and Goiaba (26.57%) still showed the germination percentage more than 70% although they were kept in the moisture content under 30%. Only S-795 variety contained moisture of 21.37% which showed bad germination under 10% (Table 1).

Germination Percentage

Germination percentage is the ability of seeds to grow becoming new plant. The germination of seeds is counted from the percentage of the normally germination. The normal seedlings showed the ability of seeds to grow become the normal plants if they are planted in adequate condition meanwhile abnormal seedlings showed the weak growth (Sudrajat *et al.*, 2017). The potential seed germination is the maximum ability of seeds to germinate into new plant. The germination percentage of six varieties of Arabica coffee as follows: Andungsari-1, BLP, Gayo-2, Kartika-1, Kayumas, and MP-3 have mean germination percentage ranging between 82.50% and 93.33% with the range of moisture

Table 1. Moisture content after storage, germination percentage (GP), percentage of abnormal germination (AG), and potential seed germination (PSG) of nine Arabica coffee varieties

Variety	Water content after storage (%)	Germination percentage (%)	Abnormal germination (%)	Potential seed germination (%)
Abesinia-3	31.44	51.67 c	10.00 b	61.67 b
Andungsari-1	39.20	85.00 a	6.67 b	91.67 a
BLP	25.65	86.67 a	9.17 b	95.83 a
Gayo-2	21.11	86.67 a	8.33 b	95.00 a
Goiaba	26.57	70.00 b	18.33 a	88.33 a
Kartika-1	26.85	89.17 a	5.00 bc	94.17 a
Kayumas	28.43	82.50 a	8.34 b	90.84 a
MP-3	32.80	93.33 a	5.84 bc	99.17 a
S-795	21.37	9.17 d	0.00 c	9.17 c
CV (%)	n.d.	11.23	23.53	8.7 a

Notes: the numbers followed by different letters in the same column show a significant difference at the 5% rate; n.d. (not determined).

content 21% until 39%, meanwhile the vareties of Abesinia-3, S-795, and Goiaba had mean germination percentage as follows 51.67%; 9.17%, and 70.00% with a moisture content range between 21% and 31%. The MP-3 Arabica coffee variety had the best germination percentage with 93.33% while S-795 variety had the worst germination percentage with 9.17%. The abnormal germination of Goiaba variety was significantly higher than the other varieties (18.33%). Generally, Arabica coffee seeds which kept in the temperature 20–22°C for six months still showed high potential seed germination (more than 85%). There was only two varieties which had lower potential seed germination that were S-795 (9.17%) and Abesinia-3 (61.67%). The low potential value of seeds to germinate in S-795 variety caused by genetic factor because the variance analysis results showed that the different varieties affect the potential value of seeds to germinate. Rosa *et al.* (2011) reported that coffee seeds stored with a moisture content of 10–11% will produce the inapropriate seeds for planting material. The purpose of examining germination is to investigate the germination potential of seed lot so the success possibility of the seedlings in the nursery can be predicted (Sudrajat *et al.*, 2017). The germination percentage is the main parameter used in the seed evaluation. Based on the regulation of Agriculture Ministry of Republic of Indonesia Number 89 (2013), the standard minimum of physiological quality of coffee seeds was 80% so that the six varieties evaluated namely Andungsari-1, BLP, Gayo-2, Kartika-1, Kayumas, and MP-3 were classified as having good physiological qualities. Physiological quality is a description of the overall characteristics of seeds that shows its suitability of growth (viability), the germination percentage, and the seed health appropriated with the specified quality requirements. In the regulation, the physiological quality standard is described by the germination percentage.

Seedling Length

HL is the length of shoot (hypocotyl) for each seedlings. RL is the length of root (radicle) for each coffee seedlings and SL is the total length of each seedlings. The observation of root length, shoot length, and total length of seedlings is conducted in the end of germination process (Silva *et al.*, 2019; Silva *et al.*, 2020). The BLP and Kartika-1 varieties have longer germination of total length (SL) than the other varieties of Arabica coffee which were 39.79 mm and 39.41 mm (Table 3). The shoot length (HL) and root length of BLP (22.52 mm; 17.27 mm) and Kartika-1 (22.09 mm; 17.32 mm) both are also significantly longer than the other varieties. It showed that BLP and Kartika-1 varieties were more vigorous than the other Arabica coffee seeds. The quality of both varieties of coffee was good. RRA is the ratio of root (radicle) and shoot (hypocotyl) for each seedling (Silva *et al.*, 2019). The Arabica coffee seeds which are germinated have longer hypocotyl than their radicle because the RRA value counted is less than 1. MP-3 variety had the lowest radicle growth because the value of RRA was 0.57 so the growth of radicle was only the half of the hypocotyl length. The germination test which counts the morphological characteristics of seedlings is based on the fact that the vigorous seedlings can produce good seedlings (Castan *et al.*, 2018). The higher value of hypocotyl and radicle length, the better viability of seeds. The seed germination can be inhibited because of the effects of fruit ripeness, harvesting, or seed processing.

Germination Time

The mean of germination time is a weighted mean of germination period. The number of seeds that was germinated in a predetermined time interval was used as the

Tabel 2. The comparison of shoot length (HL), root length (RL), total length (SL) and the ratio of root/ shoot length (RRA)

Variety	HL (mm)	RL (mm)	SL (mm)	RRA
Abesinia-3	9.66 c	8.89 cd	18.56 cd	0.93 a
Andungsari-1	8.34 c	6.60 d	14.94 d	0.79 a
BLP	22.52 a	17.27 a	39.79 a	0.77 a
Gayo-2	8.88 c	7.19 d	16.07 cd	0.82 a
Goiaba	14.58 b	11.38 bc	25.96 b	0.79 a
Kartika-1	22.09 a	17.32 a	39.41 a	0.79 a
Kayumas	17.47 b	16.72 a	34.1 9a	0.97 a
MP-3	13.74 b	7.87 d	21.61 bc	0.57 b
S-795	13.92 b	12.21 b	26.13 b	0.89 a
CV (%)	17.27	17.02	15.87	15.16

Note: the numbers followed by different letters in the same column show a significant difference at the 5% rate.

basis for calculating the germination time (Table 1). The mean germination time is the average length of time needed by seeds for maximum seed germination (Ranal & Santana, 2006). The Kartika-1 variety required a significantly faster time to germinate than the other varieties, namely 6.54 days and were not significantly different from the time to germinate the Kayumas variety (7.64 days). The BLP and Goiaba varieties took 7–8 days to germinate; the S-795 and MP3 varieties took 10 days; and the Gayo-2, Abesinia-3 and Andungsari-1 varieties required the longest germinating time of 11–12 days (Table 4). The high mean germination time indicates that the seeds need a long time to complete the germination process and otherwise. The coffee seeds germinated in the laboratory using filter paper media require a shorter time than coffee seeds germinated with soil media. Germination in the laboratory takes 2–3 weeks while germination in soil media takes 3–4 weeks until the food reserves in the cotyledons are used up for seed growth (Silva *et al.*, 2002). The germination method in the laboratory is the optimum potential for seeds to develop and grow into new plants.

The coefficient of variation of germination time shows the ratio of the standard deviation to the mean germination time (Carvalho *et al*., 2005; Ranal *et al.*, 2009; Silva *et al.*, 2019). The lower the coefficient of variation, the more precise the level of estimation is. The coefficient of variation in the germination time of the BLP variety (6.95%) was the lowest significantly compared to other Arabica coffee varieties. Seven Arabica coffee varieties namely Abesinia-3, Andungsari-1, Gayo-2, Goiaba, Kartika-1, Kayumas, and MP-3 have almost the same coefficient of variation in germination time, ranging from 20.51–25.01% and one variety (S-795) which had a coefficient of variation in germination time of 11.60%. The coefficient of variation of germination time can be used to evaluate the uniformity of germination or to determine the variability of germination related to the mean time of germination (Ranal & Santana, 2006).

Germination Rate

The mean of germination rate was inversely proportional to the mean germination time (Table 1). If the MR value increases, the MT value will decrease and it applies the opposite, but the relationship between both is not linear (Ranal & Santana, 2006). The mean germination rate can also be used to see the rate of germination so that the unit of it is per day (day^{-1}). The Kartika-1 variety has the highest mean germination rate, namely 0.153 / day and significantly higher than other varieties. The Kayumas, BLP, and Goiaba varieties have almost the same germination rate, ranging from 0.124–0.131/ day. The Andungsari-1, S-795, and MP-3 varieties had almost the same germination rate, namely 0.095–0.101/day. The Abesinia-3

Table 3. The mean germination time (MT), coefficient of variation of germination time (CVt), mean germination rate (MR) and uncertainty (U) of Arabica coffee seed germination

Variety	MT (day)	CVt (%)	MR (day^{-1})	U (bit)
Abesinia-3	11.67 a	24.01 a	0.086 d	2.644 ab
Andungsari-1	10.64 ab	25.01 a	0.095 cd	2.959 a
BLP	7.76 c	6.95 c	0.129 b	2.295 cd
Gayo-2	11.66 a	16.43 ab	0.086 d	2.577 bc
Goiaba	8.12 c	20.51 a	0.124 b	1.796 e
Kartika-1	6.54 d	20.65 a	0.153 a	2.068 de
Kayumas	7.64 cd	24.64 a	0.131 b	2.203 d
MP-3	9.98 b	20.94 a	0.101 c	2.684 ab
S-795	10.13 b	11.60 bc	0.101 c	1.251 f
CV (%)	8.41	28.95	7.85	9.96

Note: the numbers followed by different letters in the same column show a significant difference at the 5% rate.

and Gayo-2 varieties have the slowest germination rate among other varieties. High (time when the majority of seeds germinate) and low (time when a small proportion of seeds germinate) can be used as a guidelines for indicators of seed vigor (Kader, 2005). Seeds possesing good vigor performance will have a high germinating speed (seeds need a short germination time). The Kartika-1 variety has good seed vigor.

Uncertainty

Uncertainty measures the level of uncertainty related to the relative germination frequency distribution. Campbell *et al.* (2020) stated that the uncertainty value is related to the frequency and distribution of germination in the experimental group. In seed germination, the lower the uncertainty index, the more synchronized seed germination. The U value measures the rate at which germination is spread over a certain time. The low U value indicates the frequency of germination with a few peaks so that germination is more evenly distributed over a certain period of time. The unit in this index is bit which shows the binary measurement that is taken into account, between the germinated and non-germinated seeds (Labouriau & Valadares, 1976 *cit* Ranal & Santana, 2006). The uncertainty index of S-795 and Goiaba varieties (1.251 bit; 1.796 bit) are low, meanwhile the uncertainty index of Andungsari-1, Abesinia-3, Gayo-2 and MP-3 varieties are high (2.959 bit; 2.644 bit; 2.577 bit; 2.684 bit). Kayumas and Kartika-1 coffee varieties have almost the same uncertainty value, namely 2.203 bit and 2.068 bit. The BLP Arabica coffee has an uncertainty value of 2.295 bit.

CONCLUSIONS

Arabica coffee variety provides different germination performance. Among nine germinated varieties, BLP and Kartika-1 had the best seed quality. Both varieties had the highest germination percentage (> 85%) and highest seedlings growth (total length of seedlings 39.79 mm and 39.41 mm respectively). Kartika-1 and BLP varieties also need a short time (6–8 days) to complete germination. Otherwise Andungsari-1, Abesinia-3, and Gayo-2 showed low total length of seedlings and required longer time to germinate (10–12 days) even though the germination percentage of Andungsari-1 and Gayo-2 varieties were ≥ 85%. The relationship of mean germination time (MT) and mean of germination rate (MR) was inversed. A good indicator for seed vigor was if the seeds had both a low mean of germination time and high germination rate. The value of mean germination time (MT) was not related to the coefficient of variation of germination time (CVt) and the uncertainty index (U).

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