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Pre-sowing Treatment of Coffee (*Coffea arabica* L.) Seeds to Enhance Emergence and Subsequent Growth of Seedlings

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Abstract: Nursery experiment was conducted to investigate the effect of removal of parchment and soaking of coffee seeds in pure water for different durations on emergence and early growth of coffee seedlings. A split plot design of three replications was employed with seed types (seeds with parchment and clean coffee) and soaking hour (soaking seeds for 12, 24, 36, 48, 60 and 72 h and control) in pure water were assigned as main and sub-plot, respectively. Emergence of seedlings was significantly ($p<0.01$) affected by coffee seed type (clean coffee vs. coffee with in parchment) where better emergence response was obtained from clean coffee. Forty five, 75 and 90 days after sowing 18.46, 56.70 and 84.70% emergence was obtained from clean coffee, respectively while only 1.86, 15.40 and 49.41% emergence was obtained in the same period, respectively, from coffee seeds sown in parchment. Better emergence occurred from seeds soaked in water against non-soaked ones 45 and 60 days after sowing. Moreover, leaf area and total dry matter weight differed in response to different soaking hours. The seedling vigor index also showed a statistical difference ($p<0.01$) between coffee seed types where clean coffee resulted in higher value (313.13) over the parchment coffee (232.48). The same parameter resulted from coffee seeds soaked for 72 h was better ($p<0.05$) when compared to non-soaked seeds and seeds soaked for less than 72 h. The study revealed removing parchment and soaking coffee seeds in water enhances emergency and subsequent growth of coffee seedlings.

Key words: Arabica coffee, clean coffee seeds, parchment coffee seeds, soaking, emergence

INTRODUCTION

Coffee (*Coffea arabica* L.) is originated in humid high rain forest of South and Southwestern Ethiopia (Mabberley, 1997). In Ethiopia, coffee has tremendous impact on economic, social and spiritual life of people of different location and cultural background. As regards to the livelihood and employment, a quarter of the population of the nation to depends directly or indirectly on coffee for its income generation. In Sub-Saharan Africa, coffee is a mainstay of the economies of more than 20 countries and central to the livelihood of more than 20 million rural families (Rice and Ward, 1996). Arabica coffee is by far the most important economic species because of its superior quality and is responsible for 75-80% of world coffee production, with Robusta coffee supplying only the remaining 20-25%.

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For Ethiopia, coffee is a major and leading export item accounting about five percent of the Gross National Product (GNP). Currently, about 35% of the foreign exchange earning of the country was due to the crop. In the year 2008 only, greater than 1.5 billion birr was earned from coffee export (Central Statistics Authority, 2009).

As most commercially grown cultivars of Arabica coffee are largely self-pollinated and homozygous, they are normally propagated by seed. Vegetative propagation is used for multiplication of clones and hybrids at research stations (Wilson, 1999). Even though coffee seeds are capable of germinating as soon as harvested due to absence of dormancy, they lose their viability quickly (Coste, 1992). Therefore, when stored beyond two months, they reveal a decline in percentage germination and give variable and poorly developed seedlings. With its parchment, Arabica coffee seeds need 45 to more than 70 days for germination (Gopal and Ramaiah, 1971; Wrigley, 1988). From sowing to time of transplanting Arabica coffee requires 6 to 8 months at nursery in warm regions and even 12 months in higher altitudes (Cambrony, 1992). Therefore, by improving emergence and subsequent seedling growth through pre-sowing treatment of coffee seeds, it is possible to shorten the time taken to raise seedlings in the nursery and cost of nursery management can also be reduced.

So far, some works have been done in coffee for better understanding of mechanisms and regulation of coffee seed germination (Da-Silva *et al*., 2004) and find ways for effective multiplication of coffee through direct sowing of plant materials after *in vitro* propagation (Etienne-Barry *et al*., 1999). However, sowing coffee seeds in nursery and managing up to 12 months before transplanting remains the routine practice in small scale farmers and large scale commercial farms.

In Ethiopia, especially in the South and Southwestern region, the time to pick coffee and nursery operation including sowing are overlapping. Moreover, some Arabica coffee cultivars are late to ripe and hence happen to be not ready for sowing on the actual regional calendar. The fate of these seeds is inevitably storage for the coming season, which would implicitly affect the viability (Personal observation). Hence, by hastening emergency through pre-sowing treatment (removal of parchment and soaking), one can escape overlapping of activities of harvesting and sowing. Soaking seeds as a means to hasten germination and subsequent growth of seedlings remained routine practice in some crops although, reports are scarce in coffee. Ahmadi *et al.* (2007) reported that hydropriming clearly hastened emergence and increased vigor index of wheat seedlings. Similar works have also been reported on rapeseed (Bijanzadeh *et al*., 2010) and caper seed (Pascual *et al*., 2009).

In addition, removing parchment during sowing enables to sort out diseased and/or insect beaten seeds, which can hardly be detected when still in parchment. Among 100 seeds (74-40), which were ready for sowing and looked healthy with parchment, about 11 seeds were found to be defective up on the removal of parchment (Unpublished laboratory experiment). Thus, removal of parchment before sowing, apart from hastening germination, would enable to detect unfit seeds for sowing. Therefore, the objective of this study was to determine the effect of removal of parchment of Arabica coffee seeds and soaking seeds in water on the emergence rate and subsequent early growth of the seedlings.

MATERIALS AND METHODS

The trial was conducted at Jimma Agricultural Research Center from September 2005 to June 2005. The area was located at latitude of 7° 40' North and longitude of 36° 47' East with an elevation of 1753 m.a.s.l. Thirty year's mean figures reveal that the minimum and maximum temperatures are 11.3 and 26.2°C, respectively. The area received an average rainfall of 1594.5 mm per annum.

A split plot design of three replications was employed with seed types (seeds with parchment and clean coffee) and soaking hour (soaking seeds for 12, 24, 36, 48, 60 and 72 h and non-soaked) in pure water were assigned as main and sub-plot, respectively.

A recommended media (forest soil) collected from the research center, was air dried, manually crushed and passed through 2 mm sieve to remove clods, plant roots and other foreign materials (Yakob *et al*., 1998). The sieved soil was filled to black polythene bag of 12 cm wide and 25 cm length.

A single experimental unit (treatment) consisted of 16 pots. These were arranged in square fashion (4X4) on nursery bed with 60 cm spacing between experimental units and 1.5 m between replications. Red ripe cherries from coffee cultivar 74-40, which was resistant to Coffee Berry Disease (CBD), were hand picked and prepared for sowing. Seeds were soaked for the specified durations (12, 24, 36, 48, 60 and 72 h).

Four seeds were sown per polythene bag. Thinning to one seedling in a bag was done 90 days after sowing when all normal viable seedlings were expected to emerge. Every routine nursery activity was practiced uniformly to all experimental units as per the recommendation of the Jimma Agricultural Research Center (Institute of Agricultural Research, 1996). Emergency count was made from each experimental unit when just it was commenced (35 days after sowing) in three days interval. This count was extended up to 75 days after sowing. Three months after sowing, Seedling Vigor Index (SVI) data was taken to determine the variation in vigorosity among seedlings of different treatments using the formula described by Abdul-Baki and Anderson (1973) as follows:

$$SVI = TRL \times SH \times G \times E\%$$

where, SH is sample seedling height, G is girth of the sample seedling, TRL is tap root length of the sample seedling, E% is emergence percent of the treatment.

Six months after sowing, the attributes of non-destructive parameters were measured by taking two seedlings from the inner most four of each experimental unit: plant height (cm), stem diameter (cm) and leaf area (cm^2). Plant height was measured from the base to the tip of the seedling using a ruler. Stem diameter was measured at the base near the medium surface using a caliper. Leaf area per leaf was calculated using the procedure adapted by Yakob *et al*., 1998) as follows:

$$Y = K \times L \times B$$

where, Y is estimated leaf area; K is constant specific to cultivars and canopy classes (0.67); L is leaf length (cm) and B is maximum leaf breadth (cm).

Each seedling sampled for measurement of non-destructive parameters was brought to the laboratory for destructive parameters. Seedlings were cut with a scissor at collar point to separate the shoot from the root. The shoot, then, was separated to leaves and stems and fresh weight of each weighed using sensitive balance. The polythene bag containing the roots of the seedlings then, were immersed in a bucket filled with water and roots were separated carefully from the soil still being in water. The roots were subsequently washed with clean water, dried with water adsorbent cloth and fresh weight was measured following the same procedures described for the shoot. Root volume was measured using water displacement method. Finally, the entire seedling parts were oven dried at 100°C until a constant weight as described by Adjet-Twum and Solomon (1982) and dry matter determination was made using sensitive balance. The data were subject to Analysis of Variance (ANOVA) for split plot design and treatment mean separation was carried out. GenStat 10th edition was employed for statistical analysis (GenStat, 2007).

RESULTS AND DISCUSSION

Percent Emergence

There were significant differences ($p<0.01$ and $p<0.05$) observed in percent emergence between clean (seeds without parchment) coffee seeds and parchment (seeds in parchment) coffee seeds sown 45, 60 and 75 days after sowing (Table 1). Forty-five days after sowing, there was a highly significant difference ($p<0.01$) between the two treatments where clean coffee resulted in 18.46 and 1.86% from parchment coffee (Table 1). Similarly, after 60 days of sowing 56.7 and 15.4% emergence was seen from clean and parchment coffee seeds, respectively evidencing a significant difference ($p<0.05$) between the treatments. Moreover, 75 days after sowing there was a significant difference ($p<0.05$) in percent emergence between the treatments where 84.7 and 49.41% was recorded from clean and parchment seeds, respectively.

Soaking hours resulted in a significant difference ($p<0.05$) in percent emergence of 45 and 60 days after sowing (Table 1). Forty-five days after sowing, coffee seeds soaked for 12 h were better in emergence (13.28%) than non-soaked seeds (2.87%). Similarly, coffee seeds soaked for 72 h found to be better in emergence (49.80%) in comparison to non-soaked ones (22.66%). The interaction effect of coffee seed type and soaking hour was significant ($p<0.05$) in emergence percentage taken 45 and 60 days after sowing, however the interaction was not significant 75 days after sowing (Appendix 1).

Growth Parameters

There was a significant difference ($p<0.05$) in some growth parameters: leaf area, root volume, root dry weight, stem dry weight and total dry matter between clean and parchment coffee seeds (Table 2). In all parameters, clean coffee resulted in better performance over the parchment seed. Girth was superior in clean coffee (0.29 cm) over parchment seeds (0.25 cm). Leaf area was found to be better from the clean coffee (10.86 cm^2) in comparison to parchment seed (9.63cm^2). Similar result was obtained in root volume where 1.19 and 106 mL was recorded from clean and parchment coffee seeds, respectively. Root dry weight was higher in clean coffee (0.172 g) over the parchment seed (0.144 g). Stem dry weight was similarly higher in clean coffee (0.122 g) in relation to that of parchment coffee (0.092 g). Total dry

Table 1: Coffee seed emergence as affected by coffee seed type and soaking rates

	Emergence percent		
Treatment	45 days after sowing	60 days after sowing	75 days after sowing
Coffee type	**	*	*
Clean coffee	18.46a	56.70a	84.70a
Parchment	1.86b	15.40b	49.41b
SE (±)	0.21	4.30	3.46
CV (%)	41.81	54.61	23.63
Soaking rates (h)	*	*	NS
Non-soaked	2.87c	22.66a	61.72
12	13.28a	34.90b	65.46
24	8.07b	36.20b	64.06
36	10.68bc	36.98bc	69.53
48	9.38 bc	36.46b	65.63
60	8.07 b	35.68b	66.67
72	8.75b	49.80c	76.31
SE (±)	0.61	6.69	5.70
CV (%)	63.3	45.46	20.82

Means with in a column followed by the same letter (s) are not significantly different, *Significant at $p<0.05$, **Significant at $p<0.01$, NS: Non significant

Table 2: Growth of coffee seedlings as affected by coffee seed type and soaking rate six months after sowing

	Destructive and non-destructive parameters							
Coffee types and soaking rates	Height (cm)	Girth (cm)	Leaf area (cm^2)	Root volume (mL)	Leaf dry wt. (g)	Stem dry wt. (g)	Root dry wt. (g)	Total dry matter wt. (g)
Coffee type	NS	*	*	*	NS	NS	*	*
Clean coffee	11.08	0.29a	10.86a	1.19a	0.377	0.122	0.172a	0.641a
Parchment coffee	10.19	0.25b	9.63b	1.06b	0.368	0.117	0.144b	0.628b
SE (±)	0.23	0.002	0.30	0.055	0.030	0.003	0.006	0.035
CV (%)	9.47	7.52	13.29	22.13	36.96	28.04	22.11	24.90
Soakings rates (h)	NS	NS	*	NS	*	*	NS	*
Non-soaked	10.57	0.25	8.67b	0.93	0.322b	0.101b	0.114	0.537b
12	10.64	0.24	8.88b	0.97	0.371ab	0.101b	0.118	0.590b
24	11.10	0.25	10.57ab	0.97	0.321b	0.105b	0.129	0.555b
36	11.28	0.26	10.29ab	1.02	0.405a	0.138a	0.147	0.689a
48	11.65	0.26	11.02ab	1.35	0.394a	0.133a	0.163	0.690a
60	10.73	0.25	9.93ab	1.60	0.332b	0.109b	0.146	0.587b
72	11.94	0.27	12.43a	1.49	0.402a	0.150a	0.183	0.795a
SE (±)	0.43	0.008	0.75	0.165	0.062	0.014	0.024	0.093
CV (%)	9.52	7.70	18.03	35.91	40.79	28.45	41.15	35.69

Means with in a column followed by the same letter (s) are not significantly different at $p<0.05$, *Significant at $p<0.05$ NS: Non significant

matter was also found to be better in clean seeds (0.641 g) in comparison to that of parchment coffee (0.578 g). Statistical difference was not observed between the clean and parchment seeds in seedling height and leaf dry weight.

As far as soaking hours was concerned, significant different differences ($p<0.05$) were seen in leaf area, leaf dry weight, stem dry weight and total dry matter. In every parameter in which difference was observed, non-soaked seeds were inferior in performance in relation to the soaked seeds for different hours. Accordingly, in leaf area, seeds soaked for 72 h were better (12.43 cm^2) than non-soaked seeds (8.67 cm^2) and other seeds soaked for less time. Similarly, leaf and stem dry weight were inferior in non-soaked seeds 0.322 and 0.101 g, respectively. Soaked seeds for 36 and 72 h were better in leaf and stem dry weight which scored 0.405 and 0.15 g, respectively. A maximum of 0.795 g total dry matter weight was obtained in seeds soaked for 72 h in comparison to the least scoring non-soaked sees which was resulted in only 0.537g of total dry matter six months after sowing.

The interaction effect between coffee seed types and soaking hour was significant ($p<0.05$) on girth, leaf area, root volume, leaf dry weight and total dry weight however, there was no interaction effect between the two factors on plant height and root dry weight (Appendix 2).

Seedling Vigor Index (SVI)

Higher SVI was obtained from clean seed (313.13) in relation to the parchment seed (232.48) evidencing a highly significant difference ($p<0.01$) (Table 3). There was also a significant difference ($p<0.05$) between the soaking hours where seeds soaked for 72 h gave highest SIV (324.21) in comparison to seeds soaked for less time and non-soaked one, which was 233.94. The interaction effect of coffee seed type and soaking hours was significant ($p<0.05$) (Appendix 2).

Removal of parchment in coffee seeds before sowing hastened emergence. It was observed that it brought an increase in emergence by 17% at 45 days after sowing and 35.29% at 75 days after sowing. Gopal and Ramaiah (1971) stating coffee parchment delays germination of seeds as a result of physical barrier against imbibitions of water by seeds. Gordon (1988) and Taylor *et al.* (1997) also suggested the same. With regard to soaking

Table 3: Seedling vigor index as affected by coffee seed type and soaking rate

Treatment	SVI
Coffee type	**
Clean coffee	313.13a
Parchment coffee	232.48b
SE (±)	4.49
CV (%)	13.43
Soaking rate (h)	*
Non-soaked	233.943b
12	261.91b
24	264.63b
36	281.55b
48	272.55b
60	270.84b
72	324.21a
SE (±)	19.36
CV (%)	17.38

Means with in a column followed by the same letter (s) are not significantly different at $p<0.01$, *Significant at $p<0.05$, **Significant at $p<0.01$ NS: Non significant SVI: Seedling vigor index

rates, generally coffee seeds soaked in water emerged earlier than non-soaked seeds. Arin and Kiyak (2003) also reported that tomato seeds treated (soaked) in distilled water emerged faster and were better in seedling growth parameters like length, fresh weight and girth compared to non-treated seeds. Hot water (50°C) treatment before sowing also gave better germination percentage in Chinese and Mamipintar groundnuts as reported by Frimpong *et al.* (2004). All the reports are in agreement with this report. Statistically significant difference was detected in percent emergence until 75 days after sowing among different soaking hours. At 45 days after sowing, seeds soaked for 72 h were better in emergence by 15.88% than the non-soaked seeds. It was also noticed that with an increase in the number of days after sowing, the difference in emergence percent among the different soaking hours happened to be decreasing. Farooq *et al.* (2006) reported that not only water treatment affected the germination but also the duration of soaking in rice. Maximum vigor was obtained from seeds soaked for 48 h.

As far as removal of parchment is concerned, even though reports are not available in coffee that compares the quantified emergency difference between parchment coffee and clean coffee seeds, reports have been made on other seeds like rice that dehusked seed were better in germination and other early growth parameters when compared to the intact seeds (Miyoshi *et al*., 1996). The current study has no contradiction with the reports made so far as far as the effect of pre-sowing treatments to enhance emergency and subsequent growths.

The seedling vigor test also was synchronized with the emergence performance of seedlings. Accordingly, seedlings from clean coffee that showed better performance in emergence throughout the emergence period were also found to be vigorous than seedlings from parchment seeds at 90 days after sowing. In other words, coffee seedlings that emerged earlier were vigorous than late comers.

As to the growth parameters evaluated six months after sowing, statistical difference was manifested among the treatments of different coffee seeds and soaking hour. As the seedlings got older and older, the difference in vigor observed during early growth stage was seen decreasing, however the interaction effect of coffee types and soaking hour was significant. Clean coffee seeds soaked for 72 h were better in all parameters except in root volume.

Coffee farmers in Ethiopia grow coffee as a secondary or tertiary crop in addition to food crops. The time needed by coffee for nursery operation (from sowing to transplanting of seedlings) often overlaps with other farm activities like harvesting and threshing of field crops. Besides, some coffee cultivars are late to ripe so that they will not be ready for the coffee transplanting season after staying nine to ten months in nursery. The fate of those late seeds would be to be stored for one more year which would highly affect the viability of coffee seeds. Therefore, hastening of emergency of coffee seeds through different mechanism becomes important in order to minimize the operation overlap to the farmer and reduces cost of nursery operations. Large-scale (private or government) coffee farms can also make benefit from the same treatment by which they can reduce the time needed for raising a coffee seedling (usually October to July) to some extent so that they can minimize cost of coffee seedlings.

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APPENDIX

Appendix 1: Summarized analysis of variance (ANOVA) for percent emergence at different periods

		Mean squares		
Source of variation	df	45 days after sowing	60 days after sowing	75 days after sowing
Replication	2	2.2.32NS	465.040NS	415.259NS
Coffee type (A)	1	123.189**	17904.789*	13079.355*
Error	2	0.965	387.615	250.983
Soaking rate (B)	6	3.120*	362.225*	133.843NS
AXB	6	1.091*	113.870*	158.863NS
Error	24	2.208	268.544	194.976
CV (%)		63.32	45.46	20.82

*Significant at p<0.05, **Significant at p<0.01, NS: Non significant

Appendix 2: Summarized analysis of variance (ANOVA) for destructive and non-destructive parameters

		Mean squares							
Source of variation	df	Seedling vigor index	Plant height (cm)	Leaf area (cm)	Root volume (mL)	Leaf dry weight	Stem dry weight	Root dry weight	Total dry weight
						(g)			
Replication	2	8004.097 NS	4.356 NS	4.681 NS	0.584 NS	0.013 NS	0.002 NS	0.002NS	0.038NS
Coffee type (A)	1	68284.502**	0.166 NS	15.986*	0.198*	0.001 NS	0.001*	0.001*	0.002*
Error	2	422.523	1.144	1.855	0.062	0.019	0.001	0.001	0.025
Soaking rate (B)	6	4419.020*	1.675NS	9.893*	0.280NS	0.0162*	0.002*	0.004NS	0.052*
AXB	6	117.140*	1.910NS	4.403*	0.156*	0.025*	0.002*	0.002NS	0.054*
Error	24	2249.047	1.123	3.414	0.163	0.023	0.001	0.003	0.051
CV (%)		17.38	9.52	18.03	35.91	40.79	28.45	41.15	35.69

*Significant at p<0.05, **Significant at p<0.01, NS: Non significant

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