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Research Article
Viability of Shelled Rubber (*Hevea brasiliensis* Muell. Arg.) Seed Treated with PEG 6000 at Different Drying Times in Storage Period

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Abstract

Background and Objective: Storage of rubber recalcitrant seeds are very complex and dilemma, they can not be dried to under 30% moisture content and will deteriorate at low temperatures. In this condition metabolism remains active and germination process continue even in a resting state and when seeds were desiccated water content decreased, sub cellular changes began to occur, resulting in rapidly declining of seed viability. The aim of this study was to determine the viability of shelled rubber seed by PEG 6000 treatment at different drying times during storage period. **Materials and Methods:** This study was conducted using factorial design of two stage nested consisting of the factor at the first stage was the drying time from 00:00-06:00 am and 06:00-12:00 am and factor in the second stage was PEG 6000 (0, 15, 30 and 45% w/v) with two replications, where each unit consists of 75 seeds (clone PB 260). The parameters observed were: germination (%), speed of growth (%/etmal), vigor index (%) and the maximum growth potential (%) in germination phase. **Results:** At the germination phase, all variables had no significant effect, with the germination percentage were above 95% (96.58%), speed of growth were 20.87%/etmal, vigor index were 96.00% and maximum growth potential were 95.92%. **Conclusion:** The results showed that the viability of shelled rubber seed by PEG 6000 treatment at different drying times during storage period was not significantly different in the germination phase at 96.58%.

Key words: Viability seeds, shelled rubber seed, PEG 6000, drying time, storage period

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The limiting factors in the rubber recalcitrant seeds are having a very short storage period, high moisture content at mature physiology (35-70%), followed by respiration rate and high metabolic rate so that depletion food reserves continues and according Pammenter and Berjak[1], it could led to any decrease in viability and premature aging before the seed became plant material. In this case seed coating treatment is required, which is one method of enhancement. Copeland and McDonald[2] did enhancement methods to improve seed quality by the addition of chemicals to the coatings to control germination. This was supported by Ayranci and Sahin[3] and IPS[4] which stated that the PEG with molecular formula of ($HO\text{-}CH_2\text{-}(CH_2\text{-}O\text{-}CH_2)x\text{-}CH_2\text{-}OH$) is a long-chain polymer compound, unchanged (inert) and not ionic and non-toxic and does not affect the metabolism of seed and seed physiology function after periods of storage. Previously Michel and Kaufmann[5] reported that PEG 6000 has an osmotic potential can be used to create high osmotic pressure substrates without the effects of poisoning. Charloq *et al.*[6,7] has reported the results of their study that the opened shell rubber seed was to act more selectively and coated the seeds with PEG 6000 as an alternative method to induce secondary dormancy and to reduce the rate of seed metabolism in order to increase the shelf life of rubber seed in maintaining its viability. Drying or desiccating recalcitrant seeds were needed before the storage period but until this time it is a real dilemma, as reported by Chandel *et al.*[8], Bonner[9] and Wesley-Smith *et al.*[10] that all species with recalcitrant seeds were very sensitive to drying treatment and showed changes in physiological characteristics and decreased their viability dramatically. The effects of dehydration on recalcitrant seeds have been reported by Corbineau *et al.*[11] that the sensitivity of desiccation (drying) relates to the content of oligosaccharides and cell membrane materials that can trigger an increase in electrolyte leakage, deterioration and lipid peroxidation of cell membranes. The difficulty of determining desiccation according Pammenter and Berjak[1] when the seeds were dried degradation that was based on the oxidation of water activity and metabolic disorder. Therefore it is necessary to study the drying of seeds with different time and treated the shelled rubber seeds by PEG 6000, to increase storability and maintain seed viability. The purpose of this study was to determine the viability of shelled rubber seeds with PEG 6000 at different drying times during storage period.

MATERIALS AND METHODS

The materials used in this study were rubber seed (clone PB 260, moisture content 50.28%), PEG 6000 (code 8.07491.1000 1 kg, Merck Schuchardt OHG 85662 Hohenbrunn, Germany), fungicide with active ingredient (a.i) of pyraclostrobin and metiram (Cabrio Top 60 WP), insecticide (Sevin 80S), aquadest, alkohol 90%, sterilized sand. The equipments used were shell seed breaker, measuring cup, perforated plastic bag, ventilated boxes, germination box, thermohygrometer and other supporting materials. This study was conducted using factorial design of two stage nested consisting of the factor at the first stage was drying time from 00:00-06:00 am and 06:00-12:00 am and factor in the second stage was PEG 6000 (0, 15, 30, 45% w/v) with two replications, where each unit consists of 75 seeds (clones PB 260).

Research implementation: The seeds were washed three times and then drained. The seeds shell were cracked to select to see the healthy seed endosperm which should be white, dense and hard. PEG 6000 solution consists of four levels of concentration (0, 15, 30 and 45% w/v), which were dissolved in distilled water and then fungicide with a dose of 40 g/1 kg of seed was added and stirred until uniform. Seed coating was done by dipping the seeds into the solution for 10 min, then removed and drained. After that, the seed were dried at two different drying times (00:00-06:00 am and 06:00-12:00 am) in the laboratory by laying the seed, on large sheets of paper for 6 h. The temperature and humidity at the first drying time were T = 28,61 °C, RH 68% and at the second drying time were T = 29.67 °C, RH 66%. Seeds that have been treated packed with transparent perforated plastic and then put into a perforated cardboard box then stored at room temperature. The seeds were arranged randomly using factorial design nested two stage with two replications. Measurement of temperature and humidity of the room were done during 16 days of storage, three times daily i.e., morning, afternoon and evening (the average daily temperature was 28.41 with 25.4 °C min and 31.5 °C max) and the daily average of RH was 70.81% (RH min 53%-max 83%). After the 16 days storage period, seeds were germinated for 21 days on the germination vessel with sterilized sand media. Seed viability was measured by observation of germination (%), speed of growth (%/etmal), vigor index (%) and maximum growth potential (%). All data were the analyzed statistically.

RESULTS AND DISCUSSION

Results of study were arranged in Table 1. Table 1 showed that at the germination phase, all variables had no significant effect, with the germination percentage were above 95% (96.58%), speed of growth were 20.87%/etmal, vigor index were 96.00% and maximum growth potential were 95.92%. This was supported by Charloq and Chairunnisa[12] that rubber

Table 1: Observations of the averages of variable seed viability in germination phase

	Variable observation							
	Germinated seed (%)		Speed of growth (%/etmal)		Vigor index (%)		Maximum growth potential (%)	
Treatments	DT 1	DT 2	DT 1	DT 2	DT 1	DT 2	DT 1	DT 2
PEG 6000 (0% w/v)	98.67[a]	92.67[a]	27.60[a]	18.65[a]	98.00[a]	92.67[a]	98.67[a]	95.33[a]
PEG 6000 (15% w/v)	96.00[a]	98.67[a]	20.68[a]	20.43[a]	96.00[a]	97.34[a]	97.34[a]	92.67[a]
PEG 6000 (30% w/v)	100.00[a]	92.00[a]	20.92[a]	19.38[a]	98.67[a]	92.00[a]	96.00[a]	98.00[a]
PEG 6000 (45% w/v)	98.67[a]	96.00[a]	20.94[a]	18.39[a]	98.00[a]	95.34[a]	97.34[a]	92.00[a]
Average	98.33[a]	94.83[a]	22.53[a]	19.21[a]	97.67[a]	94.33[b]	97.33[a]	94.50[a]
General average	96.58	96.58	20.87	20.87	96.00	96.00	95.92	95.92

Numbers followed by the same letter in the column/row between treatments had no significant effect in Duncan's multiple range test, PEG 6000 (% w/v): Polyethylene glycol (% w/v), DT: Drying time

seeds have been selected by opening their shell and treated by coating with PEG 6000 can be protect and also different drying times and did not had different at effect on the variable observed. At drying time 1 (00:00-06:00 am) the germination percentage was 98.33 and 94.83% for the drying time 2 (06:00-12:00 am), speed of growth was 22.53%/etmal for the first drying time and 19.21%/etmal for the second drying time, maximum growth potential was 97.33% for the first drying time and 94.50% for the second drying time, while for the vigor index was 97.67% for the first drying time that was significantly different from the second drying time i.e., 94.33%.

According to King and Roberts[13], recalcitrant seed with high water content were not drought resistant and did not have a dormancy period and according to Berjak and Pammenter[14,15], handling of recalcitrant seed was on dilemma because the seeds were not resistant to drying on desiccation due to rapidly aging, but the results of this study indicated that two different drying time produced that all observations variables still have a viability above 95%, demonstrating that the viability of the seeds can be maintained quite high. This means that the biochemical mechanism that occurs in the cell membrane of seeds coated with PEG 6000, still running well enough to maintain the vitality of the seeds although it has been dried. This was supported by Charloq *et al.*[7] who reported that the water content of a recalcitrant seeds can be used an indicator in determining the resilience of the seeds where the high water content could trigger respiration rate. In the rubber seed storage, the respiration rate is very important to control because it can drain seed food reserves that led to the loss of viability. Seeds without PEG 6000 and without fungicide respiration rate of O_2 consumption increased significantly during the storage period of 16 days. After treated with PEG 6000 increase in value of the respiration rate of O_2 consumption was not significantly increased and the same with the increase in the respiration rate of CO_2 production was not increase significant. It indicated that any increase O_2 consumption and CO_2 production on respiration showed an increase of carbohydrate oxidation through respiration and the release of CO_2 is characteristic of the fast compositional changes that occur in seed tissue. The high vigor index (96.00%) indicated that storability of seeds also high, that the seeds still had a high enough energy supply that can be used by the embryo to grow and develope in the germination phase. On the other hand, rubber seed with a high moisture content seeds is highly susceptible to fungal attack in storage period, but this had been prevented the fact that the viability of the seeds was above 95%. These results supported previously by Charloq *et al.*[6] where the combination of PEG 6000 30% and fungicides 40 g/1 kg was able to maintain storability of the seeds by founding up 10.67% germination and fungal attack up to 18.00% during 16 days of storage with 96.80% germination. PEG 6000 treatment techniques combined with a fungicide (a.i., phyraclostobin-metiram) provide a dual role in suppressing the proliferation of fungi in seed storage. Chemical Company BASF[16] reported that, a.i., phyraclostridium and metiram may protected the seeds from fungal attack during storage by suppressing the development of the fungus through the mechanism of disrupt in the formation of cell walls, cell membranes, protein synthesis and the transformation of the energy associated with the mitochondrial electron transport in the fungus.

High growth rates were indication of storability of high group of seeds, it was in line with the statement of Filho[17] that the seeds that have high growth rates have a high level of vigor. Seed viability with an index that has a high vigor, can help the seeds to grow faster in the optimum and the sub-optimum environment. The maximum potential growth of seed was high enough, showing that total viability benchmarks, to demonstrate that the ability of seeds for subsistence, it was suspected that the seeds were not stressed by the conservation measures through PEG 6000 as seed preservation. This is in accordance with the standard of Don[18] that the viability and vigor was a description of a set of

properties owned by seed to determine the potential level of activity and performance of the seed or seed lot during germination and seedling emergence. The results in this study has shown a significant increase compare to a study of recalcitrant seeds of *Avicennia marina* (Forsk.) Vierh by Halimursyadah[19] when reported that seeds which were stored at room temperature of (27-28°C) and RH of 83% for 2 weeks only had germination of 36.00% from the early 89.33%.

CONCLUSION

The results showed that treatment with PEG 6000 at different drying time on storage period had seed viability that not differ significantly with the germination phase (96.58%).

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REFERENCES

1. Pammenter, N.W. and P. Berjak, 2014. Physiology of desiccation-sensitive (recalcitrant) seeds and the implications for cryopreservation. Int. J. Plant Sci., 175: 21-28.
2. Copeland, L.O. and M.B. McDonald, 2001. Principles of Seed Science and Technology. 4th Edn., Kluwer Academic Publisher, London, Pages: 467.
3. Ayranci, E. and M. Sahin, 2008. Interactions of polyethylene glycols with water studied by measurements of density and sound velocity. J. Chem. Thermodyn., 40: 1200-1207.
4. IPS., 2000. Polyethylene glycol 6000. Institute of Pharmaceutical Science and Research Centre, Bhagwant University, India, pp: 53-55. http://shodhganga.inflibnet.ac.in/bitstream/10603/28951/12/12_chapter%206.pdf
5. Michel, B.E. and M.R. Kaufmann, 1973. The osmotic potential of polyethylene glycol 6000. Plant Physiol., 51: 914-916.
6. Charloq, Z. Lubis, T.H. Siregar, J. Elisa, B.A. Sirait and N.T. Mathius, 2013. PEG 6000 ability test and fungicide efficacy in improving storability of shelled rubber (*Hevea brasilliensis* Muell. Arg.) seed. Res. J. Seed Sci., 6: 40-48.
7. Charloq, Z. Lubis, T.H. Siregar, S.B. Damanik, A. Yazid and M. Husni, 2016. Physiology changes of shelled rubber (*Hevea brasilliensis* Muell. Arg.) seed after 16 days storage with PEG 6000 30% coating to induce secondary dormancy. J. Agron., 15: 11-18.
8. Chandel, K.P.S., R. Chaudhury, J. Radhamani and S.K. Malik, 1995. Desiccation and freezing sensitivity in recalcitrant seeds of tea, cocoa and jackfruit. Ann. Bot., 76: 443-450.
9. Bonner, F.T., 1996. Responses to drying of recalcitrant seeds of *Quercus nigra* L. Ann. Bot., 78: 181-187.
10. Wesley-Smith, J., N.W. Pammenter, P. Berjak and C. Walters, 2001. The effects of two drying rates on the desiccation tolerance of embryonic axes of recalcitrant jackfruit (*Artocarpus heterophyllus* Lamk.) seeds. Ann. Bot., 88: 653-664.
11. Corbineau, F., M.A. Picard, J.A. Fougereux, F. Ladonne and D. Come, 2000. Effects of dehydration conditions on desiccation tolerance of developing pea seeds as related to oligosaccharide content and cell membrane properties. Seed Sci. Res., 10: 329-339.
12. Charloq and H.T. Chairunnisa, 2004. The effects of polyethylene glycol on the moisture content and germination of shelless rubber seeds (*Hevea brasiliensis* Muell. Arg.) during sixteen days' storage. Proceedings of the 27th International Seed Testing Association Congress Seed Symposium, May 17-19, 2004, Budapest, Hungary.
13. King, M.W. and E.H. Roberts, 1980. Maintenance of Recalcitrant Seeds in Storage. In: Recalcitrant Crop Seeds, Chin, H.F. and E.H. Roberts (Eds.). Kuala Lumpar Tropical Press, Malaysia, pp: 53-89.
14. Berjak, P. and N.W. Pammenter, 2008. From *Avicennia* to *Zizania*: Seed recalcitrance in perspective. Ann. Bot., 101: 213-228.
15. Berjak, P. and N.W. Pammenter, 2013. Implications of the lack of desiccation tolerance in recalcitrant seeds. Front. Plant Sci., Vol. 4. 10.3389/fpls.2013.00478.
16. BASF Chemicals Company, 2012. CABRIO® PLUS fungicide potato technology sheet. AgSolutions® by BASF, Ludwigshafen, Germany. https://agro.basf.ca/East/Products/Related_Files/CABRIO_PLUS_Potato_TechSheet_2012.pdf
17. Filho, J.M., 2015. Seed vigor testing: An overview of the past, present and future perspective. Scientia Agricola, 72: 363-374.
18. Don, R., 2006. ISTA Handbook on Seedling Evaluation. 3rd Edn., International Seed Testing Association, Switzerland, ISBN: 3906549399, Pages: 119.
19. Halimursyadah, 2012. [The effect of condition at various storage periods on seed viability and vigor of *Avicennia marina* (Forsk.) Vierh]. Jurnal Agrotropika, 17: 43-51, (In Indonesian).