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Ethephon Effect on Peanut Seed Dormancy Release

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Abstract: Dormancy is undesirable character for the peanut seed (*Arachis hypogeae*). KK 84-7 and KK 6 varieties are characterized to the Virginia type and its dormant period has been one to two months after harvest. The recommendation for breaking peanut seed dormancy by the International Seed Testing Association (ISTA) rules is preheat at 40 °C up to 168 h. The total germination test is 17 d, if breaking dormant seed is required. Effect of ethephon on peanut seed dormancy release was studied in this experiment to reduce the analysis time compared to preheat method. Both varieties of peanut seeds were directly mixed with 0.96% ethephon and preheated at 40 °C for 168 h. Standard germinations were conducted for all treated and untreated seeds. Ethephon was the most beneficial to release dormant seed at fresh harvest that achieved 86% and 84% normal seedlings for KK 84-7 and KK 6, respectively. The normal seedlings of preheat treatment showed 75% for KK 84-7, and 66% for KK 6. Only 6% normal seedlings were observed in untreated seeds of KK 84-7 and 56% of KK 6. After storage at 20 °C for 28 d, KK 84-7 had over 90% normal seedlings with both ethephon and preheat methods, but only 42% germination was observed in untreated seeds. In KK 6, the highest germination by 90% was found in ethephon, followed by untreated and preheated seeds that were 87% and 83%, respectively. The paired *t* test of normal seedlings between ethephon and preheat treatments demonstrated that the greater average germination was found in ethephon method for both varieties. This research suggests that ethephon is the advantageous method for breaking peanut seed dormancy. The germination test duration is more rapid, only 10 d, since preheat for 168 h is not necessary.

Key words: Peanut seed, dormancy, ethephon.

1. Introduction

Peanut (*Arachis hypogeae* L.) is the economic crop in Thailand. It is mainly used for edible food and raw material for food processing products [1]. There are generally three botanical types of peanuts following Virginia, Valencia and Spanish types. Seed dormancy is commonly found in Valencia type, and the dormant period is variable depending on its varieties [2]. Previous research found that ethylene, plant growth regulator, is able to release seed dormancy and enhance seed germination [3, 4]. Ethephon (2-chloroethy phosphonic acid) is the ethylene releaser breaking seed dormancy in numerous species such as lettuce [5], sunflower [6], and chickpea [7]. The dormant peanut seed cv. MJU60 and MJU75 were also released by 0.83×10^{-5} M ethephon, but higher concentration at 6.4×10^{-4} M was required for cv. MJU80 suggesting the variation of peanut seed varieties and depth dormancy [8]. Moreover, Wang *et al.* [9] demonstrated that ethephon resulted in increased average germination up to 97.8% of 103 accessions peanut seeds compared to water treatment. However, this technique mostly is conducted on peanut seed growing in the field. The application of ethephon treatment on seed quality testing has been rarely reported. According to seed testing rules of International Seed Testing Association (ISTA), the recommendation for breaking peanut seed is preheat at 40 °C in hot air oven up to 168 h followed by standard germination test for 10 d that is totally 17 d [10]. If the more rapid and precise method is available, it will be advantageous to reduce time for seed testing and seed certificate. Ethephon might be useful and applicable in this case. The aim of this study was to investigate the effect of ethephon on releasing dormancy of two varieties of peanut seeds.

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The comparison between breaking dormancy methods, ethephon and preheat treatments, was also determined for further application.

2. Materials and Methods

Fresh harvested peanut seeds KhonKaen 84-7 (KK 84-7) and KhonKaen 6 (KK 6) varieties were immediately processed as drying, cleaning, and then carried out to the laboratory for experiment. Peanut seed pods were packed in the woven plastic bag and stored at 20-22 °C for 28 d. Sample was randomly taken at 0, 7, 14, 21, 28 d after storage to the laboratory. Seed pods were threshed and four replicates with 100 seeds were prepared in each treatment. Threshed seeds were immediately applied to two breaking seed dormancy treatments compared with untreated seed prior to standard germination test as following;

(1) Untreated seeds (control);

(2) Preheat at 40 °C by hot air oven for 168 h (7 d) [10];

(3) 0.96% ethephon solution, prepared from 48% Ethrel diluted with distilled water in the ratio of 2 ml/L.

Thereafter, standard germination of treated and untreated peanut seeds was determined by sand method according to ISTA rules [10]. After 10 d, normal and abnormal seedlings, and fresh and dead seeds for all treatments were counted. Means of germination and fresh seed for each treatment was compared by Duncan's New Multiple Range (DMRT) Test, and the germination percentage of the two groups of breaking seed dormancy treatments was examined with the paired sample *t* test.

3. Results and Discussion

3.1 Effect of Breaking Seed Dormancy Treatments on KK 84-7 Variety

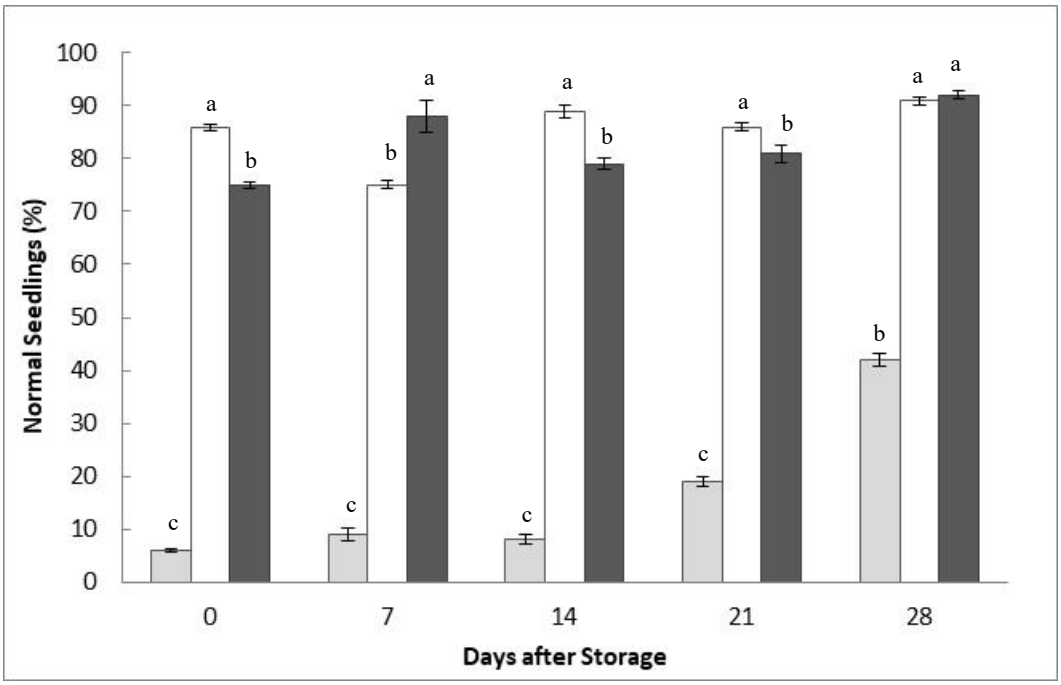
Peanut seed dormancy was released either preheated at 40 °C for 168 h or 0.96% ethephon treatments. Prior to storage, the highest normal seedling was found to be 86% in the 0.96% ethephon treatment, which was significantly different compared to preheat and untreated seeds shown 75% and 6%, respectively (Fig. 1a). The 5% of fresh seeds were observed in 0.96% ethephon that was significantly lower than other treatments. There were 11% of fresh seeds in preheat and 89% in untreated seeds (Fig. 1b).

After storage, the increased normal seedlings with the decreased fresh seeds of untreated peanut seeds were revealed with the longer storage time. At 28 d of storage, the normal seedlings and fresh seeds of untreated seeds were 42% and 32%, respectively (Figs. 1a and 1b). Meanwhile, the peanut seeds treated with 0.96% ethephon did not observe any fresh seeds and only 1% was found in preheat at 40 °C at the end of storage (Fig. 1b). The normal seedlings were not significantly different between 0.96% ethephon and preheat treatment that was 91% and 92%, respectively, but these results seemed significantly different compared to untreated peanut seeds (Fig. 1a). The confirmation of fresh seeds by 1% tetrazolium chloride was revealed in Fig. 2.

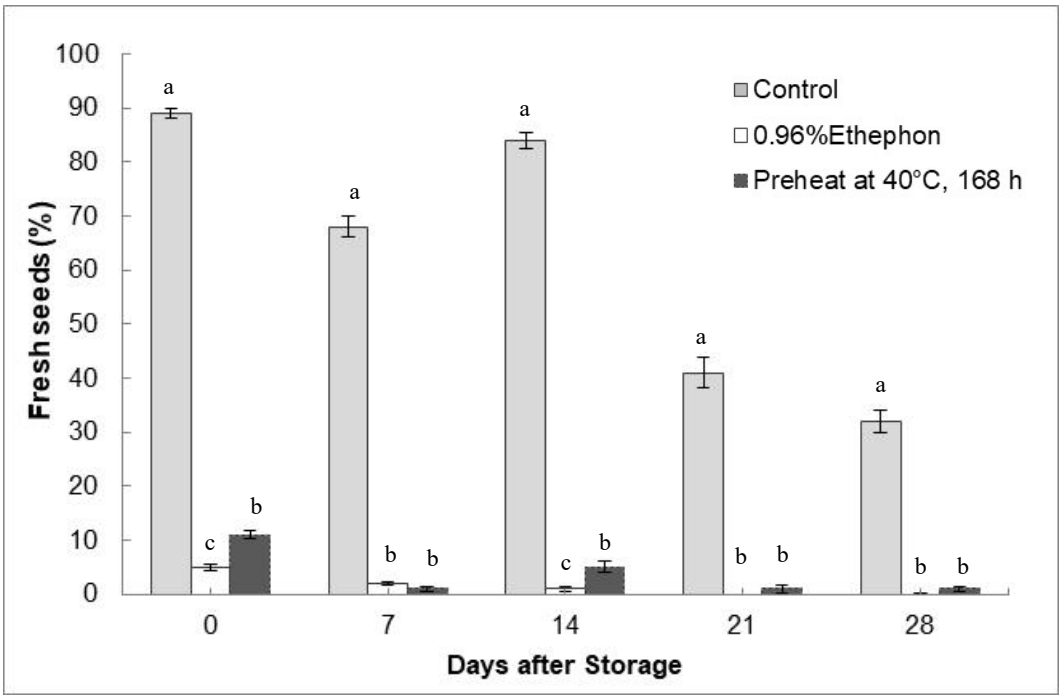
The comparison of breaking seed dormancy methods between 0.96% ethephon and preheat at 40 °C was analyzed by paired *t* test. The results showed that average normal seedlings of 0.96% ethephon were 85% and 83% for preheat at 40 °C. The paired *t* test showed non-significant difference between two methods ($p > 0.001$) that meant 0.96% ethephon can be used for breaking peanut seed dormancy instead of the preheat at 40 °C for 168 h (Table 1). The total period for ethephon breaking seed dormancy is 10 d included germination test. However, 17 d is required for preheat method in accordance with incubation at 40 °C for 7 d prior to germination test for 10 d that is needed.

3.2 Effect of Breaking Seed Dormancy Treatments on KK6 Variety

The initial standard germination of KK 6 variety was investigated before storage at 20-22 °C. The result found 56% of normal seedlings and 26% of fresh



(a)



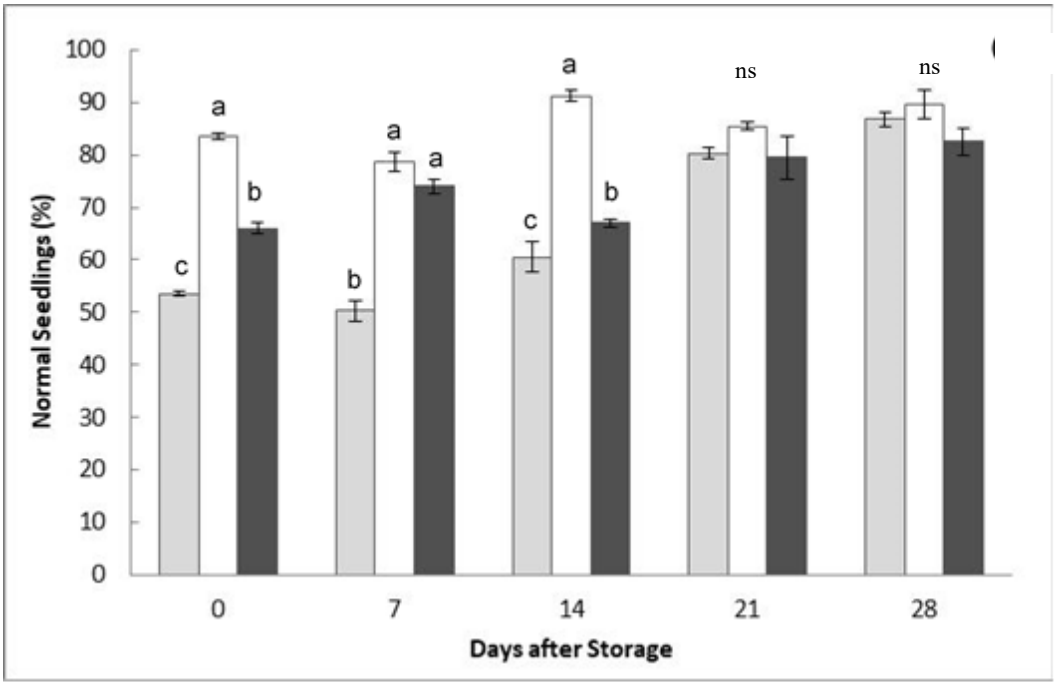
(b)

Fig. 1 Germination (% normal seedling) (a) and fresh seeds (b) of KK 84-7 peanut seed treated with 0.96% ethephon and preheat at 40 °C for 168 h compared to untreated seed (control) stored at 20 ± 2 °C for 28 d.

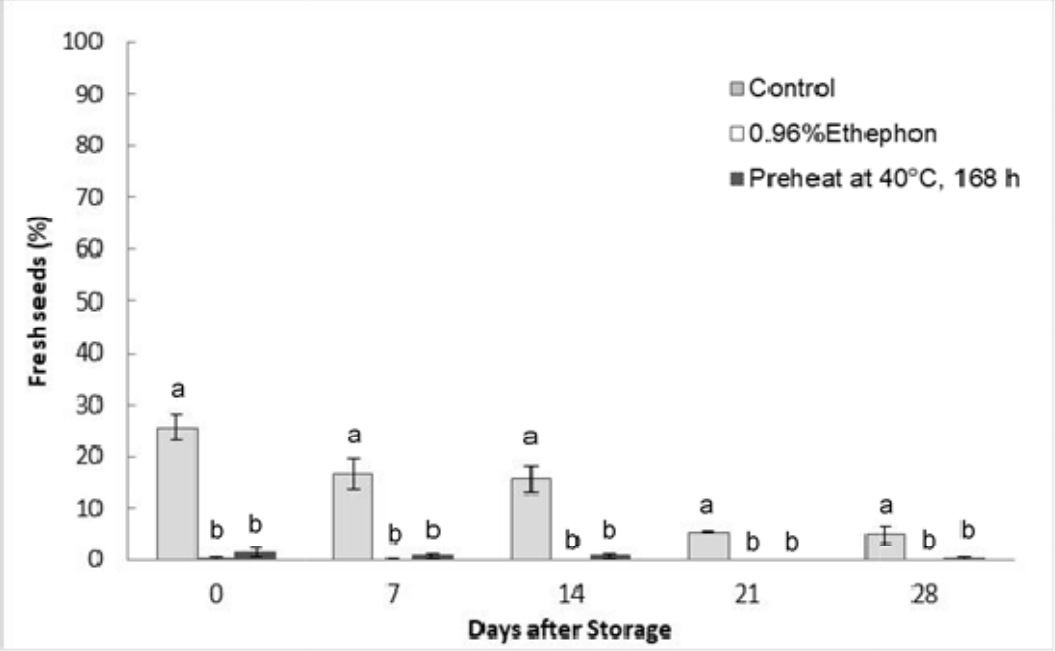
Means (± S.E.) followed by the same letter for 0, 7, 14, 21 and 28 d bars, respectively, were not different at α = 5% by DMRT test.

(a) (b)

Fig. 2 Fresh seed of peanut var. KK 84-7 without stained (a) and stained with 1% tetrazolium chloride.



(a)



(b)

Fig. 3 Germination (% normal seedling) (a) and fresh seeds (b) of KK 6 peanut seed treated with 0.96% ethephon and preheat at 40 °C for 168 h compared to untreated seed (control) stored at 20 ± 2 °C for 28 d.

Means (± S.E.) followed by the same letter for 0, 7, 14, 21 and 28 d bars, respectively, were not different at α = 5% by DMRT test. No letter was non-significant (ns) among treatments.

Table 1 The comparison of germination percentage between ethephon and preheat methods by paired sample *t* test of var. KK 84-7 and KK 6 peanut seeds.

Varieties	Comparison[a]	Mean (%)	Std. dev.	Std. error	*t* value	df	*p* value
KK 84-7	G_{Et} vs. G_{Pre}	G_{Et} 85 G_{Pre} 83	9.83	2.20	1.05	19	0.308
KK 6	G_{Et} vs. G_{Pre}	G_{Et} 86 G_{Pre} 74	8.88	1.99	5.99	19	< 0.001

[a] Germination (%) of peanut seed treated with 0.96% ethephon (G_{Et}) and preheat at 40 °C, 168 h (G_{Pre}).

seeds in untreated peanut seeds (Figs. 3a and 3b). After breaking seed dormancy by 0.96% ethephon and preheat at 40 °C for 168 h, the normal seedlings for both treatments were significantly increased compared to untreated seeds. The highest normal seedlings belonged to 0.96% ethephon that was 84%, followed by 66% in preheat treatment (Fig. 3a). Moreover, no fresh seed was observed in 0.96% ethephon and 2% fresh seeds of preheat method were revealed (Fig. 3b).

During storage, peanut seeds were taken every 7 d to monitor its germination until the end of storage. The normal seedlings of untreated seeds were 87%, and 5% fresh seeds were found at 28 d of storage (Figs. 3a and 3b). Meanwhile, seeds treated with 0.96% ethephon showed the greatest number of normal seedlings followed by preheat method that was 90% and 83%, respectively (Fig. 3a). However, these figures were not significantly different at the end of storage (Fig. 3a), and there were no fresh seeds observed for both treatments (Fig. 3b).

The comparison between normal seedlings of 0.96% ethephon and preheat at 40 °C was investigated by paired *t* test. The average normal seedlings of 0.96% ethephon were significantly higher than preheat treatments ($p < 0.001$) that were 86% and 74%, respectively (Table 1). This evidence would be clearer for the ethephon effect on breaking peanut seed dormancy and enhancing seed germination compared

to preheat method.

The dormancy of peanut seed could be related to physiological, morphological and genetic effects [9, 11]. Peanut seeds KK 84-7 and KK 6 varieties are large seed size and belong to the Virginia botanical type with dormant duration around one to two months after harvest [1, 2, 12]. This is related to a lot of fresh seeds found at beginning that was 89% for KK 84-7 and 26% for KK 6 varieties. This phenomenon may involve with the less ethylene production in embryo and seed coat [9, 13, 14], or an abscisic acid (ABA) accumulation in seed coat [11]. In this study, both ethephon and preheat treatments were clearly demonstrated to release seed dormancy since harvest. However, the greater germination with not exceeding 5% fresh seeds was investigated in ethephon treatment for both varieties. It suggests that ethylene involves in stimulating seed germination and dormancy [3, 4, 15, 16]. Moreover, ethylene counteracts ABA was reported through a regulation of ABA metabolism and signaling pathways [6, 15, 16] caused to promote seed germination. Previously, there was successfully used ethephon (2-chloroethylphosphonic acid), ethylene releasing compound, as a stimulant for dormancy release in many crops including legumes species [9, 11, 17, 18]. Wang *et al*. [9] reported that the average germination of peanut seeds applied with 10 mM ethephon showed 97.8% higher than that of 56.7% H_2O treatment. The long dormant peanut seed cv. MJU80 was released by soaking 6.4×10^{-4} M ethephon for 48 h that the average germination reached 70% and 80% after 7 and 21 d of storage, while only 5% germination was observed in control seeds [8]. This research implies that 0.96% ethephon would help to release peanut seed dormancy since fresh harvest for both varieties. This method is also fast and easy compared to preheat method according to no requirement of incubation time for 168 h.

4. Conclusion

The experiment suggests that 0.96% ethephon is the advantageous method for breaking peanut seed dormancy of KK 84-7 and KK 6 varieties according to breaking seed dormancy and promoting seed germination since fresh harvest. Analysis time, including breaking seed dormancy and germination test, was decreased from 17 to 10 d using 0.96% ethephon compared to preheat method. This method, therefore, may introduce for seed testing laboratory instead of preheat treatment.

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