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Effect of thiamethoxam and imidacloprid treatment on germination and seedling vigor of dry-heated seed of oil palm (*Elaeis guineensis* Jacq.)

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Rapid and uniform seedling growth is a pre-requisite for better establishment of oil palm plantations. Chemical pretreatments can be used to increase the rate and uniformity of seed germination. Thiamethoxam and imidacloprid are insecticides classified in the broad-spectrum systemic neonicotinoid group and their use in seed treatment has been claimed to increase seed germination and vigor in some crops. Thiamethoxam at the rate of 0, 0.1, 0.3, 0.6 and 0.9 g ai. L⁻¹ of water and imidacloprid at the rate of 0, 0.2, 0.5, 0.9 and 1.4 g ai. L⁻¹ of water were evaluated using dry-heated seed (seed with 18% moisture content heated at 40°C for 60 days) to observe the differences in germination response and seedling vigor. The results revealed that imidacloprid at the rate of 0.5 g ai. L⁻¹ of water significantly increased shoot length while thiamethoxam did not. Both neonicotinoids did not affect the germinated seed percentage, the time taken to reach 50% germination (T₅₀) and root length. It can be concluded that imidacloprid could increase seedling vigor by stimulating the shoot growth of dry-heated oil palm seed. No phytotoxicity of thiamethoxam and imidacloprid following dry-heated seed germination was observed.

Key words: Thiamethoxam, imidacloprid, germination, seedling vigor, *tenera* oil palm seed.

INTRODUCTION

Given the current situation, oil palm makes up a prominent part of the volume of international trade, due to a ceaselessly growing demand for palm oil in the world (Martine et al., 2009). In fact, palm oil production will continue to be a world leader for many years because of its variety of food and non-food uses. World production of palm oil has also seen substantial leaps in production through an expansion of the planted area that has been achieved mainly by the opening up of new land (Donough et al., 2009). Cultivation in the new and old areas still

uses germinated hybrid seeds as the plant propagation material, although tissue culture technology has been established in breeding programs (Dikin et al., 2003; Martine et al., 2009). Under natural conditions, oil palm seed germination is very slow with a low percentage that represents a major constraint for the establishment of the plantations (Rees, 1963; Martine et al., 2009).

Physical dormancy contributes to the long germination process in oil palm seed (Martine et al., 2009), with commercial *tenera* oil palm seeds requiring a pretreatment of heating at 40°C for 60 days. Fondom et al. (2010) reinvestigated the effect of heat-treatment duration on germination and subsequent seedling growth of 10 oil palm progenies and confirmed that a heat

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treatment at $39\pm 1^\circ\text{C}$ for 60 days was effective for breaking the dormancy of the oil palm seeds and encouraging successful seedling growth. Even though dry heat was developed in order to improve and speed up seed germination for a sustainable supply of germinated seeds to the planters, it appears constraints are associated with the length of time, the expense and the requirement of special installations. Many researchers have been trying various approaches to achieve a higher germination in a shorter time (Wonkyi-Appiah and Amuh, 1979; Murugesan et al., 2005; Myint et al., 2010; Chanprasert et al., 2012). Chemical pretreatment is an aspect that has been introduced into oil palm seed research (Wan and Hor, 1983; Herrera et al., 1988). Thiamethoxam and imidacloprid are insecticides classified in the broad-spectrum systemic neonicotinoid group (Moser and Obrycki, 2009) and their application in seed treatment has been claimed to increase seed germination and vigor in some crops (Horii et al., 2007; Stevens et al., 2008). Thiamethoxam accelerated the germination of soybean seed when used at the recommended level (Cataneo et al., 2010). According to Horii et al. (2007), thiamethoxam had a slightly positive effect on the seed vigor of pea, soybean and corn during germination. Imidacloprid has also been studied as a seed treatment on some crops such as wheat (Pike et al., 1993; Wilde et al., 2001), and barley (Pike et al., 1993). However, the use of thiamethoxam and imidacloprid as seed treatment in oil palm has not yet been reported. The objective of this study was to determine the effects of thiamethoxam and imidacloprid on the germination and vigor of commercial *tenera* oil palm seed following dormancy which was overcome by a dry-heat treatment.

MATERIALS AND METHODS

Seed material

The seeds derived from the oil palm cross no. 37 (Surat Thani 2) were obtained from the progenies of *Deli dura* (C2120:184D×HC133:1288D) × *LaMe pisifera* (IRH 618: 158T × HC129: 1056P) that were the products of a genetic selection program. The seeds utilized in this experiment were collected during 2009 from the palms located at the Surat Thani Oil Palm Research Center (09°07'N, 99°21'E), southern Thailand belonging to the Department of Agriculture, Ministry of Agriculture and Cooperatives.

Bunches of *tenera* cross no. 37 (Surat Thani 2) were harvested and the seeds were mechanically depericarped from the fruits, then normal practices for controlling fungal infection were conducted (that is, the seeds were washed with detergent and submerged in a mixture of surfactant and sodium hypochlorite for 20 min followed by dipping in the mixture of benomyl and thiram and left under shade for 24 h).

When the moisture content of the seed was about 18%, the air-dried seeds of each treatment were put in individual plastic bottle previously treated with alcohol and then heated at 40°C for 60 days.

After heating, the seed moisture content was determined. The dry heated seeds were divided into five groups, each treated with a different neonicotinoid concentration including the control.

Control seeds

According to the results of the seed moisture content analysis, the control seeds were soaked in water for 9 days to raise the moisture content to about 22% (dry weight basis). The soaked seeds were rinsed with water, washed with detergent then treated with a fungicide mixture, dried under shade for 2 h and then germinated in polyethylene bags measuring 37×26 cm at ambient temperature (mean temperature $28 \pm 2^\circ\text{C}$) under dark conditions.

Neonicotinoid chemical-treated seeds

Instead of 9 days soaking in water, the dry-heated seeds that were to be applied with neonicotinoids were soaked in water for 8 days then soaked in two different chemical amounts of either thiamethoxam (Cruiser® 35FS) (0.1, 0.3, 0.6 and 0.9 g ai. L^{-1} of water or 1.0, 2.5, 5.0 and 7.5 ml kg^{-1} of seed in 3 L of water) or imidacloprid (Gaucho® 70WS - powder) (0.2, 0.5, 0.9 and 1.4 g ai. L^{-1} of water or 1.0, 2.0, 4.0 and 6.0 g kg^{-1} of seed in 3 L of water) for 24 h to raise the moisture content to about 22%. The treated seeds were rinsed with water, washed with detergent then treated with a fungicide mixture, dried under shade for 2 h and then germinated in the polyethylene bags measuring 37×26 cm at ambient temperature (mean temperature $28 \pm 2^\circ\text{C}$) under dark conditions.

Experimental design and statistical analysis

The moisture content of seeds was determined each time after soaking in water. Five seeds of each treatment were weighed in order to determine their fresh weight (FW) and then dried in an oven at 105°C for 48 h to obtain a constant dry weight (DW). The moisture content was calculated on a seed dry weight basis (Panyangnoi et al., 1997). The germination percentage of the seed was evaluated twice each week, during the 40-day test period. The germination percentage was calculated as

$$\text{G\%} = (\text{number of seed germinated} / \text{number of sampled seeds}) \times 100.$$

The time to 50% germination (T_{50}) was determined following Coolbear et al. (1984),

$$T_{50} = t_i + [(((N+1)/2 - n_i) / (n_j - n_i)) \times (t_j - t_i)]$$

where t_i is the time before reaching 50%, n_i is the number of seeds emerged at time t_i , N is the final number of emerged seeds, t_j is the time next after t_i , and n_j is the number of seeds emerged at t_j .

After 40 days, the cumulative percentage of germination was determined. At the end of experiment, shoot and root length were measured in millimeters using a ruler. Treatments were arranged as a factorial combination of treatments in a completely randomized design with four replications. Forty seeds per replication were used for the germination test. Data were analyzed using an analysis of variance to detect differences between mean parameters. If significant differences were found, the least significant difference was used to determine mean differences between treatments.

Table 1. Effects of thiamethoxam and imidacloprid at five treatment rates (R_0 to R_4) on germination and time to reach 50% germination (T_{50}) of dry-heated *tenera* oil palm seed of Surat Thani 2.

Treatment rate	Germination (%)			T_{50} (days)		
	Thia	Imida	Mean	Thia	Imida	Mean
R_0	83.75	80.63	82.19	17.86	17.95	17.91
R_1	78.13	81.25	79.69	19.50	18.83	19.17
R_2	85.00	80.63	82.81	17.03	17.46	17.24
R_3	81.88	81.25	81.56	17.71	19.46	18.58
R_4	74.38	89.38	81.88	18.97	17.82	18.39
Mean	80.63	82.63		18.21	18.30	
LSD _{0.05} for						
Neonicotinoid (N)		ns*			ns	
Rate (R)		ns			ns	
N×R		ns			ns	
CV%		11.0			12.9	

*ns = non significance; Thiamethoxam (Thia) - $R_0=0$, $R_1=0.1$, $R_2=0.3$, $R_3=0.6$ and $R_4=0.9$ g ai. L^{-1} of water; Imidacloprid (Imida) - $R_0=0$, $R_1=0.2$, $R_2=0.5$, $R_3=0.9$ and $R_4=1.4$ g ai. L^{-1} of water.

RESULTS AND DISCUSSION

As shown in Table 1, the germination percentages ranged from 74.38 to 85.00% for the five concentrations of thiamethoxam and from 80.63 to 89.38% for the five concentrations of imidacloprid. There was no stimulating effect of thiamethoxam on germination in this study. Prasanna et al. (2004) also reported that thiamethoxam had no effect on seed germination in cotton, while some other studies found that seed germination was stimulated by thiamethoxam in soybean, pea and corn (Horie et al., 2007; Cataneo et al., 2010). For the effect of imidacloprid, the germination percentage was 80.63, 81.25, 80.63, 81.25 and 89.38% when treated at the rate of 0, 0.2, 0.5, 0.9 and 1.4 g ai. L^{-1} of water, respectively. However, imidacloprid did not significantly increase the germination percentage in this study. The speed of germination indicated by T_{50} was also not affected by both neonicotinoids (Table 1). These two neonicotinoids showed similar effects on T_{50} , attaining an average of 18.21 and 18.30 days for thiamethoxam and imidacloprid, respectively.

There was no significant difference in root length as affected by thiamethoxam and imidacloprid, while there was an interaction between two factors (neonicotinoid × rate) on shoot length (Table 2). No obvious effects of thiamethoxam were observed, while a more pronounced effect of imidacloprid was found. Imidacloprid at the rate of 0.5 g ai. L^{-1} of water significantly increased shoot length, while the other rates were not significantly different compared to the untreated control. One problem in oil palm seed germination is fungal infection and fungicides must be applied. In this study, a fungicide treatment procedure prior to the neonicotinoid treatment

might interact and lower the effect of the neonicotinoids. Buchholz and Nauen (2002) reported that the application of neonicotinoid as a seed treatment can maximize its uptake into plant tissues. Neonicotinoids are known to interact with the nicotinic acetylcholine receptor (nAChR) (Yamamoto et al., 1995; Matsuda et al., 2000). Binding to nAChRs is increased when the chemical is reduced by electrophilic compounds (phenolic antioxidants) to form positively-charged molecules. Such positively-charged neonicotinoids may possibly affect the electron-transport activities in plant cells that consequently improve seed vigor (Ayyappath et al., 2000; Revellin et al., 2001; Delgarde and Rouland-Lefevre, 2002; Foster et al., 2003). Therefore, the positive effects on shoot growth in this study suggest that further experiments by improving the treatment technique should be continued to attain better results of neonicotinoid seed treatment in oil palm seed germination. In this study, no phytotoxicity from the thiamethoxam and imidacloprid treatments in dry-heated oil palm seed was found, in agreement with our previous report (Chanprasert et al., 2012).

Conclusion

Two neonicotinoids, thiamethoxam at the rate of 0.1, 0.3, 0.6 and 0.9 g ai. L^{-1} of water and imidacloprid at the rate of 0.2, 0.5, 0.9 and 1.4 g ai. L^{-1} of water showed some positive effects on the seedling vigor of dry-heated oil palm seed. Imidacloprid at the rate of 0.5 g ai. L^{-1} of water significantly increased the shoot length of oil palm seedlings. No phytotoxic effect was observed in this experiment. Further research is needed to determine the appropriate treatment procedure to maximize seed

Table 2. Effects of thiamethoxam and imidacloprid at five treatment rates (R₀ to R₄) on shoot length and root length (mm) at 1 month after emergence of dry-heated *tenera* oil palm seed of Surat Thani 2.

Treatment rate	Shoot length (mm)			Root length (mm)		
	Thia	Imida	Mean	Thia	Imida	Mean
R ₀	5.28 ^{ab*}	5.38 ^{bc}	5.33	20.23	18.18	19.20
R ₁	5.43 ^{ab}	4.85 ^c	5.14	15.53	16.50	16.01
R ₂	5.63 ^{ab}	6.58 ^a	6.10	21.20	19.48	20.38
R ₃	6.23 ^a	4.58 ^c	5.40	22.18	15.13	18.65
R ₄	5.05 ^b	5.90 ^{ab}	5.48	16.85	17.85	17.35
Mean	5.52	5.46		19.20	17.43	
LSD _{0.05} for						
Neonitoid (N)		ns			ns	
Rate (R)		ns			ns	
N×R		1.02			ns	
CV%		17.9			6.7	

* Mean values followed by the same letters are not significantly different at the probability level of $p=0.05$. Thiamethoxam (Thia) - R₀= 0, R₁=0.1, R₂=0.3, R₃=0.6 and R₄=0.9 g ai. L⁻¹ of water. Imidacloprid (Imida) - R₀= 0, R₁=0.2, R₂=0.5, R₃=0.9 and R₄=1.4 g ai. L⁻¹ of water

germination and seedling vigor of oil palm.

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