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Evaluation of Entomopathogenic Fungi and Chemical to Control Termite, *Coptotermes curvignathus* on Mature Oil Palm and Supply Seedlings on Peat

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ABSTRACT

The use of chemical insecticide, fipronil and two entomotahogenic fungi, Beauveria bassiana *and* Metarhizium major *on termite control was evaluated. The experiment was conducted in high termite infested field with 10 year-old palm and young supply seedlings of six months after planting. Treatment on infested mature palm was conducted by spraying of product solution on the whole palm trunk and soil injection surrounding the palm bases. For young supply seedling, the palm was only treated by soil injection. For treatment with fipronil, all 8 treated palms were observed to be free from any termite activity at 3 and 6 months after treatment (MAT). However, at 9 MAT, one palm had collapsed due to severe damage in the inner trunk tissues caused by termite attack. For treatment with* B. bassiana*, out of 17 treated palms, no termite activity was found on 3 palms at 3 MAT, 5 palms at 6 MAT and 7 palms at 9 MAT. The number of palms which collapsed due to termite attack was only one palm, recorded at 3 MAT. For treatment with* M. major*, out of 10 treated palms, one palm was observed with no presence of termite activity at 3 MAT. While from 6 to 9 MAT, the number of palms with no termite infestation increased to 2 palms. Two palms treated with* M. major *collapsed due to severe damage of the inner trunk tissues caused by termite attack. For young supply seedlings, up to 9 MAT, only one palm treated with* B. bassiana *was infested by termite, while other palms were still healthy. Findings of this study showed that fipronil was the most effective, giving 100% control against termite infestation up to 9 MAT. The entomopathogenic fungus* B. bassiana *gave 41.18% control and* M. major *gave 20% control against termite infestation. All products were able to prevent infestation of termite on new supply seedling in high termite infestation areas up to 9 MAT. Possible factors that might have influenced the effectiveness of the two entomopathogenic fungi in controlling termite infestation were elaborated in detail. Further research is needed to develop biological products and delivery methods for effective control of termite on peat.*

ABSTRAK

Penggunaan racun serangga kimia, fipronil dan dua kulat entomopatogenik, Beauveria bassiana *dan* Metarhizium major *untuk kawalan anai-anai telah dikaji. Kajian telah dijalankan di kawasan pokok sawit matang berusia 10 tahun dan anak sawit sulam berumur 6 bulan selepas tanam. Rawatan pada pokok sawit matang dijalankan secara semburan larutan produk pada keseluruhan batang sawit dan suntikan tanah di sekitar pangkal pokok. Untuk anak sawit sulam, ia dirawat hanya dengan kaedah suntikan tanah. Hasil kajian mendapati rawatan fipronil berupaya mengawal kesemua 8 pokok sawit daripada serangan anai-anai pada 3 dan 6 bulan selepas rawatan (BSR). Walau bagaimanapun, pada 9 BSR, satu pokok telah tumbang akibat kerosakan teruk tisu dalaman batang yang disebabkan oleh serangan anai-anai. Untuk rawatan* B. bassiana*, daripada 17 pokok sawit yang dirawat, tiada aktiviti anai-anai ditemui pada 3 pokok selepas 3 BSR, 5 pokok pada 6 BSR dan 7 pokok pada 9 BSR. Bilangan pokok sawit yang tumbang akibat serangan anai-anai hanya satu pokok, yang dicatatkan pada 3 BSR. Untuk rawatan kulat* M. major*, daripada 10 pokok sawit yang dirawat, hanya satu pokok diperhatikan tanpa aktiviti anai-anai pada 3 BSR. Manakala pada 6 BSR dan 9 BSR, bilangan pokok sawit tanpa anai-anai meningkat kepada 2 pokok. Dua sawit yang dirawat dengan* M. major *telah tumbang akibat kerosakan teruk tisu dalaman batang yang disebabkan oleh serangan anai-anai. Untuk anak sawit sulam, hanya satu pokok sawit yang dirawat dengan* B. bassiana *telah diserang oleh anai-anai selepas 9 bulan rawatan. Hasil kajian ini menunjukkan bahawa fipronil merupakan rawatan yang paling berkesan, memberikan kawalan 100% terhadap serangan anai-anai sehingga 9 BSR. Kulat entomopatogenik* B. bassiana *memberikan kawalan 41.18% dan* M. major *memberikan kawalan 20% terhadap serangan anai-anai. Semua produk dapat mengawal serangan anai-anai pada anak sawit*

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sulam sehingga 9 BSR. Faktor yang menyebabkan kedua-dua kulat entomopatogenik kurang berkesan untuk mengawal anai-anai juga dihuraikan dengan lebih lanjut. Kajian lanjut bagi membangunkan produk biologi serta kaedah penggunaan di ladang untuk anai-anai perlu dijalankan.

Keywords: entomopathogenic fungi, *Coptotermes curvignathus*, fipronil, oil palm.

INTRODUCTION

Termite is one of the main insect pests that is present in oil palm plantation. The common genus that usually causes damage is *Coptotermes*. The colony can attack oil palm as early as 12 months after field planting (Lim and Silek, 2001). The infestation can cause palm death of more than 5.3% in a year (Basri *et al*., 2003). Termite management mostly relies on the use of organophosphates termiticides, especially chlorpyrifos and fipronil. These chemicals were commonly applied by drenching or spraying at the base of the infected palm (Lim and Silek, 2001; Zulkefli *et al*., 2012). However, over usage of pesticides was proved to be uneconomical and hazardous to environment and other beneficial termites such as the scavengers and wood feeders. In Malaysia, studies by Pik-Kheng *et al*. (2009) and Ramle *et al*. (2011) showed that the infection by entomopathogenic fungi *Beauveria bassiana* and *Metarhizium major* killed 100% *C. curvignathus* when treated with spore solutions between 10^7 spores ml^{-1} and 10^8 spores ml^{-1}. Therefore, the development of biological control agents such as entomopathogenic fungi is needed in order to reduce the dependency to chemical usage. This study was conducted to evaluate the effectiveness of entomopathogenic fungi and fipronil to control termite, *C. curvignathus* on healthy supply seedling and infected mature oil palm on peat in Sarawak.

MATERIALS AND METHODS

Experimental Sites

The study was conducted in Block 306 3A at Naman Estate, Ta Ann Plywood Sdn Bhd, Sarawak. The palms were 10 years old DxP planted on deep peat. The experimental block consisted of 30 palm rows with 17 planting points in each row. Prior to the treatments, census was conducted to estimate the severity of termite infestation and the results showed that there were 57 vacant oil palm planting points and 6 rotten stumps. Most of the palms in the vacant planting points had collapsed because of termite attacks. Some of collapsed palm trunks were completely decomposed. Based on this data, the infestation of termite in the experimental block was estimated at 12.4%. In all 6 rotten stumps, termite activities were still active. To replace vacant palms, the estate has planted 28 new young supply seedlings. The age of seedlings at the commencement of this experiment was about six months after planting.

Prior to treatment, the infested mature palms were census and marked. Determination of infested palm was based on observation of fresh damage and fresh mud-work on palm trunk. In some cases, a termite baiting trap was also installed either in soil or on palm trunk that exhibit infestation symptoms. There are two different types of baiting traps, namely termite detector station made from a 30 cm long PVC tube with perforated part at the bottom of the station. The station was filled with wet corrugated cardboard and buried in soil around the infected palm base. The second trap was a soap box termite trap, which was also filled with wet corrugated cardboard and tied onto the infested palm trunk.

Treatment

Three treatments were tested in this experiment, namely entomopathogenic fungi *Metarhizium major* and *Beauveria bassiana* and chemical fipronil. Palms treated with chemical fipronil were considered as positive control, as this chemical has been widely used for treatment by planters (Lim and Silek, 2001). Therefore, the treated palms were used as positive control in this experiment and have successfully prevented recurrence of new infestation. No untreated control was presented in the study because of the periodically sprayed activity done to prevent more palms from being infested.

***Preparation of* M. major.** The source of *M. major* used in the study was obtained from a commercial product Ory-X. Before application, spore solutions were prepared by mixing the product with water plus 0.02% wetting agent Tween 80 and the concentrations of spore solution was adjusted to 10^7 spores ml^{-1}.

***Preparation of* B. bassiana.** The fungus was mass produced on autoclaved rice in heat resistant transparent plastic bags and fermented continuously for two weeks. The spores were then harvested by adding 500 ml water plus 0.02% tween 80 into the bags, the bags were thoroughly shaken and the mixture poured via a 200 μm mesh sieve. The spore concentration was determined by haemocytometer and then the concentration was adjusted to 10^7 spores ml^{-1}.

Preparation of fipronil. The product used in the study is a commercial product Chalcid 5SC (fipronil 5% w/w). The product was diluted by mixing one millilitre product with ten litres of water. Plots of oil palms treated with fipronil were used as positive control.

Number of Tested Palms and Application Rate

The effectiveness of entomopathogenic fungi and fipronil to control termite was evaluated on two categories of palms; 1) infested mature palm and 2) healthy young supply seedling. The numbers of both categories of palms used in each treatment is shown in *Table 1*.

TABLE 1. NUMBERS OF INFESTED MATURE PALM AND YOUNG SUPPLY SEEDLING USED IN THE STUDY

Treatment	Infested mature palm (N)	Healthy supply seedling (N)
Metarhizium major	17	14
Beauveria bassiana	10	10
Fipronil	8	4

Calibration of Sprayer and Motorised Injector. Prior to treatment, the knapsack and motorised sprayers were calibrated to ensure uniform and accurate application of each treatment to every treated palm.

Knapsack sprayer. A 16 litres knapsack sprayer used in this study was equipped with lance and an adjustable cone-shaped nozzle. The whole palm trunk was sprayed until satisfactory water coverage on trunk surface was achieved. It was concluded that 4 minutes was required to spray 8 litres of water to achieve satisfactory wetness on palm trunk.

Motorised knapsack injector. A 20 litres motorised knapsack injector was filled with water solution mixed with white water-based dye. The solution was then injected into the soil at a depth of 15 cm, where the dispersion and penetrability of the dye solution were observed and noted. The calibration found the flow rate of the injector was at 10 seconds to deliver 1 litre of dye solution.

Application of Treatments

Treatments were applied as follows.

1. Spraying of palm trunk
2. Soil injection around the palm base.

Spraying of trunk was mainly targeted to control *C. curvignathus* colonies infesting the palm's trunk. While soil injection was used to control *C. curvignathus* in the soil and also to prevent new termite infestation. This method has been commonly practiced to control termite in urban area, but yet to be fully applied in agricultural sector, especially in oil palm plantations.

For infested mature palm, the whole palm trunk was treated by spraying at the rate of 8 litres product solution using a knapsack sprayer (*Figure 1*). The palm was then further treated by soil injection of product solution at 10 holes surrounding the palm base at the rate of 1 litre per hole (*Figure 2*). The distance of the holes from the palm base and between holes is 30 cm. For healthy supply seedling, the palm was only treated by soil injection at 5 holes around palm base (*Figure 3*). Soil injection was conducted using a motorised soil injector equipped with a nozzle attached to bottom part of a 120 cm long stainless steel rod with handle.

The effectiveness of treatments to control termite on treated palms was assessed based on the presence of termite activities at three month after treatment (MAT), 6 MAT and 9 MAT.

Figure 1. Treatment by spraying of infested palm trunk using a manual knapsack sprayer.

Figure 2. Treatment by soil injection surrounding the infested mature palms using a motorised sprayer.

Figure 3. Treatment by soil injection surrounding the healthy supply seedling using a motorised sprayer.

RESULTS

The effectiveness of entomopathogenic fungi and fipronil to control termite infesting matured palm is showed in *Table 2*. At 3 MAT, all eight palms treated with fipronil exhibited no new termite infestation as no signs of termite activities were observed. This indicated that fipronil was 100% effective in controlling termite. However, at 9 MAT, one treated palm collapsed as the inner tissues at the middle part of the palm trunk were severely damaged by termite.

TABLE 2. THE EFFECTIVENESS OF ENTOMOPATHOGENIC FUNGI AND FIPRONIL TO CONTROL TERMITE INFESTATION ON MATURE PALM

Treatment	N	Number of palms without termites activities over time			Percentage of control (%)
		3 MAT	6 MAT	9 MAT	
B. bassiana	17	3	5	7	41.2
M. major	10	2	2	2	20.0
Fipronil	8	8	8	7	90.0

Note: MAT= Month After treatment.

For *B. bassiana*, the number of palm without termite activities increased over time. At one MAT, three treated palms were confirmed to be free from termite colonies. The numbers of palms free from termites were then further increased to 5 palms at 2 MAT and 7 palms at 3 MAT. With the total number of palm treated with *B. bassiana* is 17 palms, this represents a successful control rate of 41.18%. At 3 MAT, one treated palm collapsed as the palm trunk inner tissues have been attacked by termites.

For treatment with *M. major,* out of 10 treated palms, one palm was observed to be free from termite infestation at 1 MAT, then increased to two at 2 MAT. No further increase of treated palm free from termite were observed at 3 MAT. This gives a total of 20% successful control rate for *M. major* to control termite.

For supply seedlings, out of 28 treated seedlings, only one seedling treated with *B. bassiana* died due to termite attack at 9 MAT. This reflects the ability of all treatments to prevent newly planted seedlings from subterranean termite attack.

DISCUSSION

In this study, the satisfactory performance of fipronil in controlling the subterranean termite population was expected. Many studies have found that fipronil was effective in controlling the termite population. Huang (2006) reported that baits containing fipronil had successfully reduced foraging populations of termite species *Odontotermes formosanus*. It was also found that fipronil has faster killing time and higher termite mortality as compared to indoxacarb in controlling Formosan subterranean termite, *C. formosanus* Shiraki (Hu, 2005).

The low efficacy for both entomopathogenic fungi may be due to several factors as reported in several studies. Groden (1999) reported that *B. bassiana* is a naturally-occurring fungus in soil throughout the world, and some soil-borne insects may have developed natural tolerance towards the fungus. On the other hand, this tolerance is not exhibited on foliar pests. There is a possibility that some of the insects have developed physiological mechanisms to reduce fungi infection as Cloyd (1999) reported that desert locusts *Schistocerca gregaria* produced antifungal toxin which is able to inhibit spore germination. He also reported that some insect species resorts to rapid molting to escape infection. Termites behavioural responses could avoid or repel termites to migrate to pathogen free areas, causing less effective control of the fungi against termite (Baverstock *et al*., 2010).

The soil temperature may also affect the pathogenicity of both entomopathogenic fungi. It was reported that to be effective, *B. bassiana* needs to be stored within favourable temperature and moisture conditions, with relative humidity of 92% or more (Mahr, 1997). The fungus *M. anisopliae* is also sensitive to temperature extremes, where the increment of storage temperature may lead to reduced spore viability, while at low temperature, the fungus virulence may also decrease (Cloyd, 1999).

The use of visual observation to determine the presence of termite activities on palm was limited, as this method is rendered useless when the termite colonies have already done severe damage in the palm trunk inner tissue, although externally the palm looks healthy. This method also relies on the experience and the knowledge of the census team, which contributes to deviation and infested palm not being treated. A systematic damage indices and

a uniform census method is required to ensure the estimation of termite infestation in a plantation to be more precise. A practical and affordable device to detect the termite colonies infesting the inner part of the palm trunk can also be considered.

CONCLUSION

Findings of the study showed that fipronil was the most effective to control termite infestation. Entomopathogenic fungus, *B. bassiana and M. major* gave only 41.18% successful control and 20% control, respectively against termite infestation. All products were able to prevent infestation of termite on new supply seedlings in high termite infestation areas up to nine months after planting. Further study is recommended on control strategy of termite in plantation using biological products and effective delivery techniques.

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REFERENCES

BAVERSTOCK, J; ROY, H E and PELL, K J (2010). Entomopathogenic fungi and insect behaviour: from unsuspecting host to targeted vector. *BioControl, 55*: 89-102.

BASRI, M W; NORMAN, K; IDRIS, A S; ARIFFIN, D; SHAMALA, S; RAMLE, M and RAMLAH, S A A (2003). *Handbook of Pests and Diseases of Oil Palm*. MPOB, Bangi.

CLOYD, R A (1999). The entomopathogenic fungus *Metarhizium anisopliae*. Midwest Biological Control News Online www.entomology.wisc.edu/mbcn607.html

E L VARGO, V P (2012). Effect of fipronil on subterranean termite colonies (Isoptera: Rhinotermitidae). *Journal of Economic Entomology, 2(105)*: 523-532.

GRODEN, E (1999). *Using Beauveria Bassiana for Insect Management*. Retrieved May 19, 2015 from www.ipm.uconn.edu/documents/raw2

HU, X P (2005). Evaluation of efficacy and non-repellency of indoxacarb and fipronil treated soil at various concentrations and thicknesses against two subterranean termite species (Isoptera: Rhinotermitidae). *Journal of Economic Entomology, 2(98)*: 509-517.

LIM, K H and SILEK, B (2001). Termite infestation on oil palm planted on deep peat in Sarawak: Tradewinds experience. *Proc. of the 2001 International Palm Oil Conference.* 20-22 August 2001. Kuala Lumpur.

MAHR, S (1997). The Entomopathogen *Beauveria bassiana*. Midwest Biological Control News Online www.entomology.wisc.edu/mbcn/mbcn410.html

PIK- KHENG, H; CHONG-FAH, B J; KADIR, J and AMARTALINGAM (2009). Evaluation of *Metharhizium anisopliae* (Deuteromycotina: Hyphomycete) isolates and their effects on Subterranean termite *Coptotermes curvignathus* (Isoptera: Rhinotermitidae). *American Journal of Agricultural and Biological Science 4(4)*: 287-297.

HUANG, Q Y and C L L, XUE D (2006). Field Evaluation of a Fipronil Bait Against Subterranean Termite *Odontotermes formosanus* (Isoptera:Termitidae). *Journal of Economic Entomology, 2(99)*: 455-461.

RAMLE, M; ZULKEFLI, M and MOHD BASRI W (2011). Susceptibility of termites *Coptotermes curvignathus* (Isoptera: Rhinotermitidae) sampled from oil palm tree against the entomopathogenic fungi. *Proc. Fourth IOPRI-MPOB International Seminar: Existing and Emerging Pests and Disease of Oil Palm Advances In Research and Management*. 14 November 2011. KLCC.

ZULKEFLI, M; NORMAN, K; MOHD BASRI, W and MOHD MASRI MADSALI (2012*). Termites of Oil in Malaysia: Identification and Management*. MPOB. pp 34.