

Research Article

Efficacy of insecticides in toxic baits against the subterranean termite *Microcerotermes diversus* (Isoptera: Termitidae)

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Abstract: The subterranean termite *Microcerotermes diversus* Silvestri (Isoptera: Termitidae) is a significant urban pest that infests wooden objects and trees in urban green spaces, causing substantial damage each year. In this study, the efficacy of four insecticides—azadirachtin, spinosad, emamectin benzoate plus lufenuron, and imidacloprid—was evaluated for controlling this pest under field conditions. To evaluate the antifeedant effects of the insecticides on the insects, bait consumption was determined for each treatment. The results indicated that the lowest consumption was observed for the toxic bait containing azadirachtin, which suggests an antifeedant effect of the botanical insecticide. The highest consumption was observed for the baits containing imidacloprid. To investigate the feeding activity and population status of the foraging termites, oriental beach blocks were subjected to different treatments. The greatest reduction in the weight of the blocks (an index of termite feeding activity) and the highest termite density were observed in the azadirachtin treatment, and the lowest were observed in the imidacloprid and emamectin benzoate + lufenuron treatments. Therefore, the field experiments indicated that among the different treatments, imidacloprid and emamectin benzoate plus lufenuron are the best candidates for use as toxic baits against *M. diversus*. Although azadirachtin is not an appropriate insecticide for use in toxic baits against *M. diversus* due to its antifeedant property, it can be used as an effective antifeedant compound to protect wooden objects or as a barrier against termite entry into protected environments.

Keywords: Termiticide, repellency, urban pest, baiting system, IPM

Introduction

Subterranean termites pose a serious threat to many trees, shrubs, stored products, wooden materials, and tools in agricultural, forest, urban, and household environments. (Verma *et al.*, 2009). Among the various subterranean termite species, *Microcerotermes diversus* Silvestri

(Isoptera: Termitidae) is recognized as the most destructive termite in the tropical regions of Iran, including Khuzestan Province, southwestern Iran (Habibpour *et al.*, 2010; Cheraghi *et al.*, 2013). Foraging worker termites attack different parts of the palms. The feeding activity of the pest causes small-diameter galleries throughout the wooden parts of the host plants or household

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furniture, which are filled with fine, granular feces (Habibpour *et al.*, 2010). The primary strategy for controlling this pest is to use toxic baits with insecticides or to spray with insecticides. The efficacy of insecticide application by spraying and toxic residues is often inadequate due to the pest's cryptic behavior and the location of reproductive individuals. Therefore, toxic baits containing suitable termiticides are considered practical tools for sustainably controlling termites in various environments. The trophallaxis behavior of insects can cause reproductive individuals, such as queens and king termites, to obtain sufficient active ingredients from insecticides, thereby disrupting colonies through their killing (Verma *et al.*, 2009; Yarahmadi, 2022).

Choosing the appropriate insecticide for use in toxic baits plays a critical role in controlling the effectiveness of the baits. The best choice for eliminating subterranean termites is insecticides with slow-acting and non-repellent properties. Therefore, selecting insecticides can be considered the first needed decision for sustainable control of pest colonies. Several laboratory and field studies have been performed to apply slow-acting and non-repellent insecticides to toxic baits against subterranean termites, e.g., hydramethylnon, avermectin B1, to suppress *Coptotermes formosanus* Shiraki (Isoptera: Rhinotermitidae) (Su *et al.*, 1987); sulfluramid, to control *C. formosanus* and *Reticulitermes flavipes* Kollar (Isoptera: Rhinotermitidae) (Su and Scheffrahn, 1988a); diiodomethyl para-tolyl sulfone, to eliminate *C. formosanus* (Su and Scheffrahn, 1988b); fipronil, hexaflumuron, pyriproxyfen, and imidacloprid; chlorpyrifos-methyl, to control *Amitermes vilis* Hagen (Isoptera: Termitidae) (Rashid *et al.*, 2012); and some insect growth regulators (IFRs) (flufenoxuron, buprofezin, chlorfluazuron and novaluron) against *C. curvignathus* Holmgren (Ngampongsai *et al.*, 2020).

Some insecticides have been evaluated for their ability to be applied to toxic baits to control *M. diversus* colonies. For instance, the efficiency of boric acid and hexaflumuron (Ghyourfar and Mohammadpour, 2009), Flurox[®], a chitin

synthesis inhibitor insecticide (Habibpour, 2010), and lufenuron (Al-Jassany, 2015, 2016) for the chemical control of termites using toxic baits has been previously documented.

The objective of this study was to evaluate the efficacy of four insecticides, azadirachtin, spinosad, emamectin benzoate+lufenuron (Proclaim Fit[®]), and imidacloprid, for application in toxic baits against *M. diversus* under field conditions.

Materials and Methods

Experimental design

Four locations with similar plants and environmental conditions on the east bank of the Karun River, Ahvaz, southwestern Iran, were chosen for performing the experiments. The locations included Tasmanian Blue Gum (TBG) and *Eucalyptus globulus* Labill. (Myrtaceae), and at least four active colonies of *M. diversus*. The foraging population and territory of each colony were determined using the methods of Habibpour *et al.* (2010) with some changes. In the present study, fifty blocks (5 × 5 × 20 cm) made from Oriental Beach, *Fagus orientalis* Lipsky (Fagaceae) wood, were established in the soils of each experimental location. Each wood block was placed in a wide-mouth plastic container with a diameter of 25 cm. For accessing the entrance of foraging worker termites, twelve longitudinal grooves were made on the lateral sides of each container (Fig. 1).

Insecticides and toxic baits

The selected insecticides for application in the toxic baits and the experimental treatments are presented in Table 1. The standard bait was composed of bagasse, molasses, yeast, sawdust from Oriental beach wood, and powder from date palm fibers, according to the methods of Ekhtelat *et al.* (2018), with some modifications. After weighing by a precise digital scale (AND model HR200), 20 g of each toxic bait was weighed and placed in a container, and the four containers were randomly placed in the soil of each termite

colony territory. The bait was not mixed with any insecticide in the control.

Field study of the toxic bait effects on the termite population

One Oriental beech block was located in each termite colony, according to the previously described method, after weighing was performed. After 24 days, the container was slowly removed from the soil and replaced with a new container, which included oriental beach blocks. The number of termites on each oriental beach block was recorded. Furthermore, the weight of each wood block was recorded, and the

consumed wood weight (W_c) was calculated using the following equation.

$$W_c = W_0 - W_t$$

Where W_0 and W_t are the block weights at the beginning of the trial and after 24 days, respectively.

Antifeedant effects

After 24 days, the exact weight of the remaining bait was recorded, and the amount of the consumed toxic bait was calculated to determine the possible antifeedant effect of each experimental insecticide.



Figure 1 The wide-mouth plastic container containing the oriental beach wood block.

Table 1 List of insecticides applied in the toxic baits.

Insecticide	Traditional name	Company	Mode of action	mL per L
Azadirachtin	Neem Azal® (EC 1%)	Trifoli Co., Germany	Botanical insecticide, IGR	1
Spinosad	Tracer® (SC 44%)	Dow Agrosience, USA	Biorational insecticide alters the function of nicotinic and GABA-gated ion channels.	0.2
Emamectin Benzoate + lufenuron	Proclim Fit® 50% G	Syngenta Co., Austria	Causing a continuous flow of chlorine ions in the GABA and H-Glutamate receptor sites and interfering with chitin synthesis	0.3
Imidacloprid	Confidor® (SC 35%)	Partov Nar-Iran	Interfering with the transmission of impulses in the nerve system of insects	0.5

Data analysis

The experiments were performed in accordance with a completely randomized block design with four replications. One-way analysis of variance (one-way ANOVA) was performed using SAS software, version 9.2 (SAS Institute, Cary, NC), to compare the means of the termite population and consumed blocks or toxic baits. Mean differences between different treatments were determined using Duncan's post hoc test at the 0.05 significance level. Before conducting the one-way ANOVA, the validity of the normality assumption and homogeneity of variance were confirmed using the Shapiro–Wilk and Levene's tests, respectively.

Results

Consumption of toxic baits

The weights of the remaining bait from the 20 g of toxic bait provided for each treatment are shown in Fig. 2. ANOVA indicated significant differences between the experimental treatments ($df = 4, 19$; $F = 37.71$; $P \text{ value} < 0.001$). The lowest bait consumption was observed in the azadirachtin treatment (1%). The highest bait consumption

occurred in the imidacloprid treatment group (63.15%), which was not significantly different from the bait consumption in the control group (67%). The consumption of baits in the emamectin benzoate + lufenuron and spinosad treatments was 54.25% and 36.65%, respectively.

Feeding of termites from the oriental beach blocks

The reduction in weight of the oriental beach block due to termite feeding in the different experimental treatments is presented in Fig. 3. Significant differences were observed in block consumption among the different treatments ($df = 4, 19$; $F = 4.02$; $P \text{ value} = 0.027$). The most minor feeding activity was observed in the imidacloprid treatment (18.13 g). There was no significant difference in the amount of termite feeding on the block between the azadirachtin (50.13 g) and spinosad (42 g) treatments and the control (55.43 g).

The abundances of foraging termites

The means of the recorded foraging individuals of *M. diversus* in the Oriental beach blocks across different experimental treatments are shown in Fig. 4.

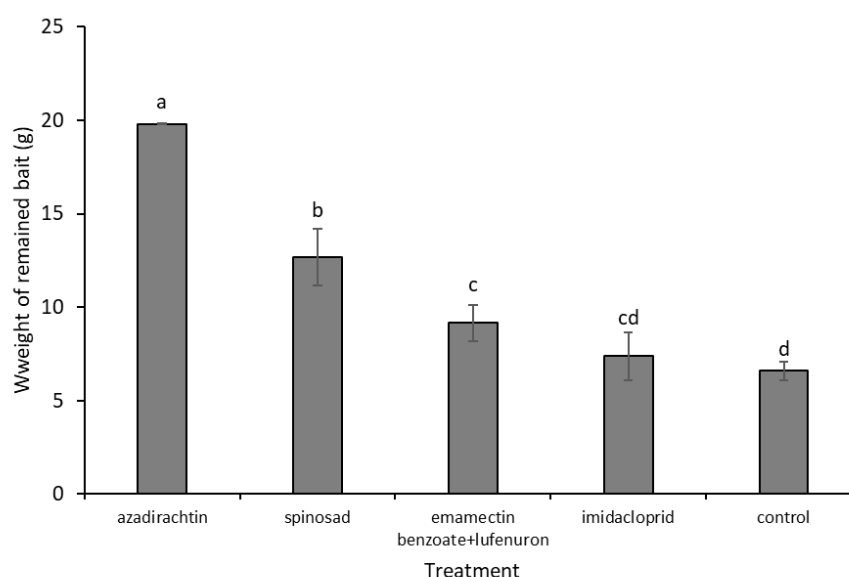


Figure 2 The mean weights of the remaining bait from 20 g of provided baits in different experimental treatments (same letter indicates non-significant difference at 0.05- Duncan post hoc test).

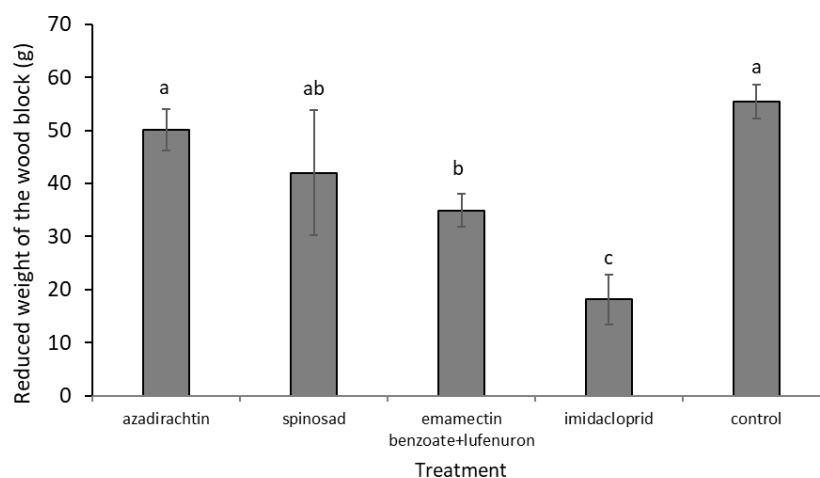


Figure 3 The reduction in the oriental beach block due to the *Microcerotermes diversus* feeding in different experimental treatments (same letter indicates non-significant difference at 0.05- Duncan post hoc test).

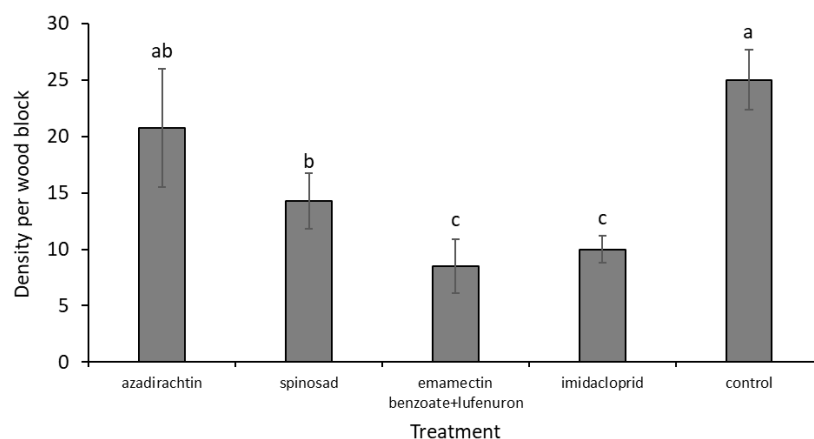


Figure 4 Means of *Microcerotermes diversus* density per the oriental beach block in different experimental treatments (same letter indicates non-significant difference at 0.05-Duncan post hoc test).

The highest and lowest termite densities were recorded in the control (25 termites) and emamectin benzoate + lufenuron treatment (8.5 termites), respectively. However, there was no significant difference in termite density between the imidacloprid (10 termites) and emamectin benzoate + lufenuron treatments.

Discussion

These findings indicated that the consumption of the toxic baits treated with azadirachtin by *M. diversus* foraging individuals was significantly

lower than that of the other treatments, suggesting the high antifeedant of the compound (Schmutterer, 1990). Azadirachtin is a tetranortriterpenoid. In addition to azadirachtin, several other tetranortriterpenoids have been extracted from the fruit and leaves of this tree, the most important of which are salanin and meliantriol, which have insecticidal properties similar to those of azadirachtin but are less active (Jones *et al.*, 1989).

The antifeedant nature of limonoid compounds present in plants belonging to the Meliaceae and Rutaceae families has been

previously reported for some termites, including *Reticulitermes speratus* Kolbe (Isoptera: Rhinotermitidae). Among the various limonoids present in these plants, two compounds, obacunone and nomilin, exhibit significant antifeedant effects, and their use is recommended to protect against termite attacks (Serit *et al.*, 1992). Himmi *et al.* (2013) also presented a semiformulated extract to protect wooden objects from *Coptotermes gestroi* Wasmann (Isoptera: Rhinotermitidae) termite feeding and damage, and their research indicated the high efficiency of this formulation in preventing the feeding activity of these termites. Another study showed that a unique formulation of neem tree extract called Margosan-O, which contains 2% azadirachtin and 14% neem oil, can effectively prevent *C. gestroi* termite damage. Although the current study showed that the azadirachtin used in this study is not suitable for use as a toxic bait against *M. diversus*. The antifeedant property of the termite *M. diversus* can be applied to protect wooden objects or as a barrier to prevent termite entry into the environment and to prevent feeding.

The highest consumption of the toxic baits was observed in the imidacloprid treatment. It appears that this combination does not impact the feeding activity of termites; in this sense, it can be considered an appropriate insecticide for use as a toxic bait to control *M. diversus* (Salehi Babarsad *et al.*, 2012).

Based on the decrease in consumption of the oriental beach blocks (as an indicator of termite activity) and the recorded population density of foraging termites in response to the different treatments, the results obtained indicated the high potential of imidacloprid for use as a toxic bait against *M. diversus* termites. One of the most desirable properties of this insecticide for use as a toxic bait against termites is its cumulative and delayed effects. These characteristics are necessary for the insecticide to spread through trophallaxis at all levels of the colony, reach sexual individuals and eliminate the colony (Rondeau *et al.*, 2014). Another advantage of this insecticide is that it does not repel insects. For the subterranean termite *R.*

virginicus Banks, it was demonstrated that sublethal residues of imidacloprid did not have any repellent effect on foraging individuals (Thorne and Breisch, 2001). Reid *et al.* (2002) reported imidacloprid as a non-repellent insecticide that controls 78% of termite-infested structures of the genus *Reticulitermes* spp. However, a study conducted on a toxic bait formulation containing imidacloprid found that this insecticide had little effect on the underground termite *C. formosanus* (Osbrink *et al.*, 2005).

Emamectin benzoate and lufenuron also had a relatively effective effect on reducing the population size and activity of *M. diversus* foraging individuals. Emamectin benzoate + lufenuron is a new insecticide that consists of 10% emamectin benzoate, which has a neurotoxic nature, and 40% lufenuron, which is a growth regulator and disrupts the formation of the endocuticle (Copping, 2000). The efficacy of Emamectin benzoate + lufenuron is the same as that of imidacloprid. Similar to our findings, experimental bioassays of certain insecticides on *Coptotermes heimi* Wasmann demonstrated that separate applications of emamectin and lufenuron led to significant mortality of the termites (Qasim *et al.*, 2024).

Spinosad, a bacterial metabolite used to manage agricultural pests, has demonstrated notable effectiveness against termites, particularly in the families Kalotermitidae and Termitidae. In controlled experiments, 25 and 50 ppm concentrations of spinosad led to over 85% mortality in *C. formosanus* after one day, reaching 100% mortality by the seventh day. Behavioral assessments revealed no repellent effects, as treated termites displayed increased grooming and movement, suggesting a potential for social behavior-driven transfer of toxins. Moreover, horizontal transfer experiments revealed more effective spinosad transmission in sand versus soil, with significant mortality observed, indicating promising efficacy for future field studies on *C. formosanus* (Bhatta, 2015; Bhatta *et al.*, 2016). Another research indicated that spinosad demonstrated superior control in preventing infestations for extended

periods in olive trees, particularly at higher concentrations. Another study indicated that spinosad exhibits superior control in preventing infestations on olive trees to *M. diversus* for extended periods, especially at higher concentrations (Abbas *et al.*, 2021).

In conclusion, field trials demonstrated that among the various treatments, imidacloprid and emamectin benzoate + lufenuron, which had the greatest effect on reducing the population and feeding activity of *M. diversus* termites, are the most effective choices for producing toxic baits against this pest. These compounds, although having the desired effects on population and feeding activity reductions, are not antifeedants and can therefore be mixed well with baits. Although azadirachtin is not a suitable insecticide for use in toxic baits due to its antifeedant property, it can be used as an effective antifeedant compound to protect wooden objects or as a barrier to avoid insect entry into a protected environment.

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کارایی حشره‌کش‌ها در طعمه مسموم علیه موریه‌ها زیرزمینی *Microcerotermes diversus* (Isoptera: Termitidae)

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چکیده: موریه‌ی زیرزمینی *Microcerotermes divesus* Silvestri (Isoptera: Termitidae) از آفات مهم شهری است که به وسایل چوبی و درختان موجود در فضای سبز شهری حمله و موجب خسارت شدیدی می‌گردد. طعمه‌ی سمی یکی از مهم‌ترین راهکارهای مقابله با کلنی‌های موریه‌ها می‌باشد. در این تحقیق، کارایی چهار حشره‌کش آزادیراختین، اسپینوزاد، امامکتین بنزوات + لوفنرون و ایمیداکلوپراید در کنترل این آفت در شرایط صحرایی مورد مطالعه قرار گرفت. در تیمار شاهد به‌جای حشره‌کش از آب استفاده شد. برای بررسی اثرات دورکنندگی سموم میزان مصرف طعمه‌های آغشته به هر تیمار تعیین شد. نتایج نشان داد که کمترین مصرف در طعمه‌های آغشته با آزادیراختین مشاهده شد که بیانگر ماهیت ضدتغذیه‌ای این حشره‌کش می‌تواند باشد. بیش‌ترین مصرف در طعمه‌های مسموم شده با ایمیداکلوپراید مشاهده شد. برای بررسی میزان فعالیت و جمعیت موریه‌ها بلوک‌های چوبی در تیمارهای مختلف به‌کار رفت. بیش‌ترین کاهش وزن بلوک‌های چوبی (شاخصی برای فعالیت موریه‌ها) و بیش‌ترین تراکم موریه‌ها در تیمار آزادیراختین و کمترین آن‌ها در تیمار ایمیداکلوپراید و امامکتین بنزوات + لوفنرون دیده شد. به‌طور کلی، آزمایشات مزرعه‌ای صورت گرفته نشان داد که در بین تیمارهای مختلف ایمیداکلوپراید و امامکتین بنزوات + لوفنرون به دلیل داشتن بیش‌ترین تأثیر روی کاهش جمعیت و فعالیت موریه‌های *M. diversus* بهترین گزینه‌ها برای به‌کارگیری در طعمه‌های مسموم علیه این آفت هستند. اگرچه آزادیراختین در این مطالعه نشان داده شد که حشره‌کشی مناسب برای به‌کارگیری در طعمه‌های مسموم به‌دلیل ماهیت ضدتغذیه‌ای نیستند، ولی می‌توانند به عنوان ترکیبی مؤثر ضدتغذیه‌ای برای جلوگیری از ورود این موریه‌ها به محیط‌های محافظت شده استفاده شود.

واژگان کلیدی: موریه‌کش، دورکنندگی، آفات شهری، سیستم طعمه‌گذاری، مدیریت تلفیقی آفات (IPM)