

Table 1: Showed days for activity of termites on wooden stakes treated with Imidacloprid and Fipronil at three different stations.

Stations	Treatment	Stakes with termite Infestation	Termites Infestation	Installation to feeding cessation(Days)	Total days
*Station 1	Fipronil	<i>P. euramericana</i> , <i>H. adenophyllum</i> , <i>C. fistula</i> , <i>A. indica</i> , <i>E. camaldulensis</i> and <i>E. subrosa</i>	<i>O. obesus</i> and <i>M. obesi</i>	175	62
*Station 2	Imidacloprid	<i>P. euramericana</i> , <i>B. variegata</i> , <i>P. roxburghii</i> , <i>H. adenophyllum</i> , <i>E. subrosa</i> , <i>M. alba</i> , <i>A. indica</i> , <i>J. mimosifolia</i> <i>C. fistula</i> , <i>A. excelsa</i> , <i>M. indica</i> , <i>D. sissoo</i> and <i>T. grandis</i>	<i>O. obesus</i> and <i>M. obesi</i>	180 days	58
*Station3		<i>P. euramericana</i> , <i>A. indica</i> , <i>P. roxburghii</i> , <i>B. monosperma</i> <i>M. alba</i> , <i>B. variegata</i> , <i>A. lebbeck</i> , <i>D. sissoo</i> , <i>H. adenophyllum</i> , <i>E. subrosa</i> , <i>S. cumini</i> , <i>M. indica</i> , <i>J. mimosifolia</i> , <i>E. camaldulensis</i> and <i>T. grandis</i>	<i>C. heimi</i>	190days	55

*Station 1: FC College Botanical garden; Station 2: FC College Botanical Garden; Station 3: Lahore canal bank

significantly ($P < 0.05$) impacted termite activity and galleries formation followed by gradual decline after treatment at week 16 and bimonthly observations up to week 32 indicated the suppression of the colony (Sep 2012-Jan.2013).The complete elimination of *C.heimi* termite colony after treatment exhibited 190 days and follow up observations after elimination were carried out up to 55 days to ensure the complete elimination of the termite colonies (Table 1).

3.6 Treatment of stakes with Fungus *M.anisopliae*-Imidacloprid combination

3.6.1 Station 1

When stakes of station 1 were treated to a combination of 0.03ppm Imidacloprid plus the entomopathogenic fungus *M.anisopliae* at 1×10^7 conidia/ml, a reduction in termite activity was observed, when compared to the control. The combination treatment of Imidacloprid and *M.anisopliae* played a significant role in decline the survivorship of *M.obesi* and *O.obesus*. In both *O.obesus* and *M.obesi*, Imidacloprid decreased Termites activity and survivorship to introduced *M.anisopliae* after 12 weeks (Sep-Nov 2012) of treatment to wooden stakes. Termites activity on different wooden stakes concurrently exposed to *M.anisopliae* and Imidacloprid was comparatively lower than the activity of termites exposed to either treatment alone. Furthermore, at last week of Sep 2012, the difference was more pronounced and hence decreasing activity in case of both termite species exposed to the highest concentration of *M.anisopliae* (1×10^7 conidia/ml-0.03ppm of Imidacloprid) in combination. After 24 week a follow up study (from Dec 2012-Mid May 2013) revealed complete absence of termite activity on all treated wooden stakes. Therefore, Lower termite activity was observed here and importantly a significant interaction between fungus and Imidacloprid with 100% termite elimination and termite activity ($F =, p < .$). The number of days to first termite activity, on any one of the bait station installed in the field, for all bait systems, ranged from a low of 49 days to high of 84 days. There was no significant difference between groups when comparing (t-test) days to first termite activity data from three sites for each baiting system ($p = 0.0, 0, 0$) (Table 2).

3.7 Treatment of stakes with Fungus *M.anisopliae*-fipronil combination

3.7.1 Station 2

When stakes of station 2 were treated to a combination of 0.03ppm fipronil plus the entomopathogenic fungus *M.anisopliae* at 1×10^7 conidia/ml, a reduction in termite activity was observed, when compared to the control. The combination treatment of fipronil and *M.anisopliae* played a significant role in declining the survivorship of *O.obesus* and *M. obesi*. In both *O.obesus* and *M. obesi*, fipronil decreased termites activity and survivorship to introduced *M.anisopliae* after 11 weeks (Sep- 3rd week of Nov 2012) of treatment to wooden stakes. Termites activity on different wooden stakes concurrently exposed to *M.anisopliae* and fipronil was comparatively lower than the activity of termites exposed to either treatment alone. Furthermore, at 3rd week of Sep 2012, the difference was more pronounced and hence decreasing activity in case of both termite species exposed to the highest concentration of *M.anisopliae* (1×10^7 conidia/ml-0.03ppm of fipronil) in combination. After 24 week a follow up study (from Dec 2012-Mid May 2013) revealed complete absence of termite activity on all treated wooden stakes. Therefore, Lower termite activity was observed here again and importantly a significant difference between fungus and fipronil with 100% termite elimination and termite activity ($F =, p < .$). The number of days to first termite activity, on any one of the bait station installed in the field, for all bait systems, ranged from a low of 46 days to high of 80 days. There was no significant difference between groups when comparing (t-test) days to first termite activity data from three sites for each baiting system ($p = 0.0, 0, 0$). These findings clearly indicate that fipronil plus fungus interaction pronounced slightly faster results than the Imidacloprid plus fungus combination. So it is evident that Imidacloprid combination with fungus is comparatively much slower in action than that of fipronil fungus interaction (Table2).

3.8 Treatment of stakes with Fungus *M.anisopliae*-Imidacloprid combination

3.8.1 Station 3

When infested stakes (*Populus euramericana* > *Azadirachta indica* > *Pinus roxburghii* > *Butea monosperma* > *Morus alba* > *Bauhinia variegata* > *Albizia lebbeck* > *Dalbergia sisoo* > *Heterophragma adenophyllum* > *Erythrina subrosa* > *Tectona grandis* > *Mangifera indica* > *Jacaranda mimosifolia*

> *Eucalyptus camaldulensis* and *Syzyium cumini* of station 3 were treated to a combination of 0.03ppm Imidacloprid plus the entomopathogenic fungus *M.anisopliae* at 1×10^7 conidia/ml, a reduction in termite activity was observed, when compared to the control. The combination treatment of Imidacloprid and *M.anisopliae* played a significant role in decline the survivorship of *C.heimi*. Imidacloprid decreased termites activity and survivorship to introduced *M.anisopliae* after 13 weeks (Sep-Nov 2012) of treatment to wooden stakes. Termites activity on different wooden stakes concurrently exposed to *M.anisopliae* and Imidacloprid was comparatively lower than the activity of termites exposed to either treatment alone. Furthermore, at 1st week of Oct 2012, the difference was much more evident and hence decreasing activity. After 24 week a follow up study (from Mar 2013-Mid May 2013) revealed complete absence of termite activity on all treated wooden stakes. These results indicated that a significant interaction between fungus and Imidacloprid exist on termite mortality with synergistic effects ($F=p<$).

The number of days to first termite activity, on any one of the bait station installed in the field, for all bait systems, ranged from a low of 52 days to high of 90 days. There was no significant difference between groups when comparing (t-test) days to first termite activity data from three sites for each baiting system ($p= 0.0, 0, 0$) (Table 2).

Table 2: Showed days for activity of termites on wooden stakes treated with Imidacloprid and Fipronil in combination with *M.anisopliae* at three different stations.

Sites/ Stations	Treatments	Termite Activity days	Infestation period	Termites
*Station 1	<i>M. anisopliae</i> - Imidacloprid	49 to 84 days	Dec 2012-Mid May 2013	<i>M. obesi</i> and <i>O. obesus</i>
*Station 2	<i>M. anisopliae</i> - Fipronil	46 to 80 days	Dec 2012-Mid May 2013	<i>M. obesi</i> and <i>O. obesus</i>
*Station 3	<i>M. anisopliae</i> - Imidacloprid	52 to 90 days	Mar 2013-Mid May 2013	<i>C. heimi</i>

*Station 1: FC College Botanical garden; Station 2: FC College Botanical Garden; Station 3: Lahore canal bank

4. Discussion

The percentage of wood materials or wood stakes with signs of termite infestation was much greater than that with foraging termites. This might be related to the termite foraging behavior, change in suitability of wood materials for termite foraging, and other factors such as weather and predators. The wooden stakes were principally infested by the following subterranean termite species: *Odontotermes obesus* (Rambur), *Microtermes obesi* and *Coptotermes heimi* (Wasmann) (Isoptera: Rhinotermitidae) at different stations. The first species was collected 89 days after the experiment was started. The highest % termite infestation was observed by *O.obesus*, 57.8% while lowest infestation was observed by *M. obesi* i.e. 16.5% respectively while *C.heimi* showed highest infestation on *P.euramericana* which was 24.7% respectively. While during infestation period at station 1 and 2 of the 45 wooden stakes 8 (17.7%) and 13 (28.8%) were attacked after 3 months. However, at station 3, of the 45

wooden stakes 15 (33.3%) were attacked by *C. heimi* after 3 months. [16] investigated the resistance of different wooden stakes on the basis of visual damage assessment in the soil for a period of 4 year; they found Termites belonged to nine species under four genera and the single family Termitidae. While *Odontotermes obesus* was the dominant species in most of the locations. [17] noted that termite infestation was higher during June to September wood degradation by termites was most during the months of April to June, moderate from July to October and less from November to March. The more degradation from April to June might be attributed due to pre monsoon showers, which provides necessary amount of moisture required for the termite survival. Degradation was less from November- March which, might be possibly due to higher atmospheric temperature and lower amount of relative humidity both act as a constraint for the foraging behaviour [18]. It is possible that the approximately one-month period between installation of the stakes and inspection was not sufficient for the termites to locate the stakes. It is more likely, however, that the termites had sufficient wood and only attacked the stakes if a subterranean foraging tunnel directly contacted them. The condition of the wood stakes may also affect feeding activity and it is possible that the stakes had to age and decay to become as attractive as bark. Decomposing wood has greater concentrations of sucrose and more associated yeasts than sound wood [19]. In addition, fungi that are associated with wood decay are often consumed by termites [20], and these fungi are rich in urea [21].

Stakes when treated with sublethal dose of Imidacloprid and Fipronil termite activity was gradually declined. Station 1 and 2 was highly infested by *O.obesus* and *M. obesi* respectively. This suggests that agonistic response may have occurred between colonies of these two species, which probably resulted in a takeover of the *O.obesus* foraging site by *M.obesi*. Interspecificity encounters among termite colonies resulting in agonistic response were observed by [22], [23]. Field observation by [24] revealed two incidents in which foraging sites of *Reticulitermes flavipes* (Kollar) were taken over by *Coptotermes formosanus* Shiraki. Whereas, *C.heimi* was dominant station 3 because vegetation cover was surrounded by *P.euramericana*. Resistance is a critical determinant of life span of tree species. Many heartwood species are known for their resistance against degradation [25]. Tree species such as *T. grandis* [26]-[30], *S. robusta* [31], *Shorea* sp. [32], *S. marcoptera*, *Dryobalanops* sp., and *Xylia dolabriformis* [33]-[35], and *Pterocarpus soyauxii* [36] are well-known for their durability from ancient times. In our study, *T.grandis* was found to be moderately resistant against degradation under Pakistani conditions.

In our study the combined formulation or application of *M.anisopliae* with Imidacloprid and Fipronil played a significant role in decline the survivorship of *M. obesi* *O.obesus* and *C.heimi* at all stations and more effective as compared to alone treatment. Similarly, the findings of [37] also explained the integration of insecticides acetamiprid, Imidacloprid and Thiamethoxam with fungal conidia on the basis of compatibility in order to enhance the biocontrol potential of entomopathogenic fungi. The nature of fungal repellency by the species within the order Isoptera has been

widely assessed in the laboratory [38]-[39]. This type of avoidance behaviour is very important for the management of insect pests like termites residing in difficult-to-reach locations, such as in underground nests. Similarly, [40] found that organic mulches supplemented with *M. anisopliae* significantly repelled foraging *C. formosanus* and reduced mulch consumption by up to 71%.

5. Conclusions

The present study clearly indicates that the addition of termiticides to *M. anisopliae* has pronounced positive effect in the control of termites in natural environment. The present investigation showed that biocontrol of termite pest with entomopathogenic fungus was impressive under experimental conditions and had high prospects in controlling termite. However, the efficacy of bio-agents is dependent on physio-chemical and biological properties of soil and climatic factors as well as the degree of susceptibility of host species [41], [42].

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