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Efficacy of Seed Extracts (*Mentha arvensis, Ocimum bacilicum, Psyllium ovata, Cichorium intybus*) Against *Coptotermes heimi* (Wasmann)

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ABSTRACT

In the present study, various concentrations of seed extracts of medicinal plants (*Mentha arvensis* (mint)*, Ocimum bacilicum* (niazbo), *Psyllium ovata* (harmal), *Cichorium intybus* (kasni)*)* were used against *Coptotermes heimi* soldiers to see their impact on survival. The seed extract of *M. arvensis* caused highest mortality i.e., 100%, 99% and 98% at 10%, 5% and 3% concentration. *Cichorium intybus* seed extract showed 92%, 88% and 85% mortality at 10%, 5% and 3% concentration. *Ocimum basilicum* extract demonstrated 80%, 78% and 76% mortality at 10%, 5% and 3% concentration. *Psyllium ovata* extract was least effective and caused 72%, 70% and 68% mortality at 10%, 5% and 3% concentration respectively. LT_{50} values for *M. arvensis* were 0.91, 0.94 and 1.13 hours for 10%, 5% and 3 % concentrations respectively. In case of *O. basilicum*, LT_{50} values at 10%, 5% and 3% concentrations were 4.3, 4.6 and 4.8 hours respectively. LT_{50} values for *P. ovata* were 5.7, 6.2 and 7.1 hours for 10%, 5% and 3% concentrations. In case of *C. intybus*, LT_{50} values were 2.4, 2.8 and 3.4 hours at 10%, 5% and 3% concentrations respectively.

Key Words: *Coptotermes heimi,* soldiers, plant extracts

INTRODUCTION

Termites cause serious damage to household materials, agricultural plants i.e., sugarcane, rice, millet and barley (Sattar *et al*., 2014; Aihetasham *et al.,* 2015). Almost 2800 species have been described throughout the world but 185 species are important which cause damage (Lewis, 1997; Verma *et al*., 2009; Aihetasham *et al.,* 2015). Fifty species of termites have been recorded from different ecological zones of Pakistan (Akhtar, 1974). Akhtar, 1983 reported about 11 species of termites which caused severe damage to wooden infrastructure, one of those, *Coptotermes heimi* (Wasmann) was said to be most destructive of all of them and reported as severe pest of agricultural fields (Manzoor & Mir, 2010; Manzoor *et al.,* 2011).

The insecticides were frequently used to suppress the activity of termite colony which are totally synthetic organochloride (Khan & Singh, 1985; Anonymous, 2000, Ahmed *et al.,* 2006). The extreme enduring effect as well as the development of insecticides resistance in target pests is considered their drawbacks. The use of excessive amount of insecticides cause serious problems i.e., environmental (if there are any leakages after heavy rain and have strong pungent odors) (Martius, 1998; Logan *et al.,* 1990) as well as a source of many disorders (chlordane cause cancer, chlorpyriphos, dimethodate and endosulfan cause genotoxicity) (Jamil *et al.,* 2005).

In recent years, many researchers found out that the use of plant extracts against the termites had high bio-efficacy value. For that purpose, many plants had been used as they had anti-termite and repellent response to termite colony such as clove bud (*Syzgium aromaticum*), vetiver oil (*Vetiveria zizaniodes*), lemon grass (*Cymbopogon citratus*), Eucalyptus (*Eucalyptus citrodora, Eucalyptus globules*), Cassia leaf (*Cinnamomum cassia*), cedar wood (*Cedrus atlantica*), (Zhu *et al*., 2001a,b), *Calotropis procera* (Singh *et al.,* 2002), isoborneol (Blaske *et al*., 2003),*Coleus amboinicus* (Singh *et al.,* 2004), *Rhazya stricta* Decne, *Lantana camara* L, *Ruta chalepensis* L, *Heliotropium bacciferum* Forssk (Alshehry *et al.,*) *Justicia adathoda* (Bala & Gabba, 2015), *Pinus roxburghii* Sargent*, Morus alba* L. and *Eucalyptus camaldulensis* Dehnh (Rasib and Aihetasham, 2016).

Present studies were undertaken to assess the toxicant potential of the seed extracts from *Mentha arvensis* (mint)*, Ocimum bacilicum* (niazbo), *Psyllium ovata* (harmal), *Cichorium intybus* (Kasni) against *C. heimi*. The objective was the Ethanolic extraction of selected plant seeds using soxhlet extractor and to study bioactivity of these seed extracts against *C. heimi* soldiers under laboratory conditions being more common in

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the colony, have more sclerotized body compared to workers and are more important source of defense of the colony.

MATERIAL AND METHODS

Collection of Termites, Seeds and Soil

Coptotermes heimi (Wasmann) soldiers were collected by installing traps of plastic bottles with tissue rolls in soil near old standing trees of *Populus euramericana*, FC college University canal road, Lahore. The soldiers were transported in glass chamber to the Department of Zoology, Entomology laboratory, University of the Punjab for further experimentation. The termites were placed in petri plates containing filter paper soaked with water and 5 gram of oven dried soil. Termites were maintained at ±26°C.

Mentha arvensis (mint)*, Ocimum bacilicum* (niazbo)*, Psyllium ovata* (harmal, isapgul), *Cichorium intybus* (Kasni) seeds were purchased from local market and confirmed by a taxonomist, Department of Botany, University of The Punjab, Lahore, Pakistan.

The soil samples were collected from the lawn of Department of Zoology, University of the Punjab, Lahore where no application of any insecticide was undertaken and was considered free of insecticidal residues. Soil was sieved through 30-mesh screen and then sterilized in a vacuum oven. The soil was used for the bioassays.

Soxhlet Extraction Process

The seeds of the medicinal plants were ground separately into fine powder. Twenty grams of each seed powder was taken separately for extraction in Soxhlet extractor using 200 ml of ethanol as solvent. Dried seeds of the plants were ground and the extract was prepared using ethanol as a solvent. Rotary evaporator was used to obtain dried residues and stored in refrigerator. Stock solution was prepared for each plant extract by taking 1 gram dried extract in 10 ml of absolute ethanol to get 10% concentration. Further dilutions i.e., 5% and 3% were prepared from it.

Anti-Termite Assay

A procedure adapted by Smith (1993) was used in experiments. Petri dishes were sterilized in the oven at 200°C. Whatman filter papers (No.1) were cut and placed at the bottom of each sterilized Petri dish. Filter papers were soaked with 1 ml of the 10%, 5% and 3% extract concentrations. The filter papers were dried at ambient temperature. Each treatment was replicated three times. Hundred soldier termites were added in each Petri dish. Hourly observations up to 12 hours were taken and the dead individuals in each Petri dish were removed. Then data for the mortality of the termite were recorded after an interval of 12 hours up to 96 hours.

Statistical Analysis

The Statistical Software Minitab 16.0 was used to evaluate differences in percentage mortality using one way ANOVA for the anti-termite tests. Probit Analysis was applied to calculate LT_{50} of termites against ethanolic seeds extracts at different concentrations i.e.10%, 5% and 3%. Results were deduced by giving input of data up to 10 hours limit and accordingly graphs were plotted.

RESULTS AND DISCUSSION

The activity of ethanolic seeds extracts of Mint (Podina) (*M. arvensis*), Niazbo (*O. basilicum*), Ispaghul (*P. ovata*) and Kasni (*C. intybus*) were applied on Whatman's filter paper at different concentrations and expressed as LT_{50} (hours) (Table I). These extracts have anti-termite activities showing significant difference from control (Table II). The activity showed concentration dependent and LT_{50} decreased with increase in concentrations as shown in Table I. Some plant and their extracts being repellent to termites, had been used as a possible alternative to control the termites activities instead of insecticides (Sbeghen *et al*., 2002).

Table I: LT_{50}values (Hours) of different seed extracts at 10%, 5% and 3% concentrations

Serial No.	Plants for Seed Extracts	10%	5%	3%
1.	*Mentha arvensis* (Podina)	0.91	0.94	1.13
2.	*Cichorium intybus* (Kasni)	2.4	2.8	3.4
3.	*Ocimum basilicum* (Niazbo)	4.3	4.6	4.8
4.	*Psyllium ovata* (Ispaghul)	5.7	6.2	7.1
5.	Control	24.3		

M. arvensis seed extract had highest anti-termite activity in the present work. The average percent mortality by *M. arvensis* at 10% concentration was 100% followed by 99% at its 5% concentration and 98% at its 3% concentration ($F=9.250$, $P<0.05$). Qureshi *et al*., 2012 the mint (*M. arvensis*) extract gave 100 % mortality of the

soldiers on exposure of 100, 50 and 25 mg of dried residue for one, two and three hours respectively While our results showed 100 % mortality at 10%, 5% and 3% seed extracts of *M. arvensis* in 3 to 4 hours.

Table II: Analysis of Variance for the effect of 4 different plant seed extracts on soldiers of *C. heimi*

Seed extracts	Source of variation	Degrees of freedom	Sum of squares	Mean squares	F-value
Mentha arvensis	Between Groups	2	8.222	4.111	9.250
	Within Groups	6	2.667	.444	
Cichorium intybus	Between Groups	2	66.889	33.444	43.000*
	Within Groups	6	4.667	.778	
Ocimum basilicum	Between Groups	2	24.000	12.000	36.000*
	Within Groups	6	2.000	.333	
Psyllium ovata	Between Groups	2	113.556	56.778	102.200*
	Within Groups	6	3.333	.556	

* = Significant at P<0.05

LT_{50} for *M arvensis* against *C. heimi* was 0.91, 0.94 and 1.13 hours for 10%, 5% and 3% concentrations respectively (Fig., 3). However, Aihetasham *et al.* (2015) reported LT_{50} values of 38.38, 67.54 and 83.19 hours for 10%, 5% and 3% concentrations for *M. arvensis* against *Heterotermes indicola* which showed *M. arvensis* quite effective against termites. The extract of *M. arvensis* may be used as the natural antitermite medicine thus protecting environment from synthetic insecticides (Qureshi *et al*., 2012).

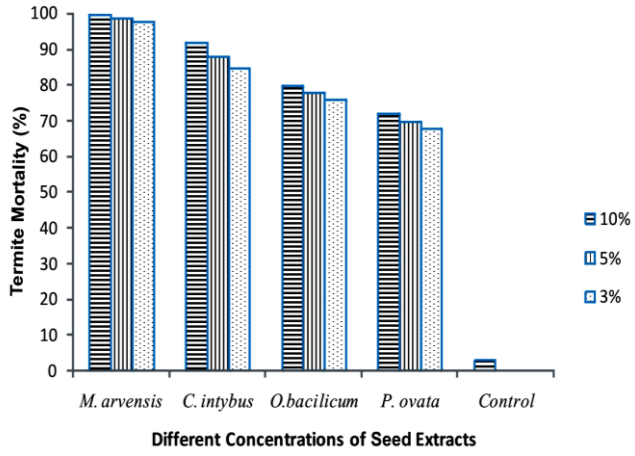

Fig., 1: Effect of different concentrations of seed extracts on *C. heimi* soldiers mortality

C. intybus seed extract showed 92 %, 88 % and 85 % mortality at 10 %, 5 % and 3 % concentrations respectively (F=43.0, P<0.05). LT_{50} values were 2.4, 2.8 and 3.4 hours at 10%, 5% and 3% concentrations respectively (Fig., 3). *C. intybus* has a rich history of use in folklore, many of its constituents have not been explored for their pharmacological potential. Several researchers reported that *C. intybus* plant extract showed good mortality results in reduction of worm beetles (Tzamaloukas *et al.,* 2006; Heckendom *et al.,* 2007; Athanasiadou *et al.,* 2007). *C. intybus* had been traditionally used as a liver toxic, cardiotoxic, diuretic, stomachic, cholagogue, inflammations, anorexia, dyspepsia, flatulence, jaundice, splenomegaly, amenorrhea, dysmenorrheal and asthma (Sala, 1994; Jabeeen *et al*., 2009; Abbas *et al.,* 2013). Toxicolgoical data on *C. intybus* is currently limited and less work has been done against termites using it.

O. basilicum seed extract illustrated 80 %, 78 % and 76 % mortality at 10 %, 5 % and 3 % concentration (F=36.0, P<0.05). According to Matovu, 2010 (leaves, Shoot, flowers and branches) of *O. basilicum* have been reported to control the insects especially termites. This plant showed great repellent and toxic effects against the dry wood termite, *Cryptotermes brevis* (Isoptera:

Kalotermitidae) (Sbeghen *et al.,* 2002). 10 %, 5 % and 3 % concentrations of *Ocimum basilicum* seemed to be enough to suppress the termite population(Fig.,2)

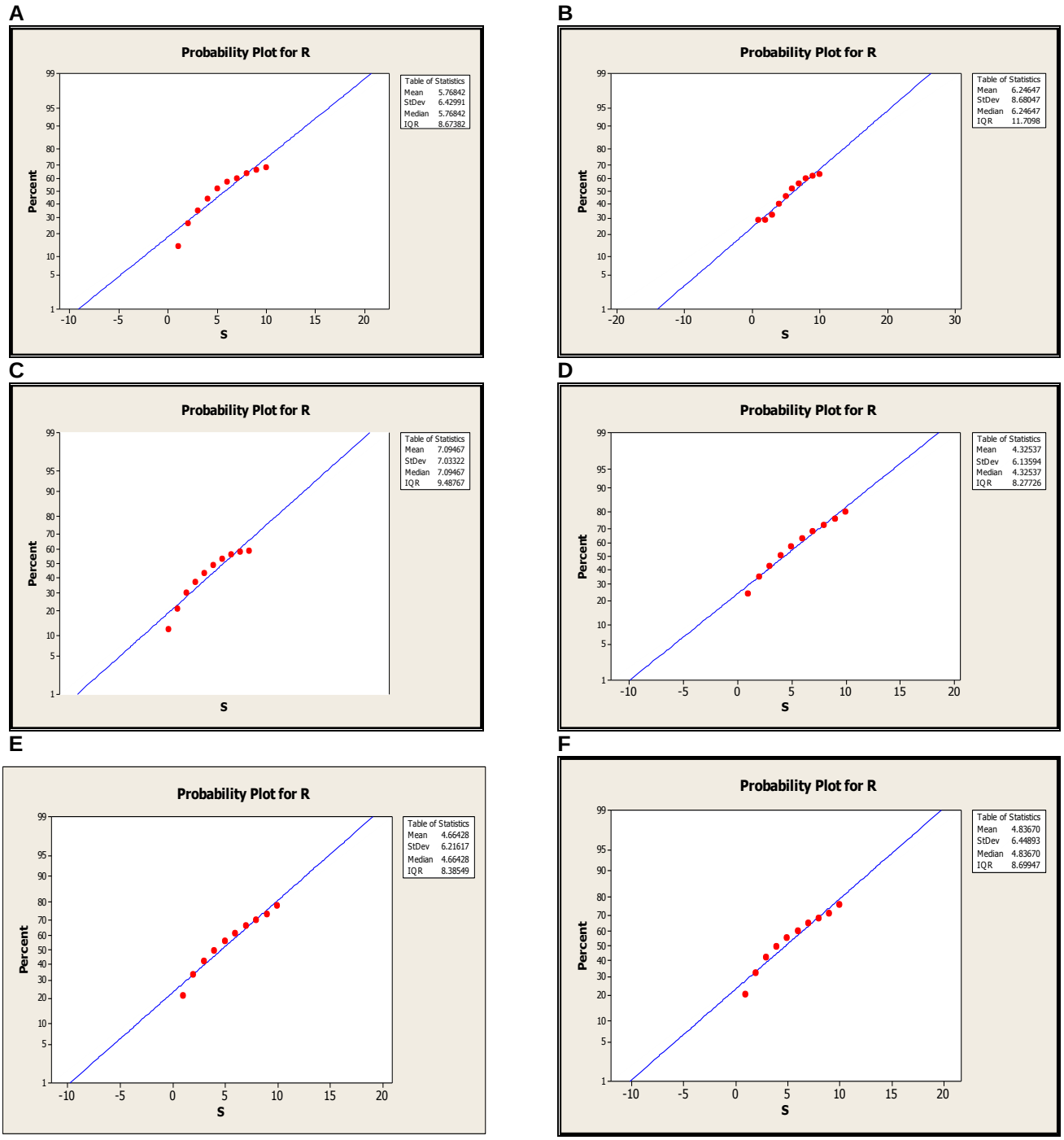

Fig., 2: (A-C) Probability plot for Regression (R) of *P. ovata* at 10%, 5% and 3% concentration. (D-F) Probability plot for Regression (R) of *O. basilicum* at 10%, 5% and 3% concentration where S indicates stress time.

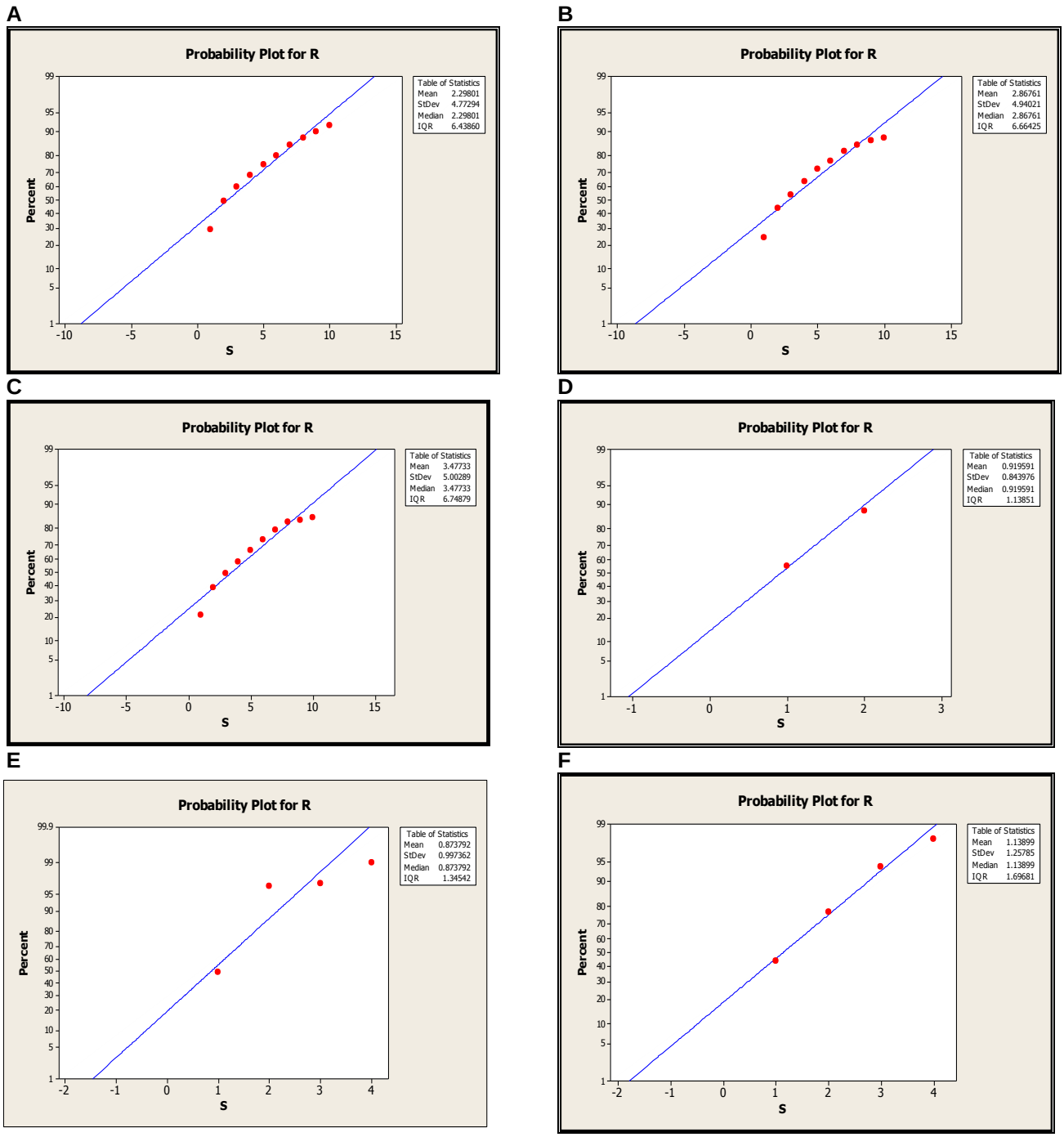

Fig., 3: (A-C) Probability plot for Regression (R) of *C. intybus* at 10%, 5% and 3% concentration. (D-F) Probability plot for Regression (R) of *M. arvensis* at 10%, 5% and 3% concentration where S indicates stress time.

P. ovata illustrated least activity on *Coptotermes heimi* mortality i.e., 72 % , 70 % and 68 % at 10 % , 5 % and 3 % concentration respectively, (Fig., 1). Abbas *et al.,* (2013) reported that *P. ovata* showed LT_{50} value above 35 hours in 10 % extract against *Odontotermes obesus*. But in the present study, LT_{50} value was less than 6 hours for 10 % concentration of *P. ovata* extract, which

showed its more effectiveness against *C. heimi* than *O. obesus* (Fig. 2). *Plantago ovata* has been known to be used as a laxative in stomach disorders and abdominal disorder (Yadav *et al.,* 2006)

CONCLUSION

The results of this study are useful regarding efficacy of *M. arvensis, O. bacilicum, P. ovata* and *C. intybus* against *C. heimi* in the agricultural fields and building structures. These seed extracts will be useful to control the population of *C. heimi*, and may be used as the alternative to insecticides. The screening results suggested that all the four plant extracts had promising capability in termite control in which *M. arvensis* showed highest %age mortality and *P. ovata* showed least %age mortality. Termiticidal compounds could be isolated from these extracts in further studies to save our huge economic loss.

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