

**Termiticidal potential of *Melia azedarach* and
Eucalyptus camaldulensis leaves extract**



By

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Faculty of Biological Sciences

Quaid-i-Azam University

Islamabad

2014

**Termiticidal potential of *Melia azedarach* and
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A thesis submitted in partial fulfillment of the requirements for the degree of

**MASTER OF PHILOSOPHY
IN
PARASITOLOGY**

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Islamabad

2014

Declaration

I hereby declare that the work presented in the following thesis is my own effort, except where otherwise acknowledged, and that the thesis is my own composition. No part of this thesis has been previously presented for any other degree.

Asma Ashraf

CERTIFICATE

This thesis submitted by Asma Ashraf is accepted in its present form by the department of Animal Sciences Quaid-i-Azam University Islamabad, as satisfying the thesis requirement for the degree of Master of Philosophy in Parasitology.

Internal Examiner: _____

(Supervisor)

External Examiner: _____

Chairman: _____



IN THE NAME OF ALLAH

THE MOST BENEFICENT

THE MOST MERCIFUL

***DEDICATED TO
MY LOVING AND CARING PARENTS
AND SUPERVISOR***

List of Abbreviations

Abbreviations	Full Names
BSA	Bovine serum albumin
TP	Total population
ODP	Observed dead population
N	Number of samples
O.R	Observed range
\bar{X}	Mean
S.D	Standard Deviation
S.E	Standard Error
C.I	Confidence interval
C.V	Coefficient of variance
HDL	High density lipid
TG	Triglycerides
mg/g	milligram/gram
mg/dl	milligram/deciliter
ml	milli litter
ppm	Parts per million
Hrs.	Hours
Spp.	Species
M	Molar
μm	Microns
WHO	World health organization
LC	Lethal concentration
LD	Lethal dose

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ABSTRACT

This study was carried out to analyze morphometric variations in two termite species i.e. *Odontotermes obesus* and *Microtermes obesi* and to determine the anti termitic potential of two local plants *Melia azedarach* and *Eucalyptus camaldulensis* against them. Soldier cast of these two termite species was collected from three different sites including Murree (Site A), Rawalpindi (Site B) and Gujarkhan (Site C). Morphometric variations in different characters like full body length, length of thorax, length of abdomen, length from head to mandible tip, length of head, width of head, length of pronotum, width of pronotum, length of postmentum, width of postmentum, length of right mandible, length of left mandible, length of antenna (scape, pedicle, flagellum), length of front, middle and hind legs (coxa, trochanter, femur, tibia, tarsus and claw) were measured and statistically analyzed for mean, standard deviation, standard error, coefficient of variability, confidence interval (95%) and analysis of variance. The mean values of the different population samples were compared with the student *t*-test, following the Minitab version 16. The results showed that majority of external characters of soldier cast were not significantly different from each other with different locations. However small numbers of characters showed variations like pedicle, scape, trochanter and tibia. To determine termiticidal activity of *Melia azedarach* and *Eucalyptus camaldulensis*, 40 termites were placed in each petri dish having filter paper soaked in different concentrations (100ppm, 200ppm, 300ppm) of water and methanol extracts of leaves. In no choice experiment percent mortality of termites was calculated after 24 hrs, 48 hrs and 72 hrs. The results were analyzed by using one way Anova and Tukey test. Among two solvent extracts tested, the maximum activity was observed in both water and methanolic extracts of *E. camaldulensis* against *M. obesi* and *O. obesus*. Protein (mg/g), Carbohydrate (mg/g) and lipid mg/dl (triglyceride, cholesterol and high density lipid) were estimated by Lowry's method, phenol sulphuric acid method and by biochemistry analyzer. Carbohydrate, lipid and protein contents of both the termite species were decreased as compared to control. The lowering of these biochemical components may be due to the insecticidal stress caused by these extract which lowered the feeding, proper digestion of food and metabolism.

INTRODUCTION

Termites are social insects (order Isoptera, class Insecta) and world widely distributed (Thorne and Carpenter, 1992). Termites are mostly present in the soil which is rich in organic materials, on lumber and decaying logs. They feed on cellulosic material but they are unable to digest it. For this purpose they have symbiotic endomicrobes (bacteria, Protozoans and fungi) in their hind gut that produce cellulases which convert cellulose into simpler organic substances absorbed by termites as energy and carbon source (Stingl *et al.*, 2005; Ikeda *et al.*, 2007).

Termites are considered as the important pest of cultivated crops and household materials. In tropical areas they are the major pests of crops and wood made things. As termites are more effective in utilization of different type of food, this ability helped them to become the wood and standing crops pest and some can attack furniture and cabinets (Akutse and Owusu, 2012). Even though termites are most unwanted pests because they damage wooden structures but they play a major role in recycling of wood and plant materials, altering soil conditions, modifying soil structure and productivity by increasing organic matter in soil, as they are competent decomposer of cellulose (Sugimoto *et al.*, 2000). They also provide food for other animals (Lee and Wood, 1971).

Seven families are included in order Isoptera including Rhinotermitidae, Hodotermitidae, Termopsidae, Termitidae, Kalotermitidae, Mastotermitidae and Semitermitidae (Noirat, 1992; Abe *et al.*, 2000). Out of seven families only Termitidae family refers to higher termite because during the course of evolution they lost flagellates from their hind gut. (Noirot, 1992). Termitidae is the largest family including eight subfamilies, about 250 genera and over 2000 species. The remaining six families are lower termites because cellulose in their diet can not be digested without the assistance of protozoan symbionts along with bacteria, archaea and fungi in their hind gut (Cleveland, 1925).

Higher termites (Termitidae) did not harbor protozoa but have only fungi, archaea and bacteria residing in their hind gut and can feed on wide range of materials like leaves,

roots, grass, manure and partially digested organic matter (humus) (Wood and Johnson, 1986). Within the family Termitidae there are two groups of termite species, one is fungus cultivating and other is non-fungus cultivating species. In non-fungus cultivating higher termite's cellulose digesting enzymes are produced by midgut and salivary gland of termite for the digestion of food material containing cellulose (Staylor, 1992; Breznak and Brune, 1994) and their intestinal bacteria does not appear to have any major role in cellulose digestion (Breznak and Brune, 1994).

Subfamily Macrotermitinae includes fungus cultivating termites. These termites construct huge fungal gardens in their nest by collecting and accumulating partially digested plant material which is further invaded and digested by fungus mycelium (Wood and Thomas, 1989). The garden fungus which is associated with termite nest belongs to the genus *Termitomyces*. Termites digest fungus mycelium which gives high nutritional content also provide that essential enzymes which are required for cellulose digestion (Martin and Martin, 1978; Martin, 1992).

In Pakistan agricultural crops are mostly attacked by fungus growing termites, *Microtermes* spp. and *Odontotermes* spp. Different termite species attack different crops in different localities. *Microtermes obesi* and *Odontotermes obesus* cause severe damage to the wheat crop. Wheat and sunflower are more badly affected by *O. obesus* because it is very serious pest of both of these. According to Pearce (1997) presence and distribution of termite species depends upon the type of vegetation in a particular region. Termites remain always present in the soil but their population increase with increase of dry period till optimum temperature (Aslam, 1994).

The Indian white termite, *O. obesus* (Rambur) has wide range of distribution in Pakistan, Bangladesh and India (Akhtar and Anwar 1975; Chhotani, 1979). It lives in huge mud mounds and is highly damaging insect. Its feeding habit usually includes cellulosic material and everything containing carbohydrate. It commonly feed on wood and cause severe damages to it. Major economic loss is caused by this termite species because it damage wooden cabinets, fuel wood, floor timber and railway tracks (Akhtar

and Anwar, 1991). Agriculture crops are more commonly attacked by this species and it effect green foliage crops. In Pakistan this species has also been recorded to damage woodwork in buildings in various ecological areas. It attacks houses in villages more commonly than in urban areas (Akhtar and Ahmed, 1991).

Genus *Microtermes* include termites with pin head size head and small mandible. Both soldier and worker are almost similar in size (personal observation). Due to small size of their head and mandibles *microtermes* soldiers are not too much effective against their predators. Due to this they mostly adopt such defensive strategies so that their predators cannot pass through tunnel passages. When their fungus comb chambers are broken they immediately repair it to avoid their predators. Where *Microtermes* found easily approachable cellulose source they over produce fungus in that area and large mushroom size fungus was found associated with their comb. They are mostly present in the mound walls of the *Macrotermes* species but not exclusively dependent on them and they are to be opportunistic when they are using already present mound of other species for their nest.

Out of the 2,600 known termite species, only 40 species has been identified in the United States (Kambhampati and Eggleton, 2000).and 53 termite species are reported in Pakistan but only 11 species were found to cause damage (Naeem and Shafqat, 2013). For identification soldier and alate cast are used and among them worker cast is morphologically similar. Mouthparts are usually used for identification. Head size, mandibles, postmentum, pronotum and length of antenna are important.

To control and prevent termite infestation and renovation processes billions of dollars are spent annually throughout the world (Su *et al.*, 1987; Su and Scheffrahn, 1990; Ahmed *et al.*, 2007). In developed countries the cost for control of these pest is considered low but in countries where population level of these pests is high, the control measure are not economically reasonable (Robinson, 1996). In developing countries like Pakistan, damages caused by termites are compensated because the expenditures to control these pests are greater than the expenditures which are required for the

replacement of wooden materials. But now the people are very anxious about the termite infestation especially in big cities of Pakistan where the living standards is raised.

To control termite population various man-made (synthetic) chemicals have been used. But these synthetic pesticides were found to be very damaging to environment, expensive and kill many other useful creatures. Synthetic pesticides have some short comings such as excessive use of these pesticides lose their effectiveness as insects develop resistance against them, some of their active compounds leaches down and raise their content in underground water table, some persist in soil and lakes, have harmful effects on non-target organisms and have health hazards associated with human life (Jitunari *et al.*, 1995; Valles and Woodson, 2002; Boue and Rania, 2003; Hu, 2005).

As synthetic pesticides which are used for the control of insects and pests, they have some serious problems. They are more expensive than natural products and they directly target the human health causing serious disorders such as immune system deformities, cancer and birth defects in new born (Nigam and Bhatt, 2001; Bounias, 2003). Therefore, there is a great need to find out such alternate methods which are based on plant products as they are less toxic and have no side effects on atmosphere. Due to their toxicity, longtime stability, food contamination and environmental pollution many synthetic compounds such as chlorinated, organophosphorus and carbamate insecticides are banned.

Biological control is one of the methods that involve the use of biologically active compounds for termite control. This method of termite control is very economical, easy to handle and environment friendly. Now a days there is an increasing global attention in developing bio active compound which can be used instead of synthetic chemical compound to control pests of remedial and commercial worth importance as these compounds can be easily degraded, nontoxic and are not harmful as synthetic pesticides are (Sharma *et al.*, 1998; Ojewole *et al.*, 2000; Sosan *et al.*, 2001; Moretti *et al.*, 2002; Cetin and Yanikogh , 2006).

Biological control involves the use of plants that have medicinal value. According to WHO a medicinal plant is any plant which can be used for therapeutic and pharmaceutical purposes and its one or more organic substances can be a precursor for the synthesis of new drugs (Sofwara, 1982). Out of 250,000 species of plants 80,000 species are reported which can have at least some medicinal values and among them 5000 plants were considered to have great therapeutic significance (Thomas and Shakira, 1998). About 52 natural plant species belonging to 25 families of angiosperms are reported in Pakistan that has medicinal values (Farooq, 1990; Siddiqui and Khan, 1991; Varaldo, 2002).

Bio-termiticides are those which are based on the active constituents of plants and possess robust termiticidal properties. Plant based insecticides can be much more effective than chemically synthesized compounds because they are more target specific, easily degraded in environment, safe to use and more efficient. Natural pesticides which are derived from the plants are more efficient, have extensive control range and reduce population of all kind of insect pest even applied in minor amount. Many plant species have been explored which shows anti-termite activities. Many organic compounds which were derived from different plants such as flavonoids (Boux and Rania, 2005), sesquiterpenes (Arihara *et al.*, 2004) and thiophenes (Fokialakis *et al.*, 2006) were found to have antitermite properties (Kinyanjui *et al.*, 2000)

Natural insecticides effect insects behavior in number of ways including growth retardation (Breuer and Schmidt, 1995), feeding inhibition (Wheeler and Isman, 2001), oviposition deterrence (Zhao *et al.*, 1998), suppression of calling behavior (Khan and Saxena, 1986) and toxicity (Hiremath *et al.*, 1997).

In field and laboratory many experiments were performed on crude plants extracts to exploit their antitermite activities. Many plant species were used in past to explore their insecticidal properties and anti-feedant activities such as *Tabebina guaycan*, *Lysitoma seemnii*, *Diospyros sylvatica*, *Pseudotusuga menziesii*, (Ganapaty *et al.*, 2004), *Curcuma aromatica* and *Euphorbia kansui* (Shi *et al.*, 2008). These extracts severely discourage termite feeding and their survival rate was also reduced.

Instead of using only rudimentary plants extracts some active components can also exhibit such activities which reduce termite growth (Kinyanjui *et al.*, 2000) and were applied on field termites to study their natural responses such as repellent, toxicity and anti-feedent behavior (Motohashi *et al.*, 2000; Blaske and Hertel, 2001; Blaske *et al.*, 2003).

Few synergists in form of poison baits were applied in combination with crude plant extract in order to increase their insecticidal properties and effectively studied the foraging, (Su and Scheffrahn, 2000) burrowing and reproductive behavior in termites (Cornelius and Lax, 2005). Few chitin synthesis inhibitors i.e. chlorfluazuron, hexaflumuron and diflubenzuron (Rojas and Morales, 2001) applied in the form of Summon disks and filter paper disks were found effective against aggregation and foraging responses in *Coptotermes* spp.

Different types of antibiotics such as ampicillin, urea and tetracycline showed increased death rate of termites and reduced number of intestinal protozoans in *R. virginicus* and *R. flavipes* (Waller, 1996). Population of symbiotic protozoans in *Coptotermes formosanus* was decreased by the application of Vetiver oil (Maistrello *et al.*, 2003). Different plants show anti-termite activities and contain certain chemicals that reduce termite growth or kill them (Adams *et al.*, 1988). These plants include clove bud, cassia leaf, vetiver oil, cedar wood, *Eucalyptus citrodora*, lemon grass, *Eucalyptus globules*, (Zhu *et al.*, 2001), *Coleus amboinicus* (Singh *et al.*, 2004), *Calotropis procera* (Shing *et al.*, 2002), *Taiwania cryptomerioides* Hayat (Chang *et al.*, 2001), *isoborneol* (Blaske *et al.*, 2003) and *Ocimum basilicum* L., *Cymbopogon winterianus* Jowitt, *Cinammomum camphora*, *Rosmarinus officinalis* (Sbeghen *et al.*, 2002),. Neem (*Azadirachta indica*) contains triterpenoid azadirachtin and it has been used to repel certain insect pests species (Ascher and Meisner, 1989; Schumutter 1986). Insecticidal properties were also reported in some species of Lantana camara and Juniper (Adam *et al.*, 1988; Verma and Verma, 2006).

M. azedarach is commonly known as Chinaberry locally recognized as Dharek. It is a deciduous plant belonging to Meliaceae family. It is a native plant of Indomalaya and Austerlia. Four other species belong to this genus *Melia* which are distributed South East Asia to northern Australia. All these plants are semi ever green. The member of family Meliaceae contain number of constituents, which show development modifying activities, insecticidal properties and antifeedant effects (Nikoletta and Filippo, 2010).

In previous studies it was noticed that extracts from different parts of *M. azedarach* plant exhibit insecticidal properties against many pests. (Abdul *et al.*, 2008). In dengue mosquito Larvicidal activity of *M. azedarach* was conducted by Wondscheer, j. and coworkers in 2004. Phagoinhibitory and antimolting activities of seed extract of *M. azedarach* in ethanol were reported against hemophagous insect *Rhodnius prolixus* which serve as a vector of chagas disease (Rani *et al.*, 1999). Phytochemistry analysis of ethanolic extracts of *M. azedarach* showed that they contain steroids and triterpenoids. Condensed tannins and alkaloids are present in both leaves and seeds. All these compounds have direct effect on development and feeding activities of insects and they also have ovicidal properties (Bahakuni, 1969). Leave extract of *M. azedarach* can be used as pregnancy interceptive (Bohnensteng *et al.*, 2002) While in some other cases it was seen that extract of roots taken in chloroform revealed a substantial contraceptive activity.

Ethanolic extracts of leaf, seed and fruit from *M. azedarach* were found effective in reducing the growth of plant and human pathogenic fungi such as *Candida albicans*, *Microsporum canis*, *Fusarium monitiform* and *Aspergillus flavus* (Carpinella *et al.*, 2002). Scopoletin, a hydroxyl coumaramin was isolated from ripe fruit seeds and was tested for its antifungal activity against *F. verticilloides* and was found effective. This isolated organic compound also showed good synergistic effets when it was applied in combination with two conventional fungicides mancozab or carboxin (Adnan *et al.*, 2009).

To test antibacterial activity different extracts of *M. azedarach* were prepared in different solvents by using Chloramphenicol, Ethyl acetate, Benzene, Methanol, Petrol.

Bacterial spp. which were used for experiments include *Staphylococcus aureus*, *Shigella flexneri*, *Proteus mirabilis*, *Shigella dysenteriae*, *Basillus subtilis* and *Plesiomonas shigellides*. Among these the extract which was taken in Ethyl acetate was found to be most effective as it blocked the growth of all experimented bacteria. To control human pathogenic bacteria some leaf extracts were used. To cure bacterial skin disease in children flower extracts of *M. azedarach* was used. The extract was prepared in methanol to make a cream. An experiment was performed to check the activity of prepared cream and neomycin skin drug. After two weeks result showed that cream is more effective than skin drug in certain cases (Saleem *et al.*, 2002). Antiviral activities of *M. azedarach* were also significant against foot and mouth disease virus (Wachsman *et at.*,1998), herpes simplex virus and polio (Wachsman *et al.*, 1982).

E. camaldulensis commonly called Red River Gum belong to the family Myrtaceae. It is a perennial plant with single stem and can be medium sized to tall tree upto 30m (Bren and Gibbs, 1986), but it can be 45m long (Boland, 1984; Brooker *et al.*, 2002). *E. camaldulensis* is mainly present in Australia and more commonly present on river side in continent (Brooker and Slee, 1996).

Genus *Eucalyptus* contain 800 species of which three or four are endemic to Australia but cosmopolitan in nature because of its ability to adapt easily and grow fastly (Coppen, 2002). *E. camaldulensis* shows significant morphological variation throughout its range that's why number of taxa has been described. Different species of this genus are implanted in different areas depending on the climatic conditions, area, soil and use. In Thailand *E. camaldulensis* Dehnh.is more commonly growing species. It is grown for pulpwood and due to its high growth rate it can be utilized at the age of 3-5 years.

Several studies on *E. camaldulensis* reported that leaf essential oils contained bioactive compounds that displayed larvicidal and mosquito repellent activities (Chang *et al.*, 2001; Watanabe *et al.*, 1993). Antibacterial (Cimanga *et al.*, 2002), analgesic and anti-inflammatory effects (Silva *et al.*, 2003), antioxidative and antiradical activities (Siramon and Ohtani, 2007) antitermitic activity (Siramon *et al.*, 2009),

As plant based insecticides are getting more importance over synthetic / chemical insecticides, this work was planned toward the estimation of termiticidal effect of the extracts from the two remedial plants *E. camaldulensis* and *M. azedarach* against two higher termite species. Now there is an utmost requirement to find out such new control methods that should remain effective even after increased resistant of insect against insecticides and drugs (Achs and Malaney, 2002).

OBJECTIVES:

1. Biodiversity of termites.
2. To sort out termite resistant plants by feeding termites on them.
3. To introduce ecologically safe and decomposable pesticides and to lessen the economic loss of wood damage caused by termites.

MATERIALS AND METHODS

2.1. Termites collection:

Two termite species *M. obesi* and *O. obesus* were collected from three different sites of pothohar region in the month of June 2013. These three sites include site A (Murree) site B (Rawalpindi) and site C (Gujarkhan). Termites were collected by using a collection trap unit as described by Sornnuwat *et al.*, (1996) with some modifications and identified with the help of taxonomic keys (Akhtar, 1983). Specimens were preserved in 70% ethanol for their morphological identification. Live material was kept in petri plates with soaked cotton plugs before using for experiments.

2.2. Identification of termites:

Termites were identified by their morphometric analysis. Specimens from the samples were picked up randomly and measured under stereoscopic binocular microscope with built in magnification changer. Measurements were taken with the help of calibrated ocular micrometer. Diagrams of the head, mandibles, postmentum, pronotum, antenna, thorax, abdomen and legs were made with the help of Olympus binocular attached camera.

2.3. Selection of Plants:

Plants which were selected for experiment on termites include following species.

Table: (a) Plants selected for experiments:

Sr.#	Botanical name	English name	Local name	Family
1	<i>Eucalyptus camaldulensis</i>	River Red gum	Safaida	Myrtaceae
2	<i>Melia azedarach</i>	China berry	Derek	Meliaceae

The leaves of *E. camaldulensis* and *M. azedarach* have been collected locally with in the locality of Quaid-i-Azam University, Islamabad, Pakistan and identified by using key (Nasir and Ali, 1977).

2.4. Extraction method:

Leaves of *E. camaldulensis* and *M. azedarach* were splashed with water to remove the accompanying organisms and attached salts. After that the leaves were dried in oven at 37 Celsius and crumpled with the aid of electric grinder. 40 and 60 mesh retained material was carefully chosen. 30g grinded leaves of the plants used for experiments were extracted in 300ml of two different solvents i.e. methanol and water for 6 to 8 hour (two cycles per hour) in a Soxhelt extraction apparatus. Whatman no.1 filter paper was used to filter the extracts. The dried residues were collected by evaporating the solvent with the help of rotary vacuum evaporator stored in a refrigerator for making stock solution.

2.5. Formulation of stock solution:

One gram of the plant residue was dissolved in 100ml of distilled water (stock solution). The stock solution was further diluted with the help of dilution formula (Jayal *et al.*, 2006).

$$C_1V_1=C_2V_2$$
$$V_1 = \frac{\text{required parts par million} \times \text{required volume}}{\text{Stock solution}}$$

2.6. Antitermites Assay:

The no choice feeding method described by Kang *et al.*, (1990) with some modifications was carried out to find out anti termite efficacy of extracts. Samples of 1, 2 and 3 ml from stock solution were dissolved in 100 ml of distilled water to make different concentrations like 100 ppm, 200 ppm and 300 ppm respectively, and 1.5 ml of each solution was applied on filter paper (Whatman No. 1). For control group filter paper were treated with solvents only. After the evaporation of solvent from filter papers by drying in air, two filter papers were placed in bottom of each petri dish and lid was provided with one. A soaked cotton plug was placed in each petri dish to maintain their moisture content. 40 active termites were put into each petri dish on filter paper, covered and placed in dark. Few drops of water were dropped daily on cotton plug. Experiment was

conducted in triplicate for each sample concentration along with the set of control and percent mortality was counted after 24 hours, 48 hours and 72 hours

$$\% \text{ mortality} = \text{ODP} \div \text{TP} \times 100$$

2.7) BIOCHEMICAL ASSAY:

After 24 hours of exposure of termites to plants extracts the termites were removed, washed with saline solution, dried out and were measured with electrical balance. Sucrose solution (0.25 molar) was used to homogenize 90mg of each sample with the help of dounce homogenizer. The homogenate was centrifuged at 13000 rpm for 15-20 minutes. The supernatant was collected and stored at 20°C for later use to determine carbohydrate, protein and lipid concentrations.

2.7.1) Protein estimation (mg/g):

Protein contents of termites were evaluated by using Lowry's method. The standard was Bovine serum albumin (BSA).

2.7.2) Carbohydrate estimation (mg/g):

Carbohydrate content of termites was estimated by Phenol Sulphuric acid method. The standard here used was Glucose solution.

2.7.3) Lipid mg/dl (triglyceride, cholesterol, high density lipid) estimation:

Triglyceride (TG), cholesterol, and high density lipid (HDL) contents were estimated through biochemistry analyzer.

2.8. Statistical analysis:

Results of morphometric analysis such as mean, standard deviation and coefficient of variance were analyzed by using student "t" test to determine the variations in different parameters. Mortality ratio percentage of termites was calculated and analyzed by using one way Anova and Tukey test. Values of $P < 0.05$ were considered significant statistically. After 24 hrs of exposure to each extract LC_{50} and LC_{90} was calculated by using Probit analysis Finney (1971).

RESULTS

Morphological variations in termites collected from three different regions of Pothohar (Pakistan).

Morphometric variations of *O. obesus*:

Two termite species collected from three site A (Murree), site B (Rawalpindi) and site C (Gujarkhan) were analyzed for variation in their external characters. Length of whole body of *O. obesus* varied from 42.54-54.01 μ m. The mean value of samples collected from three sites was 53.00 μ m, 51.83 μ m and 53.17 μ m respectively. Value of coefficient of variability of sample varied between 34.04 μ m-39.05 μ m (Table and figure 3.1). Length of abdomen varied from 17 μ m-21 μ m and mean value was 18.66 μ m, 18.66 μ m and 20.00 μ m. Value of coefficient of variability of sample varied between 19.04 μ m-21.94 μ m (Table and figure 3.1). No significant difference was found amongst the termite samples taken from various localities.

In case of length of prothorax variation between observed value range from 2.0 μ m-4.0 μ m. Three population sample i.e. A, B and C had mean value 3.00 μ m, 3.10 μ m and 3.50 μ m respectively. Value of coefficient of variability of three was 6.93 μ m-8.66 μ m (Table and figure 3.1). Length of mesothorax varied from 2.0 μ m-3.50 μ m. Three samples had mean value 3.00 μ m, 3.10 μ m and 3.00 μ m. Value of coefficient of variability of sample varied between 6.93 μ m-10.09 μ m (Table and figure 3.1). Length of metathorax varied from 2.50 μ m-4.0 μ m and mean value was 3.50 μ m, 3.76 μ m and 3.43 μ m. Value of coefficient of variability of three varied between 9.45 μ m-19.04 μ m (Table and figure 3.1). There was no significant difference in the length of thorax of termite samples taken from various localities. Similarly length from head to mandible tip varied from 20 μ m-22.5 μ m. Three sample had mean value 21.50 μ m, 22.16 μ m and 22.16 μ m respectively. Value of coefficient of variability of samples varied between 33.56 μ m-71.01 μ m (Table and figure 3.1). No significant difference was observed among them.

Length of head varied from 10.50 μ m-14.50 μ m. Three population sample i.e. A, B and C had mean value 13.00 μ m, 13.33 μ m and 11.66 μ m respectively. Value of coefficient

of variability of samples varied between 11.49 μm -41.57 μm (Table and figure 3.2). Width of head varied from 9.0 μm -11.5 μm . Three population sample i.e. A, B and C had mean value 10.73 μm , 11.00 μm and 10.66 μm . Value of coefficient of variability of samples varied between 21.92 μm -34.64 μm (Table and figure 3.2). No significant difference was present. Length of pronotum varied from 9 μm -11 μm . Three population sample i.e. A, B and C had mean value 10.16 μm , 10.16 μm and 10.16 μm respectively. Value of coefficient of variability of sample varied between 15.25 μm -20.79 μm (Table and figure 3.2). Width of pronotum varied from 4.0 μm -5.8 μm . Three population sample i.e. A, B and C had mean value 5.00 μm , 5.16 μm and 5.16 μm respectively. Value of coefficient of variability of sample varied between 8.32 μm -13.86 μm (Table and figure 3.2). No significant difference was observed. Length of postmentum varied from 7.0 μm -13 μm . Three population sample i.e. A, B and C had mean value 12.50 μm , 7.50 μm and 410.83 μm . Value of coefficient of variability of sample varied between 8.32 μm -39.84 μm (Table and figure 3.3). No significant difference was found amongst the termite samples taken from various localities. Width of postmentum varied from 8.0 μm -9.5 μm . Mean value range between 9.50 μm , 9.00 μm and 8.50 μm . Value of coefficient of variability varied between 25.98 μm -29.44 μm (Table and figure 3.2). No significant difference was found among them.

Length of right mandible varied from 8.0 μm -9.5 μm and mean value 8.50 μm , 5.83 μm and 8.66 μm respectively. Value of coefficient of variability varied between 1.80 μm -25.98 μm (Table and figure 3.3). Length of mandible varied from 8.0 μm -9.5 μm . Three population sample i.e. A, B and C had mean value 8.50 μm , 8.66 μm and 8.83 μm . Value of coefficient of variability of sample varied between 11.50 μm -47.00 μm (Table and figure 3.3). Length of tooth varied from 3.0 μm -4.0 μm and mean value varied between 3.83 μm , 3.50 μm and 3.50 μm respectively. Value of coefficient of variability of sample varied between 8.66 μm -17.00 μm (Table and figure 3.3). No significant difference was found among the three termite samples.

Length of different segments of antenna like scape, pedicle and flagellum also exhibit small differences. Observed rang in scape was 2.0 μm -3.0 μm . Three population

sample i.e. A, B and C had mean value 2.83 μ m, 2.10 μ m and 2.50 μ m respectively and value of coefficient of variability varied between 5.20 μ m-11.00 μ m (Table and figure 3.4). Observed range in pedicel was 0.5 μ m-1.0 μ m and mean value was 0.83 μ m, 0.60 μ m and 0.66 μ m respectively. Value of coefficient of variability of sample varied between 2.00 μ m-11.00 μ m (Table and figure 3.4). Observed range in flagellum was 16 μ m-19 μ m and mean value was 18.00 μ m, 15.83 μ m and 17.33 μ m respectively. Value of coefficient of variability of sample varied between 13.59 μ m-33.64 μ m (Table and figure 3.4). No significant difference was observed in different segments of antenna.

Length of different segments of front leg like coxa, trochanter, femur, tibia, tarsus and claw also exhibit small differences. Observed range in coxa was 5.5 μ m-6.5 μ m. Three population sample i.e. A, B and C had mean value 6.10 μ m, 6.00 μ m and 6.00 μ m. Value of coefficient of variability of sample varied between 17.39 μ m-27.71 μ m (Table and figure 3.5). Observed range in trochanter was 2.0 μ m-3.5 μ m and mean value was 2.60 μ m, 3.10 μ m and 3.00 μ m respectively. Coefficient of variability of sample varied between 6.93 μ m-10.09 μ m (Table 3.5). Observed range in femur was 7.5 μ m-10 μ m mean value was 9.00 μ m, 9.00 μ m and 8.66. Coefficient of variability varied between 17.39 μ m-27.71 μ m (Table and figure 3.5). Observed range in tibia was 7.0 μ m-9.0 μ m and mean value varied between 8.93 μ m, 8.50 μ m and 8.50 μ m respectively. Value of coefficient of variability varied between 25.98 μ m-34.00 μ m (Table and figure 3.5). Observed range in tarsus was 2.0 μ m-4.0 μ m and mean value was 3.03 μ m, 2.83 μ m and 3.50 μ m respectively. Value of coefficient of variability varied between 8.66 μ m-13.99 μ m (Table and figure 3.5). Observed range in Claw was 0.3 μ m-0.8 μ m and mean value was 1.43 μ m, 1.50 μ m and 1.50 μ m respectively. Value of coefficient of variability varied between 1.73 μ m-1.86 μ m (Table and figure 3.5). No significant difference was found in different parts of front leg of termite samples.

In middle legs, length of different segments of middle leg like coxa, trochanter, femur, tibia, tarsus and claw also exhibit small differences. Observed range in coxa was 5.6 μ m-7.0 μ m and sample A, B and C had mean value 5.50 μ m, 5.83 μ m and 5.00 μ m. Value of coefficient of variability of sample varied between 10.96 μ m-15.59 μ m (Table

and figure 3.6). Observed range in trochanter was 2.0 μ m-3.0 μ m and mean value was 2.50 μ m. Value of coefficient of variability of sample varied between 3.46 μ m-5.20 μ m (Table and figure 3.6). Observed range in femur was 8.0 μ m -9.5 μ m and mean value was 8.50 μ m, 8.66 μ m and 8.50 μ m. Value of coefficient of variability varied between 17.39 μ m-25.98 μ m (Table and figure 3.6). Observed range in tibia was 7.5 μ m-9.0 μ m and mean value 8.00 μ m, 8.00 μ m and 8.50 μ m respectively. Value of coefficient of variability varied between 24.25 μ m-25.98 μ m (Table and figure 3.6). Observed range in tarsus was 2.5 μ m-3.5 μ m and mean value 2.90 μ m, 3.00 μ m and 3.10 μ m. Value of coefficient of variability varied between 6.93 μ m-10.90 μ m (Table and figure 3.6). Observed range in Claw was 0.8 μ m-1.5 μ m and mean value was 1.50 μ m, 1.50 μ m and 1.33 μ m. Value of coefficient of variability varied between 1.73 μ m-2.00 μ m (Table 3.6). No significant difference was recorded in middle leg length.

Length of different segments of hind leg like coxa, trochanter, femur, tibia, tarsus and claw also show small differences. Observed range in coxa was 4 μ m-5.5 μ m. Sample A, B and C had mean value 5.00 μ m, 5.16 μ m and 4.83 μ m. Value of coefficient of variability of sample varied between 8.69 μ m-25 μ m (Table and figure 3.7). Observed range in trochanter was 1.5 μ m-2.5 μ m and mean value was 1.83 μ m, 2.0 μ m and 1.83 μ m. Value of coefficient of variability varied between 1.89 μ m-3.46 μ m (Table and figure 3.7). Observed range in femur was 7.0 μ m-9.5 μ m and mean value was 8.50 μ m, 9.00 μ m and 8.33 μ m. Value of coefficient of variability varied between 16.63 μ m-27.71 μ m (Table and figure 3.7). Observed range in tibia was 8.0 μ m-10.5 μ m. Three population sample i.e. A, B and C had mean value 9.50 μ m, 10.03 μ m and 9.23 μ m respectively. Value of coefficient of variability of sample varied between 20.95 μ m-34.70 μ m (Table and figure 3.7). Observed range in tarsus was 2.5 μ m-4.0 μ m and mean value 3.00 μ m, 3.50 μ m and 3.50 μ m respectively. Value of coefficient of variability of sample varied between 3.93 μ m -8.6 μ m (Table and figure 3.7). Observed range in Claw was 1.0 μ m-2.0 μ m and mean value 1.23 μ m, 1.43 μ m and 1.50 μ m respectively. Value of coefficient of variability of varied between 1.61 μ m-1.86 μ m (Table and figure 3.7). No significant difference was found amongst the termite samples taken from various localities.

Morphometric analysis of *O. obesus*

Table 3.1: Variations in full body length, length of thorax (Prothorax, mesothorax, metathorax) (μm) and length of abdomen (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical parameters	Full Body length			Length of abdomen			Length of prothorax			Length of mesothorax			Length of metathorax		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	52.54	49.50-52.01	52.50-54.01	17.00-19.5	17.00-19.50	19.00-21.00	2.01-4.00	2.30-3.50	2.50-4.00	2.00-4.01	2.31-3.51	2.51-3.50	2.50-4.50	3.01-4.50	2.51-4.00
\bar{X}	53.00	51.83	53.17	18.66	18.66	20.00	3.00	3.10	3.50	3.00	3.10	3.00	3.50	3.76	3.43
S.D	2.65	2.25	1.76	1.52	1.60	1.50	0.50	0.36	0.50	0.50	0.36	0.50	0.45	0.25	0.40
S.E	1.53	1.30	1.01	0.88	0.92	0.86	0.28	0.20	0.28	0.28	0.20	0.28	0.26	0.14	0.23
95% C.I	46.43-59.57	46.23-57.43	48.80-57.53	14.87-22.65	14.68-22.65	16.27-23.72	1.75-2.24	1.75-2.24	2.25-4.72	1.75-4.24	2.20-3.99	1.75-4.24	2.36-4.63	3.14-4.39	2.42-4.43
C.V	34.04	39.05	39.05	20.03	19.04	21.94	6.93	6.93	8.66	6.93	10.09	6.93	9.45	19.04	10.43
P	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.02	0.01	0.02	0.10	0.20	0.01	0.00	0.00

Table 3.2: Variations in length from head to mandible tip (μm), length of head (μm), width of head (μm), length of pronotum (μm) and width of pronotum of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical Parameters	Length from head to Mandible tip			Length of head			Width of head			Length of pronotum			Width of pronotum		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	21.50-22.00	20.00-22.50	20.00-22.50	12.31-13.50	12.50-14.50	10.51-12.00	9.01-11.21	10.51-11.51	10.01-11.51	9.51-11.00	9.01-11.02	9.01-11.50	4.50-5.50	4.51-5.81	4.50-5.80
\bar{X}	21.50	22.16	22.16	13.00	13.33	11.66	10.73	11.00	10.66	10.16	10.16	10.16	5.00	5.16	5.16
S.D	0.500	0.76	0.76	0.50	1.04	1.60	0.64	0.50	0.76	0.76	1.04	0.76	0.500	0.76	0.76
S.E	0.28	0.44	0.44	0.28	0.60	0.92	0.37	0.28	0.441	0.44	0.60	0.44	2.89	0.44	0.44
95% C.I	20.25-22.74	20.26-24.06	18.58-24.06	11.75-14.24	10.74-15.09	7.67-15.65	9.13-12.33	9.74-12.24	8.76-12.56	8.26-12.06	7.58-12.75	8.26-12.06	3.75-6.24	3.26-7.06	2.76-6.56
C.V	71.01	48.00	33.56	41.57	20.52	11.49	26.22	34.64	21.92	20.79	15.25	20.79	13.86	9.45	8.32
P	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.01

Table 3.3: Variations in length of postmentum (μm), width of postmentum (μm), length of right mandible (μm), length of left mandible (μm) and length of tooth on left mandible (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical parameters	Length of postmentum			Width of postmentum			Length of right mandible			Length of left mandible			Length of tooth on left mandible		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	12.01-13.00	7.01-8.50	10.01-12.01	9.01-11.11	8.51-9.51	8.51-9.50	8.01-9.01	8.01-9.51	8.01-9.00	8.00-9.00	8.21-9.51	8.00-9.50	3.5-4.01	3.01-4.00	3.20-4.00
\bar{X}	12.50	7.50	10.83	9.50	9.00	8.50	8.50	5.83	8.66	8.50	8.66	8.83	3.83	3.50	3.50
S.D	0.50	0.50	2.02	0.50	0.50	0.50	0.50	4.65	0.76	0.50	1.15	0.28	0.28	0.50	0.50
S.E	0.28	0.28	1.17	0.28	0.28	0.28	0.28	2.68	0.44	0.28	0.66	0.16	0.16	0.28	0.28
95% C.I	11.25-13.74	6.28-8.74	5.81-15.85	8.25-10.74	7.75-10.24	7.25-9.74	7.28-9.71	5.71-17.37	6.76-10.56	7.25-9.74	5.79-11.53	8.11-9.55	3.11-4.55	2.25-4.74	2.25-4.74
C.V	39.84	22.52	8.43	29.44	27.71	25.98	25.98	1.80	17.39	25.98	11.50	47.00	17.00	8.66	8.66
P	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.21	0.00	0.00	0.00	0.00	0.00	0.01	0.03

Table 3.4: Variations in length of antenna (scape, pedicle, flagellum) (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical parameters	Length of antenna								
	Scape			Pedicle			Flagellum		
	Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3
O.R	2.51-3.00	2.01-2.50	2.00-3.00	0.50-1.00	0.50-0.50	0.50-0.80	17.00-19.00	15.00-16.00	16.50-18.00
\bar{X}	2.83	2.10	2.50	0.83	0.60	0.66	18.00	15.83	17.33
S.D	0.28	0.31	0.500	0.28	0.17	0.28	1.00	0.76	2.08
S.E	0.16	0.20	0.28	0.16	0.10	0.16	0.57	0.44	1.20
95% C.I	2.11-3.35	1.21-2.99	1.25-3.74	0.11-1.55	0.17-1.03	0.05-1.38	15.51-20.48	13.93-17.73	12.10-22.25
C.V	11.00	5.28	5.20	11.00	4.00	2.00	29.44	33.64	13.59
P	0.00	0.03	0.03	0.42	0.05	0.18	0.00	0.05	0.03

Table 3.5: Variations in length of front legs (coxa, trochanter, femur, tibia, tarsus and claw) (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical parameters	Length of coxa			Length of trochanter			Length of femur			Length of tibia			Length of Tarsus			Length of claw		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	6.00-6.51	5.50-6.50	5.50-6.50	2.00-3.00	2.50-3.51	2.50-3.51	8.00-10.00	8.01-10.00	7.50-9.00	8.50-9.00	7.00-8.00	7.00-8.50	2.50-3.50	2.00-3.50	3.00-4.00	1.00-1.50	1.00-1.50	1.00-1.50
\bar{X}	6.10	6.00	6.00	2.60	3.10	3.00	9.00	9.00	8.66	8.93	8.50	8.50	3.03	2.83	3.50	1.43	1.50	1.50
S.D	0.36	0.50	0.50	0.361	0.36	0.500	0.50	0.50	0.76	0.40	0.50	0.50	0.25	0.28	0.50	0.40	0.50	0.50
S.E	0.20	0.28	0.28	0.20	0.20	0.28	0.28	0.28	0.44	0.23	0.28	0.28	0.14	0.16	0.28	0.23	0.28	0.28
95% C.I	5.20-6.99	4.75-7.24	4.75-7.24	1.70-3.49	2.20-3.99	1.75-4.24	7.75-10.24	7.75-10.24	6.76-10.56	7.92-9.93	7.25-9.74	7.25-9.74	2.40-3.65	2.11-3.55	2.25-4.74	0.42-2.43	0.25-2.74	0.58-2.74
C.V	24.50	17.32	17.32	7.69	10.09	6.93	27.71	27.71	17.39	34.00	25.98	25.98	13.99	11.00	8.66	1.86	1.73	1.73
P	0.00	0.00	0.00	0.01	0.01	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.20	0.22	0.22

Table 3.6: Variations in length of middle legs (coxa, trochanter, femur, tibia, tarsus and claw) (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical Parameters	Length of coxa			Length of trochanter			Length of femur			Length of tibia			Length of Tarsus			Length of claw		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	5.00-6.00	5.50-6.01	5.50-6.50	2.00-3.00	2.00-3.01	2.01-3.00	8.01-9.01	8.01-9.50	8.01-9.01	7.50-8.50	7.50-8.50	8.00-9.00	2.50-3.50	2.30-3.00	2.50-3.50	1.00-1.50	1.00-1.50	0.80-1.50
\bar{X}	5.50	5.83	5.00	2.50	2.50	2.50	8.50	8.66	8.50	8.00	8.00	8.50	2.90	3.00	3.10	1.50	1.50	1.33
S.D	0.50	0.76	0.50	0.50	0.50	0.50	0.50	0.76	1.50	0.50	0.50	0.50	0.36	0.50	0.36	0.50	0.50	0.28
S.E	0.28	0.44	0.28	0.28	0.28	0.28	0.28	0.44	0.28	0.28	0.28	0.28	0.20	0.28	0.20	0.28	0.28	0.16
95% C.I	4.25-6.74	3.93-7.73	3.75-6.25	1.25-3.74	0.75-3.24	1.25-3.24	7.25-9.74	6.76-10.56	7.25-9.74	6.75-9.24	6.75-9.24	7.25-9.74	2.00-3.79	1.75-4.24	2.20-3.99	0.25-2.74	0.25-2.74	0.61-2.05
C.V	15.95	10.96	13.86	5.20	3.46	5.20	25.98	17.39	25.98	24.25	24.25	25.98	9.13	6.93	10.09	1.73	1.73	2.00
P	0.00	0.00	0.00	0.03	0.07	0.03	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.02	0.01	0.02	0.02	0.18

Table 3.7: Variations in length of hind legs (coxa, trochanter, femur, tibia, tarsus and claw) (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical Parameters	Length of coxa			Length of trochanter			Length of femur			Length of tibia			Length of Tarsus			Length of claw		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	4.50-5.51	5.00-5.50	4.00-5.00	2.01-2.50	1.50-2.51	1.51-2.01	7.00-8.01	8.51-9.50	8.01-9.00	8.51-9.50	9.50-10.50	8.01-9.50	2.50-3.50	3.00-4.00	3.00-4.00	1.00-1.50	1.50-2.00	1.00-2.00
\bar{X}	5.00	5.16	4.83	1.93	2.00	1.83	8.50	9.00	8.33	9.50	10.03	9.23	3.00	3.50	3.50	1.23	1.43	1.50
S.D	0.50	0.28	0.76	0.76	0.50	0.56	0.50	0.50	0.76	0.50	0.45	0.68	0.50	0.50	0.50	0.25	0.40	0.50
S.E	0.28	0.16	0.44	0.44	0.28	0.32	0.28	0.28	0.44	0.28	0.26	0.39	0.28	0.28	0.28	0.14	0.23	0.28
95% C.I	3.75-6.24	4.45-5.88	2.93-6.73	0.06-3.73	0.75-3.24	0.42-3.24	7.25-9.74	7.25-9.74	6.43-10.23	8.25-10.74	8.91-11.15	7.54-10.92	1.75-4.24	2.25-4.74	2.25-4.74	0.60-1.85	0.42-2.43	0.25-2.74
C.V	13.86	25.00	8.69	1.89	3.46	2.54	25.98	27.71	16.63	29.44	34.70	20.95	3.93	8.66	8.66	1.61	1.86	1.73
P	0.00	0.00	0.01	0.19	0.00	0.12	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.01	0.00	0.25	0.20	0.22

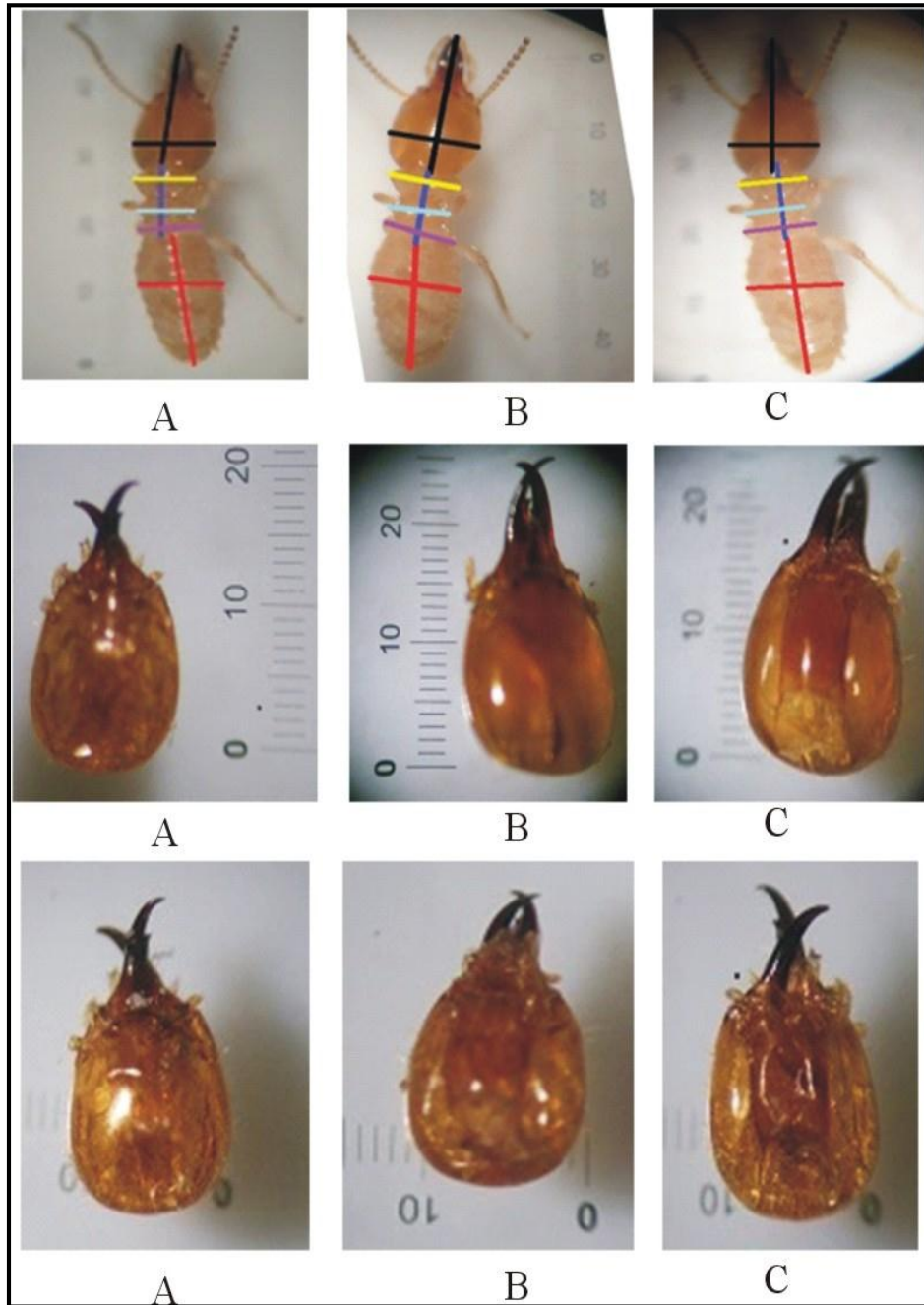


Figure 3.1: Variations in full body length, length of thorax (Prothorax, mesothorax, metathorax) (μm), length of abdomen (μm) and length from head to mandible tip (μm), length of head (μm) and width of head (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

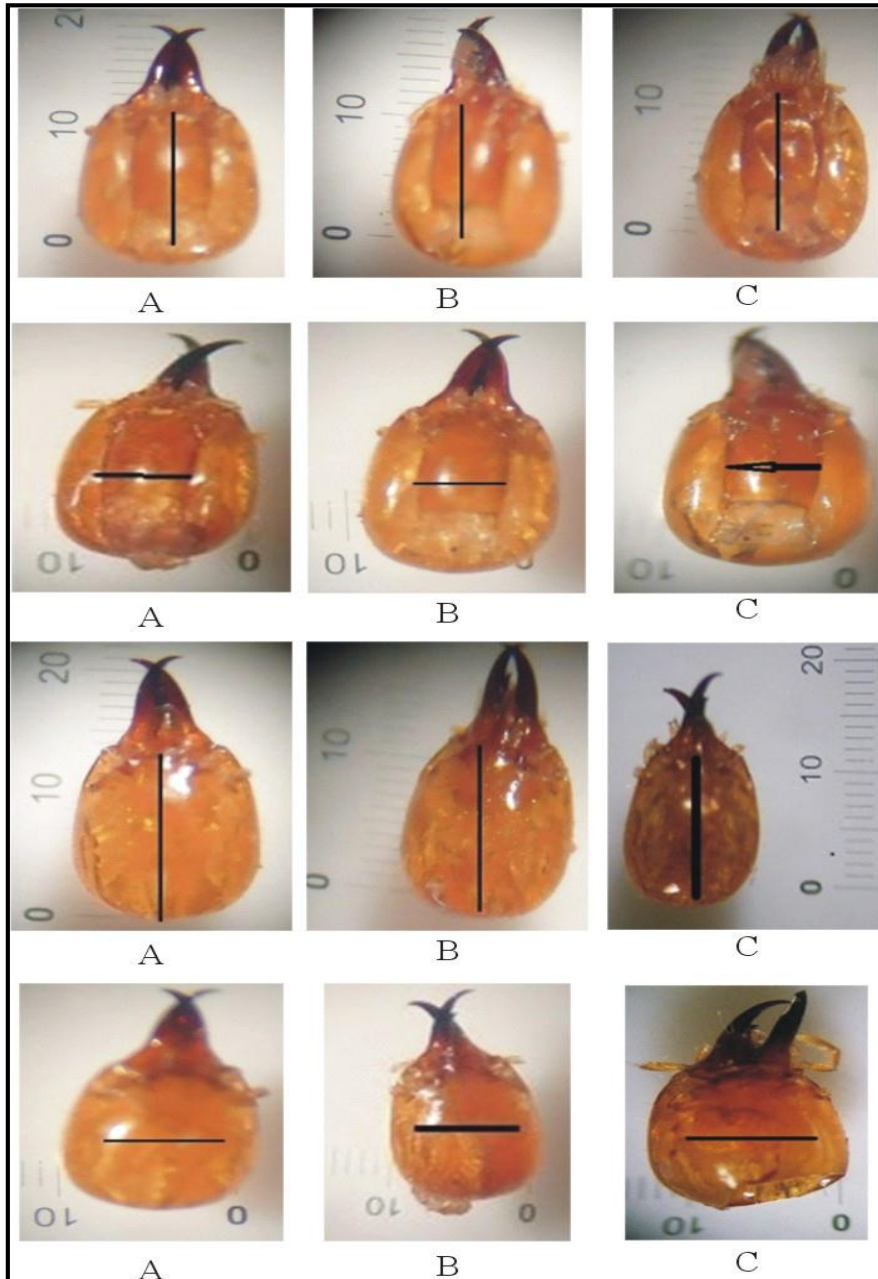


Figure 3.2: Variations in length of pronotum (μm), width of pronotum (μm), length of postmentum (μm) and width of postmentum (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

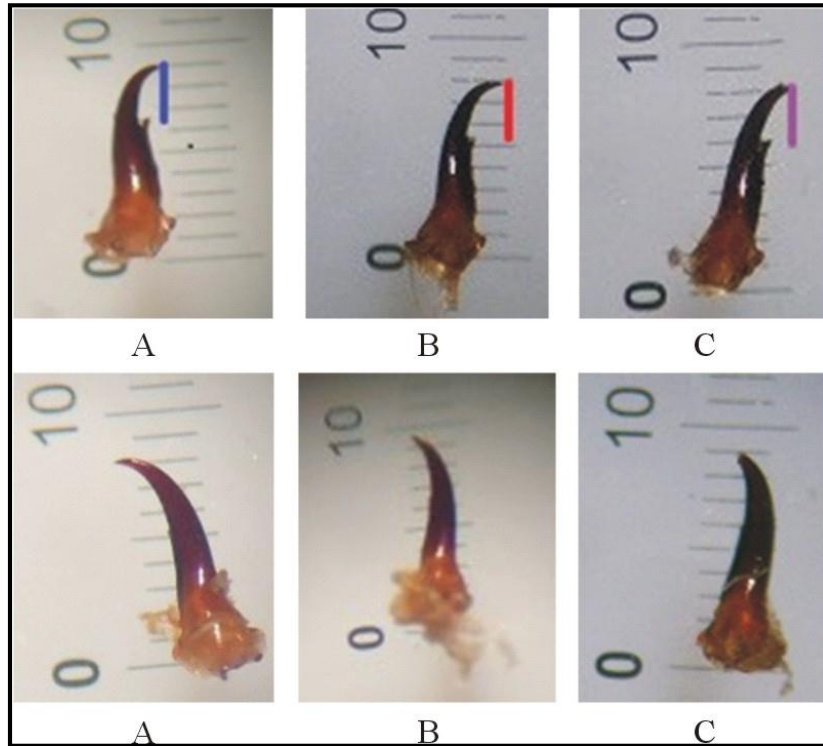


Figure 3.3: Variations in length of left mandible (μm), right mandible (μm) and length of tooth on left mandible (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

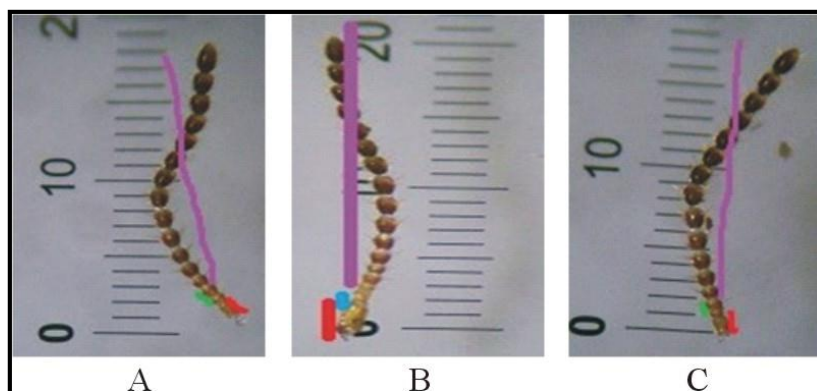


Figure 3.4: Variations in length of antenna (scape, pedicel, flagellum) (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

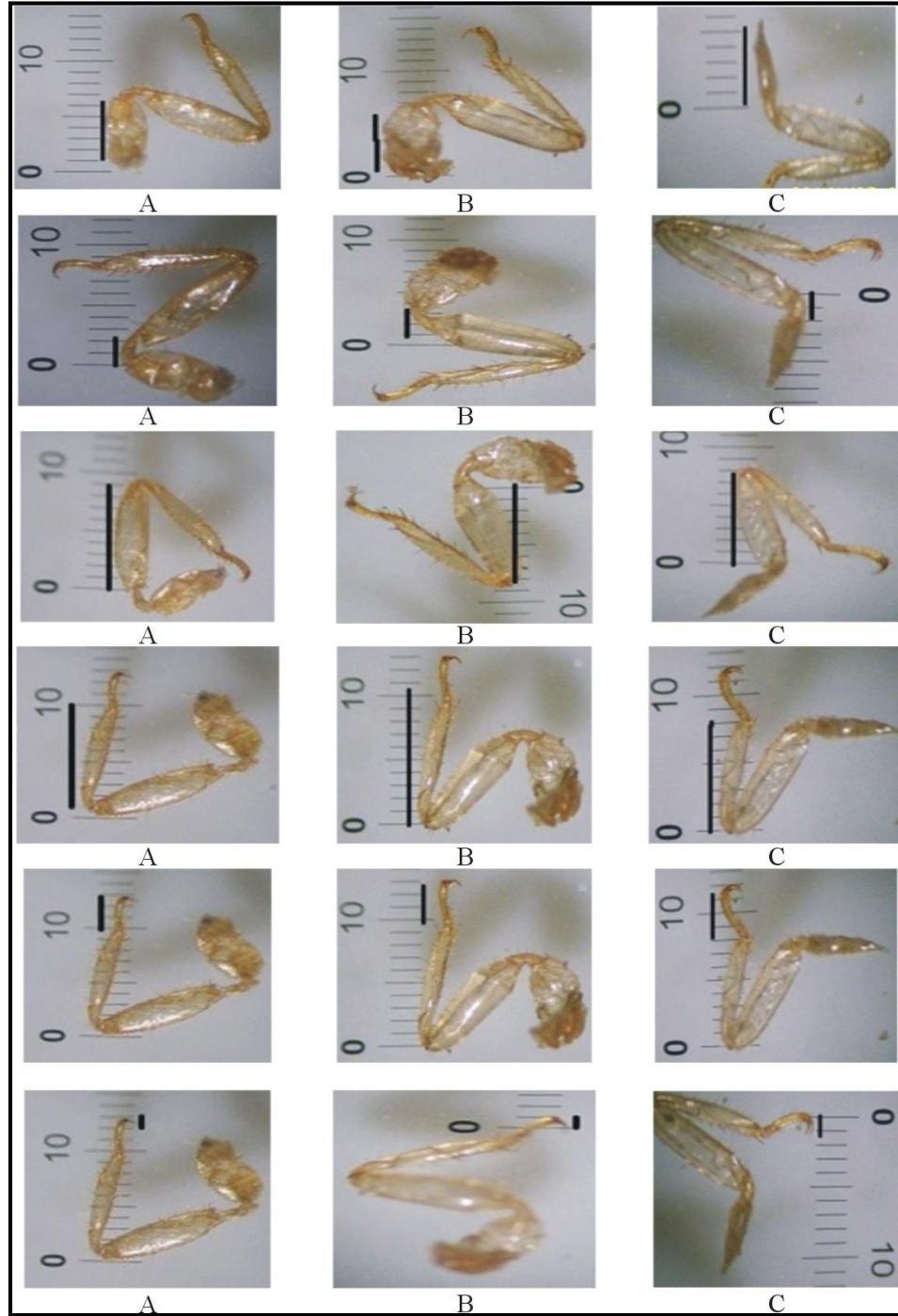


Figure 3.5: Variations in length of front leg (coxa, trochanter, femur, tibia, tarsus, claw) (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

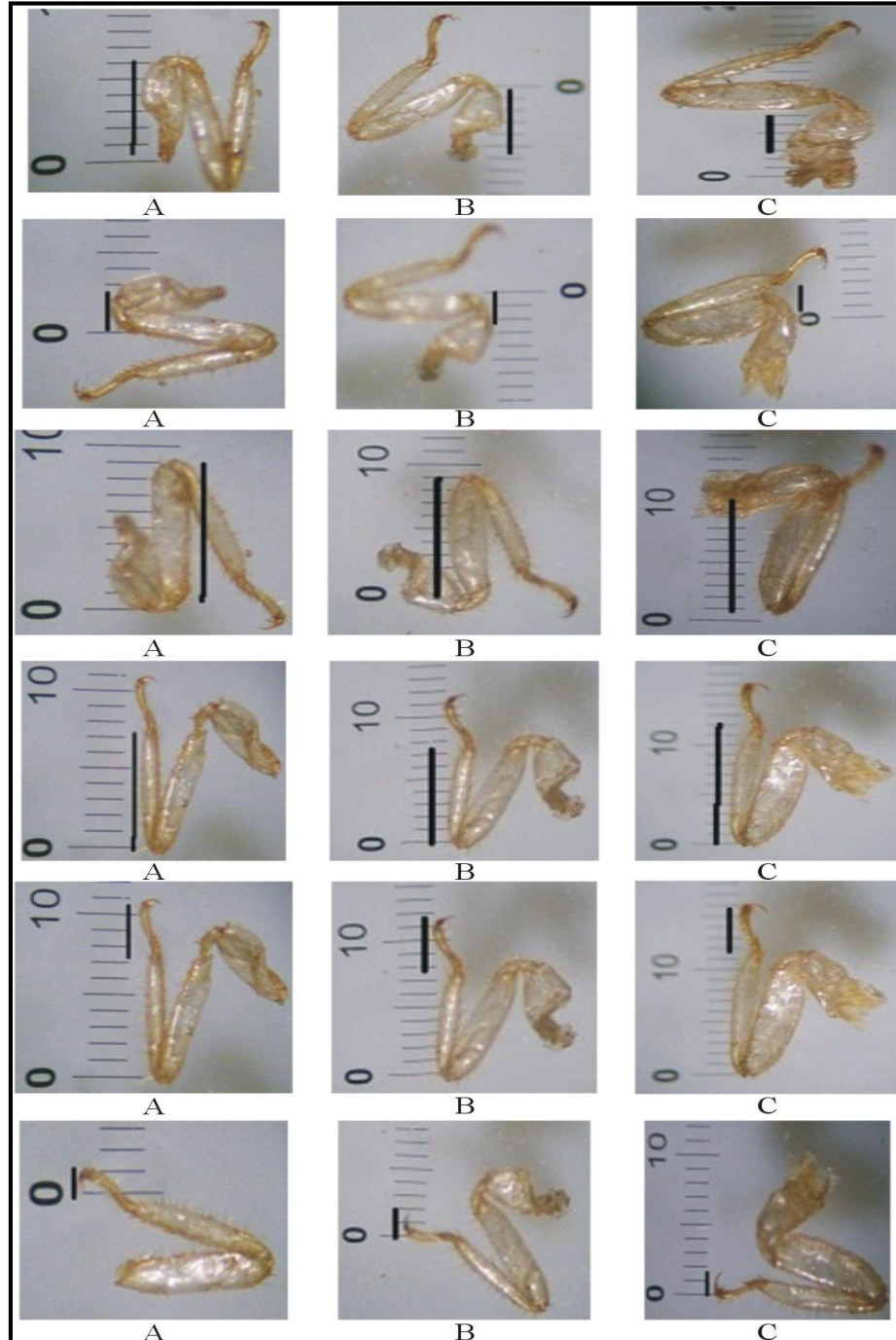


Figure 3.6: Variations in length of middle legs (coxa, trochanter, femur, tibia, tarsus, claw) (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

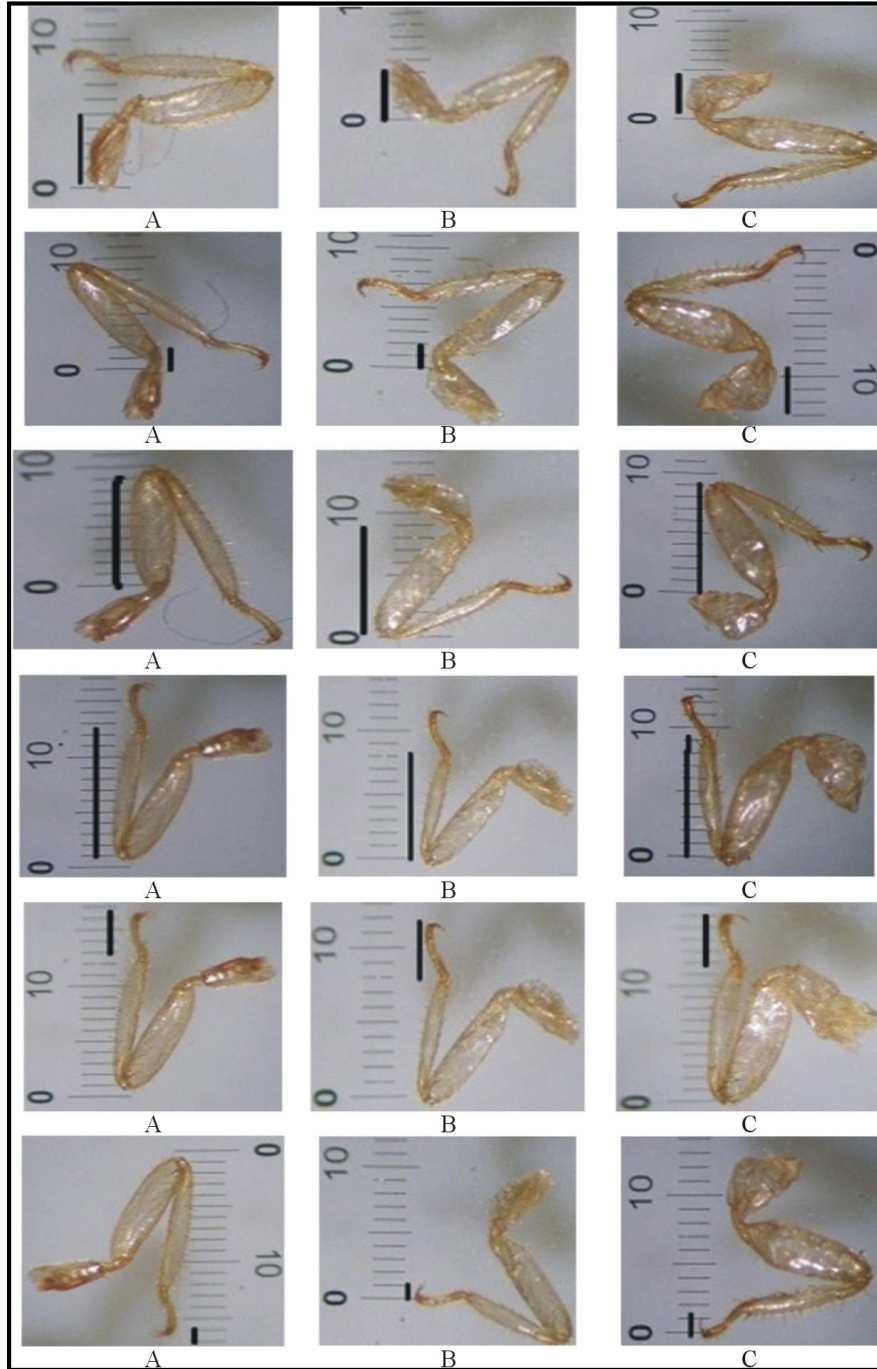


Figure 3.7: Variations in length of hind leg (coxa, trochanter, femur, tibia, tarsus, claw) (μm) of soldiers of *O. obesus* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Morphometric variations in *M. obesi*

Study of morphometric variations *M. obesi* showed that length of whole body varied from 39µm-44µm. Three population sample i.e. A, B and C had mean value 42.59µm, 40.16µm and 41.16µm. Value of coefficient of variability varied between 53.90µm-66.84µm (Table and figure 3.8). Length of abdomen varied from 15µm-22µm and had mean value 21.50µm, 17.83µm and 19.50µm. Value of coefficient of variability varied between 10.56µm-41.00µm (Table and figure 3.8). Length of Prothorax varied from 2.5µm-3.5µm and mean values 2.83µm, 3.33µm and 2.83µm. Value of coefficient of variability of sample was 11.00µm (Table and figure 3.8). Length of mesothorax varied from 2.5µm-3.5µm and mean value was 3.16µm, 3.33µm and 3.33µm. Value of coefficient of variability of sample varied between 11µm-14µm (Table and figure 3.8). Length of metathorax varied from 8.0µm-9.5µm. They had mean value of 9.00µm, 8.830µm and 8.66µm. Value of coefficient of variability of sample varied between 8.0µm-17µm (Table and figure 3.8). No significant difference was found amongst the termite samples taken from various localities.

Length from head to mandible tip varied from 15µm-22µm. The mean value was 21.50µm, 17.83µm and 19.50µm. Value of coefficient of variability of sample varied between 10.59µm-41.00µm (Table and figure 3.8). Length of head varied from 8.0µm-9.5µm and had mean value 9.0µm, 8.83µm and 8.66µm respectively. Value of coefficient of variability varied between 17.21µm-46µm (Table and figure 3.8). Width of head varied from 7.0µm-9µm. Three population sample had mean value 7.50µm, 8.33µm and 8µm. Value of coefficient of variability of varied between 22.52µm-44µm (Table and figure 3.8). No significant difference was observed in three population samples.

Length of pronotum varied from 6-9.5µm. The mean value was 7.16µm, 8.33µm and 6.8µm. Value of coefficient of variability of sample varied between 10.00µm-22.52µm (Table and figure 3.9). Width of pronotum varied from 6.0µm-9.5µm and mean value 7.16µm, 8.33µm and 6.8µm respectively. Value of coefficient of variability of

sample varied between 10.00 μm -22.52 μm (Table and figure 3.9). Length of postmentum varied from 7.0 μm -9.0 μm and had mean value 7.83 μm , 7.50 μm and 8 μm . Value of coefficient of variability of sample varied between 22.00 μm -41.00 μm (Table 3.9). Width of postmentum varied from 5.0 μm -8 μm and mean value 7.16 μm , 7.0 μm and 6.16 μm respectively. Value of coefficient of variability of sample varied between 20.00 μm -37.00 μm (Table and figure 3.9). No significant difference was present among the termite samples.

Length of right mandible varied from 5.0 μm -7 μm . Three population sample had mean value 5.83 μm , 6.00 μm and 5.83 μm respectively. Value of coefficient of variability varied between 17.00 μm -29.00 μm (Table and figure 3.10). Length of left mandible varied from 5.0 μm -7.5 μm . Three population sample i.e. A, B and C had mean value 6.88 μm , 6.34 μm and 6 μm respectively. Value of coefficient of variability of sample varied between 12.00 μm -35.00 μm (Table and figure 3.10). No significant variations were present.

Length of different segments of antenna like scape, pedicle and flagellum also exhibit small differences. Observed rang in scape was 1.0 μm -1.8 μm and mean value of 1.10 μm , 1.16 μm and 1.23 μm respectively. Value of coefficient of variability of sample varied between 1.0 μm -4 μm (Table and figure 3.11). Observed rang in pedicle was 0.5 μm -1.0 μm and mean value was 0.56 μm , 0.60 μm , and 0.56 μm . Value of coefficient of variability of sample varied between 4.0 μm -6.5 μm (Table and figure 3.11). Observed rang in flagellum was 5.0 μm -5.7 μm and mean value 0.28 μm , 0.44 μm and 0.28 μm respectively. Value of coefficient of variability varied between 3.76 μm -7.74 μm (Table and figure 3.11). No significant difference was present in different termite samples.

Length of different segments of front leg like coxa, trochanter, femur, tibia, tarsus and claw also exhibit small differences. Observed range in coxa was 3.0 μm -4.5 μm and had mean value 4.0 μm , 3.83 μm and 4.1 μm respectively. Value of coefficient of variability varied between 5.28 μm -10.39 μm (Table and figure 3.12). Observed range in trochanter was 1.0 μm -2.8 μm . Three population sample i.e. A, B and C had mean value

1.76 μm , 1.50 μm and 1.86 μm respectively. Value of coefficient of variability of sample varied between 1.78 μm -5.28 μm (Table and figure 3.12). Observed range in femur was 5.0 μm -7 μm . Three population sample i.e. A, B and C had mean value 6.5 μm , 6.0 μm and 5.8 μm respectively. Value of coefficient of variability of sample varied between 8.0 μm -19.5 μm (Table and figure 3.12). Observed range in tibia was 4.5 μm -6.0 μm . Three population sample i.e. A, B and C had mean value 5.5 μm , 5.51 μm and 5.5 μm respectively. Value of coefficient of variability of sample varied between 7.7 μm -15.9 μm (Table and figure 3.12). Observed range in tarsus was 1.0 μm -1.8 μm . Three population sample i.e. A, B and C had mean value 1.43 μm , 1.43 μm and 1.26 μm respectively. Value of coefficient of variability of three population sample varied between 1.84 μm -1.86 μm (Table and figure 3.12). Observed range in Claw was 0.3 μm -0.8 μm . Three population sample i.e. A, B and C had mean value 0.56 μm , 0.60 μm and 0.43 μm respectively (Table and figure 3.12). Value of coefficient of variability of three population sample varied between 3.61 μm -8.5 μm . No significant difference was found amongst the termite samples taken from various localities.

Length of different segments of middle leg like coxa, trochanter, femur, tibia, tarsus and claw also exhibit small differences. Observed range in coxa was 3.0 μm -4.5 μm . Three population sample i.e. A, B and C had mean value 4.00 μm , 3.83 μm and 4.1 μm respectively. Value of coefficient of variability of sample varied between 6.4 μm -10.39 μm (Table and figure 3.13). Observed range in trochanter was 1.0 μm -2.8 μm . Three population sample i.e. A, B and C had mean value 1.76 μm , 1.50 μm and 1.86 μm respectively. Value of coefficient of variability of sample varied between 1.78 μm -5.28 μm (Table and figure 3.13). Observed range in femur was 5.0 μm -7.0 μm . Three population sample i.e. A, B and C had mean value 5.8 μm , 6.0 μm and 6.5 μm respectively. Value of coefficient of variability of three population sample varied between 8.04 μm -19.05 μm (Table and figure 3.13). Observed range in tibia was 4.5 μm -6.0 μm . Three population sample i.e. A, B and C had mean value 5.50 μm , 5.51 μm and 5.50 μm respectively. Value of coefficient of variability of sample varied between 7.79 μm -15.59 μm (Table and figure 3.13). No significant difference was found amongst the

termite samples taken from various localities. Observed range in tarsus was 1.0 μ m-1.8 μ m. Three population sample i.e. A, B and C had mean value 1.42 μ m, 1.43 μ m and 1.26 μ m respectively. Value of coefficient of variability of sample varied between 1.84 μ m-1.86 μ m (Table and figure 3.13). No significant difference was found amongst the termite samples taken from various localities. Observed range in Claw was 0.3 μ m-0.8 μ m. Three population sample i.e. A, B and C had mean value 0.56 μ m, 0.60 μ m and 0.43 μ m respectively. Value of coefficient of variability of sample varied between 3.61 μ m-8.50 μ m (Table and figure 3.13). No significant difference was present.

Length of different segments of hind leg like coxa, trochanter, femur, tibia, tarsus and claw also exhibit small differences. Observed range in coxa was 3.5 μ m-5.0 μ m. Three population sample i.e. A, B and C had mean value 4.0 μ m, 4.10 μ m and 4.50 μ m respectively. Value of coefficient of variability of sample varied between 10.39 μ m-14.89 μ m (Table and figure 3.14). Observed range in trochanter was 1.8 μ m-2.8 μ m. Three population sample i.e. A, B and C had mean value 2.16 μ m, 2.10 μ m and 2.43 μ m respectively. Value of coefficient of variability of sample varied between 5.28 μ m-7.00 μ m (Table and figure 3.14). Observed range in femur was 4.0 μ m-5.5 μ m. Three population sample i.e. A, B and C had mean value 5.0 μ m, 4.6 μ m and 5.16 μ m respectively. Value of coefficient of variability of sample varied between 9.45 μ m-13.80 μ m (Table and figure 3.14). Observed range in tibia was 8.0 μ m-9.5 μ m. Three population sample i.e. A, B and C had mean value 9.0 μ m, 8.8 μ m and 8.50 μ m respectively. Value of coefficient of variability of sample varied between 17.76 μ m-27.71 μ m (Table and figure 3.14). Observed range in tarsus was 1.0 μ m-2.8 μ m. Three population sample i.e. A, B and C had mean value 2.12 μ m, 2.10 μ m and 2.43 μ m respectively. Value of coefficient of variability of sample varied between 3.46 μ m-6.41 μ m (Table and figure 3.14). Analysis of variance showed that there were non-significant differences among the samples collected from different localities.

Morphometric analysis of *M. obesi*

Table 3.8: Variations in full body length, length of thorax (Prothorax, mesothorax, metathorax) (μm) and length of abdomen (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical parameters	Full Body length			Length of abdomen			Length of prothorax			Length of mesothorax			Length of metathorax		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	41.50-44.00	39.00-41.51	40.00-42.00	20.50-22.00	15.00-20.00	18.50-21.00	2.50-3.00	2.50-3.50	2.50-3.00	3.00-3.51	2.50-3.50	2.50-3.50	3.50-4.00	2.50-3.50	2.50-3.50
\bar{X}	42.50	40.16	41.16	21.50	17.83	19.50	2.83	3.33	2.83	3.16	3.33	3.33	3.83	3.33	3.33
S.D	1.32	1.25	1.04	0.86	2.57	1.32	0.28	0.16	0.28	0.28	0.28	0.28	0.28	0.50	0.28
S.E	0.76	0.72	0.62	0.50	1.59	0.76	0.16	0.16	0.16	0.16	0.16	0.16	0.16	0.16	0.16
95% C.I	39.21-45.78	37.01-42.29	38.58-43.29	19.34-23.65	10.99-24.67	16.21-22.78	2.11-3.50	2.11-3.50	2.11-3.50	2.45-3.88	2.61-4.05	2.61-4.05	3.11-4.55	2.25-4.74	2.95-4.38
C.V	54.34	53.91	66.84	41.00	10.56	24.22	11.00	11.00	11.00	13.00	11.00	14.0	17.0	8.66	16.00
P	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.00

Table 3.9: Variations in length from head to mandible tip (μm), length of head (μm), width of head (μm), length of pronotum (μm) and width of pronotum (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical parameters	Length from head to mandible tip			Length of head			Width of head			Length of pronotum			Width of pronotum		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	11.00-13.01	11.00-14.00	12.00-13.50	8.50-9.50	8.51-9.51	8.00-9.00	7.00-8.00	8.00-8.50	7.00-9.00	6.50-7.50	8.50-9.50	6.00-7.50	6.50-7.50	8.51-9.51	6.00-7.51
\bar{X}	12.00	12.67	12.88	9.01	8.83	8.66	7.50	8.33	8.00	7.16	8.33	6.83	7.16	8.33	6.83
S.D	1.00	1.52	0.76	0.50	0.57	0.28	0.50	0.28	0.50	0.50	0.57	0.76	0.50	0.57	0.76
S.E	0.57	0.82	0.44	0.28	0.33	0.16	0.28	0.16	0.28	0.28	0.28	0.44	0.28	0.28	0.44
95% C.I	9.51-14.84	10.90-14.73	2.95-4.38	7.75-10.24	7.39-10.26	7.95-9.38	6.25-8.74	7.61-9.05	6.75-9.25	6.25-8.74	2.75-5.24	4.93-8.73	6.25-8.74	2.75-5.24	4.93-8.73
C.V	19.05	13.23	26.84	17.27	23.50	46.00	22.52	44.00	24.25	22.52	10.39	3.23	22.52	10.39	3.23
P	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.06

Table 3.10: Variations in length of postmentum (μm), width of postmentum (μm), length of right mandible (μm), length of left mandible (μm) and length of tooth on left mandible (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical Parameters	Length of postmentum			Width of postmentum			Length of right mandible			Length of left mandible		
	Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3
O.R	7.51-9.00	7.00-8.50	7.00-9.01	6.50-7.50	6.00-8.00	5.50-7.00	5.50-6.51	5.00-7.00	5.50-6.51	6.00-7.51	5.50-7.00	5.00-7.00
\bar{X}	7.83	7.51	8.01	7.16	7.00	6.16	5.83	6.00	5.83	6.88	6.33	6.00
S.D	0.28	0.50	0.50	0.28	0.50	0.28	0.28	0.50	0.28	0.28	0.67	0.50
S.E	0.16	0.28	0.28	0.16	0.28	0.16	0.16	0.82	0.16	0.16	0.44	0.28
95% C.I	7.11-8.55	6.28-8.74	6.75-9.24	6.45-7.88	6.75-8.24	6.45-7.88	5.11-6.55	4.75-7.24	5.11-6.55	6.16-7.55	4.43-8.83	4.75-7.24
C.V	41.00	82.52	24.25	37.00	20.78	37.01	29.00	17.32	29.00	35.01	12.09	17.32
P	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

Table 3.11: Variations in length of antenna (scape, pedicle, flagellum) (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical parameters	Length of antenna								
	Scape			Pedicle			Flagellum		
	Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3
O.R	1.00-1.50	1.01-1.81	1.00-1.50	0.50-0.80	0.50-1.00	0.50-0.80	5.01-6.01	5.50-7.00	6.00-7.50
\bar{X}	1.10	1.16	1.23	0.56	0.60	0.56	5.50	5.60	0.55
S.D	0.17	0.28	0.25	0.11	0.17	0.11	0.50	0.76	0.50
S.E	0.10	0.16	0.14	0.06	0.10	0.06	0.28	0.44	0.28
95% C.I	0.67-1.53	0.45-1.88	0.60-1.85	0.25-0.85	0.17-1.03	0.27-0.85	4.25-4.74	3.76-7.56	5.25-7.74
C.V	1.00	4.01	1.60	6.50	4.00	6.50	15.95	10.58	19.05
P	0.42	0.42	0.20	0.02	0.05	0.03	0.00	0.00	0.00

Table 3.12: Variations in length of front legs (coxa, trochanter, femur, tibia, tarsus and claw) (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical parameters	Length of coxa			Length of trochanter			Length of femur			Length of tibia			Length of Tarsus			Length of claw		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	3.00- 3.50	4.01- 5.01	3.50- 4.50	1.50- 2.50	2.00- 2.80	1.80- 2.50	5.80- 6.50	6.00- 6.50	5.30- 6.50	4.00- 4.50	3.50- 4.50	3.60- 4.30	2.00- 3.00	2.00- 2.50	2.50- 3.50	0.30- 1.00	0.50- 1.00	0.50- 1.00
\bar{X}	4.00	4.50	3.83	1.83	2.43	2.10	6.00	6.00	5.93	4.00	4.00	3.96	2.66	2.16	3.00	0.90	0.66	0.66
S.D	0.50	0.50	0.76	0.28	0.40	0.36	0.50	0.50	0.60	0.50	0.50	0.35	0.26	0.28	0.50	0.36	0.28	0.28
S.E	0.28	0.28	0.44	0.16	0.23	0.20	0.28	0.28	0.34	0.28	0.28	0.20	0.16	0.16	0.21	0.20	0.16	0.16
95% C.I	2.75- 5.24	3.25- 5.74	1.93- 5.73	1.11- 2.55	1.42- 3.43	1.20- 2.99	2.75- 5.24	4.75- 7.24	4.34- 7.43	2.75- 5.24	2.75- 5.24	3.09- 4.83	1.95- 3.38	1.45- 2.88	1.75- 4.24	0.00- 1.79	0.05- 1.38	0.05- 1.38
C.V	10.39	12.12	6.43	5.00	6.14	5.28	17.32	17.32	14.18	10.39	10.39	14.61	10.00	7.0	6.93	0.48	2.00	2.00
P	0.00	0.00	0.02	0.03	0.02	0.03	0.00	0.00	0.00	0.00	0.00	0.00	0.001	0.02	0.02	0.67	0.18	0.00

Table 3.13: Variations in length of middle legs (coxa, trochanter, femur, tibia, tarsus and claw) (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical Parameters	Length of coxa			Length of trochanter			Length of femur			Length of tibia			Length of Tarsus			Length of claw		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	3.50-4.50	3.00-4.51	3.51-4.51	1.50-2.00	1.00-2.01	1.50-2.81	6.50-7.00	5.5-6.5	5.00-6.50	5.00-6.00	5.00-6.00	4.50-6.01	1.00-1.80	1.00-1.81	1.00-1.50	0.50-0.50	0.50-8.80	0.30-0.60
\bar{X}	4.00	3.83	4.00	1.76	1.50	1.87	6.50	6.00	5.83	5.50	5.50	5.50	1.43	1.43	1.26	0.56	0.60	0.43
S.D	0.50	0.76	0.50	0.25	0.50	0.40	0.50	0.50	1.04	0.50	0.50	1.00	0.40	0.40	0.25	0.20	0.17	0.11
S.E	0.28	0.44	0.28	0.14	0.28	0.23	0.28	0.28	0.60	0.28	0.28	0.57	0.23	0.23	0.14	0.12	0.10	0.06
95% C.I	2.75-5.24	1.93-5.73	2.75-5,24	1.14-2.39	0.25-2.74	0.86-2.87	5.25-7.74	4.75-7.24	3.24-8.41	4.25-6.74	4.25-6.74	3.01-7.98	0.42-2.43	0.42-2.43	0.64-1.89	0.05-1.08	0.17-1.03	0.14-0.72
C.V	10.39	6.43	10.39	5.28	1.73	3.71	19.05	17.23	0.4	15.95	15.59	7.79	1.86	1.86	1.84	3.61	4.00	8.50
P	0.00	0.02	0.00	0.03	0.22	0.06	0.00	0.00	0.01	0.00	0.00	0.01	0.20	0.20	0.20	0.06	0.05	0.01

Table 3.14: Variations in length of hind legs (coxa, trochanter, femur, tibia, tarsus and claw) (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Statistical Parameters	Length of coxa			Length of trochanter			Length of femur			Length of tibia			Length of Tarsus			Length of claw		
	Sample sites			Sample sites			Sample sites			Sample sites			Sample sites			Sample sites		
	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C	A	B	C
N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O.R	3.50-4.50	3.80-4.50	3.50-5.00	2.00-2.50	1.80-2.50	2.00-2.80	4.50-5.50	4.50-5.30	4.00-4.50	8.85-9.50	9.00-9.50	8.00-9.00	1.50-2.50	1.00-2.50	2.00-2.80	0.50-0.70	0.50-0.80	0.30-0.70
\bar{X}	4.00	4.10	4.50	2.16	2.10	2.43	5.00	4.60	5.16	9.00	8.83	8.50	2.00	2.10	2.43	0.56	0.46	0.50
S.D	0.50	0.36	0.50	0.28	0.36	0.40	0.50	0.65	0.76	0.50	0.76	0.50	0.50	0.36	0.40	0.11	0.15	0.20
S.E	0.28	0.20	0.28	0.16	0.20	0.23	0.28	0.37	0.44	0.28	0.44	0.28	0.28	0.20	0.23	0.06	0.08	0.11
95% C.I	2.75-5.24	3.20-4.99	3.25-5.74	1.45-2.88	1.20-2.99	1.42-3.43	3.75-6.24	2.97-3.22	3.26-7.06	7.75-10.24	6.93-10.73	7.25-9.74	0.74-3.24	1.20-2.99	1.42-3.43	0.27-0.85	0.08-0.84	0.03-0.90
C.V	10.39	14.89	12.22	7.00	5.28	6.14	13.86	9.51	9.45	27.71	17.76	20.98	3.46	5.28	6.14	6.50	6.05	4.33
P	0.00	0.00	0.00	0.02	0.03	0.02	0.00	0.01	0.01	0.00	0.00	0.00	0.07	0.03	0.02	0.02	0.02	0.04

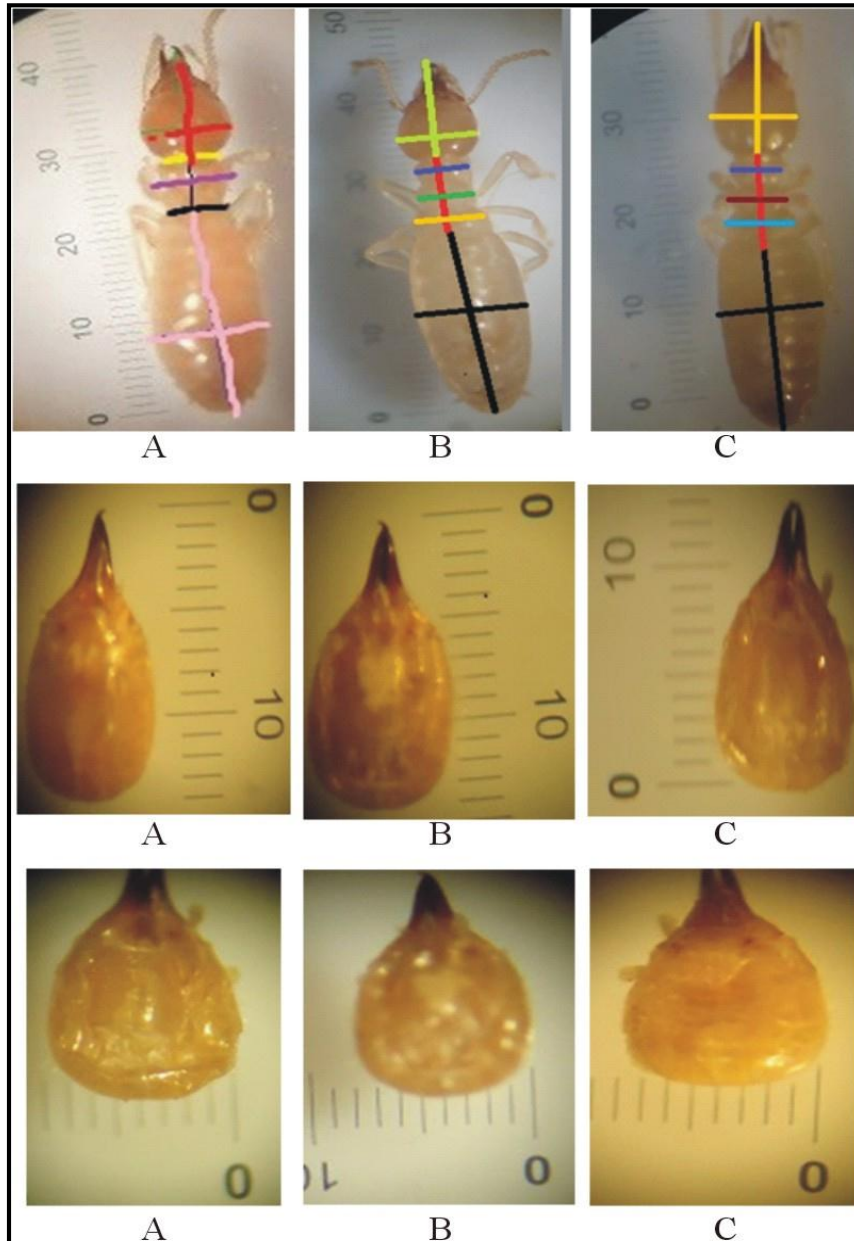


Figure 3.8: Variations in full body length, length of thorax (Prothorax, mesothorax, metathorax)(μm), length of abdomen(μm) and length of head(μm) and width of head (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan)

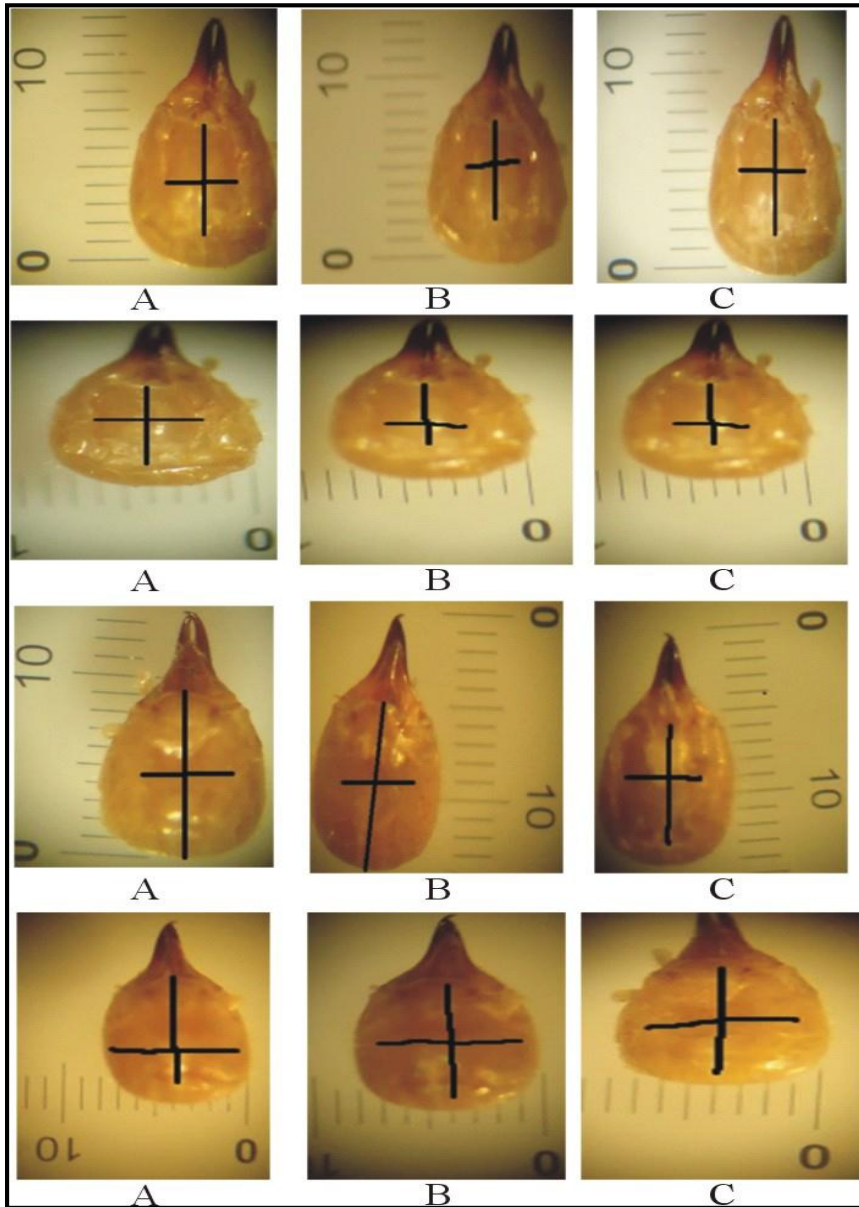


Figure 3.9: Variations in length of pronotum(μm), width of pronotum(μm), length of postmentum(μm) and width of postmentum(μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan)

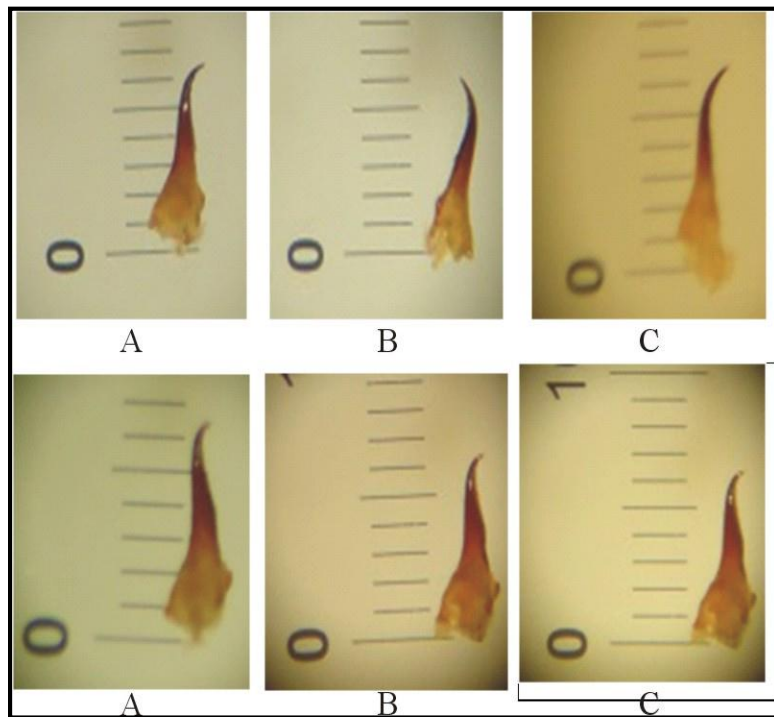


Figure 3.10: Variations in length of left and right mandible(μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

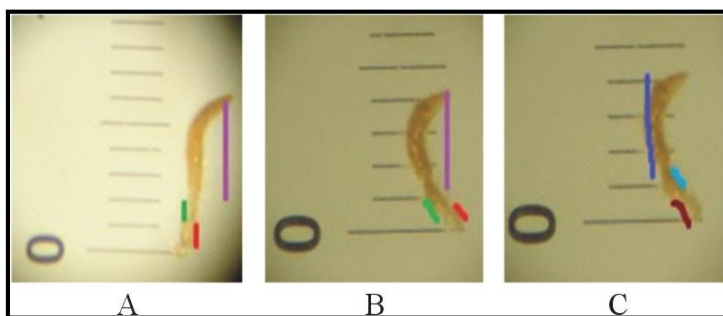


Figure 3.11: Variations in length antenna (scape, pedicel and flagellum) (μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

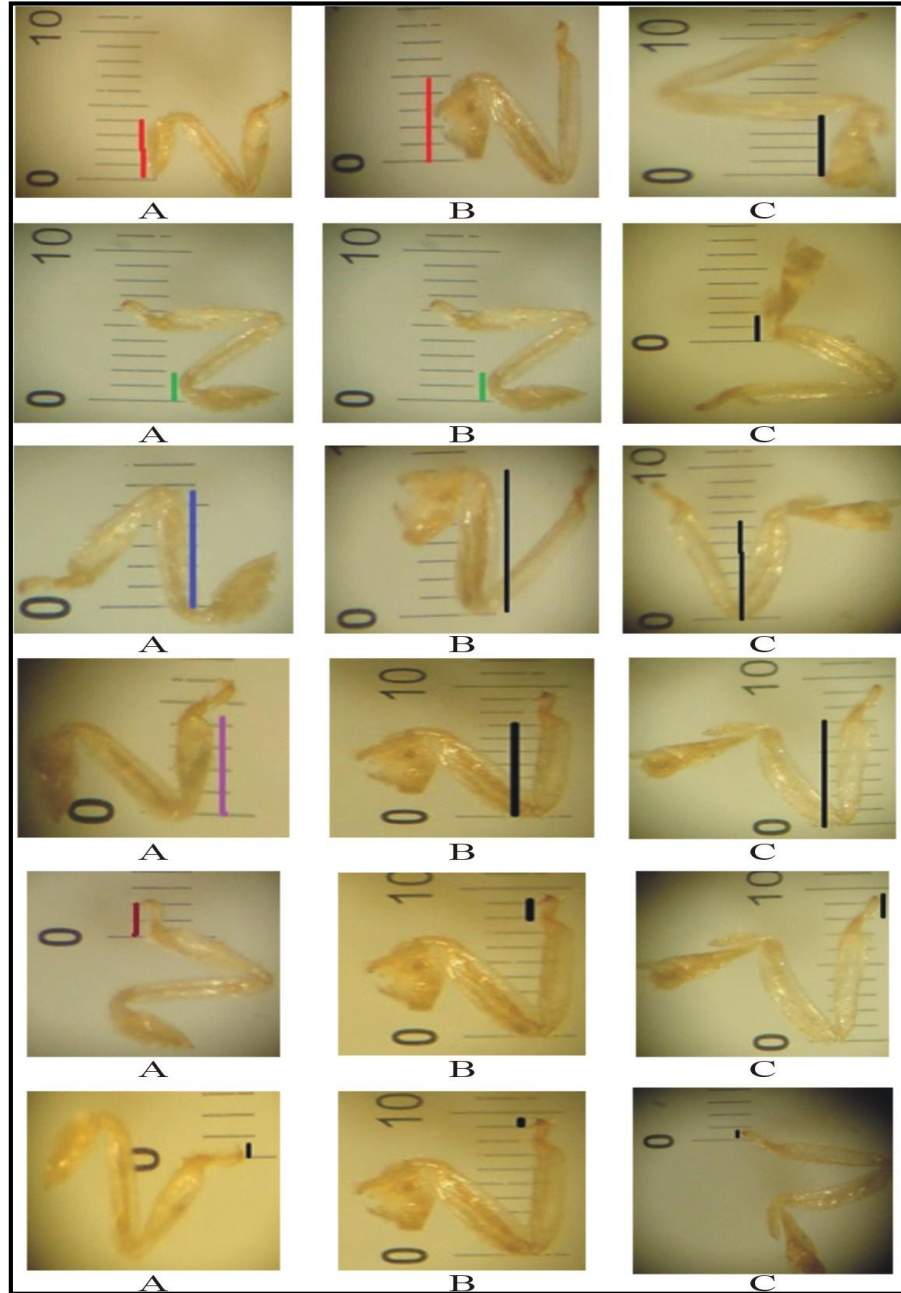


Figure 3.12: Variations in length of front leg (coxa, trochanter, femur, tibia, tarsus and claw)(μm) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

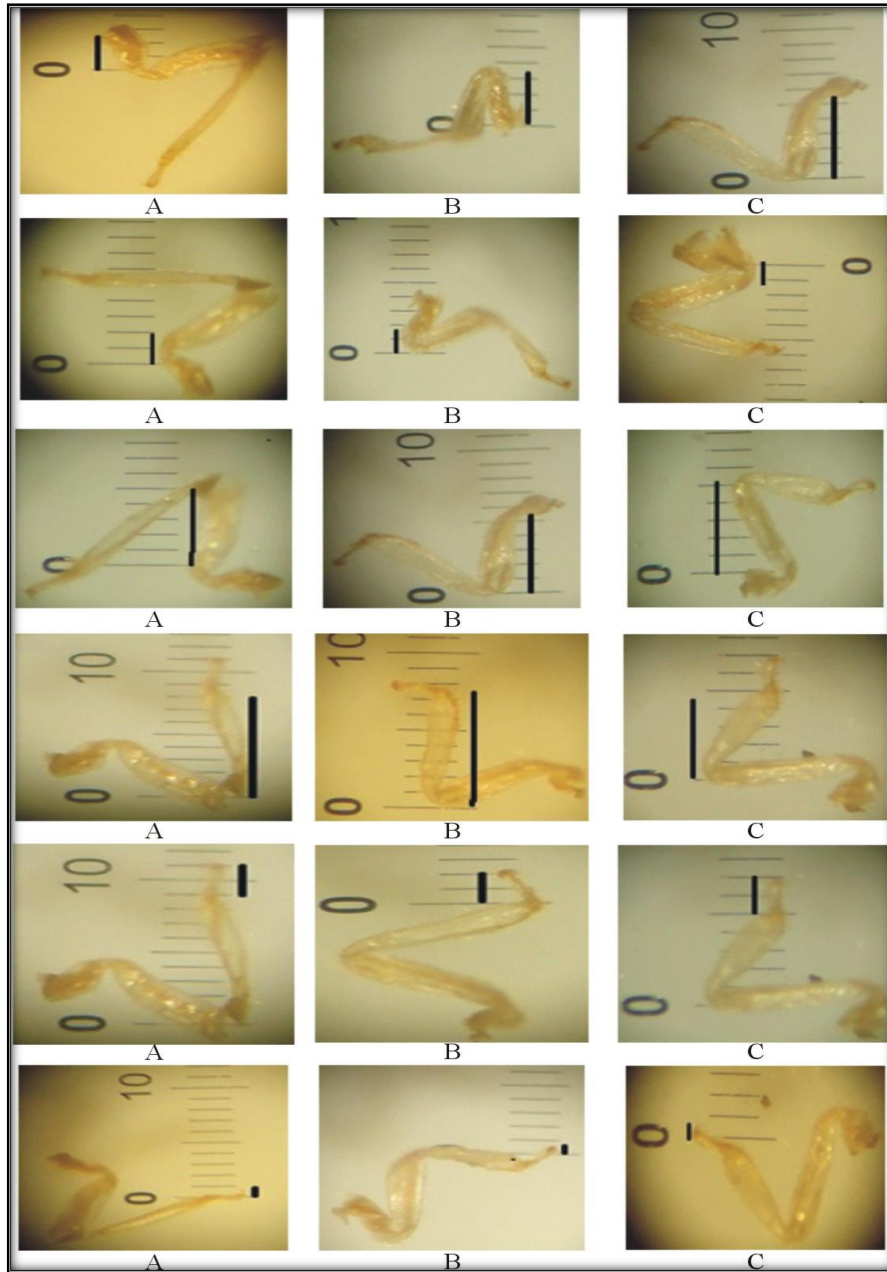


Figure 3.13: Variations in length of middle leg (coxa, trochanter, femur, tibia, tarsus and claw) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

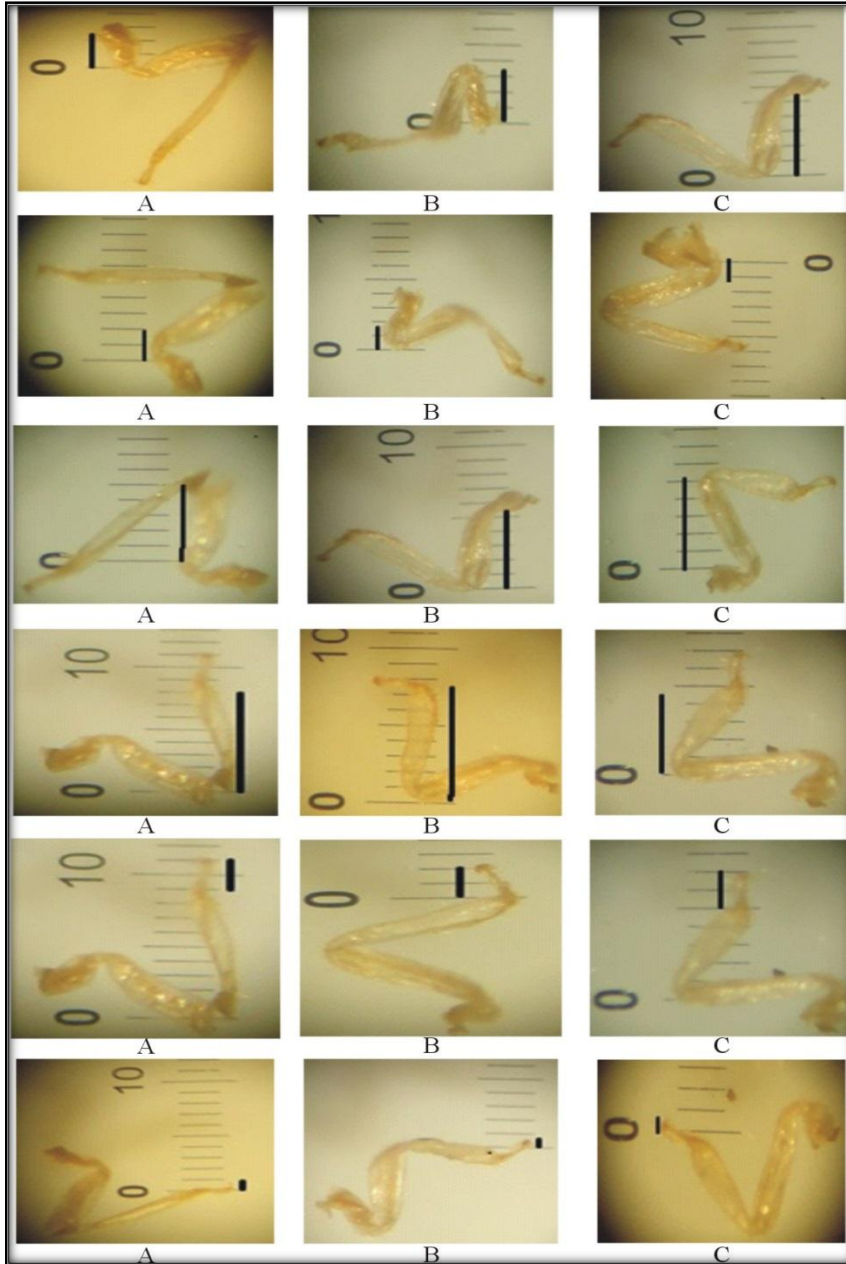


Figure 3.14: Variations in length of hind leg (coxa, trochanter, femur, tibia, tarsus and claw) of soldiers of *M. obesi* from site A (Murree), site B (Rawalpindi) and site C (Gujarkhan).

Effect of plants extract

Effect of *M. azedarach* leaves extract on *O. obesus* in water solvent:

Water soluble leaves extract of *M. azedarach* showed 7.50 ± 0.57 , 18.33 ± 0.88 , 10.20 ± 1.76 percent mortality by feeding on 100 ppm, 200 ppm and 300 ppm respectively on first day of experiment where as in control no significant mortality was observed i.e. 3.33 ± 0.33 . LC_{50} value was 5.64% and value of LC_{90} was 28.7%. On 2nd day of experiment percent mortality rate on same concentration (100 ppm, 200 ppm and 300 ppm) was 50 ± 0.24 , 69.16 ± 1.20 and 73.33 ± 1.86 while in control observed mortality was 4.16 ± 0.33 which was non significant. LC_{50} value was 0.009% and value of LC_{90} was 0.084%. Maximum mortality rate was observed on 3rd day of experiment at all above mentioned concentrations which was 93.33 ± 1.76 , 96.66 ± 0.33 , 99.16 ± 0.33 and non significant mortality rate of control was 7.5 ± 0.58 observed. LC_{50} value was 0.002 and value of LC_{90} was 0.007%. (Table and figure 3.15)

Effect of *M. azedarach* leaves extract on *O. obesus* in methanol solvent:

Leaves extract of *M. azedarach* in methanol solvent showed 13.33 ± 1.20 , 18.33 ± 1.0 and 22.5 ± 1.53 percent mortality by feeding on 100 ppm, 200 ppm and 300 ppm respectively on first day of experiment where as in control no significant mortality was observed i.e. 3.33 ± 0.88 . LC_{50} value was 0.31% and value of LC_{90} was 17.01%. On 2nd day of experiment percent mortality rate on same concentration (100 ppm, 200 ppm and 300 ppm) was 59.16 ± 1.02 , 71.66 ± 1.20 and 80.88 ± 1.76 while in control observed mortality was 5.83 ± 0.33 which was non significant. LC_{50} value was 0.006% and value of LC_{90} was 0.064%. Maximum mortality rate was observed on 3rd day of experiment at all concentrations which was 95.83 ± 1.50 , 97.5 ± 1.53 and 99.16 ± 1.20 and mortality rate of control was 8.33 ± 0.57 which was non significant. LC_{50} value was 0.0003% and value of LC_{90} was 0.004%. (Table and figure 3.16)

Effect of *E. camaldulensis* leaves extract on *O. obesus* in water solvent:

Water soluble leaves extract of *E. camaldulensis* showed 12.5 ± 1.15 , 15 ± 1.52 , 26.66 ± 1.20 percent mortality by feeding on 100 ppm, 200 ppm and 300 ppm respectively on first day of experiment where as in control no significant mortality was observed i.e. 2.5 ± 0.573 . LC_{50} value was 0.133% and value of LC_{90} was 2.04%. On 2nd day of experiment percent mortality rate on same concentration (100 ppm, 200 ppm and 300 ppm) was 56.66 ± 0.84 , 71.66 ± 2.40 and 81.66 ± 1.85 while in control observed mortality was 5.00 ± 0.57 which was non significant. LC_{50} value was 0.007% and value of LC_{90} was 0.05%. Maximum mortality rate was observed on 3rd day of experiment at all above mentioned concentrations which was 93.33 ± 1.45 , 97.5 ± 1.00 , 100 ± 0 and mortality rate of control was 10.00 ± 0.57 which was not significant. LC_{50} value was 0.002 and value of LC_{90} was 0.008%. (Table and figure 3.17)

Effect of *E. camaldulensis* leaves extract on *O. obesus* in methanol solvent:

Leaves extract of *E. camaldulensis* in methanol solvent showed 25 ± 1.15 , 34.16 ± 1.40 and 35.83 ± 0.33 percent mortality by feeding on 100 ppm, 200 ppm and 300 ppm respectively on first day of experiment where as in control no significant mortality was observed i.e. 3.33 ± 0.88 . LC_{50} value was 0.049% and value of LC_{90} was 7.57%. On 2nd day of experiment percent mortality rate on same concentration (100 ppm, 200 ppm and 300 ppm) was 72.5 ± 0.78 , 71.66 ± 1.85 and 74.16 ± 1.76 while in control observed mortality was 6.66 ± 0.88 which was non significant. LC_{50} value was 0.006% and value of LC_{90} was 0.056%. Maximum mortality rate was observed on 3rd day of experiment at all above mentioned concentrations which was 97.5 ± 0.57 , 95.83 ± 1.66 and 100 ± 0 and mortality rate of control was 9.16 ± 0.88 which was non significant. LC_{50} value was 0.008% and value of LC_{90} was 0.004%. (Table and figure 3.18)

Effect of *M. azedarach* leaves extract on *M. obesi* in water solvent:

Water soluble leaves extract of *M. azedarach* showed 20.83 ± 1.20 , 25.83 ± 1.45 and 42.5 ± 1.73 percent mortality by feeding on 100 ppm, 200 ppm and 300 ppm respectively

on first day of experiment where as in control no significant mortality was observed i.e. 4.16 ± 0.88 . LC_{50} value was 0.049% and value of LC_{90} was 0.515%. On 2nd day of experiment percent mortality rate on same concentration (100 ppm, 200 ppm and 300ppm) was 41.66 ± 1.20 , 45.83 ± 0.88 and 65.83 ± 0.4 while in control observed mortality was 9.16 ± 0.88 which was not significant. LC_{50} value was 0.017% and value of LC_{90} was 0.206%. Maximum mortality rate was observed on 3rd day of experiment at all above mentioned concentrations which was 91.66 ± 1.02 , 93.33 ± 0.88 and 99.16 ± 0.33 and mortality rate of control was 12.50 ± 0.57 which was not significant. LC_{50} value was 0.001% and value of LC_{90} was 0.009%. (Table and figure 3.19)

Effect of *M. azedarach* leaves extract on *M. obesi* in methanol solvent:

Leaves extract of *M. azedarach* in methanol solvent showed 26.5 ± 0.88 , 37.33 ± 1.73 and 41.66 ± 1.45 percent mortality by feeding on 100 ppm, 200 ppm and 300ppm respectively on first day of experiment where as in control no significant mortality was observed i.e. 3.33 ± 0.88 . LC_{50} value was 0.049% and value of LC_{90} was 1.38%. On 2nd day of experiment percent mortality rate on same concentration (100 ppm, 200 ppm and 300ppm) was 53.33 ± 1.85 , 68.33 ± 1.45 and 78.33 ± 0.85 while in control observed mortality was 8.33 ± 1.20 which is nonsignificant. LC_{50} value was 0.008% and value of LC_{90} was 0.068%. Maximum mortality rate was observed on 3rd day of experiment at all above mentioned concentrations which was 92.5 ± 1.15 , 97.5 ± 0.57 and 100 ± 0.00 respectively and a non significant mortality rate of control was observed i.e. 10.00 ± 1.15 . LC_{50} value was 0.002% and value of LC_{90} was 0.008%. (Table and figure 3.20)

Effect of *E. camaldulensis* leaves extract on *M. obesi* in water solvent:

Water soluble leaves extract of *E. camaldulensis* showed 57.5 ± 0.05 , 51.66 ± 0.88 and 66.66 ± 0.88 percent mortality by feeding on 100 ppm, 200 ppm and 300ppm respectively on first day of experiment where as in control no significant mortality was observed i.e. 0.83 ± 0.33 . LC_{50} value was 0.013% and value of LC_{90} was 0.292%. On 2nd day of experiment percent mortality rate on same concentration (100 ppm, 200 ppm and

300ppm) was 69.166 ± 1.20 , 76.66 ± 0.88 and 81.66 ± 1.76 while in control observed mortality was 3.83 ± 0.33 which was not significant. LC_{50} value was 0.002% and value of LC_{90} was 0.088%. Maximum mortality rate was observed on 3rd day of experiment at all above mentioned concentrations which was 95 ± 1.52 , 96.66 ± 0.66 and 100 ± 0.00 and mortality rate of control was 5.83 ± 0.33 i.e. non significant. LC_{50} value was 9.42 and value of LC_{90} was 0.006%. (Table and figure 3.21)

Effect of *E. camaldulensis* leaves extract on *M. obesi* in methanol solvent:

Leaves extract of *E. camaldulensis* in methanol solvent showed 30 ± 1.30 , 52.5 ± 1.73 , 57.5 ± 1.15 percent mortality by feeding on 100 ppm, 200 ppm and 300ppm respectively on first day of experiment where as in control no significant mortality was observed i.e. 3.83 ± 0.88 . LC_{50} value was 0.020% and value of LC_{90} was 0.141%. On 2nd day of experiment percent mortality rate on same concentration (100 ppm, 200 ppm and 300ppm) was 61.66 ± 1.02 , 76.66 ± 1.45 and 80 ± 1.15 while in control observed mortality was 4.16 ± 0.66 which was not significant. LC_{50} value was 0.005% and value of LC_{90} was 0.065%. Maximum mortality rate was observed on 3rd day of experiment at all above mentioned concentrations which was 97.5 ± 0.57 , 96.66 ± 0.88 and 99.16 ± 0.33 and mortality rate of control was 5.83 ± 0.33 which was non significant. LC_{50} value was 0.001% and value of LC_{90} was 0.006%. (Table and figure 3.22)

Plants extracts;

Table3.15: Percentage mortality of *O. obesus* in different concentrations of *M. azedarach* in water.

Days	Concentrations (ppm)				LC ₅₀ %	LC ₉₀ %
	Control	100	200	300		
Day 1	3.33±0.33 c	7.50±0.57bc	18.33±0.88ab	10.20±1.76 a	5.64	28.7
Day 2	4.16±0.33 c	50.00±0.24 b	69.16±1.20ab	73.33±1.86 a	0.009	0.08
Day 3	7.50±0.58 b	93.33±1.76 a	96.66±0.33 a	99.16±0.33 a	0.002	0.007

Values in the same columns with different letters are significantly different by Tukey test ($p < 0.05$).

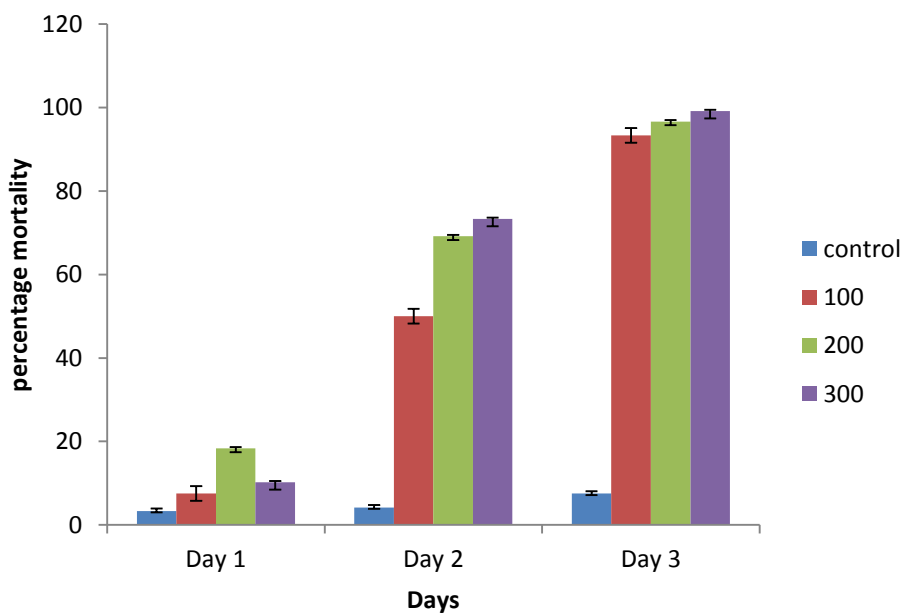


Figure 3.15: Percentage mortality of *O. obesus* in different concentrations of *M. azedarach* in water.

Table 3.16: Percentage mortality of *O. obesus* in different concentrations of *M. azedarach* in methanol.

Days	Concentrations (ppm)				LC ₅₀ %	LC ₉₀ %
	Control	100	200	300		
Day 1	3.33±0.88 b	13.33±1.20ab	18.33±1.02ab	22.5±1.53 a	0.31	17.01
Day 2	5.83±0.33 c	59.16±1.02 b	71.66±1.20ab	80.88±1.76 a	0.006	0.064
Day 3	8.33±0.57 b	95.83±1.50 a	97.5±1.53 a	99.16±1.20 a	0.0003	0.004

Values in the same columns with different letters are significantly different by Tukey test ($p < 0.05$).

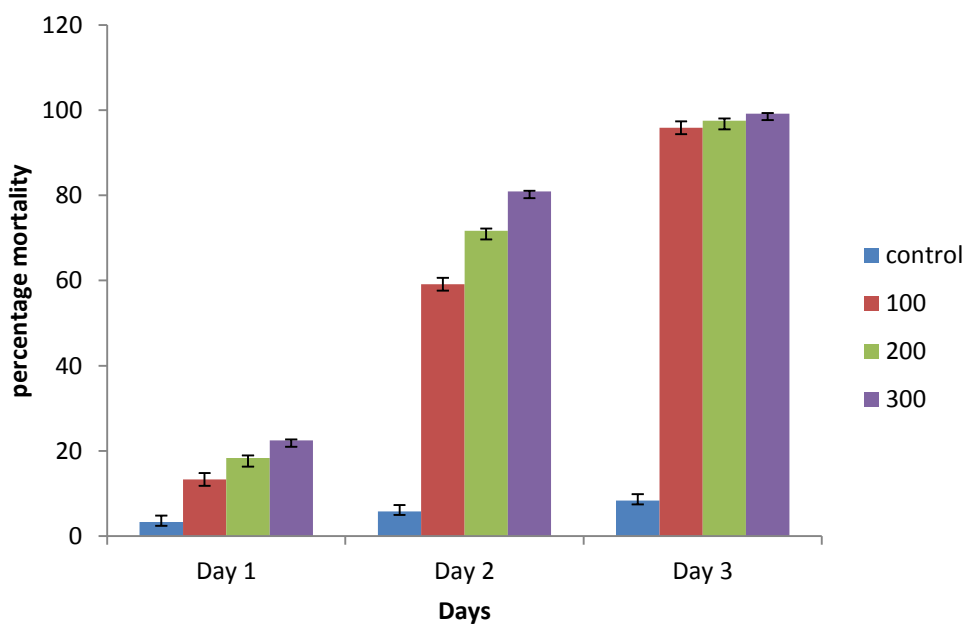


Figure 3.16: Percentage mortality of *O. obesus* in different concentrations of *M. azedarach* in methanol.

Table 3.17: Percentage mortality of *O. obesus* in different concentrations of *E. camaldulensis* in water.

Days	Concentrations (ppm)				LC ₅₀ %	LC ₉₀ %
	Control	100	200	300		
Day 1	2.50±0.57 b	12.50±1.15 b	15.0±1.52ab	26.66±1.20 a	0.133	2.04
Day 2	5.00±0.57 b	56.66±0.84 a	71.66±2.40 a	81.66±1.85 a	0.007	0.058
Day 3	10.0±0.57 b	93.33±1.45 a	97.5±1.00 a	100±0 a	0.002	0.008

Values in the same columns with different letters are significantly different by Tukey test ($p < 0.05$).

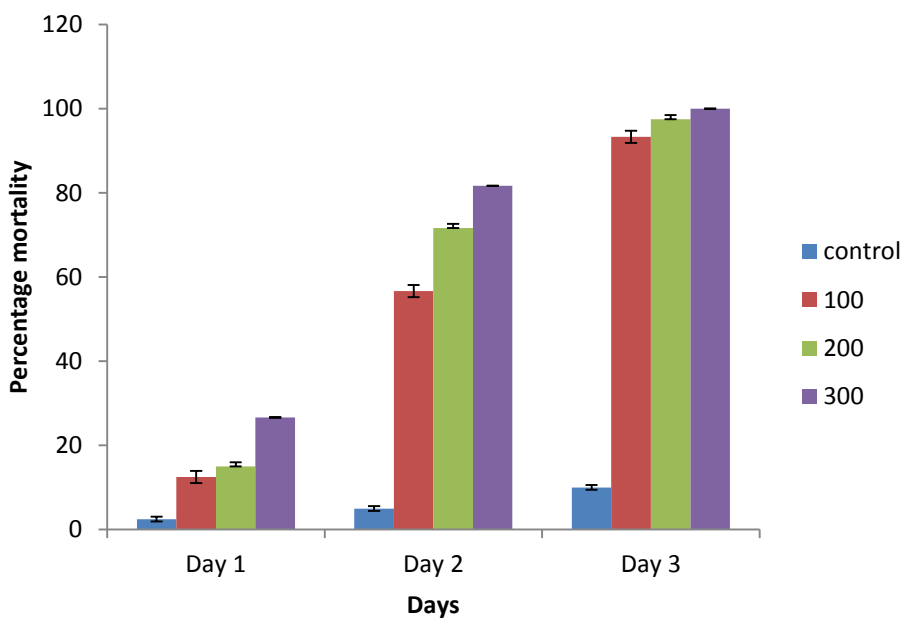


Figure 3.17: Percentage mortality of *O. obesus* in different concentrations of *E. camaldulensis* in water.

Table 3.18: Percentage mortality of *O. obesus* in different concentrations of *E. camaldulensis* in methanol.

Days	Concentration (ppm)				LC ₅₀ %	LC ₉₀ %
	Control	100	200	300		
Day 1	3.33±0.88 b	25±1.15 a	34.16±1.40 a	35.83±0.33 a	0.094	7.57
Day 2	6.66±0.88 b	62.5±0.78 a	71.66±1.85 a	84.16±1.76 a	0.006	0.056
Day 3	9.16±0.88 b	97.5±0.57 a	99.83±1.66 a	100±0 a	0.008	0.004

Values in the same columns with different letters are significantly different by Tukey test ($p < 0.05$).

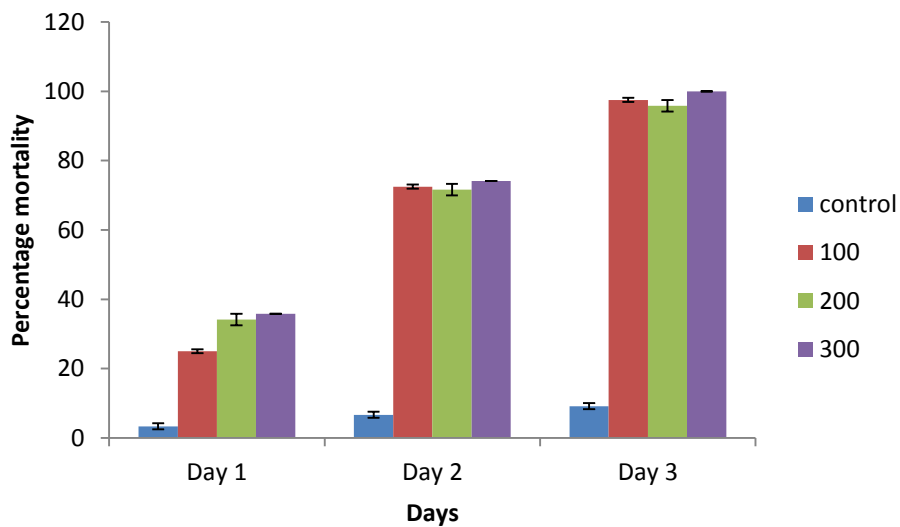


Figure 3.18: Percentage mortality of *O. obesus* in different concentrations of *E. camaldulensis* in water.

Table 3.19: Percentage mortality of *M. obesin* different concentrations of *M.azedarach* in water.

Days	Concentrations (ppm)				LC ₅₀ %	LC ₉₀ %
	Control	100	200	300		
Day 1	4.16±0.88 c	20.83±1.20 b	25.83±1.45 b	42.5±1.73 a	0.049	0.515
Day 2	9.16±0.88 c	41.66±1.20 b	45.83±0.88 b	65.83±0.40 a	0.017	0.206
Day 3	12.50±0.57 b	91.66±1.02 a	93.33±0.88 a	99.16±0.33 a	0.001	0.009

Values in the same columns with different letters are significantly different by Tukey test ($p < 0.05$).

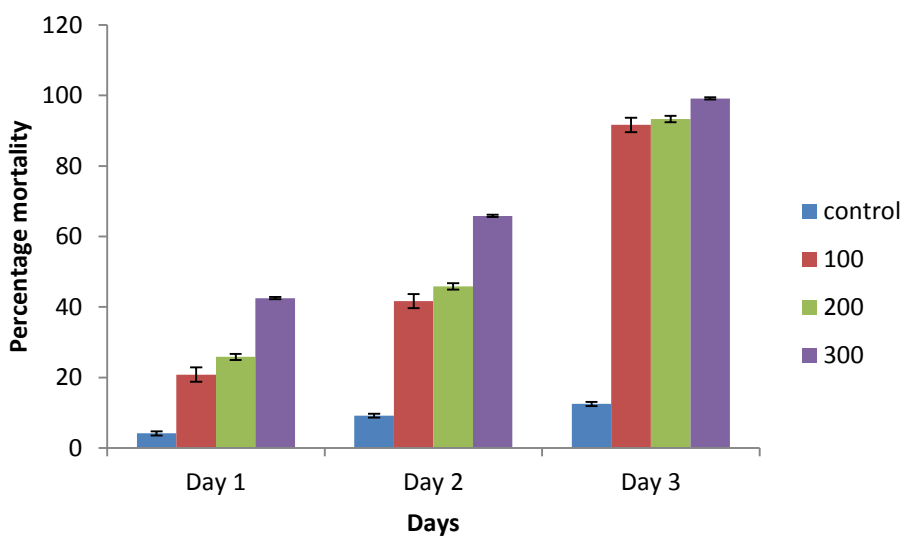


Figure 3.19: Percentage mortality of *M.obesin* different concentrations of *M.azedarach* in water.

Table 3.20: Percentage mortality of *M.obesi* in different concentrations of *M.azedarach* in methanol.

Days	Concentration (ppm)				LC ₅₀ %	LC ₉₀ %
	Control	100	200	300		
Day 1	3.33±0.88 c	26.5±0.88 b	37.33±1.73ab	41.66±1.45 a	0.049	1.385
Day 2	8.33±1.20 c	53.33±1.85 b	68.33±1.45ab	78.33±0.85 a	0.009	0.068
Day 3	10±1.15 b	92.50±1.15 a	97.50±0.57 a	100±0.00 a	0.002	0.008

Values in the same columns with different letters are significantly different by Tukey test ($p < 0.05$).

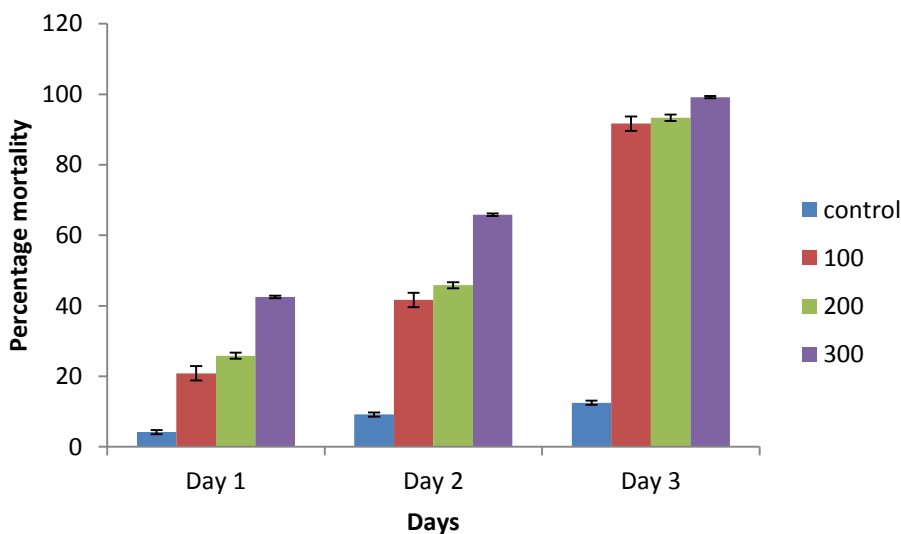


Figure 3.20: Percentage mortality of *M. obesi* in different concentrations of *M.azedarach* in water.

Table 3.21: Percentage mortality of *M. obesii* in different concentrations of *E. camaldulensis* in water.

Days	Concentrations (ppm)				LC ₅₀ %	LC ₉₀ %
	Control	100	200	300		
Day 1	0.83±0.33 b	47.50±0.05 a	51.66±0.88 a	66.66±0.88 a	0.013	0.292
Day 2	3.83±0.33 b	69.16±1.20 a	76.66±0.88 a	81.66±1.76 a	0.002	0.088
Day 3	5.833±0.33 b	95±1.52 a	96.66±0.66 a	100±0.00 a	0.001	0.006

Values in the same columns with different letters are significantly different by Tukey test ($p < 0.05$).

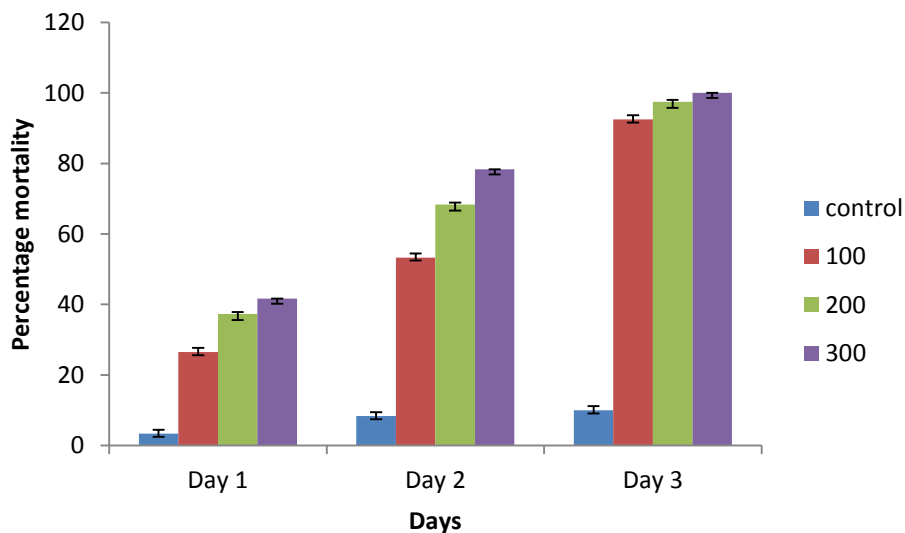


Figure 3.21: Percentage mortality of *M. obesii* in different concentrations of *E. camaldulensis* in water.

Table 3.22: Percentage mortality of *M. obesi* in different concentrations of *E. camaldulensis* in methanol.

Days	Concentrations (ppm)				LC ₅₀ %	LC ₉₀ %
	Control	100	200	300		
Day 1	3.83±0.88 c	30±1.30 b	52.5±1.73 b	57.5±1.15 a	0.020	0.141
Day 2	4.16±0.66 c	61.66±1.02 b	76.66±1.45ab	80±1.15 a	0.005	0.065
Day 3	5.83±0.33 b	94.5±0.57 a	98.66±0.88 a	99.16±0.33 a	0.001	0.006

Values in the same columns with different letters are significantly different by Tukey test ($p < 0.05$).

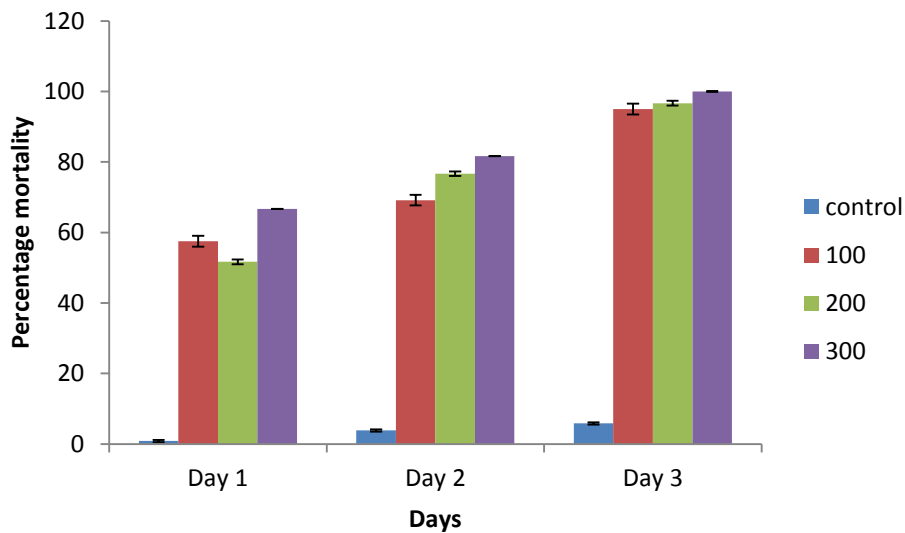


Figure 3.22: Percentage mortality of *M. obesi* in different concentrations of *E. camaldulensis* in methanol.

Biochemical test

The results showed that the protein level of *O. obesus* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 44.36 ± 0.39 and 55.08 ± 0.33 . The protein level of *O. obesus* in *E. camaldulensis* in methanol and water (solvent) at same concentration was 18.89 ± 0.99 and 50.34 ± 0.91 which was lowered from control 54.61 ± 0.05 . Protein level of *M. obesi* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 283.22 ± 0.21 and 194.33 ± 0.52 . The protein Level of *M. obesi* in *E. camaldulensis* in methanol and water (solvent) at same concentration was 211.56 ± 0.24 and 162.61 ± 0.30 lowered from control 286.21 ± 1.04 (Table and figure 3.23).

The results revealed that the Carbohydrate level of *O. obesus* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 2.243 ± 0.64 and 4.839 ± 0.03 . The carbohydrate level of *O. obesus* in *E. camaldulensis* in methanol and water (solvent) at same concentration was 3.76 ± 1.00 and 3.02 ± 1.00 which was lowered from control 4.83 ± 0.06 . The results showed that the carbohydrate level of *M. obesi* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 3.47 ± 0.61 and 3.15 ± 0.85 . The carbohydrate level of *M. obesi* in *E. camaldulensis* in methanol and water (solvent) at same concentration was 3.91 ± 0.52 and 4.84 ± 0.03 which was lowered from control 4.82 ± 0.05 . (Table and figure 3.24)

The results revealed that the Cholesterol level of *O. obesus* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 137.50 ± 1.52 and 131.20 ± 0.44 . The cholesterol level of *O. obesus* in *E. camaldulensis* in methanol and water (solvent) at same concentration was 135.70 ± 0.69 and 133.50 ± 0.37 which was lowered from control 145.60 ± 1.27 . The results revealed that the cholesterol level of *M. obesi* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 137.80 ± 0.40 and 135.10 ± 1.42 . The cholesterol level of *O. obesus* in *E. camaldulensis* in methanol and water (solvent) at same concentration was 135.90 ± 0.51 and 135.70 ± 1.51 which was lowered from control 140.60 ± 0.75 (Table and figure 3.25).

The results showed that the level of triglyceride of *O. obesus* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 38.06 ± 0.92 and 36.71 ± 1.33 . TG level of *O.*

obesus in *E. camaldulensis* in methanol and water (solvent) at same concentration was 38.53 ± 1.58 and 34.97 ± 2.63 which was lowered from control 39.48 ± 0.54 . The results revealed that the TG level of *M. obesi* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 40.9 ± 1.03 and 40.14 ± 1.28 . The TG level of *O. obesus* in *E. camaldulensis* in methanol and water (solvent) at same concentration was 36.43 ± 0.43 and 34.17 ± 0.22 lowered from control 45.57 ± 0.49 (Table and figure 3.26).

The results revealed that the HDL of *O. obesus* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 16.85 ± 1.34 and 113.55 ± 1.45 . The HDL level of *O. obesus* in *E. camaldulensis* in methanol and water (solvent) at 300 ppm was 114.29 ± 1.13 and 114.31 ± 1.79 which was lowered from control 119.57 ± 0.48 . The results revealed that the HDL level of *M. obesi* in *M. azedarach* in methanol and water (solvent) at 300 ppm was 174.41 ± 15.84 and 133.97 ± 7.26 . The HDL level of *O. obesus* in *E. camaldulensis* in methanol and water (solvent) at 300 ppm was 142.92 ± 4.31 and 141.59 ± 1.67 lowered from control 215.87 ± 2.61 (Table and figure 3.27).

Protein estimation

Table 3.23: Comparison of total protein content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M.azedarach* at 300 ppm in water and methanol solvent.

Plants extracts	Protein value (mg/g)	
	Concentration (300ppm)	
	<i>O. obesus</i>	<i>M. obesi</i>
<i>E. camaldulensis</i> in water (E)	50.347±0.91	162.619±0.30
<i>E. camaldulensis</i> in methanol (E)	18.891±0.99	211.563±0.24
<i>M. azedarach</i> in water (M)	55.081±0.33	194.333±0.52
<i>M. azedarach</i> in methanol (M)	44.361±0.39	283.229±0.21
Control	54.613±0.05	286.219±1.04

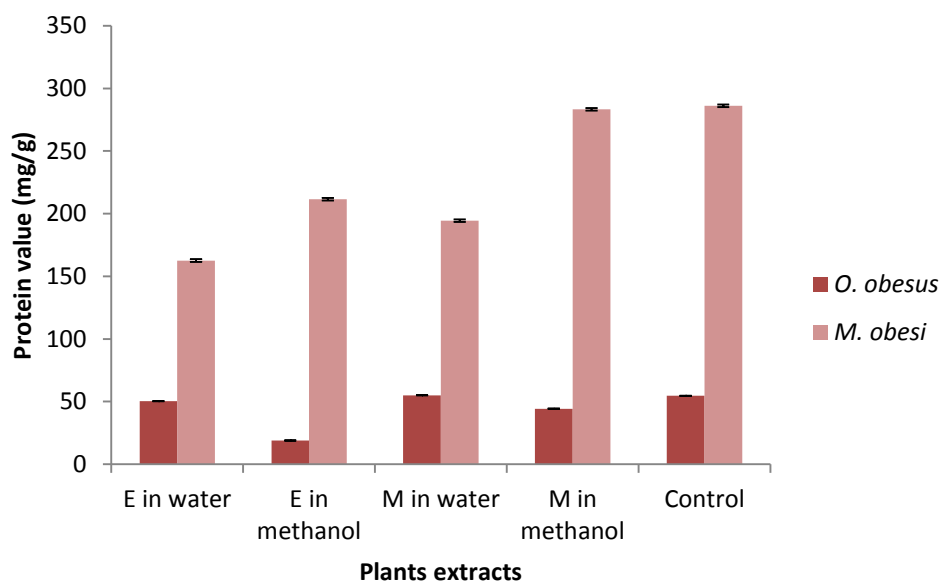


Figure 3.23: Comparison of total protein content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M.azedarach* at 300 ppm in water and methanol solvent.

Carbohydrate estimation:

Table 3.24: Comparison of total carbohydrate content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M. azedarach* at 300 ppm in water and methanol solvent.

Plants extracts	Carbohydrate mg/dl	
	Concentration (300 ppm)	
	<i>O. obesus</i>	<i>M. obesi</i>
<i>E. camaldulensis</i> in water (E)	3.021±1.00	4.845±0.03
<i>E. camaldulensis</i> in methanol (E)	3.762±1.00	3.917±0.52
<i>M. azedarach</i> in water (M)	4.839±0.03	3.154±0.85
<i>M. azedarach</i> in methanol (M)	2.243±0.64	3.479±0.61
Control	4.839±0.06	4.828±0.05

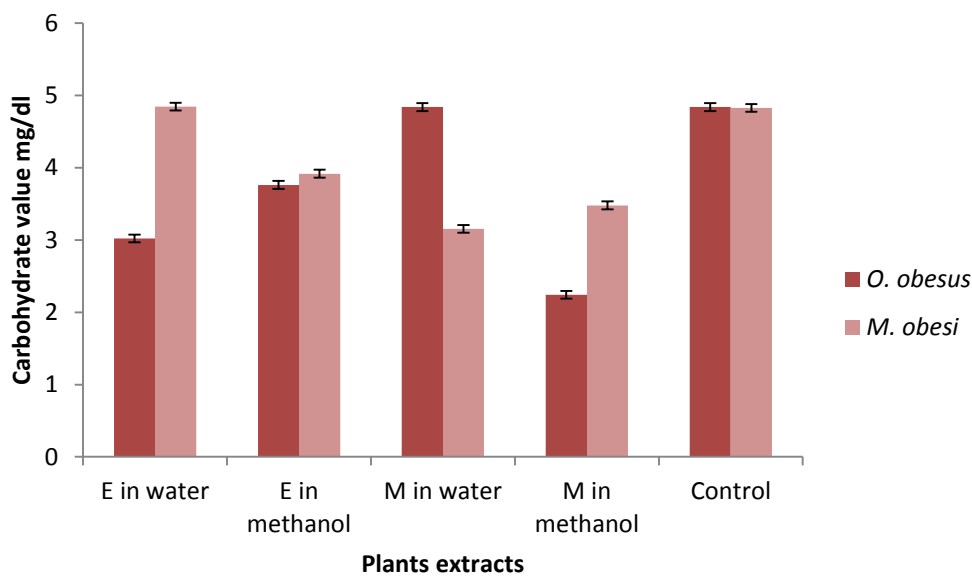


Figure 3.24: Comparison of total carbohydrate content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M. azedarach* at 300 ppm in water and methanol solvent.

Lipid estimation:

Table 3.25: Comparison of total lipid (cholesterol) content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M. azedarach* at 300 ppm in water and methanol solvent.

Plants extracts	Cholesterol mg/dl	
	<i>O. obesus</i>	<i>M. obesi</i>
<i>E. camaldulensis</i> in water (E)	133.5±0.37	135.7±1.51
<i>E. camaldulensis</i> in methanol (E)	135.7±0.69	135.9±0.51
<i>M. azedarach</i> in water (M)	131.2±0.44	135.1±1.42
<i>M. azedarach</i> in methanol (M)	137.5±1.52	137.8±0.40
Control	145.6±1.27	140.6±0.75

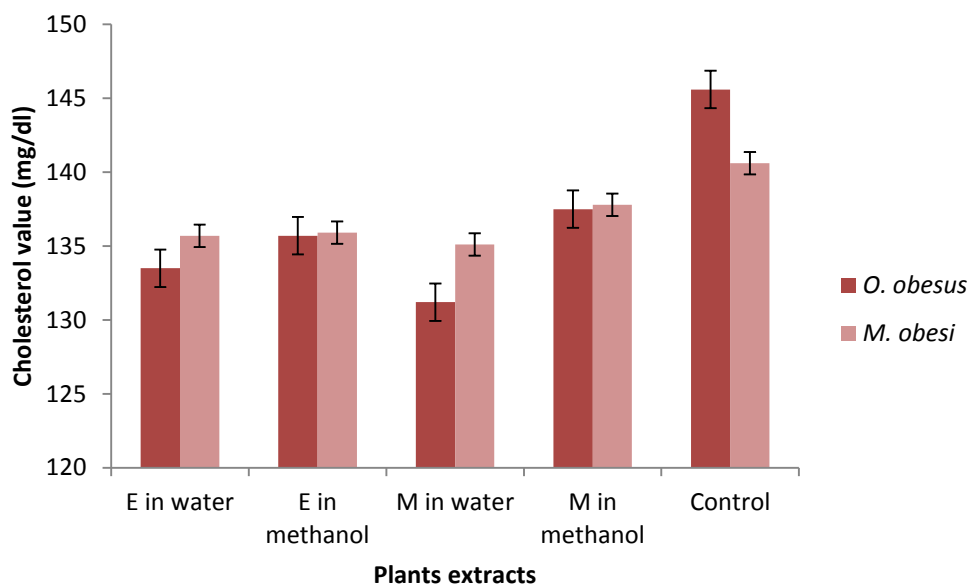


Figure 3.25: Comparison of total lipid (cholesterol) content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M. azedarach* at 300 ppm in water and methanol solvent.

Table 3.26: Comparison of total lipid (TG) content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M. azedarach* at 300 ppm in water and methanol solvent.

Plants extracts	Triglyceride mg/dl	
	<i>O. obesus</i>	<i>M. obesi</i>
<i>E. camaldulensis</i> in water	34.97±2.63	34.17±0.22
<i>E. camaldulensis</i> in methanol	38.53±1.58	36.43±0.43
<i>M. azedarach</i> in water	36.71±1.33	40.14±1.28
<i>M. azedarach</i> in methanol	38.06±0.92	40.9±1.03
Control	38.48±0.54	45.57±0.49

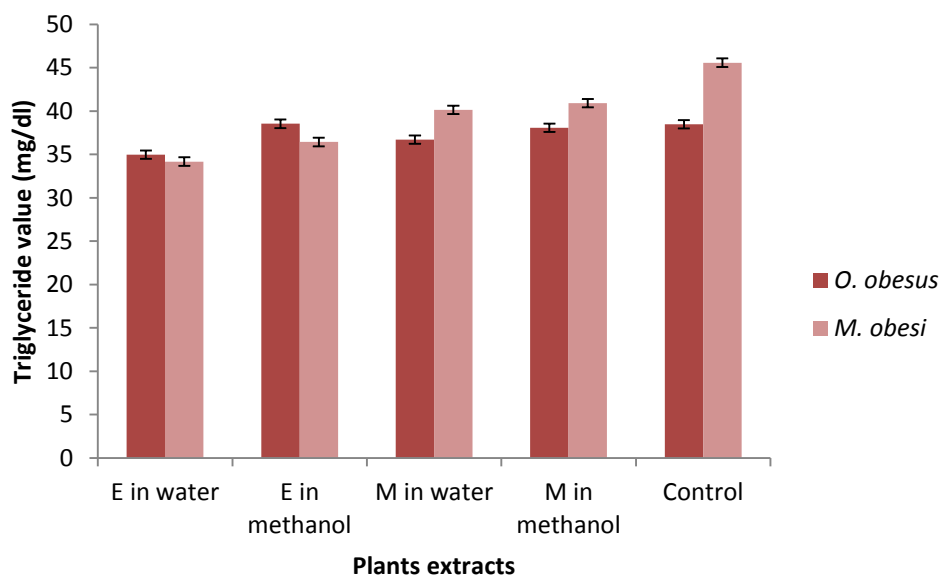


Figure 3.26: Comparison of total lipid (TG) content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M. azedarach* at 300 ppm in water and methanol solvent.

Table 3.27: Comparison of total lipid (HDL) content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M.azedarach* at 300 ppm in water and methanol solvent.

Plants extracts	HDL mg/dl	
	<i>O. obesus</i>	<i>M. obesi</i>
<i>E. camaldulensis</i> in water (E)	114.31±1.79	141.59±1.67
<i>E. camaldulensis</i> in methanol (E)	114.29±1.13	142.92±4.31
<i>M. azedarach</i> in water (M)	113.55±1.45	133.97±7.26
<i>M. azedarach</i> in methanol (M)	116.85±1.34	174.41±15.84
Control	119.57±0.48	215.87±2.61

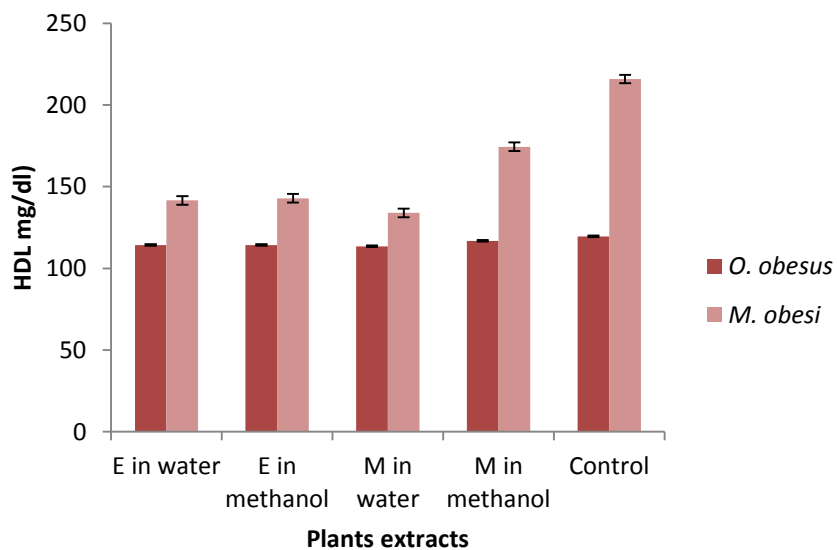


Figure 3.27: Comparison of total lipid (HDL) content of *O. obesus* and *M. obesi* in *E. camaldulensis* and *M.azedarach* at 300 ppm in water and methanol solvent.

DISCUSSION

Morphological features are important for better taxonomy, identification and classification of termites. In the present study morphometric variations in soldier caste of two species *O. obesus* and *M. obesi*, from site A (Murree), B (Rawalpindi) and C (Gujarkhan) were examined. Each species showed a little considerable variations among samples collected from different described sites, most of samples showed overlapping values in different morphological traits. Student “t” test showed that almost all external features of termites collected from three locations were not significantly different. The body length of *O. obesus* has no significant difference among samples of described sites. In abdominal length only C site (Gujarkhan) samples were larger as compared to B (Rawalpindi) and A (Murree) sites. Whereas the length of the thorax (prothorax, mesothorax and metathorax) were not significantly varied. Head’s length and width of samples slightly varied of site C (Gujarkhan) samples from site B (Rawalpindi) and site A (Murree) samples. Pronotum length and width were not varied significantly among samples of different studied sites. Postmentum length and width of samples site A (Murree) were slightly greater as compared to other sites samples. There was no significant variation in the length of left and right mandibles among described sites’ examined samples. But a little variation was found in the tooth length of left mandibles of Site A (Murree) samples as compared to other sites. Length of pedicle of antenna varied from 0.5-1.0µm in the pooled data and most of samples were overlapping. Antennal variations among samples can be used as taxonomic characteristics because antenna is used as sensor tool in termites for detecting environmental changes and it also support insects activities i.e. looking for their prey (Arifet *al.*, 2010). Variations in legs measurements can also be used as taxonomic tool for termites and used in feeding and travelling activities to access for food and shelter (Arifet *al.*, 2010). Coxa of front legs of site A (Murree) were slightly greater. While trochanter of site A (Murree) samples are smaller in length as compared to other site samples under observations. While tibia and tarsus of all different sites samples have no significant differences just the shape of claw

which is significantly different among all described sites. In middle and front legs femur is larger than tibia among all samples of described sites whereas in hind leg tibia shows significantly larger value than the femur among all the samples described sites. Similarly the tarsus and claw of hind legs were slightly larger among the samples of all described sites. So these variations in claw and tarsus show adaptive traits in different environments.

In *M. obesi* a little variation was observed among the samples from different sites (A, B, C). The whole observed range of body length 39-44µm in data pool only site A (Murree) showed slight variation. Similarly the length of abdomen varied of site A (Murree) samples a little large abdominal length as compared to other site's samples. The thorax (prothorax, metathorax, metathorax) of all sample from described sites have no significantly difference in shape, color and length. Length from head to tip, length of pronotum and postmentum, mandibles, pedicle and scape have no significant difference among the samples of understudy sites. Whereas in front leg's coxa of site B samples were 1.0µm larger in length as compared to other site's samples similarly a little bit variation was examined among trochanter of front leg of site B (Rawalpindi) among other's samples. A significant variation in tibia and femur of hind legs was observed.

Sheikhet *al* (2005) studied the morphometric variation among the *Microtermes* mycophagous (fungus growing termite) from different geographical region of Pakistan to understand the geographical range of *M. mycophagous*. Similarly this study resulted that in Pothohar region of Pakistan variations among morphological features of *M. obesi* from different sites provide a sufficient taxonomic knowledge about *M. obesi*.

To minimize the damages caused by termites various strategies have been adopted and various synthetic chemicals were used for termite control in past. During last 50 years synthetic insecticides were applied through soil for termite control and various chemicals have been identified, whose effects changes with change of ecological factors and soil structure. Synthetic insect pest repellent which are used for termite control have been proved to be toxic, and produce number of hazards associated with human health, contamination of soil, atmosphere, aquatic ecosystem and indirectly all food items grown

in soil. When these insecticides are used repeatedly than insects became resistant against them and result in insects outbreak and the cost which is used to overcome insect outbreak exceed from cost of these chemicals production and these problems which have been created by human become dangerous for plant as well as animal life(Ward *et al.*, 2008).So researchers are now directed towards the identification and applications of non toxic natural plants based insecticides for termite's control.

More than 1000 plants species have been described from many areas which have chemicals components in their seeds, stems, roots, leaves and flowers against insect pests. Among them only few plants have been used to control insects practically on commercial scale in the last few decades. Alkaloids are chemicals which are produced by plants are nitrogenous in nature. These are chemical poison of plants, are heterocyclic in nature and have direct effect on nervous system of animals. When extracts of alkaloid are applied on insects they disturb nervous system and cause desolation. Therefore these alkaloids are named as nerve poisons (Shahid, 2003). Many scientists are now paying their attention to find out such type of plant toxicants, which are environmentally safe and can control these pests in much better way(Lin, T.S. 1988;Huang, Z.Y. 1990;Parihar D.R. 1994;Hutchins R.A.1997).

For control of soil termites many synthetic compounds have been applied on soil. A number of insecticides have been used to form a barrier in the soil against subterranean termites in order to reduce their approach to food sources and tunneling activities (Ahmed *et al.*, 2010). Plant based natural pesticides are safe to use, easily biodegradable and environment friendly in contrast to synthetic pesticides (Ahmad *et al.*, 2007).

Effects of seeds and leaves extract of some plants have been reported by Ahmed *et al.*, (2007) against *M. Obesi*. In this study soil was treated with leaves and seeds extract of *Withania somnifera*, *Croton tiglium* and *Hygrophila auriculata* and different death rate was reported at different time interval and significant results ($p < 0.05$) were reported. In another study tunneling activities of *M. Obesi* were observed by using aqueous and methanolic leaves extract of *A. vasica*, *N. odorum*, *S. oleiodes*, *G. robusta* and *T.*

purpurea and it was evident from the results that tunneling activities of *M. Obesi* was reduced in both aqueous and methanolic extracts of all these plants as compared to control and mortality rate of extracts was significant and was concentration dependent (Ahmed *et al.*, 2007).

C. decidua extracts and its mixture with other compounds have been evaluated by Upadhyahet *al.*, 2010 against Indian white termite *O. obesus*. This plant shows very strong termicidal activity. And when different components were used to prepare mixtures with *C. decidua* and applied to termites their mortality rate was significantly increased and higher mortality was observed ($p < 0.05$ and 0.01).

Here the present study purpose was the use of anti termite compounds which are naturally occurring extracted from native or local plants and trees that showed encouraging results for termite Control. When termites were treated with these extracts termite mortality was observed. Here experimental plants used against termites were *E. camaldulensis* and *M. azeradach* extracted with two different solvents and targeted termites were *O. obesus* and *M. obesi*. After treating the filter paper with different concentrations of leaves extract like 100 ppm, 200 ppm and 300 ppm termite mortality was observed for consecutive three days and results was significantly different ($p < 0.05$).

In the present study filter paper bioassays was used with two different plant extracts, i.e. *E. camaldulensis* and *M. azeradach* in organic and inorganic solvent (methanol and water). Two termite species *O. obesus* and *M. obesi* were introduced in separate petri dishes having filter papers soaked in different concentration of the two plant extracts. Different mortality rate was observed in *O. obesus* and *M. obesi* in different concentrations of two plants leaves extract (*E. camaldulensis* and *M. azeradach*) in methanol and water solvent at different times. In both solvents mortality rate was higher than control ($p < 0.05$). In the leaves extract of *M. azeradach* and *E. camaldulensis* against *O. obesus* in water solvent percent mortality on 1st day at 100ppm, 200ppm and 300ppm was 7.50, 18.33 and 10.20% and 12.50, 15.00 and 26.66% respectively which was significantly different from control. Mortality rate was increased on 2nd day for same concentration and observed percent mortality was 50, 69.16, 73.33% and 56.66, 71.66,

81.66% for above mentioned both plants in water solvent. Maximum mortality rate was observed on 3rd day and it was 93.33, 96.66, 99.16% and 93.33, 97.50, 100% for both plants in water solvent. When the leaves extract of *M. azedarach* and *E. camaldulensis* were used against *O. obesus* in methanol solvent percent mortality on 1st day at 100ppm, 200ppm and 300ppm was 13.33, 18.33 and 22.50% and 25.00, 34.16 and 35.83% respectively which was significantly different from control. Mortality rate was increased on 2nd day for same concentration and observed percent mortality was 59.16, 71.66, 80.88% and 72.50, 71.66, 74.16% for above mentioned both plants in water solvent. Maximum mortality rate was observed on 3rd day and it was 95.83, 97.50, 99.16% and 97.50, 95.83, 100% for both plants in water solvent.

In the leaves extract of *M. azedarach* and *E. camaldulensis* against *M. obesi* in water solvent percent mortality on 1st day at 100ppm, 200ppm and 300ppm was 20.83, 25.83 and 42.50% and 57.50, 51.66, 66.66% respectively. Mortality rate was increased on 2nd day for same concentration and observed percent mortality was 41.66, 45.83, 65.83% and 69.16, 76.66, 81.66% for above mentioned both plants in water solvent. Maximum mortality rate was observed on 3rd day and it was 91.66, 93.33, 99.16% and 95.00, 96.66, 100% for both plants in water solvent which was significantly different from control in all concentrations even at the end of experiment. When the leaves extract of *M. azedarach* and *E. camaldulensis* were used against *M. obesi* in methanol solvent percent mortality on 1st day at 100ppm, 200ppm and 300ppm was 26.50, 37.33 and 41.66% and 30.00, 52.50, 57.50% respectively. Mortality rate was increased on 2nd day for same concentration and observed percent mortality was 53.33, 68.33, 78.33% and 61.66, 76.66 and 80.00% for above mentioned both plants in water solvent. Maximum mortality rate was observed on 3rd day and it was 92.50, 97.50, 100% and 97.50, 96.66, 99.16% for both plants in water solvent which was significantly different from control at the end day in all concentrations.

Toxicity effect of the four extracts against *M. obesi* and *O. obesus* observed was in following order *E. camaldulensis* (methanol) > *E. camaldulensis* (water) > *M. azedarach* (methanol) > *M. azedarach* (water) and all these four extracts of both plants are more effective against *M. obesi* than *O. obesus*.

When all type of insecticides are applied on insects they have some negative impact on the development and growth and also disturbs the biochemical activities and metabolic processes of insect's body. Therresults of this research showed that after treatment of termites with plants extract having termiticidal potential, the glucose level of *M. obesi* was decreased as compared to control. This decrease in glucose level of termites may be due to the insecticidal stress activated by these extracts in both of the termitespecies. Nath *et al.*, 1997 observed increase in glucose level of insects because of insecticidal stress. In this study it was observed that glucose level of *M. obesi* was greater as compared to *O. obesus* and value of glucose level of both termite species was lowered as compared to the glucose level of their control.

Level of protein in both termite species treated with extracts was considerably less. The reduction in protein contents is perhaps due to extracts having insecticidal interference of protein production regulated by hormones (Sharma *et al.*, 2009).The extracts of *E. camaldulensis* and *M. azeradach* support the effects associated with the development induced by extract in *O. obesus* and *M. obesi* and the growth inhibitory influence of the extract on the developing outline of *O. obesus* and *M. obesi* when body wall and termite tissues were started to rapture and collapsed in both the termite species. Body wall made of chitin, a protein, and other proteintious tissues were destroyed in both the species resulting in overall decrease in protein contents of the pesticide control. In the present study protein level of *M. obesi* was higher as compared to *O. obesus* nearly lower to the level of protein content present in the control in both the termites.

After treatment with extract, lipid level was deteriorated as related to control in *O. obesus*. This decrease in lipid profile displays a negative effect of the extract on lipid amount may be due to change in energy absorption and preoxidation. The decline in lipid amount may be due to the change in energy metabolism towards lipid catabolism as a result of insecticidal strain made by the extract. Lipid level was reduced in *O. obesus* and *M. obesi* treated with some plant extracts and it might be due to biological stress disorders induced by the extracts. These results are in accordance with Lohar and Wright 1993 who

reported lipid diminution in oocytes, haemolymph and fat bodies of *Tenebriomolitor* after exposing to malathion. Saket *al.*, 2006 proposed a change toward lipids catabolism from energy metabolism that results in decrease in lipid profile because of insecticidal stress.

In the present study the lipid estimation (High Density Lipid, cholesterol, triglyceride cholesterol) after treatment of extracts *E. camaldulensis* and *M. azeradach*, the lipid level (cholesterol) of *M. obesi* was higher as compared to *O. obesus* nearly lower to the level of protein content present in the control in both the plants. Lipid level (triglyceride) of *M. obesi* was higher as compared to *O. obesus* approximately lower to the level of protein content present in the control in both the plants. Lipid level (high density lipid) of *M. obesi* was higher as compared to *O. obesus* approximately lower to the level of lipid content present in the control in both the plants.

It is determined that the impacting aspects of extracts on carbohydrate, lipid and protein contents in treated termite species and specific extraction. The lowering of these biochemical components shows that these extract can lowered the feeding and proper digestion of food. They further interrupt with protein synthesis hormones causing in its decline. The significance of extracts on the metabolism of treated termite depends on their nature and maybe also on the act of different photochemicals in these extracts.

The result of this study show that overall effect of *E. camaldulensis* in methanol and water solvent was higher than *M. azedarach* in both solvents against *M. obesi* than *O. obesus* and the extract which was taken in methanol solvent was more effective than extracts in water. This may be due to the presence of some toxic compounds in *E. camaldulensis* which may not be present in *M. azedarach* or might be found in lesser quantity. Here it is concluded that our findings on two plants *E. camaldulensis* and *M. azedarach* leaf extracts against two termite species *O. obesus* and *M. obesi* were encouraging and if more attention is given on extraction and purification process of crude plants products than it is possible to increase termiticidal potential of these plants. Further studies are needed to characterize and isolate the particular chemical responsible for toxicity in insects like termite and can be put in use on commercial basis after careful experimentations of its safety for its environment and other organisms.

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