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Efficacy of plant leaf extracts on termites under laboratory conditions

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Abstract
The attack of crop plants by termites leads to serious crop damage, yield loss and contamination of products. The use of synthetic insecticides to control termites cause aquatic and environmental pollution and development of resistance by the termites. A broad range of plants were reported to insecticide properties. Therefore, the current study was aimed to determine the efficacy of locally available plants at different concentrations for the management of termites. The study was conducted under laboratory conditions to evaluate the toxic effect of five different plants such as *Azadirachta indica*, *Samadera indica*, *Allium sativum*, *Z. officinale* and *Justicia adhatoda* on termites at different concentrations like 100, 300, 600, 900 and 1000 mg/mL. Aqueous plant extracts were used for the bioassay procedure with 10 worker termites and three replications were maintained for each experiment. Form the results, it was found that *A. sativum* (86%) showed highest percentage of mortality which was followed by *Z. officinale* (82%), *A. indica* (81.33%), *S. indica* (74%) and *J. adhatoda* (72.67%) but, statistically there was no significant difference found between the plant extracts. 1000 mg/mL (97.33%) concentration of all the plant extract showed highest percentage of mortality. Based on time of exposure of termites to the extracts, 58.33% of mortality rate accrued when exposed to 20 min in the case of all extracts. It was found that 100 mg/mL was the minimum concentration required to kill 50% of the test organism (termites) for *A. indica* and *S. indica* and for *A. indica* and *S. indica*, LC_{50} was found at 300 mg/mL. Therefore, the study can be concluded that the percentage of mortality of tested termites was found concentration and time dependent as it was found to increase with an increase of the concentration and exposure time.

Keywords: Contamination, bioassay, mortality, percentage, concentration

Introduction
Termites belongs to the order Isoptera and reported as they were causing major agricultural and domestic problems in many countries (Sileshi *et al*., 2013) [21]. Among the specie *Macrotermes* sp. is the largest termite species. *Macrotermes* sp. builds nests or mounds from where they forage outwards to distances up to 50m in galleries. *Macrotermes* sp. are economically important insect pests causing serious damage up to 100% loss to agricultural crops and various domestic products. The crop damage caused by the termites lead to the secondary infection by plant pathogens which cause indirect yield loss and contamination of products with the production of aflatoxins (Osipitan and Oseyemi, 2012) [16].

A broad spectrum of persistent organochlorinated hydrocarbon insecticides were used worldwide for the management of termites. Most of them were banned from the market for harmful effects on human health and environmental pollutions. Also the synthetic insecticides were not affordable for the farmers (Sileshi *et al*., 2013) [21]. The use of synthetic insecticides in the control of termites is known to cause aquatic and environmental pollution, lethal effect on non-target organism and resistant development by the termites (Sohail *et al*., 2011) [23].

As a result of negative impacts of synthetic pesticides, research on identification of ecofriendly and locally available tools for the control of termites has been intensified by entomologists. Thus, plants with insecticidal properties could be regarded as potential alternatives to chemical pesticides. A broad range of plants are reported to have toxic, repellent, anti-feeding properties and were regarded as insecticides. Locally, plants or plant products are used by farmers to control termites. Some species of living plants are usually left inside the farms in order to deter termites from crops (Sileshi *et al*., 2008 and Mugerwa *et al*., 2014) [22, 13]. The termiticidal proprieties of plants lie mainly on their secondary compounds or organic extractives such as waxes, alkaloids, fat, gums, resins, terpenes and essential oils (Pettersen, 1984) [19].

Azadirachta indica, *Samadera indica*, *Allium sativum*, *Justicia adhatoda*, *Zingiber officinale* were reported to have antitermite activity and are inexpensive locally available plants. The repellent properties of *Z. officinale* and *A. sativum* was reported in the studies of Saljoqi *et al*., (2012) [20] and Cynthia *et al*., (2016) [6]. Addisu *et al*., (2014) [2] reported that, among many botanicals used in insect pest management, leaves and seeds of *A. indica*, bulbs of *A. sativum* and seeds of *Jatropha* sp. have been successfully used in the control of *Macrotermes* sp. *S. indica* was relatively less mentioned in literatures and have high termite controlling properties (Ahmed *et al*., 2011) [4]. A wide range of phytochemical constituents have been isolated from *J. adhatoda* which possesses activities like antitussive, abortifacient, antimicrobial, cardiovascular protection, anticholinesterase, anti-inflammatory larvicidal and other important activities (Dhankar *et al*., 2011) [7]. The chemical poisons of plants are mostly alkaloids, which are plant products and nitrogenous in nature, having strong effect on the nervous system of animals. The alkaloidal extracts when applied to the insects bring about disturbance in the nervous system and cause death (Okugawa *et al*., 1993) [15]. Therefore, the current study was aimed to determine the efficacy of locally available plants at different concentrations for the management of termites.

Materials and Methods

Collection of plants and extraction

The study was conducted at Tropical Institute of Ecological Science, Kottayam during the period of February to April 2022. Five different were collected such as *Azadirachta indica* (leaves), *Samadera indica* (leaves), *Allium sativum* (bulbs), *Z. officinale* and *Justicia adhatoda* (leaves) which were locally available. The collected samples were washed too remove the dust particles, air dried and aqueous extracts of each samples were prepared separately in different concentrations (100, 300, 600, 900 and 1000 mg/mL).

Collection and establishment of test organism

Termites were collected from a termitarium at the campus. Termite mound was dug up using shovel and soil containing termites was collected on plastic sheets. Termites were isolated from the plastic sheets using camel hair brush and placed in plastic containers as described by Addisu *et al*., (2014) [2]. Termites were fed with dry wood inside the container and the top of the container was covered with muslin cloth to allow free flow of air also to prevent the termites from escaping. The containers carrying the termites were carried to the laboratory and placed in a cool dark area until needed.

Bioassay to determine the toxicity of extracts on termites

Filter papers of 9 cm diameter were placed in sterile petriplates and treated separately with 2mL of extracts of each concentrations. Ten workers from the collected termites were released to the plates. Control plates were also maintained without adding plant extracts. Three replications were maintained for each experiment. The plates were maintained at 70% humidity and under controlled room temperature of 27-30 ºC. The termites were allowed to feed and data was recorded at 30 min interval till 100% mortality of termites was achieved in all the treated dishes. Live and dead termites were counted and percentage mortality was calculated according to the following equation,

$$\text{Percentage mortality} = \frac{\text{Number of dead termites}}{\text{Total number of termites}} \times \frac{100}{1}$$

Statistical analysis

The treatment means were compared by ANOVA one way at 5% level of significance (significant at $p<0.05$) and the ranking of treatments denoted by letters using the SPSS 20.0 statistical package.

Results

It was found that the extracts of *A. sativum* (86%) showed highest percentage of mortality which was followed by *Z. officinale* (82%), *A. indica* (81.33%), *S. indica* (74%) and *J. adhatoda* (72.67%). Statistical analysis revealed that there was no significant difference in the percentage of mortality of termites based on the plant extracts at $p\leq0.05$. Concentration wise analysis revealed that 1000 mg/mL (97.33%) concentration of all the plant extract showed highest percentage of mortality which was followed by 900 mg/mL (86.67%), 600 mg/mL (79.33%), 300 mg/mL (74.0%) and 100 mg/mL (58.67%). There was no significant difference found between 300 and 600 mg/mL concentration of plant extracts. The results were shown in table 1.

Analysis of mortality rate of termites with respect to time revealed that there was no effect observed within 10minin the case of all plant extracts in all the concentrations. It was found that there was no significant difference found in the mortality rate of termites on the basis of plants at $p\leq0.05$. Based on concentration 1000 mg/mL of all plants showed highest percentage of mortality (32.44%) which was followed by 900 mg/mL (28.89%), 600 mg/mL (26.44%), 300mg/mL (26.67%) and 100 mg/mL (19.56%). There was no significant difference found between 1000 and 900mg/mL concentrations and between 600 and 300mg/mL concentrations at $p\leq0.05$. Based on time of exposure to the extracts, 58.33% of mortality rate accrued when exposed to 20min in the case of all extracts and complete death of termites were found at 30min. The mortality rate at 20min was significantly higher than the mortality rate at 30min at $p\leq0.05$. The results were shown in table 2.

The results for the analysis for minimum lethal concentration (LC_{50}) on the mortality of termite were presented in figure 1. It was found that 100mg/mL was the minimum concentration required to kill 50% of the test organism (termites) for each extract except *A. indica* and *S. indica*. For *A. indica* and *S. indica*, LC50 was found at 300 mg/mL. The lethal concentration (LC50), was found significantly different only from 100 mg/mL at $p\leq0.05$.

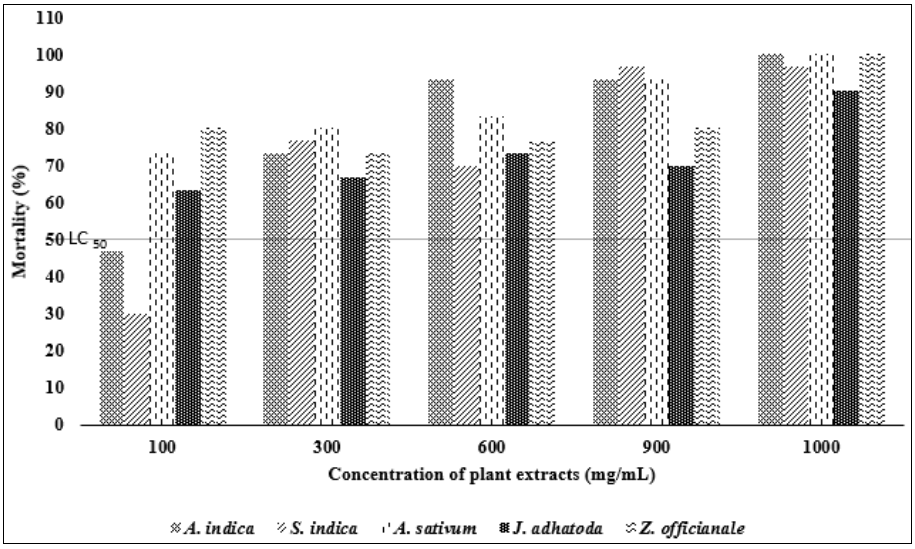

Fig 1: Minimum lethal concentration LC50 for Each Plant Extract

Discussion

The effect of *Z. officinale* and *A. sativum* on mortality of *Macrotermes* sp. was reported in the studies of Cynthia *et al*., (2016) [6]. He states that the efficacy of the plant materials as insecticide or pesticide depends on the active ingredients present in the plant. The active constituents of *A. sativum* includes phenols, saponins and sapogeins (Mona and Yasser, 2009) [11] and *Z. officinale* includes gingerol, shogaol and zingerone. Addisu *et al*., (2014) [2] reported that azadirachtin present in the *A. indica* acts a contact poison and has systemic effect when applied to the foliage. Azadiractin has a broad activity which was working as a feeding deterrent, insect-growth regulator, repellent, sterilant and it may also inhibit the oviposition of insect pests (Isman, 2006) [8]. The substances present in the *A. indica* has synergetic effects on a wide range of insects and pests (Morgan, 2009) [12]. Ahmed *et al*, (2011) [4] reported in his studies that the leave of *S. indica* has termiticidal property. The main components present in *S. indica* were alkaloids, flavonoids, saponins, carbohydrates, proteins, phenols, steroids, and tannins (Nair *et al*., 2018) [14].

The leaves of *J. adhatoda* were toxic to 'all forms of lower life' and have insecticidal effects (Agrawal *et al*., 1986) [3]. The phytochemical analysis showed that phenols, tannins, alkaloids, anthraquinone, saponins, flavonoids and reducing sugars were found in the leaves of *J. adhatoda* (Pathak, 1970) [18]. But the pharmacologically most studied chemical component in *J. adhatoda* was the alkaloid, vasicine (1, 2, 3, 9-tetrahydropyrrole [2, 1-b] quinozolin-3-ol, $C_{11}H_{12}N_2O$which is present in the leaves, roots and flowers. Besides vasicine, the leaves contain several alkaloids (Vasicinone, Vasicinol, Adhatodine, Adhatonine, Adhvasinone, Anisotine and Hydroxypeganine), betaine, steroids and alkanes (Lahiri and Prahdan, 1964 and Chowdhury and Bhattacharyya, 1987) [10, 5]. All the selected plants in this study were rich source of alkaloids. Mao and Henderson (2007) [10] reported that alkaloids had a strong antifeedant effect against Formosan subterranean termite by affecting the nervous system of animals.

Cynthia *et al*., (2016) [6] reported that the constituents present in the plant might probably get into the body system of the insect and interfere with the normal development causing mortality of the termite. His finding was also in consonance with the finding of who evaluated the antifeedant, toxicant and growth regulatory effects of the acetone leaf extracts of *Curcuma longa* and *A. sativum* against the adults of *Tribolium castaneum*. Achio *et al*., (2012) [1] reported that water extracts of plant materials were more effective than their powders or other solvent extracts.

From this study, it was found that 100mg/mL was the minimum concentration required to kill 50% of the test organism (termites) for each extract except *A. indica* and *S. indica*. For *A. indica* and *S. indica*, LC50 was found at 300mg/mL. Addisu *et al*., (2014) [2] also reported that 10-35% concentration of plant extracts was effective to obtain LC_{50} in minimum time of exposure. His study revealed that 10% concentration *of A. indica* tested was able to cause mortality on *Macrotermes* sp. with less lethal time (LT50) than the untreated control. He also states that as time of exposure and concentrations of the botanicals increases the efficacy of the botanicals also increases. Higher mortality of termites due to the application of the plant extracts at different concentrations was observed as the exposure time of the pest to the treatment increased. As exposure time increased there was a progressive increase in the toxicity of the botanicals to the termites recording considerable control of termites. Besides the more the concentration was the better the efficacy of each botanical. Therefore, it can be said that mortality percentage of tested termites due to the tested plant extracts was found concentration and time dependent as it was found to increase with an increase of the concentration and exposure time. Similar results was found in the studies of Addisu *et al*., (2014) [2]. Plants with repellent effects cause little or no negative impact in the environment when used for pest management because they drive pest by stimulating their sensory organ before the damage is caused to plants. These plant extracts could serve as alternatives to synthetic insecticides in termite management practices because they are biodegradable, cheap, easy to prepare and readily available in common markets. This indicates that naturally occurring anti-termite compound extracted from locally available plant have potentials for managing the population of termites (Cynthia *et al*., 2016) [6].

Table 1: Percentage of mortality of termites exposed to different concentrations of plant extracts

Plant extracts	Mortality (%)	Concentration (mg/mL)	Mortality (%)
A. indica	81.33^{ab}	100	58.67^{d}
S. indica	74.00^{bc}	300	74.00^{c}
A. sativum	86.00^{a}	600	79.33b^{c}
J. adhatoda	72.67^{c}	900	86.67^{b}
Z. officinale	82.00^{ab}	1000	97.33^{a}
P value	0.008	*P* value	0.000

Values followed by the same letter were not significantly different at $p \leq 0.05$

Table 2: Percentage of mortality of termites with respect to time

Plant extracts	Mortality (%)	Concentration (mg/mL)	Mortality (%)	Time interval	Mortality (%)
A. indica	27.11^{ab}	100	19.56^{d}	10 min	0.00^{c}
S. indica	24.67^{ab}	300	24.67^{c}	20 min	58.53^{a}
A. sativum	28.67^{a}	600	26.44b^{c}	30 min	20.67^{b}
J. adhatoda	24.22^{b}	900	28.89^{ab}		
Z. officinale	27.33^{ab}	1000	32.44^{a}		
P value	0.107	*P* value	0.000	*P* value	0.000

Values followed by the same letter were not significantly different at $p \leq 0.05$

Conclusion

The results revealed that the extracts of *A. sativum* (86%) showed highest percentage of mortality which was followed by *Z. officinale* (82%), *A. indica* (81.33%), *S. indica* (74%) and *J. adhatoda* (72.67%) but, statistically there was no significant difference found between the plant extracts in the percentage of mortality of termites. 1000mg/mL (97.33%) concentration of all the plant extract showed highest percentage of mortality. Based on time of exposure of termites to the extracts, 58.33% of mortality rate accrued when exposed to 20min in the case of all extracts and complete death of termites were found at 30min. It was found that 100mg/mL was the minimum concentration required to kill 50% of the test organism (termites) for each extract except *A. indica* and *S. indica*. For *A. indica* and *S. indica*, LC50 was found at 300 mg/mL. The percentage of mortality of tested termites due to the tested plant extracts was found concentration and time dependent as it was found to increase with an increase of the concentration and exposure time. The present findings demonstrated that all of the plant extracts tested against termites possess termiticidal properties that can be used in the management of *Macrotermes sp.*

References

1. Achio S, Ameko E, Kutsanedzie F, Alhassan S. Insecticidal effects of various neem preparations against some insects of agricultural and public health concern, International journal of research in biosciences. 2012;1(2):11-19.
2. Addisu S, Mohammed D, Waktole S. Efficacy of Botanical Extracts against Termites, *Macrotermes* spp., (Isoptera: Termitidae) Under Laboratory conditions. International Journal of Agricultural Research. 2014;9:60-73.
3. Agrawal S, Chauhan S, Mathur R. Antifertility effects of Embelin in male rats. Andrologia. 1986;18:125-131.
4. Ahmed S, Zafar MI, Hussain A, Riaz MA, Shahid M. Evaluation of plant extracts on mortality and tunnelling activities of subterranean termites in Pakistan, Pesticides in the modern world – Pests Control and Pesticides Exposure and Toxicity Assessment; c2011. p. 39-54.
5. Chowdhury BK and Bhattacharyya P. Adhavasinone: A new quinazolone alkaloid from *Adhatoda vasica* Nees. Chem. Ind., (London). 1987;1:35-36.
6. Cynthia OC, Precious O, Lynda OS. The toxicity and repellency of some plant extracts applied as individual and mixed extracts against termites (*Macrotermes bellicosus*), Journal of Entomology and Zoology Studies. 2016;4(1):406-418.
7. Dhankhar S, Kaur R, Ruhil S, Balhara M, Dhankhar S, Chhillar AK. A review on *Justicia adhatoda:* A potential source of natural medicine, African Journal of Plant Science. 2011;5(11):620-627.
8. Isman MB. Botanical insecticides, deterrents and repellents in modern agriculture and an increasingly regulated world, Ann. Rev. Entamol. 2006;51:46-66.
9. Lahiri PK, Prahdan SN. Pharmacological investigation of Vasicinol- an alkaloid from *Adhatoda vasica* Nees. Indian J Exp. Biol. 1964;2:219-223.
10. Mao L, Henderson G. Anti-feed ant activity and acute and residual toxicity of alkaloids from *Sophora flavescens* (leguminosae) against formosan subterranean termites (Isoptera: Rhinotermitidae). J Econ Entomol. 2007;100(3):866-870.
11. Mona FA, Yasser AE. Toxicity and biochemical efficacy of six essential oil against *Tribolium confusum* (Du Val) (Coleoptera: Tenebrionidae). Egypt academic journal of biological science. 2009;2(2):1-11.
12. Morgan, ED. Azadirachtin, a scientific gold mine, Bioorganic Med. Chem. 2009:17:4096-4105.
13. Mugerwa S, Mpairwe D, Zziwa E, Swaans K, Peden D. Integrated termite management for improved rainwater management: A synthesis of selected African experiences. NBDC Technical Report 9. Nairobi, Kenya: ILRI; c2014, 33.
14. Nair M, Shincymol VV, Ansary PY. Preliminary phytochemical analysis of leaves of guchakaranja (*Quassia indica/Samadera indica* gaertn). International Journal of Ayurveda and Pharma Research. 2018;6(9):43-48.
15. Okugava H, Ueda R, Matsumoto K, Kawanishi K, Kato A. Effects of agar wood extracts on the central nervous systems in mice. Planta Med. 1993;59:32-36.
16. Osipitan AA, Oseyemi AE. Evaluation of the bio-insecticidal potential of some tropical plant extracts against termite (Termitidae: Isoptera) in Ogun state, Nigeria Journal of Entamology. 2012;9:257-265.
17. Osipitan AA, Jegede TO, Adekanmbi DI, Ogunbanwo

IA. Assessment of *Datura metel*, local soap and garlic (*Allium sativum*) in the management of termite (Termidae: Isoptera). Munis Entomology and Zoology. 2013;8(1):407-414.
18. Pathak RP. Therapeutic Guide to Ayurvedic Medicine (A handbook on Ayurvedic medicine) Shri Ramdayal Joshi Memorial Ayurvedic Research Institute. 1970;1:121.
19. Pettersen RC. The Chemical Composition of Wood. In: RM Rowell (ed), The Chemistry of Solid Wood. Madison: American Chemical Society Washington DC, USA; c1984. p. 57-126.
20. Saljoqi AR, Khan MA, Huma Z, Sattar A, Ullah M, Khan F. Behavioral Changes of *Heterotermes indicola (*Isoptera: Rhinotermitidae*)* Against Some Natural Products. Pakistan J Zool. 2012:44(6):1613-1622.
21. Sileshi A, Sori W, Dawd M. Laboratory evaluation of entamopathogenic fungi *Metarhizium anisopliae* and *Beuvaria bassiana* against termites, *Macrotermes* (Isoptera: Termitidae), Asian journal of plant sciences. 2013;12(1):1-10.
22. Sileshi G, Kuntashula E, Matakala P, Nkunika PO. Farmers' perceptions of pests and pest management practices in agroforestry in Malawi, Mozambique and Zambia. Agrofor. Syst. 2008;72(2):87-101.
23. Sohail A, Mazhar IZ, Abid H, Muhammad AR. Muhammad S. Evaluation of plant extracts on mortality and tunnelling activities of subterranean termites in Pakistan. Pesticides in the modern world pest control and pesticides exposure and toxicity assessment. 2011;1:39-54.