

THE EFFECT OF SOME PLANTS EXTRACTS AND ESSENTIAL OILS ON THE WORKERS OF TERMITES LABORATORY *MICROCEROTERMIS GABRILES* (ISOPTERA: TERMITIDAE)

Sanaa Jameel Thamer

Department of Biology , College of Sciences, University of Basrah, Basrah, Iraq

(Received, Accepted)

Keywords: Plant Extract, Mortality, Oil.

ABSTRACT

The activity of water and ethanol extracts of *Ruta graveolens* and *Cuminum* and the essential oils of the groups A and H were evaluated on the termites workers of *Microcerotermis gabriles* in the laboratory with different concentrations.

Result showed that ethanol extracts of *R. graveolens* , *C. cyminum* were the most effective in mortality significant differences from other extracts in filter paper treatment which cause 71.6%, 73% respectively in concentration 10% in 24 hour and increase to 100% after 72 hour of treatment Direct method contact showed that powder of *C. cyminum* was the most effective in mortality with rates 66.6 % for period 10 minutes and increase to 100% after 72 hour of the same period. All tested plants extracts was repellent to termites and the essential oil of *C. cyminum* was the effective once.

INTRODUCTION

The termites (Isoptera) is a social highly organized insects, live in colonies as a nest in soil or woods , and it was a pest when they damage products of value to man: timbers used for buildings , transmission poles, trees and plants in agriculture (1).

Many commercial termiticides are potential environment contaminants with long persistence , therefore ,recently there is interested directed toward plants compounds as alternatives of insecticides in pest control programs because of their low mammalian toxicity and safe to environment (2) .some studies have referred that some plants have anti-termic properties such as the vetiver oil (3) , the leaves of *Flourensia cernua* (4) ,the plant *Azadirachta excelsa* (5) and the plant *Calotropis* sp (6) .

Regarding the environmental importance of termites, the important of some plants as a cheap source of natural pesticides and the little studies of testing the activity of some plants against termites, therefore our study was to test the effect of some plants extracts on these insects.

MATERIALS AND METHODS

Collecting and classification of insects samples :

The collecting of termites was done as described by Myles (7) by using plastic tubes (30 cm length and 15 cm diameter) with plastic cover.

A pieces of rolled carton were added to it .the tubes were buried in the soil that surrounds the infected trees with termites and wetting the soil with water ,left in soil for 3-4 weeks , after that the traps were checked if infected and the cartoon containing termites incubated in 25- 30 c under dark condition with highly humidity.

All the specimens were prepared and identified in biology department , collage of sciences.

Preparation of plants extracts :

water extracts:

Twenty grams of each dried plants (*Ruta graveolens* or *Cuminum cyminum*) was weighted and added to 200 ml of distilled water in a temperature 60 c .The mixture was mixed with electrical mixer for 5 minutes , then filtered by cloth with small pores , the filtered was separated by centrifugation in a speed 3000 course/minute for ten minutes, the extracts was put in Petri dishes in oven in 45c for 18 hours for drying (8).All extracts were kept as a powder in refrigerator until use.

Alcohol extracts :

Twenty grams of milled dried plants was taken using rolled filter paper and put in soxhlet extractor in 50 c with 200 ml of ethanol for 24 hours . The operation was repeated many times in order to get enough amount of extracts , the extracts was dried by oven in 45 c for one hour (8) and kept in refrigerator until use.

The essential oils:

The groups (A ,H) essential oils was obtained from the researches center of biology department / collage of sciences /Quait university , and used in the current study in concentration 10%,5% and 1%.

Group A: containing the essential oils of plants: *Artemisia*,
Ocimum, *Citrus*, *Mintha* and *Patchouli*

Group H : containing the essential oils of plants : *Cinnamon* , *Citronella*, *Eucalyptus* , *Mintha*,
Thymus .

Preparing the concentration

Stock solution was prepared for each plant extract by weighting 2.5 mg and melting in 25 ml of sterilized distilled water to get a solution of 10 % concentration and all concentration was prepared from it 10%, 5%, 1% .

Extraction essential oil from *Cuminum cyminum*

Fifty grams of powder was put in a flask containing 500 ml of distilled water and exposed to source of heat . the rising steam from the sample was condensed by condenser connected with glass cylinder to collect the resultant water of the evaporation .the oil layer accumulating on the surface of water and obtained by separating funnel (8) , the oil was kept in refrigerator until use.

Testing plants extracts

Forice feeding (filter paper treatment) for termites

Filter paper (Whateman No.1: diameter :5.5cm) was treated with 0.5 ml of water or ethanol extracts of each plant in the concentration 10%, 5% and 1% in three replication of each concentration .the paper left drying and then put in a plastic Petri dish with diameter 5.5 cm and 20 workers (termites or ants) were enter in it , untreated filter paper was stuck to the lid of each Petri dishes and wetted in amount of distilled water , the dishes moved to into the incubator in 25-30 c .

the percentage of killed insects was scored after the 24,48 and 72 hours with noticing wetting the papers in the lid of each dishes to keep up the moisture .

In order to know the conduct of of termites to consuming the treated papers , 0.5 ml of Neutral red was added with concentration of 0.1 % to each treated paper , It is possible to see the coloring of the guts of termites with red when feeding (9). In the control it consist of two treatments : the one : filter papers treated with distilled water and the other : filter papers treated with distilled water and Neutral red . To know the percentage of death insects in case of non-feeding on the filter papers treated with plants extracts , starvation treatment for insects was used with 1gm of sterilized sand instead of filter papers, the sand wetted with water as a source of moisture.

Direct contact method

Soft layer of plant powder was spreaded on a plastic Petri dish (5.5 cm) , 20 workers of termites were entered into dish and they were left to move on it in different periods of times 1, 5 and 10 minutes separately for each period with three replicates for each time. After that the dish was shaken to cover the insects with powder ,then it was emptied on filter paper to let the insects shed the surplus powder . the insects were taken into plastic dishes containing clean filter paper , they wetted and incubated in 25-30 c , the percentage of dead termites was scored after 24, 48 and 72 hours .(10) .

Repellent and attractive effects of plants extracts:

Filter paper of diameter 9 cm divided into two equal halves were used .the firs half was treated with 0.5 ml of plant extract apart ,the second half was left with out treatment . After the drying the treated half it was put with untreated one in a plastic Petri dish (9cm) and 20 workers of insect were entered into it with three replicates for each extracts .The number and location of the existing insects on the treated half were scored during the periods 15,30 ,45,60,90,120,180,240 minutes . repellent or attractive effect of plants extracts depending on the number of existing insects on the treated half of filter paper , in case the number less than 50 % of insects number , the extract was repellent and if the number more than 50% , the extract was attractive .(11) . in control : one half of filter paper was treated with distend water and the other with out treatment.

Statistical analysis:

The percentage data were corrected by Abbot formula (Schneider and Oraell formula) (12) and analyzed by general liner modile (ANOVA) , the means was compared by R.L.S.D in $p < 0.01$ (13).

RESULT AND DISCUSSION

Forice treatment for termites :the result of statistical analysis in a level of possibility ($p < 0.01$) shape 1: referred the high effect of ethanol extract of *Ruta graveolens* to cause high mortality in the workers of termites with different significant from the other extracts , the mortality rate of the concentrations 10%,5% was 71.6% ,35% after 24 hour and increased to100% after 72 hour for treatment in the same concentration .followed by the ethanol extract of *Cuminum cyminum* with mortality rate 73%, 13.3 % for concentration 10% ,5% after 24 hour and reached to 100% ,57.8%

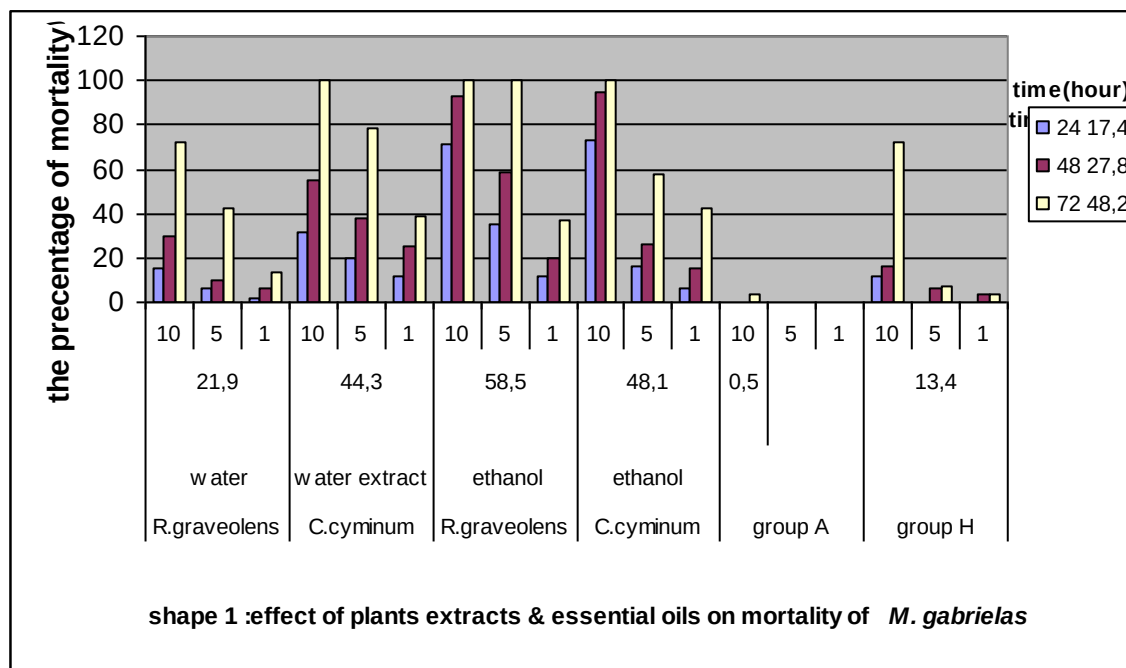
for same concentration after 72 hour .the ethanol extract of *Cuminum cyminum* did not different from it s water extract in mortality rate which was 100% ,78.9% for concentration 10%, 5 % after 72 hour of treatment . the water extract of *Ruta graveolens* and the essential oils of group H scored lesser effectiveness in mortality rate of termites which was 71.9% ,71.8% for concentration 10% respectively after 72 hour of treatment , while the essential oils of the group A was very low effect in mortality with rate 3.5 % for concentration 10% after 72 hour.

It was clear from the statistical analysis for the interfere among the extracts ,concentrations and the times referred to significant interfere between them and increase mortality rate of workers of termites in concentration 10% after 72 hours from treatments except the essential oils of the group A which have the lest effect in mortality . the result of statistical analysis of interfere referred to significant differences between the extracts and concentration, the extracts and times and between the concentration and the times.

The filter papers treated with water or ethanol extracts of showed no feeding activity of termites on the treated papers by noticing the non color of their guts with Neutral red so that the effectiveness of the extracts was either from the repellent or deterrent effect but it does not cause mortality in workers as it was showed in control starvation which the mortality was very low (14,15) so the effect would be done by a direct toxic effect to insects (15,16).The filter papers treated with essential oils of the group A or H showed feeding activity of termites therefore the high effect of the essential oils of the group H is due to either on effect on the microorganisms in the gut or on the epithelial cells of the intestine in addition to the direct toxic effect to insects . the lower effect of the essential oils of group A may be related to differences in nature of compounds of the oil which consequently difference in effect.

The different effect of ethanol extracts from the water extract related to the type of solvent used in extract therefore different in the types of compounds extracted from the plants (8) . the study(15)showed that the water, ethanol extracts of *Nicotina tabacum* cause 100% mortality to workers of *M.diversus* in filter paper treatment in concentration 1%, followed by ethanol acetate extract of *Nerium oleander*, hexan of *Ricinus communis* ,ethanol of *Citrullus colocynthis* , crude extract of *Melia azedarach* which cause 96.1 % ,97.3%,89.74%,and 96.04%.

In a study (17) about the effect of water extracts of leaves , flowers of *Polygonum hydropiper* on the workers of termites *Heterotermes indicola* , *Coptotermes heimi* in treating of filter paper for 10 days which cause mortality rate 28%, 52% for first species and rate 28%, 74.7% for the second species ,in the control the mortality rate was 10.7%, 12% , while the extracts of leaves ,seeds of *Cannabis sativa* cause mortality rate 54.7%, 64% for *H. indicola* and 58.7% ,70.7% for *C. heimi* in filter paper treatment for 10 days.



R.L.S.D for extracts ($p < 0.01$) = 4.10

R.L.S.D for concentration ($p < 0.01$) = 2.9

R.L.S.D for times ($p < 0.01$) = 2.9

R.L.S.D for extracts x concentration ($p < 0.01$) = 7.11

R.L.S.D for extracts x times ($p < 0.01$) = 7.52

R.L.S.D for concentration x times ($p < 0.01$) = 5.34

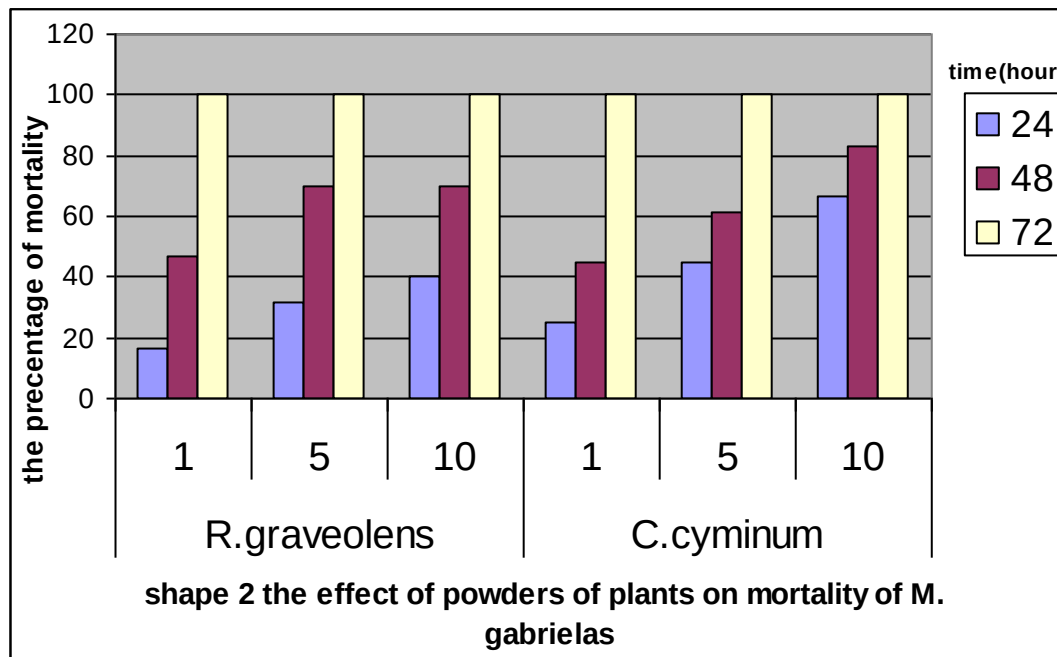
R.L.S.D for extracts x concentration x times ($p < 0.01$) = 13.57

Direct contact method: the result of statistical analysis in a

level of possibility ($p < 0.01$) shape 2 : referred the high effect of the powder of *Cuminum cyminum* which cause high mortality in the workers of termites with different significant from the powder of *Ruta graveolens*, the mortality rate (treatment for 10 minutes) was 66.6% and increase 83.3% after 48 hour and 100% after 72 hour. the powder of *Ruta graveolens* cause mortality rate 40% after 24 hour and increase to 70% ,100% after 48 and 72 hours of treatment.

The statistical analysis showed significant differences between the exposure periods of the powders to the workers and significant differences between the times 24, 48 and 72 hours.

The high effect of the tested plants powders may be due to the toxic effect by contact to the surface of the body of insects and penetration of compounds to the cuticles or spiracles and effect to nervous or digestive system (18).the study (15) showed that powders of *N.Tabacum*, *N. oleander*, and *R.communis* cause 100%, 98.7%,and 92.4% mortality after 72 hour of treatment with powder for 10 minutes.



R.L.S.D for extracts ($p < 0.01$) = 3.39

R.L.S.D for exposure periods ($p < 0.01$) = 5.44

R.L.S.D for times ($p < 0.01$) = 5.44

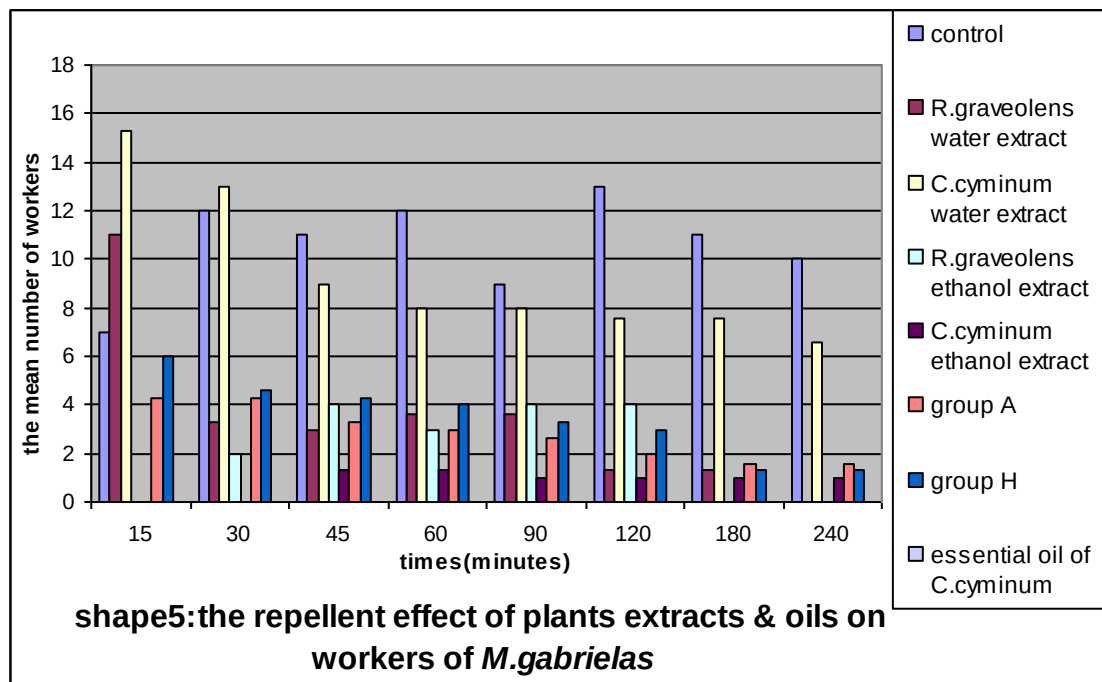
R.L.S.D for extracts x exposure periods ($p < 0.01$) = 6.40

R.L.S.D for extracts x times ($p < 0.01$) = 7.72

R.L.S.D for exposure periods x times ($p < 0.01$) = 9.42

Repellent effect of plants extracts :

The result shape 3 showed all tested plants have repellent effect against termites and the most effective was the essential oils of *Cuminum cyminum* which no rate was scored for insects extence on the treated part of the filter paper and cause mortality to insects on the untreated filter paper. Followed by the water and ethanol extracts of *Ruta graveolens*, *Cuminum cyminum* ,the rate of insects extence on the treated part was 1%,0% for termites and 2% ,0% for ants after 4 hours of treatment , and the essential oils of the group H and A was 1.6%,1.3% for termites and 2% ,0% for ants after the same of exposure time . the activity of the essential oils as insecticides Against termites was suggest from some studies (3) and some essential oils extracted from cedar wood (19) and from *Lisea cubeba* (20) and *Cinnamemum sp.* (21) was repellent to termites ,and many essential oils containing terpenoids like citronellol ,citral,geraniol ,eugenol have repellent properties to termites (22), the terpenoides in the vetiver oil was repellent to termites (23) , crude extract of *Nerium oleander* was repellent to *Microcerotermes. diversus* (15) .



تأثير بعض المستخلصات النباتية والزيوت العطرية في نسب هلاك عاملات حشرة الارضه *Microcerotermus gabrielis* (Isoptera:termitidae) مختبريا

سناء جميل ثامر

قسم علوم الحياة، كلية العلوم ، جامعة البصرة ، البصرة ، العراق

الخلاصة

أختبرت فعالية المستخلصات المائية والكحولية للسذاب والكمون ومستخلصات الزيوت العطرية للمجموعتين A و H في عاملات حشرة الارضه مختبريا وبتركيز مختلفه . وقد بينت النتائج وجود تأثير لهذه المستخلصات في هلاك عاملات الارضه ، فقد تفوق المستخلص الكحولي لكل من السذاب والكمون وبفارق معنوي عن بقية المستخلصات في احداث الهلاكات العاليه للعاملات عن طريق معاملة اوراق الترشيح ونسب هلاك 71.6% و 73% على التوالي للتركيز 10% خلال 24 ساعه وارتفع التأثير الى 100% بعد 72 ساعه من المعامله. ولطريقة معاملة تلويث العاملات بالمساحيق النباتيه فقد تفوق مسحوق الكمون في الهلاكات والتي بلغت 66.6% للفترة 10 دقائق وارتفعت الى 100% لنفس الفتره بعد 72 ساعه . واطهرت جميع النباتات المختبره تأثير طارد لعاملات الارضه وكان اكثرها فعاليه الزيت العطري (الطيار) للكمون اذ لم تسجل اي نسبه لتواجد العاملات على الجزء المعامل.

REFERENCES

1. Al-Alawy, Sady Abd –Al mehsin (1987). Classification and ecological studies of termites in Iraq, Phd. Atheists , collage of agriculture ,Baghdad university .323 pp.(Arabic)
2. Cornelius, M. L. ; Grace, J. K.; Ford, P. W. and Davidson, B. S. (1995).Toxicity and repellency of semiochemicals extracted from adichoderine ant (Hymenoptera : Formicidae) to the Formosan subterranean termites (Isoptera : Rhinoteimitidae). Environ. Entomol. 24 : 1263 – 1269.

3. Zhu, B.C.R., G. Henderson, F. Chen, H.X. Fei, and R.A. Laine. 2001a. Evaluation of vetiver oil and seven insect-active essential oils against the Formosan subterranean termite. J. Chem. Ecol. 27: 1617-1625.
4. Tellez, M., Rick Estell, Ed Fredrickson, et al. 2001. Extracts of *Flourensia cernua* (L): Volatile Constituents and Antifungal, Antialgal, and Antitermite Bioactivities, Journal of Chemical Ecology, 27(11): 2263-2273
5. Sajap, A. S.; Aloysius, F. 2000. Effects of leaf extracts of *Azadirachta excelsa* on *Coptotermes curvignathus* (Isoptera: Rhinotermitidae), Sociobiology, 36(3): 497- 503
6. Singh, D. and A-K. Singh.(1991). Repellent and insecticidal properties of essential oils against house fly, *Musca domestica* L. Insect.Sc.Applic.12:487-491.
7. Myles, T.G.(1994). Use of disodium octoborate tetrahydrate to protect aspen wafer-board from termites. Forest Products Journal 44,33-36.
8. Harborne, J. B. (1984). Phytochemical methods. Chapman and Hall. New York. 2nd ed. 288 pp.
9. Blaske, V. -U. and Hertel, H. (2001). Repellent and toxic effect of plant extracts on subterranean termites (Isoptera : Rhinotermitidae). J. Econ. Entomol. 94 : 1200 – 1208.
10. Pearce, M. J. (1997). Termites : Biology and pest management. CAB International USA. 172 pp.
11. Hutchins, R. A. (1997). Evaluation of natural antitermitic properties of *Aleurites fordii* (Tungtree) Extracts. J. Mississippi Academy Sci., 42: 165-172.
12. Abbot, C. I. (1925). A method of computing the effectiveness of an insecticide. J. Econ. Entomol., 13: 65 – 67.
13. Al-Rawi, Khasha Mahmud and Abd Alaziz ,Khalf Ala(1980). Statistical analysis of agriculture. dar al kutub for printer. Mohosal university. 488pp.(Arabic)
14. Grace, J. K. ; Wood, D. L. and Frankie, G. W. (1989). Behavior and survival of *Reticulitermes Hesperus* Banks (Isoptera : Rhinotermitidae) on Selected sawdust and wood extracts. J. Chem. Ecol. 15: 129-139.
15. Thamer, S.J.(2005). Studying the effect of some plants extracts against workers of subterranean termites *Microcerotermes diversus* silvestri (1901). MSc. a thesis , collage of sciences , university of Basrah.(Arabic)

16. Bultman, J. D.; Beal, R. H. and Ampong, F. F. K. (1979). Natural resistance of some tropical African woods to *Coptotermes formosanus* shiraki. Forest Prod. J. 29 : 46 – 51.
17. Badshah, H., Khan, A. S., Farid, A. and Khan, A. (2005). Toxic effects of *Palpoluck Polygonum Hydropepper* L. and Bhang *Cannabis sativa* L. plants extracts against termites *Heterotermes indicola* (Wasmann) and *Coptotermes heimi* (Wasmann) (Isoptera: Rhinotermitidae). Songklanakarin J. Sci. Technol., 27(4): 705-710.
18. Al-saady, T. A. a. (2001). The effect of some plants extracts on the survival and production of adult of *Callosobrachus maculatus* (Forbicius) (Coleoptera : Bruchidae). Msc. degree thesis, Agriculture collage, Basrah university, 85pp. (Arabic)
- 19- Grace, J. K. and R. T. Yamamoto. 1994. Natural resistance of Alaska-cedar, redwood, and teak to Formosan subterranean termites. Forest Prod. J. 44(3): 41-45.
- 20- Lin, T-S. and H-W Yin. 1995(a). Effects of *Litsea cubeba* press oils on the control of termite *Coptotermes formosanus* Shiraki. Taiwan For. Res. Inst. New Series 10: 59-63.
- 21- Lin, T-S. and H-W Yin. 1995(b). The effects of *Cinnamomum spp.* oils on the control of the termite *Coptotermes formosanus* Shiraki. Taiwan For. Res. Inst. New Series 10: 459-464.
- 22- Cornelius, M. L. and Grace, J. K. (1997). Effect of termite soldiers on the foraging behavior of *Coptotermes formosanus* (Isoptera : Rhinotermitidae) in the presence of predatory ants. Sociobiology 29 : 247 – 253.
19. Henderson, G., D. O. Heumann, R. A. Laine, L. Maistrello, B. C. R. Zhu, F. Chen. 2001. Extracts of vetiver oil as a repellent and toxicant to ants, ticks and cockroaches. US Patent disclosure, serial number 09/932,555.