Available online on www.ijtpr.com
International Journal of Toxicological and Pharmacological Research 2017; 9(3); 199-205

ISSN: 0975-5160

Research Article

Analysis of Bioactive Chemical Compounds of *Trogoderma granarium* (Insecta: Coleoptera: Dermestidae) Using Gas Chromatography – Mass Spectrometry

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Available Online:25th July, 2017

ABSTRACT

Methanolic extract of bioactive compounds of *Trogoderma granarium* was assayed. GC-MS analysis of *Trogoderma granarium* revealed the existence of the Pentanoic acid , 1,1-dimethylpropyl ester , (1H)-Pyrimidinone , 5-chloro-4,6-diphenyl, Cyclobutanemethanol , α-methyl- , Nitro-2-methyl-1,3-propanediol , Hydroxylamine ,O-(2-methylpropyl)- , Uridine , 2',3'-O-(phenylmethylene)- ,Acetic acid ,2-benzoylthio-,2-oxo-2-phenylethyl ester , methylpropyl)- , Uridine , 2',3'-O-(phenylmethylene)- , 5'-(4-methylbenzenesulfo , Indolinol , 1-benzoyl-, Benzeneethanol , β-methyl-,(s)- , Acetic acid ,2-benzoylthio-,2-oxo-2-phenylethyl ester , Phenacyl thiocyanate , Deoxy-L-ribose-2,5-dibenzoate , Methenamine , Alanine , N-methyl-n-propargyloxycarbonyl-, decyl ester , Benzoyl chloride , Thiophene-2-ol , benzoate , Ethanone , -(5-nitrotetrazol-2-yl)-1-phenyl- , 2,5-Dimethylhexane-2,5-dihydroperoxide , Benzamide , N-(3-benzylthio-1,2,4-thiadiazol-5-yl)- , Methyl p-(2-phenyl-1-benzimidazolyl)benzoate , Methyl-2-phenoxyethylamine , Pentaborane(11) , cis-Methoxy-5-trans-methyl-1R-cyclohexanol , Nitro-1-phenyl-3-(tetrahydropyran-2-yloxy)propan-1-one , cis-Methoxy-5-trans-methyl-1R-cyclohexanol. *Trogoderma granarium* produce many important secondary metabolites with high biological activities.

Keyword: gas chromatography – mass spectrometry, bioactive chemical compounds, *Trogoderma granarium*.

INTRODUCTION

The khapra beetle, *Trogoderma granarium* Everts, is one of the world's most feared stored-product pests. In fact, it has been described as one of the 100 worst invasive species worldwide[1,2]. *Trogoderma granarium* is a serious pest of stored products under hot dry conditions. Reproduction may be so rapid that larvae are found in large numbers in the surface layers of binned grain. Its discovery in a non-infested area usually leads to an immediate quarantine of suspected goods and an expensive eradication and control effort[3-9]. *T. granarium* contaminates non-host cargo and packing. The insect also infests spices, dried gums, seeds, dried fruits and other dried proteinaceous plant materials. The Khapra beetle occurs in very low numbers and can survive for a long period as an inactive state. Larvae feed on a wide variety of stored products and dried foods[10-19]. They prefer whole grain and cereal products such as wheat, barley, and rice, but larvae have been recorded on the following: oats, rye, corn, dried blood, dried milk, fishmeal, ground nuts, flour, bran, malt, flax seed, alfalfa seed, tomato seed, pinto beans, black-eyed cowpeas, sorghum seed, grain straw, alfalfa hay, noodles, cottonseed meal, dried fruits, lima beans, coconuts, garbanzos, lentils, powdered yeast, and many others. Because of its refuge seeking behavior, it is extremely difficult to eradicate this insect from premises or transport facilities, and the existence of a special type of recurrent dormancy in the larval stage enables populations to exploit times of food abundance and to survive long periods of deprivation[20-25]. In addition, this pest appears relatively tolerant to insecticides and many fumigants especially at larval stage and has been steadily increasing its geographic range during the last century[26]. Established infestations are difficult to control because of the beetle's ability to live without food for long periods of time and to survive on foods of low moisture content. These beetles tend to crawl into tiny cracks and crevices and remain there for long periods, making them relatively tolerant to many surface insecticides and fumigants[27].

MATERIALS AND METHODS

Laboratory culture

Larvae (3 mm length) of *T. granarium* were obtained from same-age rearing. Insects were fed with soft wheat flour and beer yeast (95:5) and incubated in darkness at a constant temperature of 30°C and a relative humidity of 70%. Parent adults were provided by the college of science for woman university of Babylon[28-33]. The extraction was performed by adding 100 ml methanol to the whole body insect powder. Methanol was used as solvent control.

Gas chromatography – mass spectrum analysis

The GC-MS analysis of the *Trogoderma granarium* extract was made in a (Agilent 789 A) instrument under

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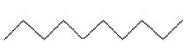
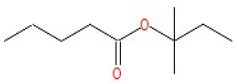
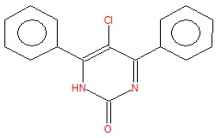


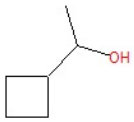
Decade
Rt=3.247
Mw=142.172151



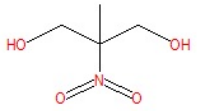
Pentanoic acid , 1,1-dimethylpropyl ester
Rt=3.356
MW=172.14633



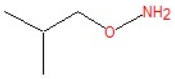
2(1H)-Pyrimidinone , 5-chloro-4,6-diphenyl-
RT=3.636
MW=282.05599



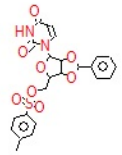
Cyclobutanemethanol , α-methyl-
RT=3.739
MW=100.088815



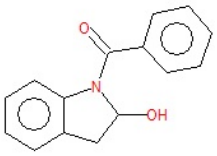
2-Nitro-2-methyl-1,3-propanediol
RT=3.813
MW=135.053158



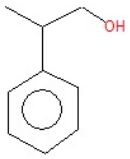
Hydroxylamine ,O-(2-methylpropyl)-
RT=4.311
MW=89.084064



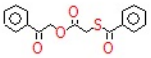
Uridine , 2',3'-O (phenylmethylene)- , 5'-(4-methylbenzenesulfo
RT=5.582
MW=486.109686



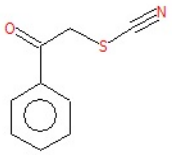
2-Indolinol , 1-benzoyl-
RT=5.725
MW=239.094628



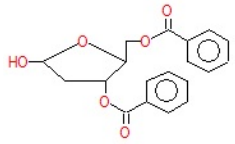
Benzeneethanol , β-methyl-,(s)-
RT=6.045
MW=136.088815



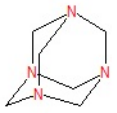
Acetic acid ,2-benzoylthio-,2-oxo-2-phenylethyl ester
RT=6.159
MW=314.06128



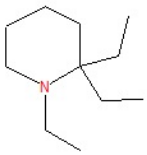
Phenacyl thiocyanate
RT=6.514
MW=177.024835



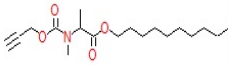
3-Deoxy-L-ribose-2,5-dibenzoate
RT=6.571
MW=342.110338



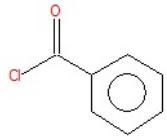
Methenamine
RT=6.755
MW=140.106196



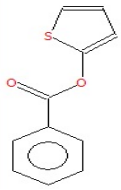
2,2-Diethyl-N-ethylpiperidine
RT=6.743
MW=169.18305



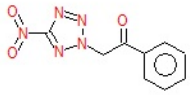
Alanine , N-methyl-n-propargyloxycarbonyl-, decyl ester
RT=6.852
MW=325.22531



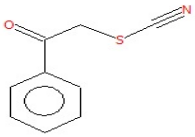
Benzoyl chloride
Rt=6.995
Mw=140.0028925



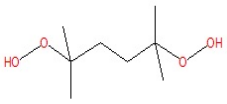
Thiophene-2-ol , benzoate
RT=7.264
MW=204.024501



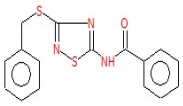
Ethanone , -(5-nitrotetrazol-2-yl)-1-phenyl-
RT=7.767
MW=233.054889



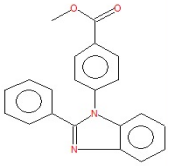
Phenacyl thiocyanate
RT=8.082
MW=177.024835



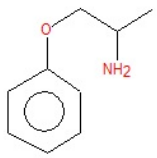
2,5-Dimethylhexane-2,5-dihydroperoxide
RT=8.174
MW=178.120509



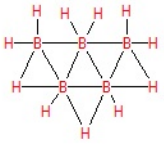
Benzamide , N-(3-benzylthio-1,2,4-thiadiazol-5-yl)-
RT=8.511
MW=327.050005



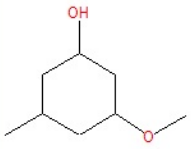
Methyl p-(2-phenyl-1-benzimidazolyl)benzoate
RT=8.700
MW=328.121178



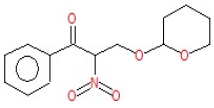
1-Methyl-2-phenoxyethylamine
RT=9.123
MW=151.099714



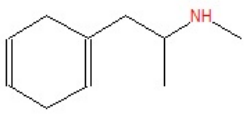
Pentaborane(11)
RT=9.066
MW=66.132602



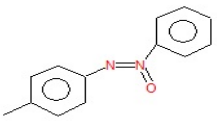
3-cis-Methoxy-5-trans-methyl-1R-cyclohexanol
RT=9.015
MW=144.115029



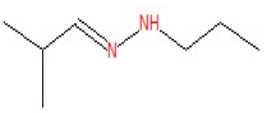
2-Nitro-1-phenyl-3-(tetrahydropyran-2-yloxy)propan-1-one
RT=9.392
MW=279.110672



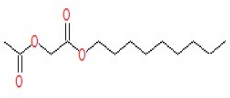
1-(1,4-cyclohexadienyl)-2-methylaminopropane
RT=9.638
MW=151.1361



1-Phenyl-2-(4-methylphenyl)-diazene 1-oxide
RT=10.520
MW=212.094963



Isobutyraldehyde , propylhydrazone
RT=11.498
MW=128.131349



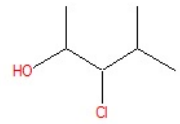
Acetoxyacetic acid ,nonyl ester
RT=11.727
MW=244.1674595

computer control at 70 eV about 1µL of the methanol extract was injected into the GC-MS using a micro syringe and the scanning was done for 45 minutes. The time from when the injection was made (Initial time) to when elution occurred referred to as the Retention time (RT). Helium gas was used as a carrier as well as an eluent. The flow rate of helium was set to 1ml per minute[34-71]. Compounds were identified by comparing their spectra to those of the Wiley and NIST/EPA/NIH mass spectral libraries.

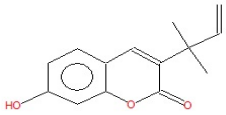
RESULTS AND DISCUSSION

Gas chromatography and mass spectroscopy analysis of

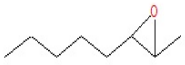
, 1,1-dimethylpropyl ester , (1H)-Pyrimidinone , 5-chloro-



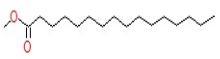
2-Pentanol , 3-chloro-4-methyl -,(R*.R*)-(+)
Rt=11.967
Mw=136.065493



7-Hydroxy-3-(1,1-dimethylprop-2-enyl)coumarin
RT=12.425
MW=230.094295

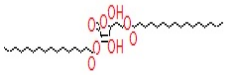


Trans-2,3-Epoxyoctane
RT=13.375
MW=13.375
MW=128.120115

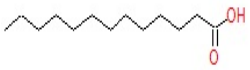


Hexadecanoic acid , methyl ester
RT=14.628
MW=270.25588

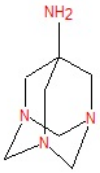
Cyclopentaneundecanoic acid , methyl ester
RT=14.908
MW=268.24023



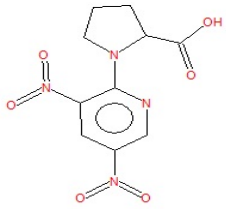
l-(+)-Ascorbic acid 2,6-dihexadecanoate
RT=15.263
MW=652.49142



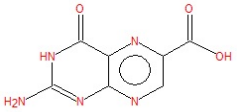
Tridecanoic acid
RT=15.389
MW=214.19328



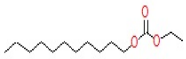
7-Amino-1,3,5-triazaadamantane
RT=15.618
MW=154.121846



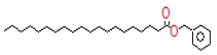
N-[3,5-Dinitropyridin-2-yl]proline
RT=15.698
MW=282.060034



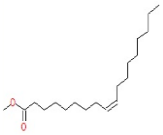
Pterin-6-carboxylic acid
RT=15.601
MW=207.039239



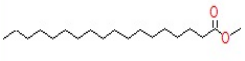
Ethyl undecyl carbonate
RT=15.864
MW=15.864



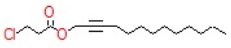
Eicosanoic acid ,phenylmethyl ester
RT=16.047
MW=402.349781



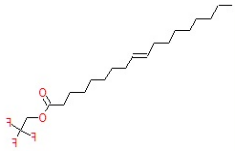
9-Octadecenoic acid ,(Z)-, methyl ester
RT=16.247
MW=296.27153



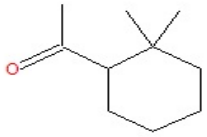
Methyl stearate
RT=16.470
MW=298.28718



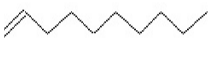
3-Chloropropionic acid , tridec-2-ynyl ester
RT=16.648
MW=286.169958



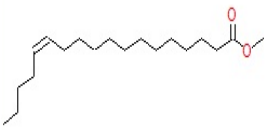
9-octadecenoic acid ,2,2,2,-trifluoroethyl ester
RT=16.745
MW=364.258915



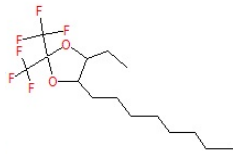
Ketone , 2,2-dimethylcyclohexyl methyl
RT=15.973
MW=154.135765



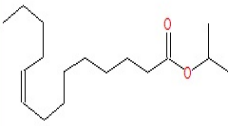
2-Undecene , (Z)-
RT=16.087
MW=154.172151



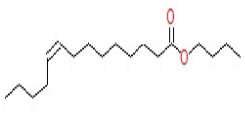
Cis – 13-Octadecenoic acid , methyl ester
RT=16.253
MW=296.27153



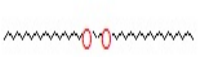
1,3-Dioxolane , 4-ethyl-5-octyl-2,2-bis(trifluoromethyl)-,cis-
RT=17.071
MW=350.168049



i-Propyl 9-tetradecenoate
RT=17.512
MW=268.24023



Butyl 9-tetradecenoate
RT=17.638
MW=282.25588



Octadecane , 1,1'-[1,3-propanediylbis(oxy)]bis-
RT=21.059
MW=580.615833

Table 1. Bioactive chemical compounds identified in methanolic extract of *Trogoderma granarium*.

compounds was carried out in methanolic extract of *Trogoderma granarium*, shown in Table 1. The GC-MS chromatogram of the peaks of the compounds detected was shown in Figure 1. Chromatogram GC-MS analysis of the methanol extract of *Trogoderma granarium* showed the presence of fifty three major peaks and the components corresponding to the peaks were determined as follows. The First set up peak were determined to be Pentanoic acid 4,6-diphenyl, Cyclobutanemethanol , α-methyl- , Nitro-2-methyl-1,3-propanediol , Hydroxylamine ,O-(2-methylpropyl)- , Uridine , 2',3'-O-(phenylmethylene)- ,Acetic acid ,2-benzoylthio-,2-oxo-2-phenylethyl ester , methylpropyl)- , Uridine , 2',3'-O-(phenylmethylene)- , 5'-(4-methylbenzenesulfo , Indolinol , 1-benzoyl-, Benzeneethanol , β-methyl-,(s)- , Acetic acid ,2-benzoylthio-,2-oxo-2-phenylethyl ester , Phenacyl

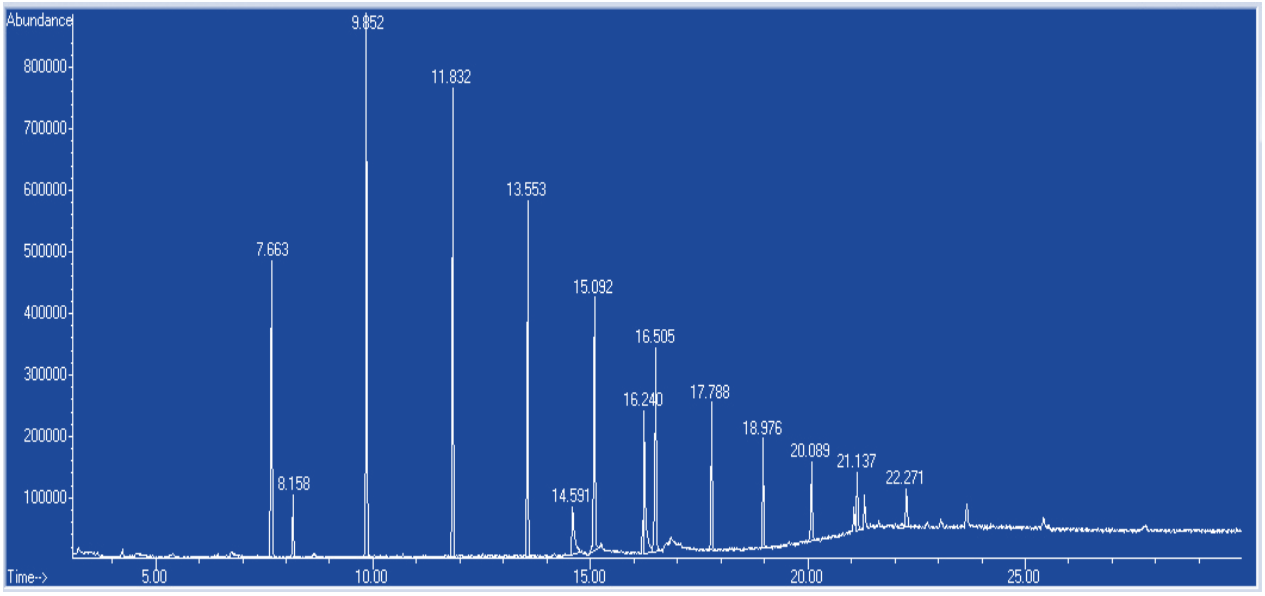


Figure 1: GC-MS chromatogram of methanolic leaves extract of *Trogoderma granarium*

thiocyanate , Deoxy-L-ribose-2,5-dibenzoate , Methenamine , Alanine , N-methyl-n-propargyloxycarbonyl-, decyl ester , Benzoyl chloride , Thiophene-2-ol , benzoate , Ethanone , -(5-nitrotetrazol-2-yl)-1-phenyl- , 2,5-Dimethylhexane-2,5-dihydroperoxide , Benzamide , N-(3-benzylthio-1,2,4-thiadiazol-5-yl)- , Methyl p-(2-phenyl-1-benzimidazolyl)benzoate , Methyl-2-phenoxyethylamine , Pentaborane(11) , cis-Methoxy-5-trans-methyl-1R-cyclohexanol , Nitro-1-phenyl-3-(tetrahydropyran-2-yloxy)propan-1-one , cis-Methoxy-5-trans-methyl-1R-cyclohexanol. The Khapra beetle, *Trogoderma granarium* (Coleoptera, Dermestidae) is considered to be one of the most serious pests of stored grain products, various leguminous crops, rice, oat, barley, and rye throughout the world. It is originally occurred in India, and spread to Africa, Europe, South America and East Asia[71]. According to FAO estimate, 10 to 25% of the world harvested food is destroyed annually due to insects and rodent pests. Losses caused by *Trogoderma granarium* have been reported to range from 0.2 to 2.9% over a period of 1 to10.5 months[72]. Chemical insecticides such as malathian, cypermethrin, bifenthrin are used for rapid control, but are expensive, not readily available and may be poisonous to humans and environment. Moreover, malathian and cypermethrin have gone ineffective due to development of resistance in insect pests of stored grain, particularly in *Trogoderma granarium*[73-75].

CONCLUSION

Fifty three bioactive chemical constituents have been identified from methanolic extract of the *Trogoderma granarium* by GC-MS technique.

ACKNOWLGEMENTS

The authors thank the department of Medical Science in college of nursing for providing all necessary facilities to conduct this study.

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