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Isolation and Identification of volatiles compounds from Rice Weevil, *Sitophilus oryzae* (coleptera: Curculionidae) in Iran with Solid Phase Micro Extraction (SPME) and Headspace Chromatography

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ABSTRACT

Rice weevil *Sitophilus oryzae*(L), cosmopolit Insect which today is scattered in all parts of the world .This pest often makes much damage on rice and other stored grains. Pesticides residue cause restriction use of these chemicals in some places such as warehouses and store locations. As Integrated Pest Management is new strategy for pest control ,Identification of specific volatiles compounds is necessary. Some studies have shown that pheromones compounds of a specific species could have some changes in different environments or nutritional conditions and this is effective at the efficacy of pheromones. This study were conducted to isolation and determination of volatile chemicals by SPME-HS Chromatography. Test Insects grew up in 27 ± 1 Celsius degree temperature and 65 ± 5 percent relative humidity. Volatile extraction were done in four different times duration with three replicates for each of them and virgin weevils used with median age 1-10 days after transferring them to sterile glass vials. SPME fibers contacted to head space of the vials then analyzed by (Gas Chromatography/Mass Spectrometry(GC/MS) system. Detected compounds checked by Willy and NIST database software. Results showed 16 different compounds .most of them were similar with Detected compounds by other researchers but some of them were new. This difference may be for different geographical or nutritional condition, or may be for difference in extraction time. More study is needed for better determination and application of these compounds for IPM.

Key words : Coleoptera, Curculionidae ,Pheromone, *Sitophilus oryzae*, SPME, GC/MS.

INTRODUCTION

There are near one billion hungry in today's world in other view more than five hundred millions of grains annually destroyed due to pests and plants disease .Stored products protection based on Scientific and technical principles is important and economic. Therefore, pest identification and use suitable methods to their control is also noteworthy. Rice weevil is one of the major and dangerous

Pests of rice and some other grains. This pest has 2-3.5 mm length with reddish brown color, there are a lot of pointed pits on its pronotum and four round area with light red color on the elytra are important characters for its definition [1,2]. These pests consume internal endosperm and cause decrease in the quality and quantity of the commodities. Pest monitoring is the first step of their control and it may be done by pheromones or other traps. Pheromones can be used for Integrated Pest Management (IPM) that is a non chemical method for pest control [3,4,5].

Pheromones are a group of volatile compounds that are secreted to the environment by an adult insect and cause a kind of behavior by another adult that receives these materials. These chemicals have low molecular weight and their effects cause inter specific reaction such as sex or aggregation.

These materials are divided into two groups: primary pheromones and releaser pheromones [6]. The pheromones have not any harmful effect for plants or environment. These materials are used as a tool for pest monitoring, mating disruption, alarming and attraction & killing. There are some detected volatile similar between rice weevil and other stored weevils such as wheat and maize weevils and reported one of them is 2-methyl-3-hydroxy-4-pyrone. This compound is isolated by tenax trap [7].

Sitophilus zeamys aggregation pheromone extracted by tenax trap and washing it in hexan solvent [8]. Burkholder et al 1981 extracted rice weevil aggregation pheromone with attractive disks and collected them with sufficient solvent, one reported compound is 5-hydroxyl-4-methyl-3-heptanone. In this study isolation of volatile compounds was done by SPME and they identified with GC/MS.

MATERIALS AND METHODS

Insects Rearing :

Test insects collected from some Tehran local stored places and transferred to laboratory and grown in cylindrical jars with 13 cm height and 5.8 cm diameter. Healthy wheat mixed with a portion of broken them were as food for this growth, with 200 gr for each jar. Growth condition was $27 \pm 1^\circ\text{C}$ temperature and, relative humidity 65 ± 5 percent, light and darkness duration was 16:8.

After five generations virgin adults separated for pheromone and volatiles isolation.

Pheromone Collection :

One to ten days virgin adults selected for volatile extraction and extraction, so emerged adults alienated and held in separated sterile vials. First tests passed in four time treatment with three replicates for each of them. In second tests treatment established based on different volume of adult insects.

The condition of treatments and replicates provided in table 1.

Table 1: Treatment and other conditions in experiments

Vials	size treatment	volume virgin adults	replicates	extraction time
	50	50	3	48
	50	100	3	24
	100	100	3	18
	50	100	3	12

In this study use SPME fibers (polydimethyl siloxane) PDMS with 65 μm film thickness provided from SUPELCO .Selected Insects transferred to some glass sterilized vials and properly sealed with plastic covers. SPME fibers entered to upside of these vials after conditioning in GC injector on 280°C Temperature .

Analytical Methods:

After sufficient contact of fibers with head space of the vials they injected to a GC/MS system for analyses of compounds that may bond with the fiber.GC/MS was HP6890 series with DB5 column Helium was the carrier gas with 1.5 mm/min flow.Temperature program was consist of :Injector 200°C Detector 160 °C column initial temperature was 40°C with 6 min hold ,column temperature ramp with 10 °C /min to 210°C ,and 5 minute hold on 210 °C .Two factors were the base of study,retention time and mass spectra. The survey of detected spectra done by WILEY 275.L and NIST for determination and comparison.

RESULTS AND DISCUSSION

All of results shown I tables 2 and 3,in table 2 detected alkanes can be seen that consists of nine compounds.

Table 2 all of alkanes detected in the study

Compounds	Retention time	P
Octan	6.85	91
Decan	12.67	95
Undecane	15.45	90
Dodecane	18.07	95
Tetradecane	22.84	99
Nonadecane	24.34	98
Pentadecane	27.06	97
Hexadecane	27.76	91
Octadecane	31,02	99

Table 3 Pheromones detected in the study

Group	compounds
A	Hexadecanoic acid/Methyl ester
B	Hexadecanoic acid (Cas) Palmitic
C	2-Hexadecen-1-
D	3,7,11,15-tetramethyl-2-hexadecen-1-ol
E	9,12- octadecadionioic acid
F	7/10/13-Hexadecatrienoic acid /methyl ester
G	Ethyl Linoleolate

Compound A which detected in this study has some similarity with sitophenone that known by Burkholder(1981) ,Schmuff (1984),and Walgenbach(1987) But it has some difference with them That may be from geographical or nutritional position[8,9,10,]. CompoundsD,E,and F are similar with Maltol,pental and vanillin reported by Phillips(1993) but their alkanic chain is a little longer[11] .

The G compound is very similar with aggregation pheromone for granary weevil reported by Phillips.

There is a little different between these two compounds that may be for nutritional condition[12].

CONCLUSION

The results showed there are some difference between determined volatiles in our study and other reports so pheromone determination in different geographical location must be done to detect compounds correctly. Application SPME method for pheromone isolation is a useful and important way that could increase recovery in this studies. This is a fast and environmental friendly method without any use of solvents. In this test the volume of insects and the time of exposure surveyed and showed time must not be more than 12 hours and the best time is 5 hours ,because the test insects begins to dead behavior.

REFERENCES

- [1] R. G. Boot ; M. L. Cox; R.B, Madye. II Guides to insect of Importance to man3. Coleoptera. International Institute of Entomology (An Institute of C.A.B International) the Natural History Museum, **1990**, pp.193-194.
- [2] P. G. Koehler, Rice weevils *sitophilus oryzae* (Col:Curculionidae) IFAS Extension. University of florida available in <http://www.edis.ifas.ufl.edu/IG1994>.
- [3] G.DalBello; S.padin; C.Lopez Lostra; M. Fabrizio. Laboratory evaluation of chemiced-biological control. **2001**.
- [4] B.C. Longstaff. *J. Stored. Prod. Res*, **1981**, 17: 73-76.
- [5] L. M. Hanks; J. G. Millar; J. A. Moreira; J. D. Barbour; E. S. Lacey; J.S. Mcelfresh; F. R. Reuter; A. M. Ray. *J.Chem. Ecol*, **2007**, 33:889-907.
- [6] A. Cork; D.R.Hall; J. L. Mullings; O.T.Jones. A new PVC resin formulation for controlled release of insect pheromones. In: proceedings of 6th International symposium on controlled Release of Bioactive Materials, **1989**, Chicago, 9-10. Lincolnshire, Illinois: controlled Release.Inc.
- [7] J.k. Phillips ; J.F. Andersen; W. E. Burkholder. Determination of the enantiomeric composition of (R*, S*)-1- ethyl propyl-2-methyl-3-hydroxypentanoate. The male- product aggregation pheromone of *sitophilus granaries*. *Experimentalis Applicata*, **1989**, 51, 149-153.
- [8] C.A. Walgenbach; J. K. Phillips; W. E. Burkholder; G.S. King; K.N. Slessor; K. Mori. *J. Chem. Ecol*, **1987**, 13: 2159-2169.
- [9] N. R.Schuff; J.K.Phillips; W.E. Burkholder; H.M. Fales; C.W.Chen; P.P. Roller; M. Ma. *Tetrahedron Lett*, **1984**, 25: 1533-1534.
- [10] W.E. Burkholder. Biomonitoring for stored-product insect in management of insect pests with semiochemicals: concepts and practice. Ed. E. R. Mitchell, **1981**, 4: 29-40 Newyork/ London: plenum 514pp.
- [11] T.W,Phillips; M. R, Strand. Larval secretion and food odors affect orientation in female plo of Tehran dia interpunctella. *Entomol. Exp*, **1993**, Appl. 71: 185-192.
- [12] J.K. Phillips; P. F. Miller; J. F. Andersen; W. E, Burkholde. *Tetraherdron letters*, **1987**, 28: 6145- 6146.s