## PESTICIDAL POTENTIAL OF SOME SELECTED TROPICAL PLANT EXTRACTS AGAINST CALLOSOBRUCHUS MACULATES (F) AND CALLOSOBRUCHUS CHINENSIS (L) (COLEOPTERA: BRUCHIDAE)

RHS Rajapakse\* and D Ratnasekera Department of Agricultural Biology, Faculty of Agriculture, University of Ruhuna, Mapalana, Kamburupitiya, Sri Lanka

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## **ABSTRACT**

Plant oils obtained from leaves and other parts of 20 different plant species were bioassayed under laboratory conditions for their ability to protect stored legumes from damage by cowpea weevil (*Callosobruchus maculatus*) and adzuki bean seed weevil (*Callosobruchus chinensis*). Three plant oil extracts showed some bioactivity, nine plant oil extracts caused significant adult mortality in both species and eight had none. Six plant oil extracts, black pepper, lemon grass, clove seeds, neem, custard apple, and sacred basil inflicted between 41 to 100% egg mortality in both species in the order of 60, 60-67, 70, 90, 91 and 100% respectively.

## INTRODUCTION

The cowpea weevil, *Callosobruchus maculatus*, can reproduce both in the field and in storage. In order to protect legumes in warehouses especially in third world countries we often have recourse to synthetic pesticides with their attendant dangers of user safety, high cost, development of resistant strains and toxic residues. To avoid such undesirable consequences, scientists have focussed their attention on the use of less hazardous practices or substances to protect stored products.

Historical usage of nicotine and pyrethrum has encouraged scientists to focus their attention on alkaloids, flavonoids, terpenoids and other secondary compounds to be used as pest control agents (Harbourne 1977). The effectiveness of plant oils in controlling insect pest infestations in grain legumes has been effectively demonstrated by several researchers in Asia (Mummigatti and Raghunathan 1977; Varma & Pandey 1978). The oils extracted from the kernels of the Mee plant (Madhuca longifolia) significantly lowers the oviposition and egg hatchability of Callosobruchus maculatus in Sri Lanka (Ranasinghe & Dharmasena 1987). Natural compounds are likely to provide effective, environmentally safe, easily biodegradable and narrow specturm pesticides (Attri & Prasad, 1980; Saxena, 1987). Previous research on plants has indicated the efficacy of solvent extracts (Lambert et al 1985) in the preservation of cowpea against insect pests. In this study the results are reported of the laboratory evaluation of the efficacy of the plant oils of 20 different plant species against cowpea weevil (C. maculatus) and adzuki bean seed weevil (C. chinensis) in Sri Lanka.

Twenty different plant species belonging to different families were selected for the present

study (Table 1) based on the following characteristics: relative absence of insect damage, taxonomic closeness to families known to possess biologically active compounds, usage of plants as pesticides in rural agriculture, traditional knowledge as reported by extension workers.

The Cowpea variety MI 35 used in this study were purchased from the Department of Agriculture and handpicked to remove infested seeds and other debris. Seeds that showed no visible signs of weevil attack were separated, bulked and transferred into a kilner jar (IL) covered with fine mesh for aeration and equilibrated at ambient temperature (25-30°C) and RH (70-80%) for 3 weeks to remove all evidence of prior infestation by the cowpea weevil before bioassay.

The culture of *C. maculatus was* collected from domestic stores and reared in the laboratory as described by Olaifa & Erhun, 1988. Adults emerged from this culture were used for the bioassay within 48h of emergence. The remaining insects were used to start a new culture so that cultures with emerging adults are available continuously. A similar procedure was adopted to culture *C. chinensis*.

Fresh green leaves of the selected plants were chopped in a blender, weighed and transferred to conical flasks containing 95% ethanol at the ratio of 10g/150ml. The flasks were held at room temperature for 5 days with occasional daily shaking before filtering the extract through Whatman No. 1 filter paper. The leaf residues were washed twice with 5-ml ethanol, and the pooled filtrate was concentrated to 0.5 ml in a rotary evaporator, weighed and transferred to a volumetric flask. A 10% (w/v) emulsion of the extract was obtained by addition of water containing 0.5% (v/v) Triton X-100 stored in a refrigerator and used within 2 days.

<sup>\*</sup>Corresponding author

Table 1: Toxicity of the Ethanol Extracts of the leaves of 20 plants to C. maculatus and C. chinensis

Plant Species	% Corrected Mortality					
	Day 1		Day 2		Day 3	
	C.c	C.m	<i>C.c</i>	C.m	C.c	C.m
Capsicum frutescens	35	40	45	48	60	60
Myristica fragrans	00	00	00	00	00	00
Piper nigrum	35	40	48	50	55	60
Citrus reticulata	10	12	10	08	12	14
Cymbopogan citratus	50	60	50	60	65	67
Artocarpus heterophyllus	05	08	12	11	15	17
Gliricidia sepium	00	00	00	00	00	00
Eugenia caryophyllata	50	60	60	60	71	70
Ricinus communis	00	00	00	00	00	00
Dillenia retusa	40	44	50	56	50	58
Azadirachta indica	50	60	60	65	80	90
Cajaanus cajan	00	00	00	00	00	00
Cassia occidentalis	06	09	12	13	15	18
Anona reticulata	80	80	80	90	90	91
Mangifera indica	00	00	00	00	00	00
Eupatorium odorantum	00	00	00	00	00	00
Ocimum sanctum	80	80	80	90	100	100
Capsicum annum	30	35	43	50	48	45
Dioscorea polygonoides wild	00	00	00	00	00	00
Hibiscus rosasinensis	00	00	00	00	00	00

The beetles were sprayed directly with 1 ml of different concentration of a crude extract under Potters Precision tower at a pressure of 2.5 kg/cm<sup>2</sup>. The controls were sprayed with water. Mortality was recorded every 24h for 5 days and corrected by Abbot's formulae (Busvine, 1972).

Different volumes (0.5, 1.5, 3) µl plant oils of O. sanctum, A. reticulata and A. indica obtained by extraction as described above were made up to 30 ul with redistilled acetone in a covered Petri dish (9cm diameter) by shaking the Petri dish for a few seconds and immediately the seeds were introduced into it. The solution was used to coat uniformly on 50 seeds of the cowpea. After 10 min. when the acetone had dried up, the seeds were transferred into 150 ml translucent plastic cups with firm cover, and 5 males and 5 females of freshly emerged C. maculatus (0-24h) were introduced. The seeds were coated with ordinary acetone in the control treatment. There were three replicates for each treatment and all the cups were incubated at ambient temperature for 18 days. Egg count, the number of emerged adults and other parameters were determined.

Crude Ethanol (CE) extracts of 20 different plant species showed biological activity as reflected by mortality inflicted upon the treated *Callosobruchus* spp. (Table 1).

Extracts of Myristica fragrans, Gliricidia sepium, Ricinus communis, Cajanus cajan, Mangifera indica, Eupatorium odorantum, Dioscorea polygonoides, and Hibiscus rosasinensis showed no toxicity while those of Citrus reticulate, Artocarpus heterophyllus and Cassia occidentalis had little toxicity. Capsicum annum and Dillenia retusa plant extracts were slightly toxic causing 45% and 58% mortality in the C. maculatus group. C. frutescens and Piper nigrum were moderately toxic while those of Eugenia caryophyllata caused fairly high (70%) beetle mortality. The highest bioactivity (90 – 100% mortality) was manifested by the extracts of Azadirachta indica, Anona reticulata and Ocimum sanctum.

Mortality of the beetles caused by the bioactivity was gradually reaching a maximum in 72h. The absence of low level of toxicity exerted by most plant extracts may not be the true reflection of the bioactive potentials of the plants as a whole. The distribution of bioactive compounds often varies in different plant parts. Neem kernel has more azadirachtin than leaves and other tissues (Kossou 1989) while leaves of *N. tabacum* have six times more bioactive compounds than root, stalk or inflorescences. (Rao and Chakraborty1982).

The possession of highly bioactive compounds of plants such as *O. sanctum, A. indica* and *A. reticulata* which are grown widely in Sri Lanka offer an opportunity for developing them as alternatives to hazardous pesticides. Ketoh *et al* (2000) reported that *C. schoenanthus* oil contained up to 61% of piperitone and 23.4% careen- 2 as major constituents. *Cymbopogan citratus* oil is rich in citral (*gevanial* 34.6% and *neral* 50.9%). *Citronnellal* (35.4%), *Citronnellol* (9.7%), *Geraniol* (23.5%) and *occidentalol* (6.5%) were the major components of *C. nardus* oil (Ketoh *et al* 2000).

Data obtained with these 3 plant oils indicated that oils of *O. sanctum* at 1.5 µl and *A. reticulata* at 3.0 µl completely inhibited oviposition and adult emergence (Table 2). A mean of 8.2 eggs were laid on seeds treated with *O. sanctum* oils at 0.5 µl and a comparable number of emerged adults were recorded. *A. indica* oils did not inhibit oviposition completely at any of the 3 doses tested. This indi-

Table 2: The Effect of Plant Oils on egg deposition and adult emergence of *C. maculatus* 

Oils	Dose(µl)/ 50 seeds	Mean no of Eggs	.Mean no. adults	of%damaged seeds		
		(emergence)				
Ocimum	0.5	$8.2 \pm 2.1$	$7.0 \pm 2.7$	$14.0 \pm 2.5$		
sanctum	1.5	00	00	nd		
	3.0	00	00	nd		
Anona	0.5	$10.1 \pm 2.0$	$9.4 \pm 2.1$	$16.0 \pm 2.1$		
reticulata	1.5	$5.2 \pm 2.1$	$3.4 \pm 1.6$	$9.0 \pm 2.2$		
	3.0	00	00	nd		
Azadirachta	0.5	$10.2 \pm 2.1$	$11.2 \pm 2.2$	$14.0 \pm 1.1$		
indica	1.5	$6.8 \pm 1.1$	$3.2 \pm 1.6$	$6.0 \pm 1.4$		
	3.0	$2.8 \pm 0.9$	$2.1 \pm 1.1$	$6.0 \pm 0.9$		
Control		$76.6 \pm 4.8$	$356.8 \pm 2.6$	59.4 3.8		

nd = no determination

<sup>\*</sup> C. maculates (C.m) C. chinensis (C.c.)

cated that the volatile oils of *O. sanctum* and *A. reticulata* effectively protected stored cowpea from infestation by *C. maculatus* and insecticidal activities of these two plants used traditionally for preserving cowpea appear to reside in the ethanolic extracts. It was found that insecticidal activity of *O. sanctum* observed is due to the presence of linalool in the leaves.

Schoonhoven (1978) indicated that 100 ml of selected vegetable oils effectively protected cowpea against the pulse beetle *Zabrotes subfasciatus*. Plant oils used in this study were more efficient than the vegetable oils reported by Messina & Renwick (1983) and Pereira (1983). The non-bitter state of a plant oil used is an added advantage over neem oil which is known for its bitter taste.

The relatively small amounts of oils required their effectiveness and the simple technology of extraction will make these plant oils a better candidate for seed dressing purposes for cowpea storage. With increase of prevailing prices of insecticides, the application of plant oils would be an inexpensive control method against *C. maculatus* and *C. chinensis*.

## **REFERENCES:**

- Attri BS, Prasad R 1980 Neem oil extractive an effective mosquito larvacide. *Indian Journal of Entomology* 42(3): 371 374
- Busvine JR 1972 A Critical Review of the Technique for Testing Insecticides. 2<sup>nd</sup> edition. *Commonwealth Agricultural Bureaux*. London. UK.
- Harbourne JB 1977 Introduction to Ecological Biochemistry. Academic Press. New York. 104 129
- Kossou DK 1989 Evaluation des different productits du neem *Azadirachta indica* A Juss pour le controle de Sitophilus zeamais Motsch..sur le mais en post recolte. *Insect Science and Application* 10 : 365-372

- Ketoh GK, Glitho AI, Koumaglo KH, Garnen FX (2000). Evaluation of essential oils from six aromatic plants in Togo for *Callosobruchus maculatus* pest control. *Insect Science and Application* 20: 45-49
- Lambert JDH, Gale J, Arnason JT, Philogene BJR (1985). Bruchid control with traditionally used insecticidal plants *Hyptis spicigera* and *Cassia nigricans*. *Insect Science and Application* 6; 167-170
- Messina FJ, Renwick JA (1983). Effectiveness of oils in protecting stored cowpeas from the cowpea weevil (Coleoptera: Bruchidae) *Journal of Economic Entomology* 76;634-636
- Mummigatti SG, Raghunathan A N 1977 Inhibition of multiplication of *C.Chinensis* (L.) by vegetable oils. *Journal of Food Science and Technology* 14: 184-185
- Olaifa JI, Erhun WO 1988 Laboratory evaluation of Piper guineense for the Protection of cowpea against Callosobruchus maculatus. Insect Science and Application 9: 55-59
- Pereira J 1983 The effectiveness of six vegetable oils as protectants of cowpeas and bambara groundnuts against infestation by *C. maculatus* (Coleoptera: Bruchidae). *Journal Stored Product Research* 19: 57-62
- Ranasinghe MASK, Dharmasena CMD 1987 Effect of three vegetable oils on oviposition and egg hatchability of cowpea bruchid *Callosobruchus maculatus Sri Lanka Journal of Agricultural Science* 24: 42-48
- Rao CVN, Chakraborty MK 1982 Tobacco cultivation, improvement and utilization of tobacco waste for production of nicotine. In *Cultivation and Utilization of* Medical Plants (Eds Atal CK and Kapur BM). 668-679. United Printing Press. Harinagar, New Delhi, India
- Saxena RC 1987 Antifeedants in tropical pest management. *Insect Science Application* 18:731-736
- Schoonhoven AV 1978 Use of vegetable oils to protect stored beans from bruchid attack. *Journal of Economic Entomology* 71: 254-256
- Varma BK, Pandey OP 1978 Treatment of stored greengram seed with edible oils for protection from Callosobruchus maculates, Indian Journal of Agricultural Science 48: 23-30