Agricultural Sciences, 7(1):59-63 (2002)

Potential of Omani Flora as Source of Natural Products for Control of Pulse Beetle, *Callosobruchus chinensis*

H.T. Al-Lawati*[1], K.M. Azam[2], and M.L. Deadman[2]

[1]*Agriculture Research Station, Ministry of Agriculture and Fisheries, Rumais, P.O. Box 50, A'Seeb 121, Sultanate of Oman*
[2]*Department of Crop Sciences, College of Agricultural and Marine Sciences, Sultan Qaboos University, P.O. Box 34, Al Khod 123, Sultanate of Oman*

إمكانية استخدام النباتات العمانية المحلية كمصدر لمنتجات طبيعية لمكافحة خنفساء البقوليات (*Callosobruchus chinensis*)

حسن اللواتي وخاجة عزام ومايكل ديدمان

خلاصة: تم اختبار ثمانية مستخلصات من النباتات العمانية المحلية لدراسة تأثيرها على خنفساء حبوب البقوليات المخزنة من حيث وضع البيض ونمو الحشرة الكاملة وكذلك نسبة موت الحشرات. تمت ملاحظة عدد البيض الذي تضعه الحشرات على بذور الفاصوليا المعاملة وعدد الحشرات التي تخرج من البذور وموت الحشرات المتعرضة للبذور المعاملة. تفاوت عدد البيض على البذور المعاملة وخروج حشرات الجيل الجديد منها تفاوتا ملحوظا فقد كانا في مستخلصات الإيثانول أقل منها في مستخلصات الميثانول، ولوحظ أيضا أن نسبة الموت في الحشرات المعرضة للبذور المعاملة بمستخلص المستعفل كانت عالية جدا حيث وصلت النسبة فيها إلى ١٠٠% خلال يومين و ستة أيام في مستخلصات الإيثانول ومستخلصات الميثانول على التوالي.

ABSTRACT: Extracts of eight plants from the Omani flora were tested for their effects on oviposition, adult emergence from eggs and mortality of the pulse beetle, *Callosobruchus chinensis*. Observations were made on the number of eggs laid on kidney bean seeds treated with extracts, adult emergence from seeds and mortality of adults exposed to treated seeds. The number of eggs laid and the adults emerged from seeds treated with extracts prepared from ethanol were significantly less than from seeds treated with extracts prepared from methanol. The mortality of beetles released on grains treated with *A. squamosa* was 100% within 2 and 6 days in ethanol- and methanol-based extracts, respectively.

Keywords: flora, natural products, pulse beetle, control, *Callosobruchus chinensis*.

With a greater awareness of the hazards associated with the use of synthetic organic insecticides, there has been an increased need to explore suitable alternative methods of pest control. A number of plant-derived products are potential candidates. This paper reports the results of research into the effects of extracts of plants from the Omani flora on various stages in the life cycle of the pulse beetle, *Callosobruchus chinensis*. (Coleoptera: Bruchidae). Although most of the plants used in this study are indigenous to Oman, little has been reported on their efficacy against insect pests.

Pandey *et al.* (1986) studied the effect of plant extracts against pulse beetle, *Callosobruchus chinensis*. The results indicated that extracts of neem, *Lantana camera*, *Ageratum conzoides*, *Thevtia nerifolia* and *Ipomoea carnea* mixed with seeds of green gram (*Phaseolus aurenus*) highly inhibited the oviposition and protected against infestation. Islam (1987) studied the optimal doses and application methods of four insecticidal plants and to reviewuate their antifeedant and growth regulating properties. The result showed that extracts of *Azadirachta indica*, *Melia azedarach*, *Amoora rohituka*, *Annona reticulata* and *A. squamosa* were the most effective.

Reddy *et al.* (1994) studied the effect of edible and non–edible oils on the development of *C. chinensis* and on viability of mungbean seeds. Pre-storage seed treatment of mungbean (*Vigna radiata*) with oils of neem (*Azadirachta indica*), karanj (*Pongamia pinnata*), Indian mustard, groundnut and castor (*Ricinus communis*) at 2.5, 5.0 and 10.0 ml/kg seed. The results showed that karanj oil, mustard oil and castor oil (10 ml/kg seed) were found to be effective in halting the embryonic development in *C. chinensis*.

*Corresponding author.

TABLE 1

Scientific names, common names of plants, location and solvents used to study insecticidal properties against the pulse beetle.

Scientific Name	Common Name	Location	Solvents Used
Acacia nilotica	*Qarat*	Endemic to Oman	Methanol and Ethanol
Annona squamosa	*Mustafal*	Cultured in Oman	Methanol and Ethanol
Azadirachta indica	Shereesh	Widely cultivated in Oman	Methanol and Ethanol
Boswellia sacra	*Luban*	Indigenous to Oman	Methanol and Ethanol
Crotolaria juncea	Kheshkhash	Widely cultivated in Oman	Methanol and Ethanol
Jatropha dhofarica	*Zebrot*	Indigenous to Oman	Methanol and Ethanol
Myrtus communis	*Yas*	Indigenous to Oman	Methanol and Ethanol
Suaeda aegyptiaca	*Suwwad*	Indigenous to Oman	Methanol and Ethanol

The effect of seven different plant oils (*Cymbopogon citratus*, *Bassi longifolia*, *Ricinus communis*, *Cocos nucifera*, *Arachis hypogaea*, *Glycine max* and *Azadirachta indica*) on oviposition by *Callosobruchus chinensis* was studied by Rajapakse and Senanayake (1997). All oils significantly affected oviposition at all concentrations tested. The oils tested did not affect seed germination.

The current study was designed to assess the potential of Omani flora as sources of natural products that could be used for the protection of stored products against storage insect pests.

Materials and Methods

Sub-tropical plants indigenous to Oman or introduced and cultivated (Table 1) were tested for their growth inhibition properties against the beetles *Callosobruchus chinensis*.

Leaves of *B. sacra, J. dhofarica, M. communis* and *S. aegyptiaca*, and seeds of *A. nilotica, A. squamosa, A. indica* and *C. juncea* were collected from the plants and air-dried and powdered. 12.5 g of the powdered material from each plant was soaked separately, in the dark, in a solution of 12.5 ml water and 50 ml solvent (methanol or ethanol). After one day the solutions were filtered and stored in the refrigerator prior to use.

The extracts were tested for their effects on three life cycle stage of *C. chinensis*: oviposition (number of eggs laid), percent adult emergence and mortality. For each plant extract, twenty bean seeds were shaken thoroughly with 2 ml extracts. Control seeds were mixed with the same amount of methanol or ethanol. After mixing, all seeds were allowed to dry, leaving a film of extract. Into glass jars containing 20 seeds coated with each extract, five pairs of insects were released. Jars were closed by cheesecloth and sealed with rubber band. Four replications were used for each of the ten treatments (nine plant extracts, one control). The mortality of adults was recorded in each of the treatments. Egg numbers were counted weekly until the emergence of adults. After adult emergence, the numbers of adults were counted weekly. Newly emerged adults were removed from the jars. Observations continued until nine weeks after the release of adults into the jars.

TABLE 2

Number of eggs laid on seeds coated with methanol- and ethanol-based plant extracts.

Treatments	Number of Eggs Laid on Seed Treatment with Methanol-based Plant Extracts [1,2]	Number of Eggs Laid on Seed Treatment with Ethanol-based Plant Extracts [1,2]
Acacia nilotica	109.3 a	17.5 b
Annona squamosa	11.0 b	3.0 b
Azadirachta indica	38.8 b	32.5 b
Boswellia sacra	48.8 b	20.0 b
Crotolaria juncea	123.0 a	36.3 b
Jatropha dhofarica	98.3 a	23.5 b
Myrtus communis	81.3 a	31.0 b
Suaeda aegyptiaca	111.5 a	35.0 b
Commercial neem	93.5 a	25.8 b
Control	121.0 a	24.8 b

[1]Mean number of eggs laid in 4 replications.
[2]Within solvents, values without letters in common are significantly different at $P<0.05$.

Results

NUMBER OF EGGS LAID: The number of eggs laid on grains in each jar was counted over the 9-week experimental period (Table 2). Overall the number of eggs laid on grains treated with plant extracts in ethanol was significantly lower ($P<0.05$) than for the methanol treatments. The mean number of eggs laid was the lowest for the *A. squamosa* extracts, being 11 and 3 from 5 pairs of adult beetles in methanol and ethanol extracts respectively. Grains treated with other plant extracts and the controls (solvent only) had large mean number of eggs. For example, *A. nilotica*, had 109.3 and 17.5 for methanol and ethanol solvents, respectively. For *C. juncea*, the egg numbers were 123.0 and 36.3 for the same solvents. In the case of *S. aegyptiaca* the numbers of eggs were 11.5 and 35.0. For the solvents the number of eggs laid on the seed were 121.0 (methanol) and 24.8 (ethanol).

Analysis of variance showed that the mean number of eggs laid on *A. indica*, *A. squamosa* and *B. sacra*

TABLE 3

Number of eggs laid on seeds coated with methanol- and ethanol-based plant extracts.

Treatments	Number of Eggs Laid on Seed Treatment with Methanol-based Plant Extracts [1,2]	Number of Eggs Laid on Seed Treatment with Ethanol-based Plant Extracts [1,2]
Acacia nilotica	67.3 a	14.3 b
Annona squamosa	0.8 c	0.8 c
Azadirachta indica	12.3 c	6.3 b
Boswellia sacra	32.5 b	16.5 b
Crotolaria juncea	33.3 b	11.5 b
Jatropha dhofarica	59.3 a	19.0 b
Myrtus communis	36.8 b	25.8 b
Suaeda aegyptiaca	67.5 a	26.3 b
Commercial neem	17.5 b	16.5 b
Control	63.3 a	20.0 b

[1]Mean number of eggs laid in 4 replications.
[2]Within solvents, values without letters in common are significantly different at P<0.05.

were significantly less (P<0.05) than for other extracts based on methanol. There was no significant difference between seeds treated with the ethanol-based plant extracts.

ADULT EMERGENCE : Adult emergence was counted in each jar at weekly interval until week 9, after which there was no more further adult emergence. The data (Table 3) shows the mean number of adults emergence for the four replications of the extract treated seeds and the controls. Overall, adult emergence was less in ethanol-based plant extracts compared to methanol-based plant extracts. In the case of extracts from *A. squamosa* there was negligible adult emergence whatever the solvent used. From the methanol-based extracts, *A. squamosa* and *A. indica* had significantly less adult emergence (P<0.05) from the seed compared to the other extracts. Seed treated with methanol-based extracts of *A. nilotica*, *J. dhofarica*, *S. aegyptiaca* had significantly greater emergence rates (P < 0.05).

However, for the ethanol-based extracts, *A. squamosa* (mean emergence 0.8) and *A. indica* (mean emergence 6.3) induced less adult emergence than the other extracts (P < 0.05). Other plant extracts were not significantly different from each other in their effects (P > 0.05, Table 3).

The mean numbers of eggs laid and the overall adult emergence from treated seeds was significantly less (P<0.05) for the ethanol-based extracts than for those treated with methanol-based extracts. However, the percentage adult emergence (per egg laid) was higher (except for *A. squamosa* and *A. indica*) for the ethanol-based extracts (Table 4). This was probably due to significantly fewer eggs being laid on seeds treated with the ethanol-based extracts.

TABLE 4

Percent adult emergence (per number of eggs laid) from seeds treated with methanol- and ethanol-based plant extracts.

Treatments	Methanol	Ethanol
Acacia nilotica	61.6	81.4
Annona squamosa	6.8	25.0
Azadirachta indica	31.6	19.2
Boswellia sacra	66.7	75.0
Crotolaria juncea	27.0	31.7
Jatropha dhofarica	60.3	80.9
Myrtus communis	45.2	83.1
Suaeda aegyptiaca	60.5	75.0
Commercial neem	18.7	64.1
Control	52.3	80.8

INSECT MORTALITY: The cumulative percentage mortality of the adult insects released into the jars containing seeds treated with methanol- and ethanol-based plant extracts are shown in tables 5 and 6. The mortality data for the methanol-based extracts (Table 5) show the distinct effect of *A. squamosa* in causing high mortality. Other treatments that caused high mortality were *A. nilotica* (67.5%), *B. sacra* (70%), *C. juncea* (67.5%), *J. dhofarica* (80%), *M. communis* (82.5%), *S. aegyptiaca* (77.5%), and control (80%). The mortality data for the ethanol-based extracts (Table 6) show that all 40 beetles had died within 2 days in the treatment using *A. squamosa* as the test plant. Treatments for which 100% mortality was recorded after 8 days were *A. nilotica*, *B. sacra*, *C. juncea* and the solvent control.

Analysis of variance shows the significant (P < 0.05) mortality-inducing effect of *A. squamosa* plant extracts in both solvents. In the methanol-based extracts, the other treatments showed little differences between them in terms of mortality effects effects. In the ethanol-

TABLE 5

Cumulative mortality of C. chinensis in the presence of seed treated with methanol-based plant extracts.

Treatments	Intervals: Days after release (%) 2	4	6	8	10
Acacia nilotica	22.5	50.0	67.5	90.0	100.0
Annona squamosa	87.5	95.0	100.0	-	-
Azadirachta indica	22.5	37.5	50.0	85.0	87.5
Boswellia sacra	15.0	40.0	70.0	77.5	97.5
Crotolaria juncea	27.5	52.5	67.5	85.0	97.5
Jatropha dhofarica	20.0	55.0	80.0	87.5	92.5
Myrtus communis	30.0	67.5	82.5	92.5	100.0
Suaeda aegyptiaca	37.5	60.0	77.5	92.5	100.0
Commercial neem	22.5	37.5	55.0	80.0	100.0
Control	27.5	47.5	80.0	92.5	100.0

* Total of 4 replications.

TABLE 6

Cumulative mortality of C. chinensis in the presence of seed treated with ethanol-based plant extracts.

Treatments	Intervals				
	Days after release (%)				
	2	4	6	8	10
Acacia nilotica	72.5	82.5	92.5	100.0	
Annona squamosa	100.0	-	-	-	-
Azadirachta indica	30.0	60.0	85.0	95.0	100.0
Boswellia sacra	35.0	65.0	85.0	100.0	-
Crotolaria juncea	40.0	67.5	92.5	100.0	-
Jatropha dhofarica	42.5	62.5	85.0	92.5	100.0
Myrtus communis	22.5	62.5	82.5	97.5	-
Suaeda aegyptiaca	47.5	62.5	72.5	90.0	100.0
Commercial neem	35.0	65.0	85.0	95.0	100.0
Control	37.5	7.5	95.0	100.0	-

* Total of 4 replications.

based extracts, mortality caused by *A. squamosa* was significantly greater ($P<0.05$) than the other plants. Mortality caused by *A. nilotica* was also significantly higher ($P<0.05$) than for the remaining plant extracts.

Discussion

Several workers have reported growth and development inhibition properties of plant extracts on pulse beetle, *C. chinensis*. Much of this literature refers to the effects of neem oil. Ketkar (1986) reviewed the effect of neem oil along with three other non-edible oils against *C. chinensis*. It was found that the growth index with neem was the lowest; neem also had the greatest ovicidal effect. Pandey *et al.* (1986) reported that a petroleum ether extract of neem leaves and twigs mixed with green gram seeds inhibited the oviposition of *C. chinensis*. Das (1989) studied the effects of chickpea storage duration on oviposition of *C. chinensis* when the seeds were treated with neem oil. Khaire *et al.* (1993) studied the effect of 10 vegetable oils, including neem, on ovipositional preference and egg hatching of *C. chinensis*. They reported that all treatments affected ovipositional preference. Kachare *et al.* (1994) found that no hatching of eggs of *C. chinensis* took place during the storage of pigeonpea for 33 days when the seeds had been treated with neem oil. Reddy (1994) reported that mungbean seeds treated with neem oil halted the embryonic development of *C. chinensis*, protecting stored seed for a period of 12 months. Studies conducted by Chiranjeevi and Sudhakar (1996) revealed that neem seed powder, mixed with mungbean seed, completely prevented the development of *C. chinensis*. Rouf *et al.* (1996) reported that mixing of neem leaf powder with lentil seeds resulted in reduced oviposition and adult emergence in the same beetle. Studies reported by Rajapakse and Senanayake (1997) showed that seeds treated with neem oil significantly reduced oviposition of *C. chinensis*. There is far less literature on the effects of other plant extracts on the development of *C. chinensis*.

In the current study plant extracts from native Omani flora, or introduced plants that are extensively cultivated were tested for their effectiveness in disrupting the life cycle of *C. chinensis*. The effects were compared with those induced by a commercial neem preparation and those produced by the solvents used to extract the natural plant products. Although seed dressed with commercial neem caused fewer eggs to be laid compared with the ethanol control, the effect was less than that caused by a fresh neem preparation and an extract of *A. squamosa* in ethanol. There is little previously reported information on the life cycle disrupting properties of *A. squamosa*. Although Islam (1987) reported that extracts of *Anonna squamosa* were highly effective as antifeedant and as growth regulators, no mention is made of the oviposition deterrent effects.

A. squamosa extracted in methanol also significantly reduced the level of adult emergence from those eggs laid. The extracts of *Crotolaria juncea* also had a similar effect. Interestingly, previous reports (Azam *et al.*, unpublished) have shown that extracts of A. squmaosa attract adult *C. chinensis* beetles. Furthermore, the data presented here and elsewhere (Azam *et al.*, unpublished), show A. squamosa to have a significant toxicological effect, causing increased mortality in adult beetles. Together, these results would indicate a significant potential for this plant as a possible source of natural products that could be used as an alternative to synthetic insecticides.

References

Chiranjeevi, C. and T.R. Sudhakar. 1996. Effect of indigenous plant materials on the fecundity, adult emergence and development of pulse beetle *Callosobruchus chinensis* (L.) in black gram. *Journal of Research APAU* 24:57-61.

Das, G.P. 1989. Effect of the duration of storing chickpea seeds treated with neem oil on the oviposition of the bruchid *Callosobruchus chinensis* Linn. (Bruchidae: Coleoptera). *Bangladesh Journal of Zoology* 17:199-201.

Islam, B.N. 1987. Use of some extacts from Meliaceae and Annonaceae for control of rice hispa, *Dicladispa armigera*, and the pulse beetle, *Callosobruchus chinensis*. In: *Natural Pesticides from the Neem Tree (Azadirachta indica A. Juss.) and Other Tropical Plants. Proceedings of the 3rd International Neem Conference*, ed. R. Schmutterer and K.R.S. Ascher, 217-242, Nairobi, Kenya.

Kachare, B.V., V.M .Khaire, and U.N. Mote. 1994. Efficacy of different vegetable oils as seed treatment in increasing storage age ability of pigeonpea seed against pulse beetle, *Callosobruchus chinensis* Linn. *Indian Journal of Entomology* 56:58-62.

Ketkar, C. M. 1986. Use of tree-derived non-edible oils as surface protectants for legumes against *Callosobruchus maculatus* and *C. chinensis*. In: In: *Natural Pesticides from the Neem Tree (Azadirachta indica* A. Juss.*) and Other Tropical Plants. Proceedings of the 3rd International Neem Conference*, ed. R. Schmutterer and K.R.S. Ascher, 535-542, Nairobi, Kenya.

Khaire, V.M., B.V. Kachare, and U.N. Mote. 1993. Effect of different vegetable oils on ovipositional preference and egg hatching of *Callosobruchus chinensis* Linn. on pigeonpea seeds. *Department of Entomology, Mahatma Phule Agricultural University, Seed Research* 21:128-130.

Pandey, N.D., K.K. Mathur, S. Pandey, and R.A. Tripathi. 1986. Effect of some plant extracts against pulse beetle, *Callosobruchus chinensis* Linnaeus. *Indian Journal of Entomology* 48:85-90.

Rajapakse, R.H.S. and S.G.J.N. Senanayake. 1997. Effectiveness of seven vegetable oils against *Callosobruchus chinensis* L. *Entomon* 22:179-183.

Reddy, V.S., T.R. Babu, S.H. Gussaini, and B.M. Reddy. 1994. Effect of edible and non-edible oils on the development of pulse beetle *Callosobruchus chinensis* L. and on viability of mungbean seeds. *Pest Management and Economic Zoology* 2:15-17.

Rouf, F. M. A., M.A. Sardar, and K.S. Ahmed. 1996. Individual and combined effect of some plant mterials for protection of lentil seeds against pulse beetle *Callosobruchus chinensis* L. *Bangladesh Journal of Entomology* 6:13-21.

Received June 2001.
Accepted May 2002.