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Biocidal activity of some castor extracts against the whitefly *Bemisia tabaci* (Genn) (Homoptera: *Aleyrpdidae*)

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ABSTRACT

The biocidal activity of castor oil plant (*Ricinus communis*) in controlling the whitefly *Bemisia tabaci* (Genn) (Homoptera: *Aleyrpdidae***)** was examined**.** Castor oil was extracted from seeds using different solvents, hexane, acetonitril and methanol. Assays were performed with adults and larvae. Mortality of the pest increased with increasing concentrations of castor oil. The lethal concentrations of the castor oil extracts was higher for adult fly than the immature stage. The insecticidal values of castor oils were compared with other formulations based on Azadirachtin (neemix, 4.5% EC) and Pyrethrum 5% SC. Emulsions of castor seed oil gave effective control against the whitefly *Bemisia tabaci* (Genn).

Key wards: *Ricinus communis*, oil, Azadirachtin, Pyrethrum, *Bemisia tabaci* (Genn)

INTRODUCTION

Whitefly *Bemisia tabaci* (Genn) is a very important pest because of the damage they cause. Control methods using castor oil have been evolved and practiced with varying degrees of success (Abbas, 2003; Baldin *et al*. 2007). Many researchers have taken the Azadirachtin and pyrethrum for comparison in their work. Also in the present study they were used for comparison and to ascertain the activities of castor oil.

Azadirachtin and Pyrethrum have a long history for use as insecticides. Azadirachtin exhibits good efficacy against key pests such as whiteflies, leaf miners, fungus gnats, thrips, aphids, and many leaf-eating caterpillars (Immaraju, 1998). Its use is recommended for insecticide resistance control, integrated pest control, and organic pest control programs. Over 400 species of insects have been shown to be susceptible to the neuroendocrine effects of azadirachtin, exhibiting classic symptoms of abnormal molts, larval–adult intermediates, mortality at ecdysis, and delayed molts, often resulting in greatly extended instar lengths. Many studies on a wide variety of insect taxa have allowed detailed descriptions of symptoms to be recorded that show consistency between all insects tested and an ED50 (effective dose for 50% of the population) of between 1 and 4 mg/g bodyweight (Koul and Isman, 1991; Mordue (Luntz) and Blackwell, 1993; Riba *et al*., 2003).

Pyrethrum is one of the first insecticides with a century-long history of safe use (Katsuda, 1999). The active compounds of pyrethrum extract are six esters, commonly referred to as the pyrethrins. Most insects are still susceptible to pyrethrum at low concentrations, and its action is against a wide variety of different insect species (Silcox and Roth, 1994).

MATERIALS AND METHODS

Collection of the plant seeds and preparation of the plant oil

Seeds of castor were collected from El-Minufiya Governorate during the summer of 2007 and 2008. The seeds of the *Ricinus* species were collected from dry fruits still on the plants, and were thus ready for further processing when collected.

The oil of *Ricinus communis* was extracted and obtained by grounding the seeds using blender, then using different solvents for extraction starting with hexane for three weeks, after each week filtration was carried out and the oil was collected and obtained, so three samples of the oil were collected. After hexane, acetonitril was added and the oil was colleted. Repeating the experiment starting with hexane and then followed by methanol. So oils extracted by hexane, acetonitril, and methanol were obtained. Humidity and any water can be removed using calcium chloride anhydrous. Density of the three oils was calculated and found nearly the same and estimated as 0.97, 0.96 and 0.96 gm/ cm^3 for the oils, respectively.

Insect used

Laboratory strain of whitefly *Bemisia tabaci* (Genn.) was obtained from Syngenta Agro Egypt Co., and maintained without exposure to insecticides for about three (3) years in the rearing laboratories of the Central Agricultural Pesticides Laboratory. The insects were confined with plotted cotton, which placed in an environmental chamber under conditions of 27 $\pm$ 2 ºC, 55 $\pm$ 5% R H. and a photoperiod of 18 L: 6 D.

The cotton plants *(Gossyium hirsutum* L., Giza-83) were grown in plastic pots containing mixture of peatmoss, sand and clay soil at the rate 1: 1 and maintained in another environmental chamber until the seedlings reached 15 – 20 cm high. Thereafter, these seedlings were introduced to the insects (Coudriet *et al.,* 1985).

Emulsifier and surfactant used

1- Genapol OAO: white cloudy liquid, nonionic surfactant.

2- Colsogen 4814: Browinish viscous liquid, anionic emulsifier of n- C12 alkylbenzene sulphonate calsium salt in 2- ethylhexanol.

The emulsifier used is a mixture of these two compounds with a ratio 1: 3

Bioassay

The bioassay method for obtaining concentration – mortality lines was previously described by Prabhaker *et al.* (1985). Attached cotton leaves were dipped for 5 Sec. in 100 ml of the desired concentrations of each insecticide and allowed to dry. Twenty adult whiteflies were exposed to the treated leaves confined in small cages (by an aspirator). Mortality was recorded 48 hrs after treatment. At least 5 concentraions were tested for each insecticide, and each test replicated 5 times. The treated leaves with the adults were kept at constant laboratory conditions of 27 $\pm$ 2 ºC, 55 $\pm$ 5% R H. and a photoperiod of 18 L: 6 D. The data were analysed statistically using probit analysis.

Commercial botanical insecticides formulations used:

- Neemix 4.5% EC (W/V): (formulated by Certis biopesticides company-USA
- Pyrethrum 5 % SC (W/V): (formulated by Agropharm)

RESULTS AND DISCUSSION

Extracts of castor (*Ricinus communis*) seeds prepared using various solvents, hexane, Acetonitrile and Methanol, were studied against the whitefly, *Bemisia tabaci* (Homoptera: Aleyrodidae). The insecticidal values of castor oils were compared with other formulations based on Azadirachtin (neemix 4.5% EC) and Pyrethrum (Reptide 5% SC). The current work was undertaken to establish the effectiveness of the crude castor oils as an insecticide against whitefly. The crude oils were treated with an emulsifier to facilitate its mix with water. It formed an emulsion directly when mixed with water. In addition, nonionic surfactants was used which preferred for best results. The nonionic surfactants are surface-active compounds, which do not ionize in water solution.The ratio of the surfactant to the emulsifier was 1: 3. The emulsifier content never exceeded 10% of the oil content by any way to not affect on the results obtained.

Water was used in our investigations as a preferred carrier. A direct spray was used so that upper and lower leaf surfaces are contacted. The spray was applied in a manner to provide complete and uniform coverage of infested plants.

The data for mortality of Azadirachtin (neemix 4.5% EC), Pyrethrum 5% SC and crude castor oils are shown in Tables 1 to 5. Tables 6 to 10 show the mortality related to the active ingredient percentage and Figures 1 to 5 show their regresion lines.

Table 1: LC_{50} and data analysis for Neemix 4.5 % (W/V) against white fly *Bemisia tabaci*

Larval instar	LC_{50} (mg/L)	95% confidence limits (mg/L)		Slope ± Standard Error (SE)	Chi square (χ^2)
		Lower	Upper		
1st Instar	1.12	0.15	2.46	1.70±0.16	20.45
2nd Instar	1.76	1.49	2.03	2.34±0.24	0.08
3rd Instar	2.12	1.71	2.63	1.27±0.14	2.59
4th Instar	4.34	3.33	6.29	1.11±0.20	3.37

Table 2: LC_{50} and data analysis for Pyrethrum 5% SC (W/V) against white fly *Bemisia tabaci*

Larval instar	LC_{50} (mg/L)	95% confidence limits (mg/L)		Slope ± Standard Error (SE)	Chi square (χ^2)
		Lower	Upper		
1st Instar	1.86	1.50	2.22	1.77±0.21	3.77
2nd Instar	3.48	2.67	4.79	1.10±0.19	0.42
3rd Instar	4.05	2.42	11.08	1.27±0.15	6.56
4th Instar	5.47	3.22	28.26	1.81±0.22	3.85

Table 3: LC_{50} and data analysis for castor oil seed extracted by acetonitrile) against white fly *Bemisia tabaci*

Larval instar	LC_{50} (mg/L)	95% confidence limits (mg/L)		Slope ± Standard Error (SE)	Chi square (χ^2)
		Lower	Upper		
1st Instar	0.50	0.36	0.65	2.92±0.24	10.78
2nd Instar	0.71	0.48	0.98	2.49±0.18	13.94
3rd Instar	1.47	0.95	2.14	2.50±0.19	9.82
4th Instar	3.01	1.75	5.55	2.47±0.23	4.98

Table 4: LC_{50} and data analysis for castor oil seed extracted by methanol against white fly *Bemisia tabaci*

Larval instar	LC_{50} (mg/L)	95% confidence limits (mg/L)		Slope ± Standard Error (SE)	Chi square (χ^2)
		Lower	Upper		
1st Instar	0.35	0.22	0.46	2.45±0.24	8.31
2nd Instar	0.54	0.46	0.62	2.29±0.18	5.26
3rd Instar	1.63	1.00	2.56	2.45±0.19	12.39
4th Instar	2.49	2.16	2.86	2.29±0.22	1.23

Table 5: LC_{50} and data analysis for castor oil seed extracted by hexane) against white fly *Bemisia tabaci*

Larval instar	LC_{50} (mg/L)	95% confidence limits (mg/L)		Slope ± Standard Error (SE)	Chi square (χ^2)
		Lower	Upper		
1st Instar	0.84	0.62	1.11	2.59±0.18	11.05
2nd Instar	1.69	1.08	2.62	2.49±0.19	11.66
3rd Instar	2.56	2.12	3.15	1.42±0.15	3.30
4th Instar	3.79	3.19	4.60	1.77±0.21	0.66

Table 6: LC_{50} and data analysis for Neemix 4.5 % (W/V) against white fly *Bemisia tabaci* related to the active ingredient

No	Line name	LC_{50}	Lower limit	Upper limit	Slope	Slope +/-
1	Nemix 1st instar	50,548			1,701	0,161
2	Nemix 2nd instar	80,128	66,833	93,489	2,127	0,235
3	Nemix 3rd instar	95,429	77,112	118,676	1,27	0,144
4	Nemix 4th instar	195,432	149,862	282,76	1,114	0,196

Table 7: LC_{50} and data analysis for pyrethrum 5% SC against white fly *Bemisia tabaci* related to the active ingredient

No	Line name	LC_{50}	Lower limit	Upper limit	Slope	Slope +/-
1	Pyrethrum 1st instar	93,017	75,209	110,887	1,772	0,212
2	Pyrethrum 2st instar	174,303	133,718	239,164	1,101	0,194
3	Pyrethrum 3rd instar	202,314	161,115	270,385	1,272	0,149
4	Pyrethrum 4th instar	273,621	226,719	350,441	1,81	0,22

Table 8: LC_{50} and data analysis for Castor Oil extracted by Acetonitril against white fly *emisia tabaci* related to the active ingredient

No	Line name	LC_{50}	Lower limit	Upper limit	Slope	Slope +/-
1	Oil by acetonitril 1st instar	448,855	279,624	636,815	2,922	0,24
2	Oil by acetonitril 2nd instar	640,979	374,615	999,853	2,442	0,188
3	Oil by acetonitril 3rd instar	1371,712	1190,218	1586,834	2,215	0,22
4	Oil by acetonitril 4th instar	2709,076	2378,408	3093,078	2,469	0,229

Table 9: LC_{50} and data analysis for Castor Oil extracted by MeOH against white fly *Bemisia tabaci* related to the active ingredient

No	Line name	LC_{50}	Lower limit	Upper limit	Slope	Slope +/-
1	Oil by MeOH 1st instar	312,411	167,18	425,696	2,446	0,235
2	Oil by MeOH 2nd instar	483,541	414,649	554,929	2,252	0,189
3	Oil by MeOH 3rd instar	1569,683	1354,077	1841,822	2,099	0,218
4	Oil by MeOH 4th instar	2257,118	1954,538	2591,837	2,278	0,223

Table 10: LC_{50} and data analysis for Castor Oil extracted by Hexane against white fly *Bemisia tabaci* related to the active ingredient

No	Line name	LC_{50}	Lower limit	Upper limit	Slope	Slope +/-
1	Oil by hexane 1st instar	776,069	681,553	881,767	2,393	0,187
2	Oil by hexane 2nd instar	1639,466	1415,406	1926,577	2,119	0,219
3	Oil by hexane 3rd instar	2306,406	1906,846	2835,202	1,424	0,148
4	Oil by hexane 4th instar	3409,513	2871,359	4151,552	1,77	0,209

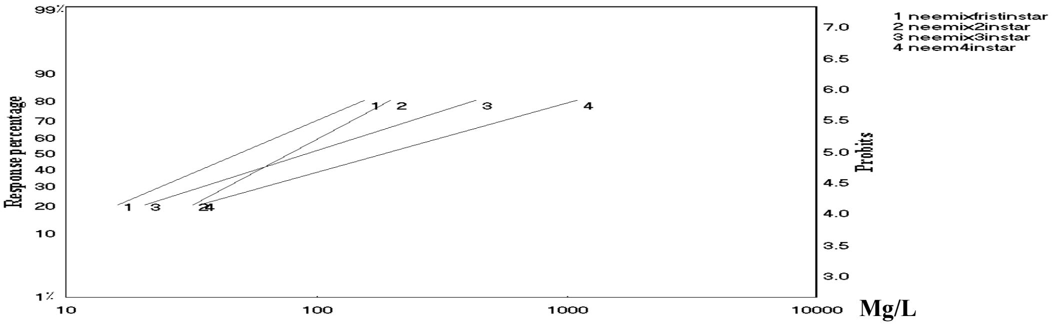

Fig. 1: LC_{50} – probits analysis for Neemix 4.5 % EC (W/V) against white fly *Bemisia tabaci* related to the active ingredient

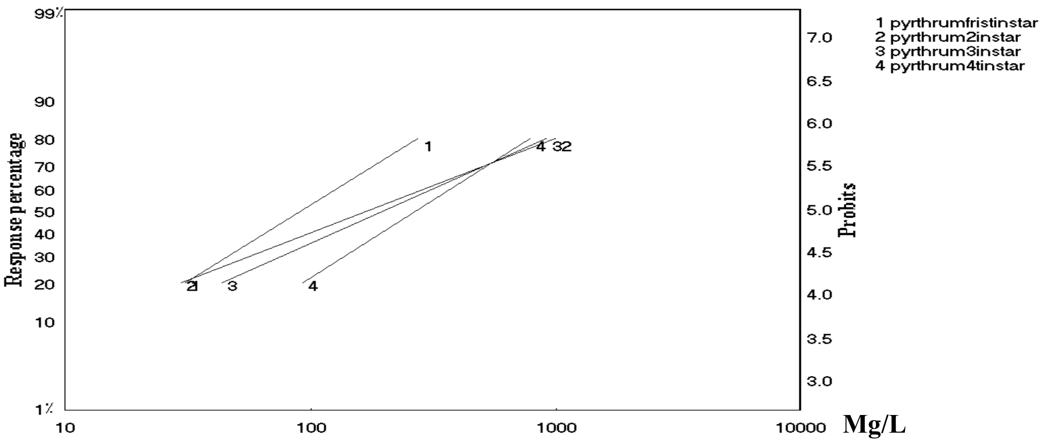

Fig. 2: LC_{50}–probits analysis for pyrethrum 5% SC (W/V) against white fly *Bemisia tabaci* related to the active ingredient.

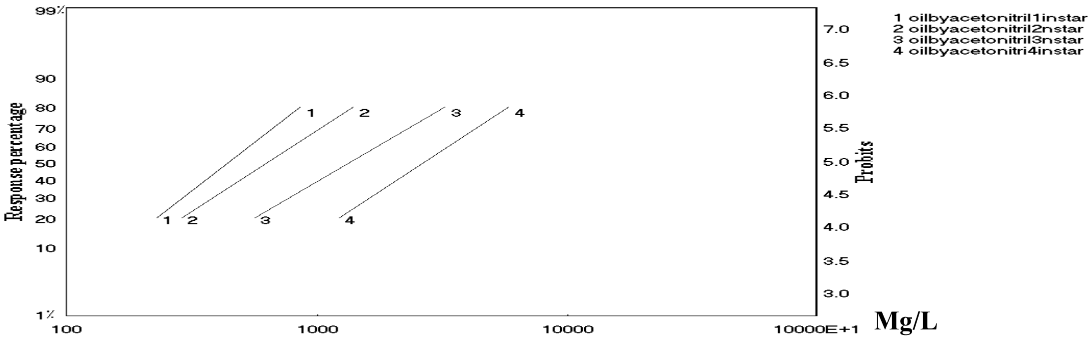

Fig. 3: LC_{50} – probits analysis for Castor Oil extracted by Acetonitril against white fly *Bemisia tabaci* related to the active ingredient.

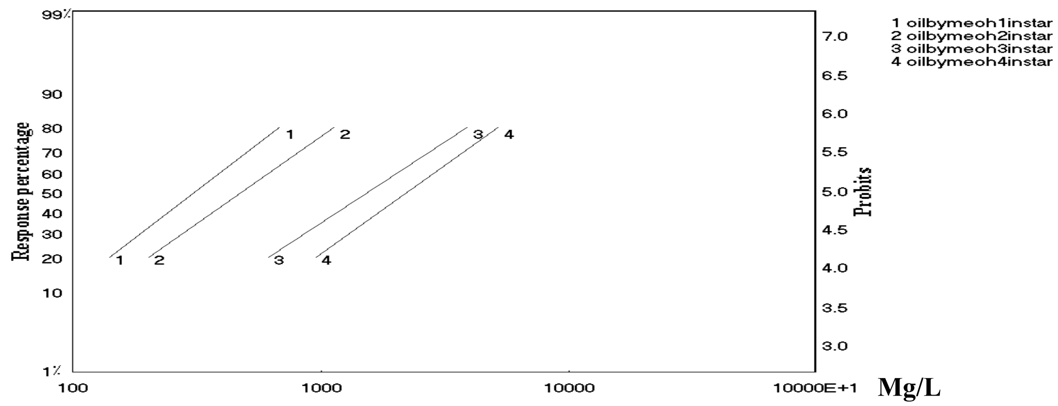

Fig. 4: LC_{50} – probits analysis for Castor Oil extracted by MeOH against white fly *Bemisia tabaci* related to the active ingredient.

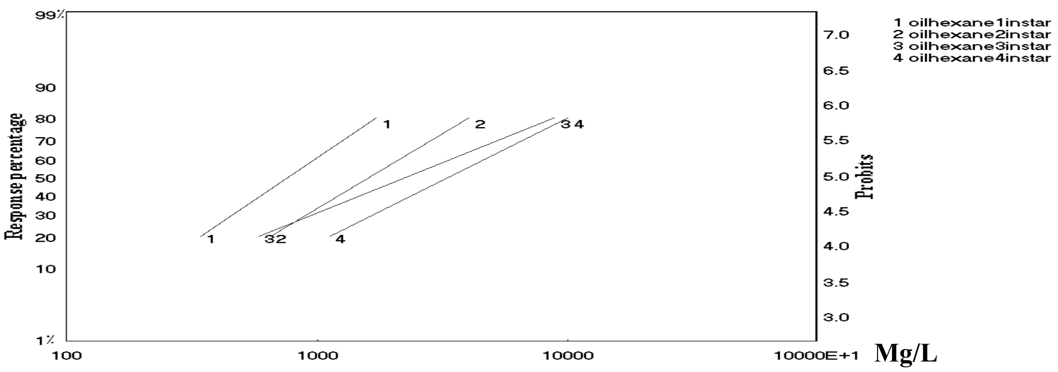

Fig. 5: LC_{50} – probits analysis for Castor Oil extracted by Hexane against white fly *Bemisia tabaci* related to the active ingredient.

The results showed that mortality of the pest (whitefly, *Bemisia tabaci*) increased with increasing concentrations of castor oil. The lethal concentration of the castor oil extracts was higher for the adult fly than immature. A methanol oil extract was the most potent and had the highest contact toxicity. Other extracts produced toxicities which were slightly lower. The order of decreasing toxicity was methanol, acetontrile and hexane extracts, respectively. This close of mortality with different extracts which gave different product yield compounds support the mode of action of oils that is partially attributed to interference in normal respiration, resulting in suffocation (Schoonhoven, 1978). The most likely mechanism of activity of a refined mineral oil on *S. grunarius* adults was that of blocking the tracheal system, causing beetle death by anoxia (Hewlett, 1975).

However, factors other than oxygen starvation probably also play a role in their mode of action. Repellent and ovicidal properties of different oils have also been observed (Varma and Pandey, 1978; Babu *et al.,* 1989). Larew and Locke (1990) proposed that the oils may involve removal of insect cuticle wax, physical action, repellency or cell membrane disruption. Egg mortality has been attributed to toxic components (Su *et al.,* 1972) and also to physical properties, which cause changes in surface tension and oxygen tension within the egg (Singh *et al.*, 1978). It is also thought that oils exert some lethal action on developing embryos or 1st instar larvae, e.g. by the reduction in rate of gaseous exchange due to a "barrier" effect and/or direct toxicity by penetrated oil fractions (Don Pedro, 1989). Another hypothesis is that oil infiltration under the operculum may block respiration or disrupt the water balance of eggs and developing embryos (Messina and Renwick, 1983). It has been postulated that fatty acid chain lengths are related to the amount of insect control and or insecticidal activity has also been attributed to the triglyceride component and to the oleic acid content (Hill and Schoonhoven, 198l a, b). These postulation and hypothesis can be proposed with our experiment for oils of *Ricinus communis* where the GC/Mass analysis showed that the oils comprised largely of fatty acids and ricinoleic esters content especially with acetonitrile extract. Ricinoleic acid, the major fatty acid present in castor oil, has a variety of effects on the gastrointestinal tract, including inhibition of water and electrolyte absorption (Donowitz, 1979).

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ARABIC SUMMARY

النشاط الابادي الحيوى لبعض مستخلصات نبات الخروع ضد آفة الذبابة البيضاء

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استهدفت هذه الدراسة بحث و تقييم كفاءة بعض المستخلصات النباتية و بعض المبيدات الحيوية النباتية ضد بعض الآفات الحشرية. حيث تم دراسة تاثير المستخلصات الزيتية الناتجة من بذور نبات الخروع باستخدام مذيبات مختلفة مثل الهكسان و الاسيتونيتريل و الميثانول ضد الذبابة البيضاء وتم مقارنتها ببعض المستحضرات النباتية التجارية التي تحتوى على مادة الأزاديراكتين والبيريثرم كمواد فعاله. أثبتت الدراسة كفاءة استخدام زيت بذرة الخروع ضد الذبابة البيضاء. وتم اضافة بعض المواد التي تساعد على استحلاب هذه الزيوت وذلك لسهولة امتزاجها بالماء مع مراعاة عدم زيادة هذه المواد عن ١٠% حتى لا توثر على النتائج.

نسب الموت في الذبابة البيضاء ارتفعت مع زيادة التركيزات المستخدمة وكانت الجرعة نصف المميتة في الأطوار الأولى اقل منها في الأطوار الكبيرة لليرقات. أظهر مستخلص الميثانول تأثيرا أعلى نسبيا من المذيبات الأخرى وان كانت أظهرت تأثيرا متقاربا مع الميثانول أيضا. اظهر التحليل الكيماوي بواسطة جهاز كروماتوجرافى الغاز احتواء هذه الزيوت على نسبه عاليه من الاحماض الدهنيه واسترات الريسينين.