Susceptibility of African Bollworm *(Helicoverpa armigera)* Populations to Synthetic Insecticides in Ethiopia

***Geremew Terefe*[1] *and Surachate Jamornmarn*[2]**
[1]*Ethiopian Agricultural Research Organization, Werer Agricultural Research Center P.O. Box 2003, Addis Ababa, Ethiopia*
[2] *Department of Entomology, Kasetsart University, 50 Pahoyothin Road, Bangkok 10900, Thailand*

Abstract

Six field-collected populations of third-instar African bollworm, *Helicoverpa armigera* (Hubner), (Lepidoptera: Noctuidae) were tested for their susceptibility to endosulfan, profenofos, lambdacyhalothrin and methomyl applied as topical spray. Populations were established by collecting eggs and larvae from four major cotton and two chickpea growing areas of the country to generate baseline data on the level of resistance or susceptibility to commonly used insecticides. LD_{50}s were determined using probit analysis. High LD_{50} (73.47 µg/g) was recorded for samples collected from Arbaminch and the lowest for those from Dukem (21.28). The calculated resistance ratio (RR) of samples from Arbaminch for endosulfan was 3.45. The Dubti population recorded the highest LD_{50} and RR for profenofos. The Arbaminch population was found highly susceptible to lambdacyhalothrin with LD_{50} of 0.05 µg/g larval weight. High LD_{50} (0.52 µg/g) was recorded for Dubti population with RR of 10.40. LD_{50} of methomyl were very low in all populations indicating susceptibility of populations to this insecticide.

KEY WORDS: African bollworm, Ethiopia, susceptibility, synthetic insecticides

Running title: Susceptibility of African bollworm to synthetic insecticides

Introduction

The African bollworm, *H. armigera* is the most economically important insect pest of cotton in Ethiopia and accounts for more chemical control applications than any other pest (Tessema et al. 1980). It is reported to have developed resistance to a range of insecticides in many areas of the world and many resistance problems with this species arise on cotton and other high-value commercial crops, where insecticides are extensively used (Wolfenbanger et al. 1981). The development of resistance in many insects of importance in agriculture and public health has led to bioassay of many species for the purpose of determining their level of susceptibility to insecticides during the course of control program (Hoskins & Craig 1962).

Once resistance becomes noticeable as a loss of field efficiency, there are few practical alternatives other than abandoning the use of the insecticide or accepting increased damage. Recent theoretical considerations of insecticide resistance have concluded that the changes in gene frequency must be detected at early stage for corrective measures to be applied (Muggleton 1984).

In Ethiopia, insecticide application against cotton insect pests dates back to the period of 1928–1933 (Read 1941). Since then, with the growing mechanization of commercial farming, the quantity and kinds of insecticides used has increased several folds. Consequently, the high selection pressure from frequent insecticide application is anticipated to result in the development of resistance to insecticides used. This study was undertaken to determine the status of *H. armigera* susceptibility to endosulfan, profenofos, lambdacyhalothrin and methomyl, the insecticides frequently used to control *H. armigera*

Materials and Methods

H.armigera larvae and eggs were collected from six geographically separate locations Werer, Arbaminch, Dukem, Dubti, Humera and Zemea (Fig. 1) from December 2001 to October 2002 for population establishment.

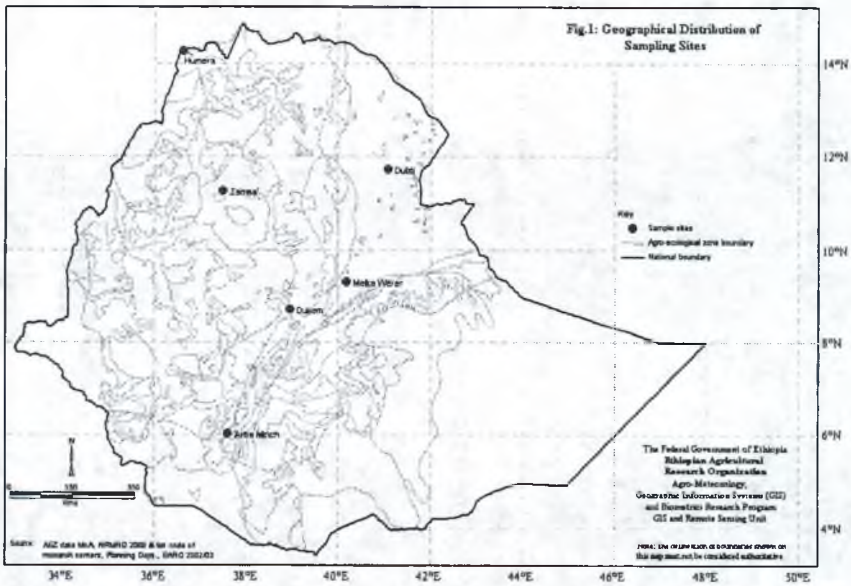

Fig 1. Location of the study sites

Insecticide use history (how long it was used, against what insect pests and with what application frequency) on cotton was obtained from respective state farms in the sampling sites. The sources for chickpea growing areas were bureaus of the Ministry of Agriculture. The larvae were reared on semi artificial diet prepared on soybean flour (125 g), agar (12.5 g), bread yeast (10 g), ascorbic acid (3 g), sorbic acid (1.5 g), casein salt (3 g), methyl 4-hydroxybenzoate (2.5 g), formalin 40% (2 ml), benomyl 50%WP (1 g), distilled water (750 ml), and vitamin stock solution (10 ml), prepared following the procedures of Taekle & Jenson (1985). All populations were maintained under laboratory conditions (30 $\pm$ 3°C; photoperiod 10L:14D at Werer Agricultural Research Center (WARC).

Three frequently used insecticides (endosulfan, profenofos and lambdacyhalothrin) and one not-so-frequently used insecticide (methomyl) against insect pests of cotton were selected. The technical grade insecticides used in the bioassay were endosulfan (95%) supplied by Adamitulu Pesticide Processing and Packing Share Company, Ethiopia; Profenofos (88.7%) and lambdacyhalothrin (84. 9%) supplied by SYNGENTA Bangkok, Thailand and Methomyl (89.6%) from Aventis Crop Science, Bangkok, Thailand. Serial dilutions of technical grade insecticides were prepared in analytical grade acetone. The test concentrations were 0, 250, 500, 1000, 2000 and 4000 ppm for endosulfan; 0, 3.9, 7.8, 15.6, 31.3, 62.5, 125 and 250 ppm for lambdacyhalothrin; 0, 31.25, 62.5, 125, 250, 500 and 1000 ppm for profenofos, and 0, 62.5, 125, 250, 500 and 1000 ppm for methomyl.

The LD_{50}, of these insecticides against third-inster *H .armigera* of comparable ages was determined using topical application method as recommended by the Entomological Society of America (Anonymous 1970). The insecticides were applied to the larvae using a Hamilton® repeating dispenser fitted with 50-µl syringe and 1µl drop was applied to the thoracic dorsum of the larvae (weighing 30–40 mg). Control insects were treated with acetone alone. For each control and test group, the insecticides were replicated 3–4 times with 10–12 larvae in each replication. The larvae were maintained in 12cs multi-wells with lid or in 45 x 30 x 25mm plastic cups with fresh diet, before the trials ensued.

Mortality was recorded by counting dead or moribund insects after 24, 48 and 72 hours. The larvae that could not respond to pencil tip prodding were considered dead. The LD 50's were estimated by probit analysis as described by Finney (1971). Natural mortality was corrected by using Abbott's formula (Abbott 1925). The LC_{50} and the 95% Fiducial Limits (FL) were calculated using the SAS probit analysis software (SAS Institute, 1985). The resulting lethal concentrations (LC_{50}'s) were calculated by 1000 to convert the ppm to microgram (µg) and the result was again divided by average larval weight (0.03g), which was then expressed in microgram per gram (µg/g) body weight. The major criterion used to discriminate between resistant and susceptible populations was failure of the 95% Fiducial limits (FL) to overlap at LD_{50} ($P < 0.05$).

Among field populations tested, a population with low LD_{50} value was selected and used as susceptible check. Accordingly, the Dukem population was selected as susceptible check for endosulfan, profenofos and methomyl while the Arbaminch population was used for lambdacyhalothrin. Thus, resistance ratios (RR) for endosulfan, profenofos and methomyl were calculated by dividing the LD_{50} of Arbaminch, Dubti, Humera, Zemea and Werer populations by LD_{50} of Dukem population. Resistance ratio of lambdacyhalothrin was obtained by dividing LD_{50} of Dubti, Dukem, Humera, Werer and Zemea by LD_{50} of Arbaminch population

Results

Insecticide use

Insecticide use varied both within sampling regions and from one year to another. In the chickpea-growing regions, the farmers use insecticides seldom for armyworm control. The use of either of these chemicals for African bollworm control on chickpea was not reported. In all major cotton growing regions, the use of endosulfan is a common practice and the number of applications ranged from 1 to 6 per season, depending on pest infestation and attack. Currently, the use of lambdacyhalothrin (Karate®) on cotton is restricted to only 1 to 2 applications in a season and the use of methomyl for bollworm control was reported from Middle Awash Agricultural Development Enterprise for the period 1982 only.

Endosulfan

Results of dose-mortality response of the probit analysis indicated low LD_{50} (21.28) μg/g larval body weight for Dukem population (Table 1). The Zemea population has also recorded low LD_{50} (24.57) μg/g larval bodies weight with a slope of 2.83. High LD_{50} (73.47) was recorded for Arbaminch population. The 95% FL did not overlap for the Arbaminch population. The highest RR was calculated for Arbaminch population (3.54) and the lowest for Zemea (1.15). The recorded RR for populations from the other fields was very low and ranged from 1.40 to 1.62 with slope values ranging between 1.66 and 2.83. Heterogeneity was not significant for all populations tested (χ^2, $P < 0.05$).

Table 1. Probit analysis of dose-mortality response of *H. armigera* populations to endosulfan

Population	n[a]	LD_{50} (μg/g)	Fiducial limit (95%)	Slope ± SE	χ^2	RR[b]
Arbaminch	1536	73.47	47.26 – 95.58	2.82 ± 0.55	0.544	3.452
Dubti	490	34.58	25.17 – 44.29	2.4 ± 0.398	0.235	1.625
Dukem	540	21.28	16.82 – 27.56	2.34 ± 0.328	4.958	-
Humera	390	30.32	11.78 – 25.15	1.66 ± 0.44	1.440	1.425
Werer	780	29.84	14.05 – 42.53	2.58 ± 0.598	3.267	1.402
Zemea	1860	24.57	18.99 – 30.08	2.83±0.389	1.259	1.154

[a] = Number of larvae tested; [b] RR (Resistance Ratio) = LD_{50} of test population/LD_{50} of Dukem population

Profenofos

The overall results, of bioassay of profenofos indicated that the LD_{50} value for Dukem population was 3.87. The highest LD_{50} (7.74) was recorded for the Dubti population (Table 2). The LD_{50} values for populations from Arbaminch, Humera, Werer, and Zemea were 4.83, 4.04, 4.06, and 4.49 μg/g larval body weight, respectively. In general, the LD_{50} and RR values were very low for all treatments and ranged from one to two fold. The slopes of the probit equations for all treatments were greater than 2 except for the Werer population (1.26) and, the FL overlapped for all populations. Generally, all populations tested for profenofos resistance were found susceptible as indicated by overlapping FL at LD_{50} values.

Table 2. Probit analysis of dose-mortality response of *H. armigera* populations to profenofos

Population	n[a]	LD_{50} (μg/g)	Fiducial limit (95%)	Slope ± SE	χ^2	RR[b]
Arbaminch	1020	4.834	3.87 – 6.00	2.58 ± 0.31	1.28[4]	1.249
Dubti	462	7.741	4.99 – 10.48	2.57 ± 0.42	1.48[3]	2.00
Dukem	630	3.871	1.79 – 5.68	2.45 ± 0.52	0.19[3]	–
Humera	700	4.042	2.56 – 5.57	2.55 ± 0.40	2.50[3]	1.044
Werer	1400	4.068	3.33 ± 4.93	1.26 ± 0.15	2.86[4]	1.051
Zemea	1456	4.498	3.78 ± 5.34	2.92 ± 0.30	1.78[4]	1.162

[a] = Number of larvae tested; [b] RR (Resistance Ratio) = LD_{50} of test population/LD_{50} of Dukem population

Lambdacyhalothrin

The probit mortality analysis indicated that the Arbaminch population was significantly lower ($P< 0.05$) than that of populations from other sites (Table 3). The steep log dose-probit (Ldp) line slope (2.43), low LD_{50} value (0.05) and none overlapping FL (0.03–0.08) (Table 3) indicate that the Arbaminch population was susceptible to lambdacyhalothrin. On the contrary, the high and non- overlapping FL and high LD_{50} value (0.52) are indicators for presence of resistance in Dubti population. The highest RR (10.4) was obtained for Dubti population. The recorded RR for other field populations was low (2.68 to 4.24) with slope values ranging from 1.62 to 2.69. The Ldp lines of all of the six populations had non-significant heterogeneity (Heterogeneity χ^2, $P < 0.05$). However, the pooled χ^2 (0.287) over populations was found significant at site (Heterogeneity $\chi^2_{[4]}$, $P > 0.05$), indicating the presence of mixed resistant phenotypes.

Table 3. Probit analysis of dose-mortality response of *H. armigera* populations to lambdacyhalothrin

Population	n[a]	LD_{50} (μg/g)	Fiducial limit (95%)	Slope ± SE	χ^2	RR[b]
Arbaminch	570	0.050	0.030 – 0.080	2.43 ± 0.56	1.442 ns	-
Dubti	240	0.520	0.410 – 0.640	2.52 ± 0.29	3.232 ns	10.40
Dukem	420	0.212	0.015 – 0.270	2.53 ± 0.4	1.378 ns	4.24
Humera	210	0.134	0.081 – 0.177	2.69 ± 0.58	0.369 ns	2.68
Werer	1392	0.180	0.100 – 0.250	1.62 ± 0.25	1.202 ns	3.60
Zemea	1600	0.150	0.090 – 0.210	1.78 ± 0.26	3.865 ns	3.00

[a] = Number of larvae tested; [b] RR (Resistance Ratio) = LD_{50} of test population/LD_{50} of Arbaminch population

Methomyl

The lowest LD_{50} (8.57) and (8.66) μg/g larval body weight was recorded for Dubti and Dukem populations, respectively and the highest (15.61) for Arbaminch population. The LD_{50} values for Humera and Zemea populations were 11.40 and 13.27μg/g larval body weight, respectively. The slope values of all populations ranged from 1.57 to 2.61. The resulting RR was not significant and ranged from 1.01 to 1.82 (Table 4). Due to overlapping of FL and low LD_{50} values, the populations were found to be susceptible to methomyl.

Table 4. Probit analysis of dose-mortality response of *H. armigera* populations to methomyl

Population	n[a]	LD_{50} (μg/g)	Fiducial limit (95%)	Slope ± SE	χ^2	RR[b]
Arbaminch	900	15.61	11.47 - 23.69	1.698 ± 0.295	0.010	1.80
Dubti	180	8.57	6.132 – 12.05	1.577 ± 0.701	2.594	0.99
Dukem	540	8.66	1.54 – 13.78	2.611 ± 0.877	3.079	-
Humera	364	11.40	2.06 – 18.22	1.966 ± 0.668	1.093	1.32
Werer	1152	10.45	2.12 – 19.22	1.580 ± 0.445	0.772	1.21
Zemea	1296	13.27	7.86 – 17.69	2.396 ± 0.561	0.848	1.53

[a] = Number of larvae tested; [b] RR (Resistance Ratio) =(LD_{50} of test population/LD_{50} of Dukem population

Discussion

Endosulfan

H. armigera populations in major cotton growing areas of Ethiopia (Dubti, Humera and Werer) did not develop significant level of resistance to endosulfan. However, the variations observed among field populations for endosullfan sensitivity indicate the presence of some level of resistance. The low LD_{50} values for Dukem (21.28) and Zemea (24.57) populations indicate either their true susceptibility to endosulfan, or presence of low incipient resistance level. Similar LD_{50} value (26.4) was recorded for the susceptible BK77 strain in Cote d'Ivore (Martin et al. 2002).

Pesticide use history suggests that the use of endosulfan on chickpea is likely. However, the recorded incipient level of resistance for populations from Dukem area could be a result of gene flow via migration of moths during dry season from cotton farms to horticultural crops and vice versa (Daly & Gregg 1985). On the other hand, the observed non-overlapping fiducial limit, coupled with high LD_{50}, (73.47) indicate the presence of resistance in Arbaminch population. At Arbaminch and along Lake Abaya (Umolante, Lante and Mierab Abaya), the use of endosulfan on cotton and tomato is a common practice. Therefore, the results of this study indicate the presence of resistance to endosulfan in the Arbaminch population.

Generally, the data indicate that *H. armigera* populations in Ethiopia are developing resistance to endosulfan. Similar results (RR < 30) were reported on pyrethroid resistant populations of *H. armigera* in Thailand (Ahmad & McCaffery 1988), India (McCaffery et al. 1989) Armes et al. 1992), and Australia (Forrester et al. 1993), Cote d'Ivore (Martin et al. 2002) and Pakistan (Ahmad *et al.* 1995)

Profenofos

The LD_{50} values and resistance ratios showed similar responses for all populations and no significant difference was found (P 0.05) The low LD_{50} recorded for Dukem and populations are true as the populations had no history of exposure to profenofos. The low LD_{50} values (3.87 to 7.74) for all tested populations could be used as estimates of the baseline response of *H. armigera* to profenofos. Therefore, it was difficult to delineate the field populations into resistant and susceptible populations due to lack of standard susceptible strain. However, the low LD_{50} recorded for Dukem population was comparable with the LD_{50} of susceptible strain from Cote d'Ivore (Martin et al. 2002). The West African susceptible strain (BK77) had 6.84µg, while the Dukem population had 3.87µg active ingredient per gram of larvae (Martin et al. 2002). This concurs with the results of Ahmad et al. (1995) in Pakistan, where he had reported 0.7–24 resistance ratios. Similarly, low level of resistance to profenofos was reported in Australia (Gunning & Easton 1993, and McCaffery 1998). However, the low LD_{50} expressed by populations from major cotton producing areas of Ethiopia (Arbaminch, Dubti, Humera and Werer) could be due to the low selection pressure of profenofos

Lambdacyhalothrin

The low LD_{50} of lambdacyhalothrin against *H. armigera* from Arbaminch was comparable with similar results obtained by different investigators elsewhere. Martin et al. (2000) recorded 0.046–0.069 LD_{50} for deltamethrin on the susceptible (BK77) strain in Cote d'Ivore. Ahmad (1994), and Ahmad & McCafferey (1988) reported LD_{50} ranging from 0.009–0.031 for cypermethrin and 0.015 μg/g for fenvalerate from the susceptible Thailand species. Therefore, it is possible to consider the Arbaminch population as baseline susceptible population.

The slop of the regression lines for all groups was low (1.62–2.69), indicating the presence of genetic heterogeneity among populations for resistance trait (Aldosari et al. 1996). The result of the current work indicated that all field-collected *H. armigera* populations were susceptible to lambdacyhalothrin, except the Dubti population for which high and non-overlapping fiducial limit and resistance ratio of 10 fold was recorded. This could be the result of repeated use of lambdacyhalothrin (Karate 0.8% ULV at the rate of 2 l/ha) for whiteflies and bollworm control at Dubti. In other areas, however, the use of lambdacyhalothrin against bollworm and other insect pests in cotton farms is almost similar, except for the two chickpea growing areas, where pyrethroid use pattern is not exactly known. Upper Awash Agro-industry Enterprise applies lambdacyhalothrin only at the end of the cotton growing season, while at Middle Awash Agricultural Development Enterprise, the use of lambdacyhalothrin is restricted to the period 90–120 days. In chickpea growing areas, the use of pyrethroids is seldom and if at all, it would probably be only once per season. Therefore, it is possible to consider the mosaic use pattern in small-scale farming systems and the low selection pressure on large-scale commercial cotton farms as main factors for maintaining susceptibility of *H. armigera* to λ-cyhalothrin in Ethiopia. However, it is essential to monitor continuously or exert selection pressure for many generations to be sure of the pyrethroid resistance, as the insect is known to migrate long distances as observed in Australia and West Africa (Daly & Gregg 1985).

Methomyl

The LD_{50} of methomyl against bollworm populations collected from all sites was low. The resistance ratios were also low and ranged from 1 to 1.82. The lower slopes of the Ldp-line and the overlapping fiducial limits indicate similarities of field populations in response to methomyl. According to the results of this study, there was no significant difference between populations from the six geographic locations in their susceptibility to methomyl. This lack of variation in response indicates the absence of selection pressure for methomyl in all populations tested. Therefore, methomyl could be used in future insecticide resistance management program as one of the insecticides in the alteration scheme.

Conclusion

The LD_{50} values recorded for all the technical grade insecticides during this study were low compared to results obtained in Australia, West Africa and Asia. However, due to lack of standard susceptible strain, it was difficult to delineate the field populations into resistant and susceptible strains. To overcome difficulties in evaluating field populations in the absence of susceptible strain, it is widely recommended to continuously monitor field populations over broad geographic areas. Laboratory strains provide the advantage of totally susceptible benchmark for calculating resistance ratios, but these values have little relevance to any true field levels. Therefore, even in the absence of laboratory reared susceptible strain, it is possible to generate dose-mortality data, which can be used as baseline for future monitoring works.

Currently, endosulfan is the major insecticide used for the control of cotton bollworm in all commercial farms in Ethiopia. The Arbaminch population has expressed presence of resistance to endosulfan. At the same time, resistance in populations from other location to endosulfan is also in the rise. Thus, to prolong the active life of this insecticide, its use must be limited to only one application per season. In the mean time, other alternative control methods must be developed and/or alternative insecticide must be screened.

Results of this study showed that *H. armigera* populations from Arbaminch, Humera and Werer did not develop significant level of resistance to lambdacyhalothrin. The Dubti population had shown a degree of resistance to this insecticide and thus, the use of this product in the area must be limited. All *H. armigera* populations tested were found susceptible to methomyl, λ-cyhalothrin and profenofos. It appears, however, that resistance may increase with time and space as insecticide application continues. Thus, the continuous monitoring of insecticide resistance in cotton pests must form an integral part of the chemical control of pests to enable the detection of resistance as early as possible so that its economic, toxicological, and biological consequences may be obviated. Therefore, it is essential to periodically monitor resistance level in order to avoid use of wasted insecticide and to prolong the usefulness of endosulfan, lambdacyhalothrin and profenofos. Once resistance to an insecticide is detected, the use of the product in that area must be suspended or abandoned altogether. To ensure the effective life of available insecticides, their use must be limited and resistance management strategy must be developed and implemented.

The present study was a step towards the development of a monitoring program designed to detect changes in susceptibility that may result from repeated and prolonged exposure to a chemical. It is expected that the data would serve as baseline for future insecticide resistance monitoring and management works. It is also hoped that this paper will stimulate further studies on resistance monitoring on the most economically important agricultural insect pests, which are controlled by extensive application of insecticides.

Acknowledgements

We thank the Adamitulu Pesticide Processing and Packing Share Company, Aventis Crop Science, and SYNGENTA for supplying technical grade insecticides. We also gratefully acknowledge the technical support of staff of Entomology and Pathology Sections, Werer Agricultural Research Center and the financial support of the Ethiopian Agricultural Research Organization.

References

Abbott SW. 1925. A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology* 18: 265–267.

Ahmad M, MI Arif and Z Ahmad. 1995. Monitoring insecticide resistance of *Helicoverpa armigera* (Lepidoptera: Noctuidae) in Pakistan. *Journal of Economic Entomology* 88: 771–778.

Ahmad M. 1994. Analysis of insecticide resistance in *Helicoverpa armigera* in Thailand. FAO Plant Protection Bulletin. 42: 63–69.

Ahmad M and AR McCaffery. 1988. Resistance to insecticides in a Thailand population of *H. armigera* (Hubner) (Lepidoptera: Noctuidae). *Journal of Economic Entomology* 81: 45–48.

Aldosari SA, TF Watson, S Sivasupramaniam and AA Osman. 1996. Susceptibility of field populations of Beet armyworm

(Lepidoptera: Noctuidae) to cyfluthrin, methomyl and profenofos and selection for resistance to cyfluthrin. *Journal of Economic Entomology* 89: 1359–1363.

Anonymous. 1970. Standard method for detection of insecticides resistance in *Heliothis zea* (Boddie) and *H virescens* (F.). Bulletin of Entomological Society of America 16: 147–153.

Armes NJ, DR Jadhav, GS Bond and ABS King. 1992. Insecticide resistance in *Helicoverpa armigera* in South India. Pesticide Science. 34: 355–364.

Daly JC and P Gregg. 1985. Genetic variation in *Heliothis* in Australia: Species identification and gene flow in the two pest species. *H. armigera* (Hubner) and *H. punctigera* Wallengren (Lepidoptera: Noctuidae). Bulletin of Entomological Research. 75: 169–184.

Finney DJ. 1971. Probit analyses, (3rd ed.) Cambridge University Press, Cambridge, London.

Forrestert NW, M Cahill, LJ Bird and JK Layland. 1993. Management of pyrethroid and endosulfan resistance in *Helicoverpa armigera* (Lepidoptera: Noctuidae) in Australia. Bulletin of Entomological Research, Supplement Series No. 1: 132.

Gunning RV and CS Easton. 1993. Resistance to organophosphate insecticides in *Helicoverpa armigera* (Hubner) (Lepidoptera: Noctuidae) in Australia. General and Applied Entomology 25: 27–34.

Hoskins WM and R Craig. 1962. Uses of bioassay in entomology. Annual Review of Entomology. 7: 437–464.

Martin T, GO Ochou, NF Hala, JM Vassal and M Vaissayre. 2000. Pyrethroid resistance in the cotton bollworm, *Helicoverpa armigera* (Hubner), in West Africa. Pest Management Science 56: 549–554.

Martin T, GO Ochou, M Vaissayre and D Fournier. 2002. Positive and negative cross-resistance to pyrethroids in *Helicoverpa armigera* from West Africa. Resistance Pest Management Newsletter 12: 16–19.

McCaffery AR. 1998. Resistance to insecticides in heliothine Lepidoptera: A global view. Philos Trans Royal Society. London. B. 353: 1735–1750.

McCaffery AR, ABS King, AJ Walker and H El-Nayir. 1989. Resistance to synthetic pyrethroids in the bollworm, *Heliothis armigera* from Andhara, India. Pesticides Science 27: 65–76.

Muggleton J. 1984. The evolution of insecticide resistance and its relevance to control strategy. In: *Proceedings of British Crop Protection Conference*, Croydon, U.K, BCPC Publications. 585–592 pp.

Read PD Jr. 1941. Cotton in Ethiopia, Eritrea and The Somalilands. *In*: Cotton in Africa. National Cotton Council of America, Memphis 1, Tennessee, USA

SAS Institute. 1985. SAS users guide: Statistics, version 5, SAS Institute, Cary, N. C. USA.

Shorey HH and RL Hale. 1965. Mass-rearing of the larvae of nine noctuid species on a simple artificial medium. *Journal of Economic Entomology* 58: 522–524.

Taekle ER and JM Jenson. 1985. *Heliothis punctigera*. *In*: Singh P. and R F Moore (eds) Handbook of insect rearing. Vol. II. Pp. 312–322. Elsevier, Amsterdam.

Tessema Megenassa, Taddesse Gebremedhin and Tesedeke Abate. 1980. Review of the state of agricultural pesticides use in Ethiopia. *Ethiopian Journal of Agricultural Science* 2: 29–37.

Wolfenbarger DA, PRV Bodegas and RG Flores. 1981. Development of resistance in *Heliothis* spp. in the Americas, Australia, Africa and Asia. Bulletin of Entomological Society of America **27**: 181–185.