VOL. 1 NO. 2 DECEMBER 1994 ISSN 0128 - 7141

BRC JOURNAL

Journal of the Bureau of Research and Consultancy, MARA Institute of Technology.
(Jurnal Biro Penyelidikan dan Perundingan, Institut Teknologi MARA)

BUREAU OF RESEARCH & CONSULTANCY
INSTITUT TEKNOLOGI MARA

A PRELIMINARY STUDY ON THE RESIDUE OF CARBOFURAN IN SWEET POTATO IN PERLIS

by
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ABSTRACT

The aim of this research was primarily to identify the correct dose of Furadan, a systemic pesticide, in the control of sweet potato weevil. Different doses of the chemical, viz., 0, 0.5, 1.0, 1.5, 2.0, 2.5, and 3.0 kg a.i. Furadan 3 G / ha, were applied to the soil, and their effects on the yield and residue in the plant and soil were investigated. Gas chromatography was used in the residue studies. The solid phase extraction method, a modification of the techniques used by Beachamp *et al.* and Getzin *et al.* in 1989, was used in isolating pure carbofuran (to be used for the preparation of standard solution for residue analysis) from the commercial product, Furadan 3 G. No residue was detected from sweet potato tubers treated with different doses of the chemical. However, carbofuran persisted in soils from polybags for 30 days. The rate 1.5 kg a.i. / ha, showed a phytotonic effect on tuber formation and a highest average yield in Empat Bulan (yellow-flesh variety of sweet potato). It also recorded the lowest *Cylas* damage of 15% in Serdang 1 variety, when compared to the 50% damage in the no chemical treatment. Hence, it could be selected as one of the pesticides in the control of *Cylas* sp. in sweet potato.

Keywords: Residue, carbofuran, sweet potato, Perlis.

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INTRODUCTION

Carbofuran (trade name Furadan) is a systemic insecticide, acaricide and nematicide, used against both foliage and soil pests. It is toxic (LD50 oral : 5 - 14 mg/kg) and it persists in soil for 30 - 60 days (Hill & Waller, 1984). The chemical is not bioaccumulative in animal tissue (Brown, 1987), but it can lower the cholenesterase level in blood samples of farm workers frequently exposed to the chemical (Ngatia & Mgnani, 1980).

The permissible level of carbofuran residue in root-crops is 0.5 ppm, while in non-leafy vegetables, it is 0.1 ppm (Pesticide residue, 16th Schedule, 1985). The chemical, which is granular in form, is widely used by Perlis farmers in the control of padi pests. Its 'easy to apply' nature, encourages some farmers to use it indiscriminately in the cultivation of their supplementary cash crops like sweet potato, and chilly.

The aim of this research was primarily to identify the correct dose of Furadan in the control of sweet potato weevil (*Cylas formicarius*. F. Coleoptera: Cucurlionidae) a major pest of the crop during harvest.

MATERIALS AND METHODS

Studies *in vivo*

Two varieties of sweet potato (*Ipomeoea batatas;* Family: Convolvulaceae) viz., "Empat bulan" (yellow flesh) and "Serdang I" (white flesh) were selected for the field experiments. The field trials were conducted in plots with sandy loam soil suitable for sweet potato cultivation. Raised bunds (10 m long, 1 m wide, 30 cm high) using the RCBD (randomized complete block design) with 4 replicates were planted with 30 cm long *Cylas* sp. infested stem-cuttings obtained from a farmer's plot. The distance between each bund was 1 m. Varying doses of Furadan 3 G: 0.5 kg a.i./ha, 1 kg a.i./ha, 1.5 kg a.i./ha; 2.0 kg a.i./ha; 2.5 kg a.i./ha and 3.0 kg a.i./ha, were applied to bunds in each replicate, 2 weeks after planting. To reduce the interrow effect (Rui & Giraldi 1958) untreated control bunds were alternated with chemically treated bunds. Each bund was planted with 20 stem cuttings (each 45 cm apart). Furadan 3 G granules were applied in small furrows (radius: 10 cm; depth: 2 cm) along each planting point. These furrows were covered with soil after administering the chemical treatments.

For residue studies in soil, sweet potato cuttings were planted and treated with Furadan in black nursery polybags measuring 30 cm x 23 cm, using the RCBD design with 3 replicates. For each of the six chemical treatments plus the control, six polybags were used. Soil samples from the polybags were analysed at monthly intervals for residue studies. Throughout the experiments, the regular agronomic practices for sweet potato cultivation were observed and besides Furadan no other pesticide was used. The ICI Fertilizer administration used was: CIRP - 200 kg/ha at planting, ICI Fertilizer no 45 (12:12:17:2) - 200 kg/ha, 1 week and 4 weeks after planting. For residue studies in the plants, sweet potato was harvested 3 months after planting and plant samples were sent to the analytical laboratories of the University Science, Pulau Pinang for residue analysis.

The *Cylas* sp. damage in sweet potato was observed during harvest. Twenty tubers of sweet potato were selected at random from each harvested bund. Each tuber was then cut horizontally with a sharp knife and observed for the bored tunnels caused by the *Cylas* sp.

Studies *in vitro*

Methods of analyzing the residue of carbofuran in plants and soil include: gas chromatography (Cook *et al.* 1969); gas liquid chromatography, UV spectrophotometry, immunization technique and anti-cholenterasw enzyme technique (Gould 1971); thin-layer chromatography (TLC) and radio-assay technique (Huque 1972). In studies on residue of pesticides, TLC is time-saving and can be used in varying situations (Khan 1966).

The residue analyses for carbofuran in sweet potato were done in the analytical laboratories of the Chemistry Department at the University of Science, Pulau Pinang, using the solid phase extraction method, a modification of the method of Beauchamp *et al.* 1989 and Getzin *et al.* 1989.

Split mode technique of gas chromatography (model Hewlett Packard 5880 A series: SPB 20 capillary column, – 30 m long; gas – Helium; oven temperature – 220°C; injection temperature 270°C; marker temperature in gas chromatography – 270°C) was employed in the residue analysis. The solid phase extraction (SPE) column (3 ml) was packed with 500 mg of silica gel using a vacuum pump.

The chemicals used in gas chromatography were: Acetone, A.R. (further purified). Dichloromethane, A.R. (further purified). Benzene, and Hexane.

The buffers employed were: HCl – KCl buffer pH 2.0 (106 ml 0.2 M HCl mixed with 500 ml 0.2 M KCl); Carbonate – bicarbonate buffer pH 10.7 (450 ml 0.2 M Natrium carbonate mixed with 50 ml 0.2 M Natrium bicarbonate).

The standard solutions used in the collaboration of the apparatus were 2 ppm, 4 ppm, 6 ppm, 8 ppm, and 10 ppm concentrations obtained from the stock solution of 500 ug/ml carbofuran isolated from Furadan 3 G and later cross-checked with the pure carbofuran standard solution obtained from the Head Office, Department of Agriculture, Kuala Lumpur, Malaysia.

The isolation of carbofuran from the various samples was done by the following methods:

i) The samples under study were cleaned, digested and dried in the oven at 95°C.

ii) 200 g of the crushed sample were put into a dark bottle and treated with 300 ml acetone and later with 35 ml HCl-KCl buffer pH 2.0. The bottle was then sealed tightly and put on a mechanical shaker overnight.

iii) The next day the sample was allowed to settle. It was then filtered using a Whatman N. 42 filter paper to obtain a clear extract which was collected in a 250 ml conical flask.

iv) 100 ml of the above extract was put into a 250 ml separation flask. A total of 75 ml of dichloromethane was mixed with the separated extract followed later by 25 ml carbonate – bicarbonate buffer pH 10. 7. The above mixture was shaken to obtain 2 layers.

v) The bottom layer containing a mixture of acetone and dichloromethane was separated out and filtered into a 250 ml conical flask through anhydrous natrium sulphate. The bottom layer of the extract still containing the mixture of acetone and dichloromethane was further treated with 25 ml dichloromethane and refiltered. The extract obtained was pooled with the earlier extract.

vi) The extract was then placed on a steam bath and concentrated to a volume of about 5 ml using a rotary evaporator. The concentrated extract was stored in a small bottle to be used later in *solid phase extraction* or "SPE - clean up".

vii) In the SPE clean up drops of benzene were put into the 3ml SPE column which was packed with 500 mg silica gel and connected to a vacuum pump. Before the column could dry up, a drop of the sample extract under study was passed through SPE column at a flow rate of 2 ml/min. After this the column was washed with benzene, dried and rinsed again with drops of a mixture of hexane and acetone (19 + 1, v/v).

viii) The SPE column was then operated with hexane acetone mixture (1 + 1, v/v) and the extract solution collected in a small bottle to be further concentrated and dried. One ml of acetone was added to the above product when gas chromatography analysis was to be done. For carbofuran extracted from Furadan, CHN test analysis was done to check on the purity of the sample.

ix) The gas chromatography apparatus was standardized by obtaining chromatographs with purified acetone and carbofuran standard solution before spectra for the test sample were analysed. The volume used for each injection was 5.0 *ul*

Based on the time table used in the *in vivo* studies, the test samples were analysed at different time intervals.

RESULTS

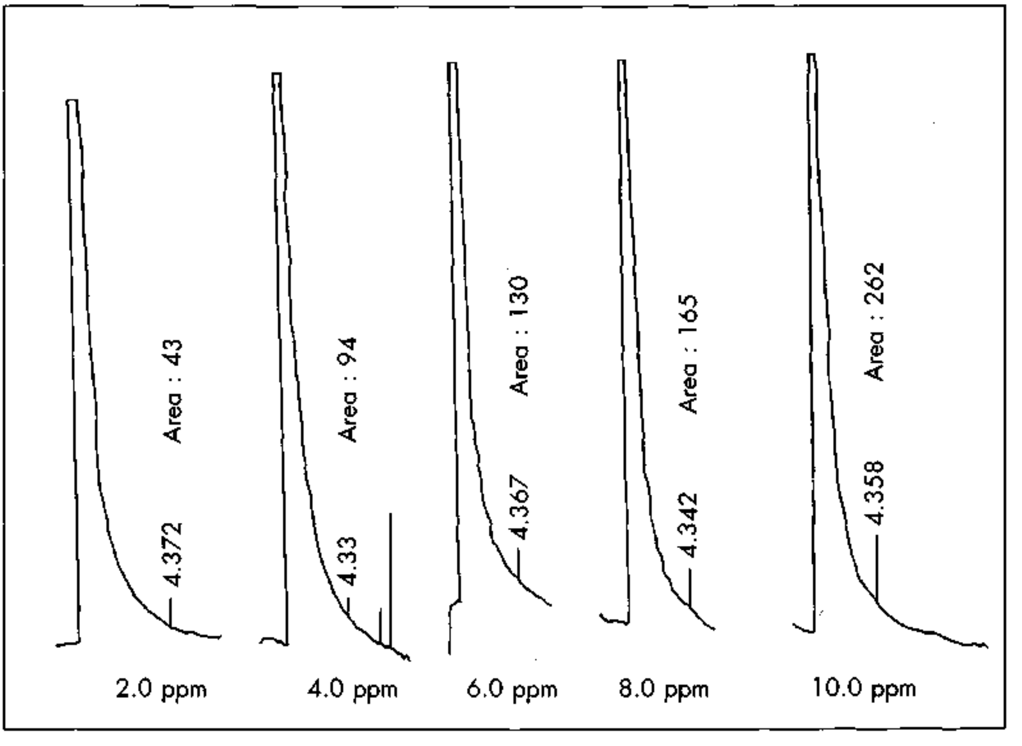
The CHN test analysis used to determine the purity of carbofuran extracted from the commercial product Furadan 3 G is presented in Table 1.

Table 1 CHN analysis for carbofuran extracted from Furadan

Sample reference	%C	%H	%N
Carbofuran extract	65.14	6.89	6.23
Theoretical value	65.16	7.79	6.33

This method used in the isolation of carbofuran from Furadan was highly successful, since the purity of the extracted sample was nearly 100%. The chromatograph of the sample is shown in Figure 1.

Figure 1 Gas Chromatograph of Standard Solution of Carbofuran Isolated from Furadan



Field studies in March 1988 using the "Empat Bulan" variety showed that 1.5 kg carbofuran/ha provided the highest average yield (Fig. 2) when compared with other treatments, but this treatment was not significantly different from treatments 4, 5 and 6. However, this group of treatments significantly increased yield as compared to the control T_0 ($p = 0.05$; df = 18; MSE = 1.132). Furadan 3 G also has a phytotonic effect on the root development of sweet potato. This effect was shown in treatment 3. Residue studies of carbofuran in sweet potato were not done.

Studies on the residue of carbofuran in the soil from polybags planted with sweet potato showed that the chemical is persistent in the polybag soil for more than 30 days.

The gas chromatography spectra with various treatments did not show a clean peak (Fig. 3). Standard solution of carbofuran at various ppm concentrations were added at a ratio of 1 : 1 to those Furadan treatments which showed tiny peaks so as to determine their concentrations as tabulated in Table 2.

Figure 2 Yield in Empat Bulan Variety of Sweet Potato after Treatment with Various Doses of Furadan

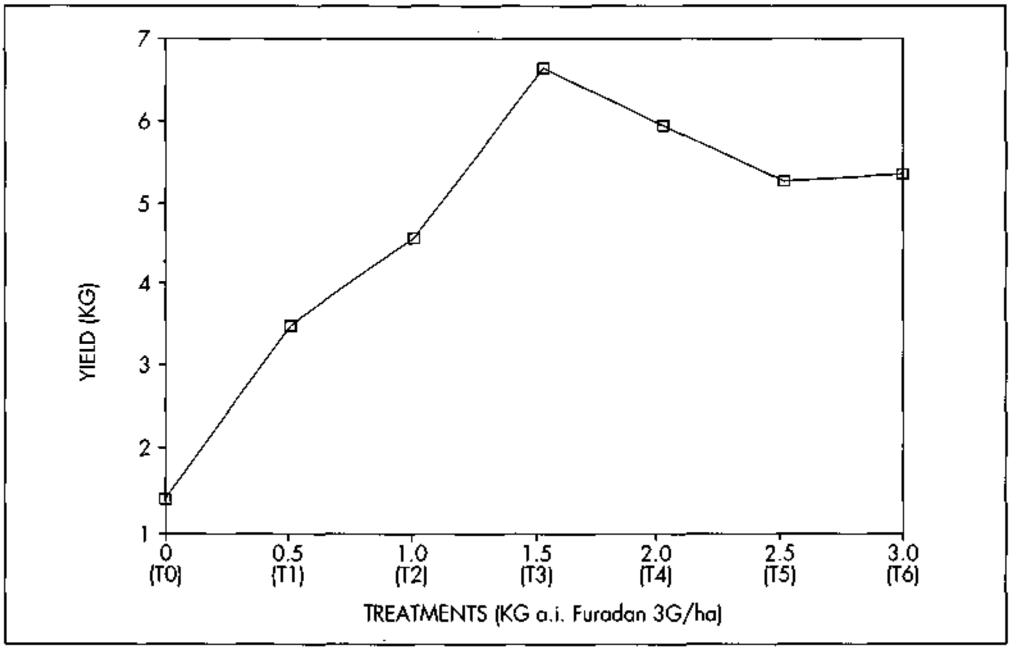


Figure 3 Gas Chromatograph of Soil sample 2 after adding 6 ppm Standard Solution

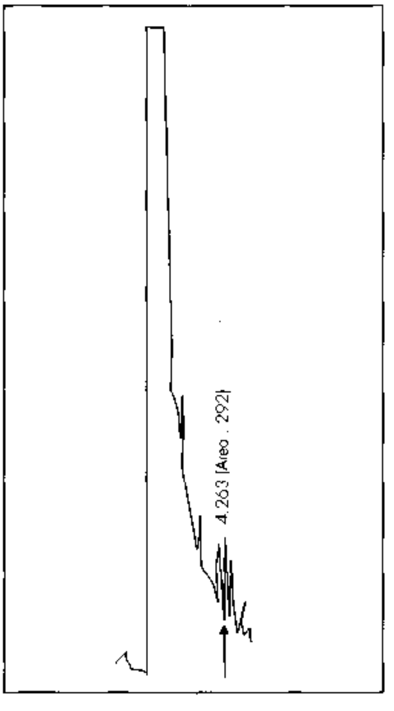


Table 2 Concentration of Carbofuran after adding the standard solution

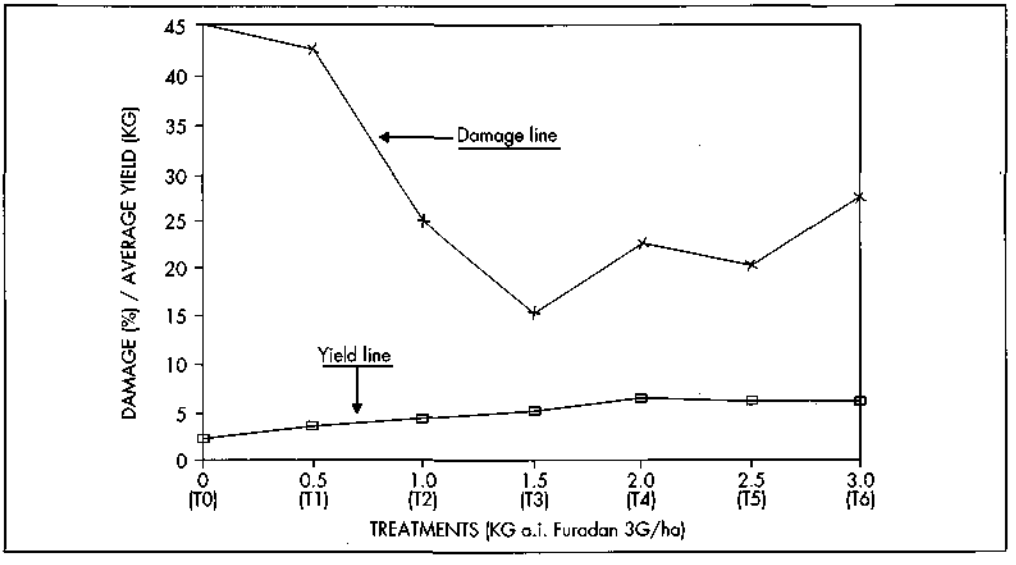
Sample	Treatment	Carbofuran conc. (ppm)
0	Treat 0 (control)	0
1	Treat 1	-
2	Treat 2	1.8
3	Treat 3	-
4	Treat 4	3.85
5	Treat 5	11.50
6	Treat 6	-

Carbofuran residues in ppm were detected from treatments 2, 4, and 6. Residues were not detected in the other treatments. This may be due to the leaching, drying or other form of degradation which might have occurred in the polybag soil which was predominantly sandy-loam in texture. Even though the results from Table 2 are not conclusive, they do demonstrate that the chemical persists in the soil for more than 30 days but less than 60 days. The test samples were analysed to the minimal range of 1 ppm. No residues were detected in roots, stems and leaves of sweet potato grown in polybags.

Another field trial was conducted in September 1990 to study the Furadan dose which could control the *Cylas* pest effectively. Serdang 1 variety infested by the *Cylas* pest, was planted in this trial.

As in the previous trial, the group of treatments 3, 4, 5, and 6 showed a significant difference over the control ($p = 0.05$; df = 18 MSE = 1.22) when their yields were compared (Fig. 4). The population of *Cylas* sp. in the bund treated with 1.5 kg/ha was low. Gas chromatography analysis showed no residue of carbofuran in plants, viz., the tubers treated with various doses of Furadan.

Figure 4 Effect of Furadan in Serdang 1 variety of Sweet Potato.



DISCUSSION

The difficulty in obtaining a pure carbofuran solution for the gas chromatography studies in the early stages of the research, sparked our interest in isolating the chemical in a pure form from the commercial product Furadan 3 G. The solid phase extraction, a modification of the methods of Beauchamp *et al.* 1989 and Getzin *et al.* 1989 was successful, as the carbofuran extracted from Furadan was almost 100% pure.

Carbofuran persists in the soil for about 90 days (Miller & Berg 1969). The result in the present study, based on sandy loam soil in polybags, however showed that the pesticide remained in the soil for about 30 days. The polybags were selected in the present study, not only to ensure the uniformity of the soil structure, but also for convenience in the experimentation. More than 50% carbofuran is lost after 30 – 60 days in the soil (Lichtenstein 1972). This phenomenon was also observed in the present study from soil in polybags.

Cylas formicarius F. is an important pest in sweet potato (Yunus & Balasubramaniam 1981). The weevil is small and resembles an ant. The elytra is dark and shiny green with the prothorax yellow in colour. Its life cycle is between 20 – 30 days. The damage in sweet potato is done both by the grubs and adults which tunnel into the stems and tubers. The attacked tubers become discoloured internally and the tunnels are filled with faecal matter. This pest can be controlled through crop rotation, by keeping the soil mounded around the vines or by drenching infested soil with 0.1% a.i. of either heptachlor or dieldrin. Carbofuran was used for the first time in the present study to control *Cylas* sp in sweet potato.

Carbofuran has been a popular pesticide in pest management of several crops (FMC Pamphlet 1983). Its usage, based on the FMC information is summarized in Table 3.

Table 3 Use of Furadan in various crops

Crop	Dosage (a.i.)	Country	Pest
Tobacco	2 kg/ha	Argentina	Nematodes
Padi	1.5 - 2.0 kg/ha	Philippines	General insects
Maize	1.5 kg/ha(Furadan 3G)	Nigeria	Stem borers
Sugarcane	3 kg/ha	Brazil	Soil insects
Banana	2.5 kg/ha	St. Lucia	Soil insects
Coffee	1.5 kg/ha	Costa Rica	Soil insects
Potato	2.5 kg/ha	Venezuela	Soil insects
Potato	1.8 kg/ha	Spain	Colorado beetle

The field trial studies showed that 1.5 kg a.i. Furadan 3 G/ha, can be considered as the best dosage, since it controlled the *Cylas* sp. population in sweet potato by more than 80%, provided a good yield, and exhibited a phytotonic effect on the formation of tubers. This treatment also did not leave any residue in sweet potato after harvest. The present research is preliminary in nature. However, a suitable method to isolate carbofuran from Furadan, the commercial product of the pesticide has been developed. The solid phase extraction method could be further employed in other studies on the residue of pesticides in agricultural crops.

ACKNOWLEDGEMENT

The authors wish to express their thanks to: The Bureau of Research and Consultancy MARA Institute of Technology, Shah Alam, Selangor, Malaysia for providing a research grant for this project. The University of Science Malaysia for providing facilities to do the analysis of carbofuran residue at its analytical laboratories. The Agriculture Department of Malaysia for providing the standard solution of carbofuran for gas chromatography. The Principal, MIT Perlis for allowing to conduct field trials at the campus farms and all those who contributed directly or indirectly to the success of this research.

References

Beauchamp, K.W., Liu D.D.W., and E.J. Kitka. 1989. Determination of carbofuran and its metabolites in rice paddy water by using solid and liquid chromatography. J. Assoc. Off. Anal. Chem. 72: 845-847.

Brown, A.W.A. 1978. Insecticide residues and biotic food chains. In : Ecology of pesticide. John Wiley & Sons. New York: 270-319.

Cook, R.E., R.P. Stanowick and C.C. Cassil. 1969. Determination of carbofuran and its carbamate residue in corn using a nitrogen specific gas chromatographic detector. J. Agr. Food Chem. 17: 277-282.

FMC Phamphlet. 1983. Furadan (Insecticide/nematicide), world-wide summary. All crops. FMC Corporation, Philadelphia, U.S.A.

Getzin, L.W., C.G. Cogger, and P.R. Bristow. 1989. Simultaneous gas chromatographic determination of carbofuran, metalaxyl, and simazine in soils. J. Assoc. Off. Anal. Chem. 72: 316-364.

Gould, R.F. 1971. Pesticide identification at the residue level. In: Advances in Chemistry Series 104, American Chemical Society, Washington. 182 p.

Hill, D.S. and J.M. Waller. 1982. Pest and diseases of tropical crops. Vol. 1. Longman, London. 175 p.

Huque, N. 1972. Preliminary report on the residues of cabaryl granules in rice plants. In: Panel Proceeding Service, International Atomic Energy, Vienna.

Khan, M.A.Q. 1966. Pesticides in awuatic environments. Plenum Press, New York. 257 p.

Lichtenstein, E.P. 1972. Environmental factors affecting fate of pesticides. Degradation Syn. Org. Mol. Biosphere, Proc. Conf. 1971 (Pub. 1972). Nat. Acd. Sci. Washington, U.S.A.: 190-205 (Eng).

Miller, M.W. and G.G. Berg. 1969. Chemical fallout: Current research on persistent pesticides. Springfield, ill., Thomas,1969.

Ngatia, J. and A. Mgani. 1980. The effects of continuous exposure to organophosphorous and carbamate insecticides on the cholinesterase (CHE) levels in humans. In: Environmental Science 7. Elsevier Scientific Publ. Co. Amsterdam.

Rui, D. and G. Giraldi. 1985. Nematode fitoparasite and nematicidi. Annali della Spesimentazioni Agraria 12: 481-502.

Yunus, A. and A. Bakasubramaniam. 1981. Major crop pests in Peninsular Malaysia. Bulletin No. 138. Ministry of Agriculture Malaysia. 190 p.