

CHAPTER TWO

Evaluation of suitable insecticide baits for armoured bush cricket (Orthoptera: Hetrodinae) control in Botswana.

ABSTRACT

Cereal bran, maize meal and crushed crickets were evaluated for preference as bait carriers against the armoured bush cricket (ABC) *Acanthopplus discoidalis* (Walker). The quantities of maize bran, sorghum bran, millet bran, crushed crickets or maize meal consumed by ABC were not significantly different ($p < 0.05$) from each other. Farmers can therefore use locally available cereal bran for bait formulation. Bran-based baits formulated using eight different insecticides were laboratory-tested for oral toxicity against the ABC. Fipronil was the most toxic of the eight insecticides tested and its LD_{50} value ($0.05 \mu\text{g } \mu\text{l}^{-1}$) was significantly lower than that of the other insecticides. The toxicity of cyfluthrin was not significantly different from that of imidacloprid, cypermethrin and chlorpyrifos. Similarly, the LD_{50} of carbaryl did not differ significantly from those of imidacloprid, cypermethrin and chlorpyrifos. The least toxic of the insecticides tested against ABC were gamma-BHC and malathion, for which there was no significant difference ($p < 0.05$) in oral toxicity. The order of toxicity amongst these eight insecticides was: fipronil > cyfluthrin > imidacloprid > cypermethrin > chlorpyrifos > carbaryl > gamma-BHC > malathion. This study provides a guide to the potential performance of these eight insecticides and suggests appropriate dosages for use in bait against ABC. The results suggest that several insecticides could be suitable toxicants for bran baits against ABC. A subsequent study tested carbaryl and cypermethrin for mortality due to secondary poisoning. The primary mortality of both carbaryl (3 g a.i./kg bran) and cypermethrin (0.09 g a.i./kg of bran) was 100 %. Carbaryl gave 65 % mortality and cypermethrin gave 10 % mortality in secondary poisoning. Both carbaryl and cypermethrin did not kill any crickets fed with carcasses from secondary poisoning.

These results gave carbaryl an additional advantage to be used in formulating baits for control of armoured bush crickets in Botswana.

Key words: *Acanthoplus discoidalis*, insecticide baits, bait carriers, Botswana, LD₅₀, primary mortality, primary toxicity, Secondary cannibalistic poisoning.

INTRODUCTION

The armoured bush cricket (ABC), *Acanthopplus discoidalis* (Walker), belongs to the order Orthoptera, family Tettigoniidae, subfamily Hetrodinae. It is an omnivorous insect that feeds on a variety of wild grasses and broadleaved plants. It also attacks maize, sorghum and millet panicles and to a lesser extent cowpea, sunflower and other crop plants. In addition, ABC also cannibalise their own species. The ABC is considered the second most economically important cereal pest, after quelea birds, in eastern Botswana (Matsaert *et al.* 2000). Bashir *et al.* (1991) estimated a total yield loss of 40 % in sorghum fields during a serious ABC outbreak in six agricultural regions of Botswana. Damage levels of 34 % were observed in sorghum in this study.

Ingram *et al.* (1973) and Manthe (2000) reported ABC as a destructive pest, which is difficult for resource poor farmers to control. Insecticides have been the main means of controlling the ABC. However, resource poor farmers, in Botswana as elsewhere, often do not use insecticides, which are expensive and perceived as hazardous (Matsaert *et al.* 2000). This has prompted a search for affordable and easily implemented alternative strategies to control the ABC.

Insecticides formulated as baits have been used in the past to control a range of crop pests, locusts in particular, until the advent of organochlorine insecticides (Peregrine 1973). During the 1930's and early 1940's, baits using edible bran or saw dust as carriers was considered the most effective and economical method of protecting crops from rangeland grasshoppers in North America (Paul 1942; Shotwell 1942) and migratory locusts in Africa (Peregrine 1973; Steedman 1990). In many situations, baiting is effective because baits can be formulated and deployed so that they efficiently target one particular pest. Compared to liquid spray, which has the tendency to drift off target and affect non-target organisms, insecticide baits are cheaper and more target-specific in presenting toxic doses to target pests (Mukerji *et al.* 1981). Carbaryl- and malathion-treated bran baits have proved very effective against rangeland grasshoppers in North America, causing up to 85 % population reduction (Quinn *et al.* 1989). Besides being effective, the use of baits also offers considerable environmental and economic advantages compared to conventional spraying because less active ingredient is needed to

achieve a desirable population reduction (Mukerji *et al.* 1981; Ewen & Mukerji 1987; McGuire *et al.* 1991). Courshee (1983) suggested that bait formulations are by far the most efficient means of using insecticides, approaching 50 % of the theoretical maximum use of the active ingredient against the target pest. So far as ABC is concerned, Wohlleber (1996; 2000) suggested that by using a bait formulation to control ABC in Namibia, it is possible to reduce the amount of pesticide per unit area by up to 75 % compared to a ground application with a sprayer. Baits are an attractive option when environmental contamination and insecticide hazard, in addition to ABC control efficacy, are important considerations (Price & Brown 1997). This was found true for small-scale locust control programmes in the U.S.A. when locust populations were small (Courshee 1983; Capinera & Hibbard 1987).

Following the discovery of organochlorines in the late 1940's, the use of baits declined apart from isolated uses such as controlling rangeland grasshoppers in the Western USA (Onsager *et al.* 1980). Organochlorines were easy to apply and sprays could uniformly cover a large area of pest infestation due to drift. Comparatively, the use of baits on large-scale pest infestations is deemed impracticable due to logistical and other inherent problems associated with the use of the baits (McGuire & Shasha 1992). However, due to the increasing costs of modern insecticides and their risk to the environment, bait formulations are now being re-examined as a pest control measure.

Because ABC predictably move into crop fields from the surrounding scrub at the time of panicle emergence/heading, deployment of baits at field edges just before the field invasion would appear to be a feasible ABC control method. Wohlleber (2000) reported promising results against ABC in Namibia using baits formulated with deltamethrin and beta-cyfluthrin, although no control treatment data was presented for comparison. Given that insecticide availability to farmers varies considerably from area to area throughout the region of southern Africa, more insecticides need to be evaluated for bait formulation against ABC so that farmers can make the best choice from the range of insecticides that is available to them and use them at the correct dosages.

The aims of this study therefore, were to evaluate locally available material as bait carriers and to test efficacy of different insecticides formulated as baits for the ABC.

MATERIALS AND METHODS

Test insects

ABC later instar nymphs and young adults were collected from the field and kept in a screen cage measuring 8 m long x 3 m wide x 2.5 m in height. The natural grass in the cage was regularly supplemented with fresh sorghum panicles. Water was provided in moistened cotton wool contained in shallow petri dishes. Insects were starved for 3 hours before the experiment but were provided with water to drink. Experiments were conducted under laboratory conditions at 25⁰ C and 50 % RH with a 12:12 LD light cycle to mimic field conditions.

Evaluation of possible bait carriers

Maize bran, sorghum bran and millet bran, crushed ABC and maize meal were evaluated for feeding preference by adult ABC. The carriers were mixed with tap water using a wooden paddle. Different utensils were used for each carrier. The food and the container were weighed before being placed in a honey jar covered with cheesecloth held in place with rubber bands. One cricket was placed in each jar. Ten insects were used for each food type exposure. Food was presented to the ABC in small bottle tops of 2.5 cm diameter and 1.5 cm depth (Fig. 2.1). A dried branch of *Acacia tortilis* was placed in each jar for perching. Most crickets fed on the bait as soon as they were released in the jars. Some wandered around inside the jars before settling to feed. Controls were prepared similarly but no insects were introduced in the control jar. The insects were allowed to feed for 24 hours and the remaining food was re-weighed. The whole preparation was replicated four times. Reduction in weight loss due to moisture loss from the food was corrected for by subtracting the moisture loss value for the control experiment. Dead crickets were disregarded in data analysis.

Evaluation of insecticide baits

An experiment was conducted in the laboratory in which eight insecticides, representing six different chemical groups, were tested to determine their oral toxicity to adult ABC. The insecticide concentrations, trade names, chemical groups and

formulations used are listed in Tables 2.0 & 2.1. The percentage active ingredient of baits was calculated on a dry weight basis of the bran. Several insecticides were tested in order to provide farmers with a wider choice of insecticides to use in bait preparation.

The insecticides were tested at four or more concentrations, based around existing LD₅₀ information (BCPC 1991). At each concentration, for each insecticide, three replicates of 10 adult crickets were exposed to bran bait carrier containing different insecticides. Insects were weighed and only insects of both sexes weighing 3.5 ± 0.5 g were used in the experiment. The bait mixtures were prepared by thoroughly mixing tap water volume/mass with dry maize bran. The bait carrier was selected on the basis of bioassays conducted in the laboratory. The insecticide solution and bran were mixed together thoroughly just before each trial and the freshly prepared moistened bait was used immediately. The bait moisture content was as described by Price & Brown (1997). Moist crumbly bait from which a drop or two of water could still be squeezed when compressed between the fingers. The bioassays were carried out in wooden rearing cages measuring 45 cm long x 30 cm wide x 30 cm high covered with a sliding glass top. Bait was presented inside bottle tops of 2.5 cm diameter and 1.5 cm depth, so as to reduce the chance of body contact of the insects with the bait and thereby help ensure that mortality observed was due to ingestion of the bait. Each cricket was allowed to feed for 10 minutes and then removed and placed in clean cages and provided with untreated food. The 10 minutes feeding duration was earlier determined in the laboratory. Most crickets fed continuously before they stopped and wandered away. Crickets were inspected to ensure that there was no bait adhering to the insect body. Crickets were disqualified from the experiment if they did not feed at all during 30 minutes of exposure to the food. ABC were monitored subsequently and mortality recorded at 24 hours post treatment to compare the relative oral toxicities of the insecticides used. When scoring mortality, each insect was examined individually. The criterion for death was failure of the crickets to move any part of its body when probed with a soft paintbrush. Experiments were conducted under laboratory conditions at 25⁰ C and 50 % RH with a 12:12 LD light cycle to mimic field conditions.

Secondary cannibalistic poisoning effects

Opportunistic cannibalism by ABC is likely to be a commonplace in the field where bait is used, so secondary cannibalistic poisoning effects in an important consideration. Carbaryl and cypermethrin were also evaluated for residual toxicity on ABC. In this study, primary toxicity was defined as ABC mortality resulting from feeding directly on the bait. To obtain primary toxicity, 10 crickets were fed on bran bait formulated using LD₉₀ values of either carbaryl or cypermethrin. Crickets were allowed to feed for 10 minutes to determine primary toxicity. Secondary toxicity resulted from ABC that fed for 10 minutes on cricket cadavers resulting from primary toxicity. Crickets that died at 24 hours post treatment at each level of poisoning were fed to live crickets. Crickets were allowed to feed for 10 minutes on dead crickets before they were removed and put in cages and fed with bran. Each experiment was replicated three times and mortality in this case was recorded at 72 hours post treatment.

Mortality due to insecticide treatment was determined by correcting for natural mortality using Abbot's (1925) formula. The dose-response curves were calculated using probit analysis (dose transformed into log₁₀ and mortality into probits). The analysis yielded LD₅₀ and LD₉₀ values plus the slopes of the dosage-mortality regression lines.

RESULTS

Evaluation of possible bait carriers

Crickets consumed similar amounts of the different food types. There was no difference in feeding rates of the ABC on different cereal bran as well as crushed crickets and maize meal (Fig. 2.2: F value = 3.89, df = 4, $p > 0.693$). Crickets were observed to locate the different food types without difficulty despite different colours and tastes of the food materials. There was no sign of ABC reluctance to feed on any of the bait carriers. Once the crickets have started feeding, they would continue until satiated.

Evaluation of insecticide baits

Mortality data from bait bioassays with the eight insecticides against ABC are presented in Table 2.2. The order of decreasing toxicity of these eight insecticides was

fipronil > cyfluthrin > imidacloprid > cypermethrin > chlorpyrifos > carbaryl > gamma-BHC > malathion. Results showed that fipronil had the highest oral toxicity, with an LD_{50} value of $0.05 \mu\text{g } \mu\text{g}^{-1}$ which was significantly lower than that of the other products tested. The LD_{50} of cyfluthrin was not significantly different from that of imidacloprid, cypermethrin and chlorpyrifos. Similarly, toxicity of carbaryl was not significantly different from imidacloprid, cypermethrin and chlorpyrifos. Lastly, there was no significant difference in toxicities of gamma-BHC and malathion, the two insecticides that were least toxic.

The slopes of fipronil, cyfluthrin, imidacloprid, cypermethrin and chlorpyrifos were not significantly different from each other. Likewise, the slopes of carbaryl and malathion were not significantly different from each other. Insecticides with similar slopes show that products have got similar rate of toxic effects on the ABC. Gamma-BHC had a steeper slope which was significantly different from all other compounds (Table 2.2). This means that an additional unit dose of gamma-BHC gives a significant unit increase in mortality rate of the ABC.

Secondary cannibalistic poisoning effects

The primary mortality from both carbaryl and cypermethrin were 100 % at 24 hours post treatment. When crickets were released to feed on cadavers, they fed either on the neck or legs. Feeding started immediately after release and most crickets fed through the entire 10 minutes of observation. Carbaryl resulted in 65 % mortality while cypermethrin resulted in 10 % mortality in secondary toxicity at 72 hours post feeding. In tertiary toxicity, both carbaryl and cypermethrin gave no mortality of ABC 72 hours post feeding (Fig. 2.3).

DISCUSSION

Lack of significant differences between feeding rates on different bait carriers means that farmers can use any of the tested products as carriers to prepare insecticide baits against the ABC. Wohlleber (2000), reported on other bait carriers evaluated against ABC. Dried crickets, a mixture of millet, whole meal and fishmeal, and millet, whole meal and maize meal were most preferred compared to dehulled millet. The present study

complements these earlier findings in that cereal bran (a readily available material) were evaluated in comparison with maize meal. Cereal bran is locally available in most farming households. Different cereal bran are sold from local millers both in rural and urban areas in Botswana for US \$1.50 per 50 kg bag and is affordable for most small scale farmers.

The present experiments were designed to test a range of insecticides for their oral toxicity to ABC and to suggest dosages that are practical for use in the field. In the present studies, mortality after 24 hours was selected as an appropriate indicator for bait effectiveness. Considering that the bait will in many cases be applied inside a trench from which the ABC actively seek to escape (see Chapter 4), a fast acting insecticide would be preferable. As a result, those farmers who use the baited-trenches will see the dead ABC inside the trenches and be encouraged, rather than tempted to repeat baiting, thinking that the first application was ineffective. Farmers however, need to take necessary precautions to protect themselves from pesticides injuries.

This study has provided a guide to the potential performance of eight insecticides. The results of the experiments presented here suggest that several insecticides tested here are suitable toxicants for bait preparation against ABC. Efficacy studies in the laboratory cannot wholly substitute for the real situation in field trials, however. There are variable situations in the field such as temperature and humidity variation, photo degradation, rainfall, all of which may affect insecticide activity.

The practical use of any of these chemicals, however, depends on their relative safety and economic benefits. Fipronil SC (Regent®) is acutely toxic to ABC and other insects at very low concentrations ($LD_{50} = 0.05 \mu\text{g g}^{-1}$). Furthermore, it remains active, both by contact and ingestion, for relatively long periods with a half-life of 122 – 128 days in oxygenated sandy loam soil (Colliot *et al.* 1982). Hence, whilst it is an extremely effective ABC killing agent it also poses a sustained environmental hazard to other invertebrates. This aspect is further assessed in chapter 6. Another practical consideration is that working with such extremely low concentrations (see Table 2.0), as are required with fipronil 200 SC, may present farmers with calibration problems during bait preparation. Currently fipronil is not readily available to resource poor farmers in Botswana. Whilst pyrethroids are environmentally less persistent under field conditions,

they are expensive and cannot be afforded by small-scale farmers. Imidacloprid (Gaucho®) has moderately sustained activity and it is relatively benign environmentally (Pfuger & Schumuck 1991). It is not, however, readily available in the local markets in Botswana. Carbaryl (Karbaprays®) is widely used in insect control. It is stable, active at low doses, cheap and readily available to farmers in local markets in Botswana (Mosupi 1990; Mosupi 1992; Koosimile & Mosupi 1995). It appears to be the most viable of the candidate insecticides for use in an ABC bra bait mix. Chlorpyrifos is the most toxic of the two organophosphates tested with an LD₅₀ of 4.50 µg g⁻¹. It is widely used in agriculture and against household pests in southern Africa, but can have a serious impact on non-target organisms. Malathion is locally available and quite widely used in Botswana. However due to its pungent smell farmers are very reluctant to use it if another alternative chemical is available. Therefore other pesticides are likely to be more acceptable for use in an ABC bait mix. Gamma-BHC is widely available and cheap, so is used by many farmers and government agencies in southern Africa. The steep slope of gamma-BHC indicates that very small quantities are needed to increase mortality between dosage rates as compared to the other products tested. Organochlorines are persistent in the environment and accumulate in the food chain. However, gamma-BHC is still an appropriate insecticide for use in ABC bait preparation.

The high levels of secondary mortality resulting from carbaryl 85 WP (Karbaprays®) makes this insecticide additionally suitable for use in targeted baiting. At a dosage rate of 2.5 g a.i./kg of bait, it gave 65 % secondary mortality in crickets eating other bait-killed ABC. Therefore carbaryl is the most promising insecticide for future work on baits in Botswana. Targeted application of carbaryl-bran bait is evaluated in Chapter 4, using a baited-trench method.

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Table 2.0. Active ingredients, trade names and concentrations of pesticides used to test for oral toxicity against the ABC

Active ingredient	Trade name	Dose ($\mu\text{g/g}$) of carrier
Fipronil	Reagent [®]	0.125, 0.4, 0.9, 1.25, 4
Cyfluthrin	Baythroid [®]	1, 3, 6, 10, 25
Imidacloprid	Gaucho [®]	0.3, 0.9, 10, 50, 100
Cypermethrin	Avi sipermethrin [®]	4,9,12.5,40,90
Chlorpyrifos	Dursban [®]	4, 5, 20, 40, 50
Carbaryl	Karbaspray [®]	10, 60, 100, 600, 800
Gamma-BHC	Gamma Benhex [®]	30, 60, 110, 230, 560
Mercaptothion	malathion [®]	5, 10, 21, 80, 210, 420

Table 2.1. Chemical groups, formulations, chemical names, amount of active ingredient and toxicities of the insecticides tested as baits against the ABC.

Common name	Formulation	Chemical name	Chemical group	LD ₅₀ (rat) Oral mg/kg
Malathion	500 g/l EC	Diethyl (dimethoxy thioxo phosphoryl-thio) succinate	Organophosphate	2 800
Chlorpyrifos	480 g/l SC	0,0-diethyl 0-(3,5,6-trichloro-2-pyridyl) phosphorothioate	Organophosphate	135
Cypermethrin	200 g/l EC	(RS)-Cyano(3-phenoxyphenyl)methyl(IRS)-cis-trans-3-(2,2-dichloroethenyl)-2,2-dimethyl Cyclopropane carboxylate	Pyrethroid	251-4123
Cyfluthrin	100g/kg WP	Cyano(4-fluor-3-phenoxyphenyl) methyl-3-(2,2,-dichloroethenyl)-2-2-dimethyl-cyclo-propane carboxylate	Pyrethroid	900
Gamma-BHC	200 g/kg DS	1,2,3,4,5,6-Hexachlorocyclohexane	Organochlorine	88-270
Carbaryl	850 g/kg WP	1-Naphthyl methyl carbamate	Carbamate	500
Fipronil	200 g/l SC	(±)-5-Amino-1-(2,6-dichloro-4-trifluoro-methyl phenyl)-4-trifluoromethyl-sulfinylpyrazole-3-carbonitrile	Pyrazole	100
Imidacloprid	700 g/kg WS	1-(6-chloro-3-pyridylemethyl)-N-nitroimidazoladin-2-ylidenamine	Chloronicotynils	450

Table 2.2. Summary of the dose-responses with different insecticides against adult armoured bush crickets.

Insecticide	LD ₅₀ (95% fiducial limits) (µg/g)	LD ₉₀ (µg/g)	Regression Equation	Slope (SE)
Fipronil	0.05 (0.009-0.1)a	0.23	$Y = 7.33 + 2.00x$	2.00 (0.30)b
Cyfluthrin	0.52 (0.25-0.92)b	2.60	$Y = 5.50 + 1.83x$	1.83 (0.24)b
Imidacloprid	1.24 (0.54-2.83)bc	7.60	$Y = 4.85 + 1.63x$	1.63 (0.28)bcd
Cypermethrin	2.30 (0.87-3.40)bc	12.6	$Y = 4.37 + 1.74x$	1.74 (0.32)bc
Chlorpyrifos	4.50 (0.58-16.3)bc	23.0	$Y = 3.80 + 1.80x$	1.80 (0.36)bc
Carbaryl	5.0 (2.6-8.9)c	56.0	$Y = 4.12 + 1.25x$	1.25 (0.16)d
Gamma-BHC	20.0 (17.0-25.0)d	60.0	$Y = 1.40 + 2.75x$	2.75 (0.35)a
Malathion	21.1 (19-24)d	197	$Y = 3.20 + 1.34x$	1.30 (0.17)cd

The LD₉₀ values of the tested insecticides are recommended for field-testing against the ABC. Means within columns followed by same letter do not differ significantly ($p < 0.05$).

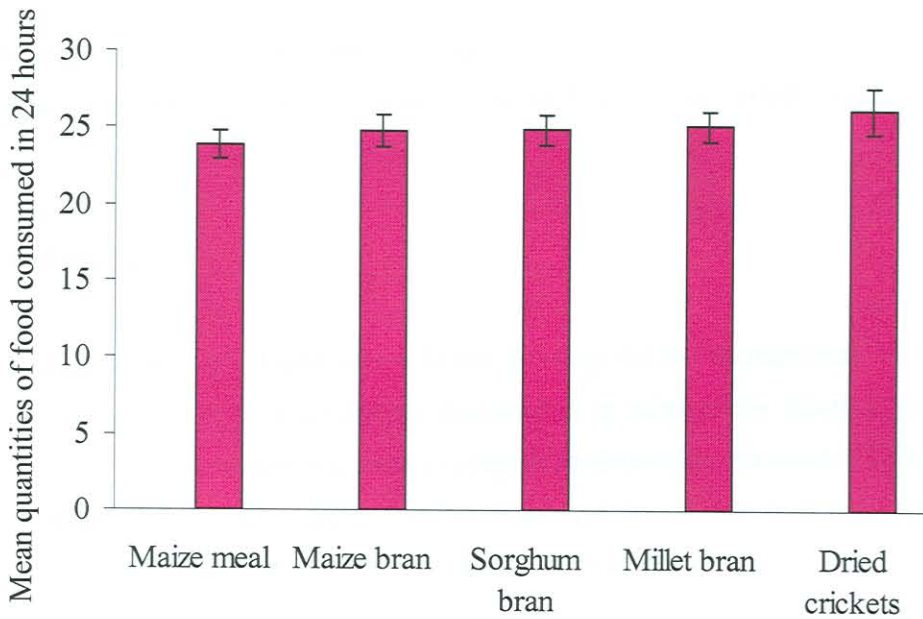


Fig. 2.2. Average amounts (g) of different food types consumed by armoured bush crickets in a period of 24 hours.

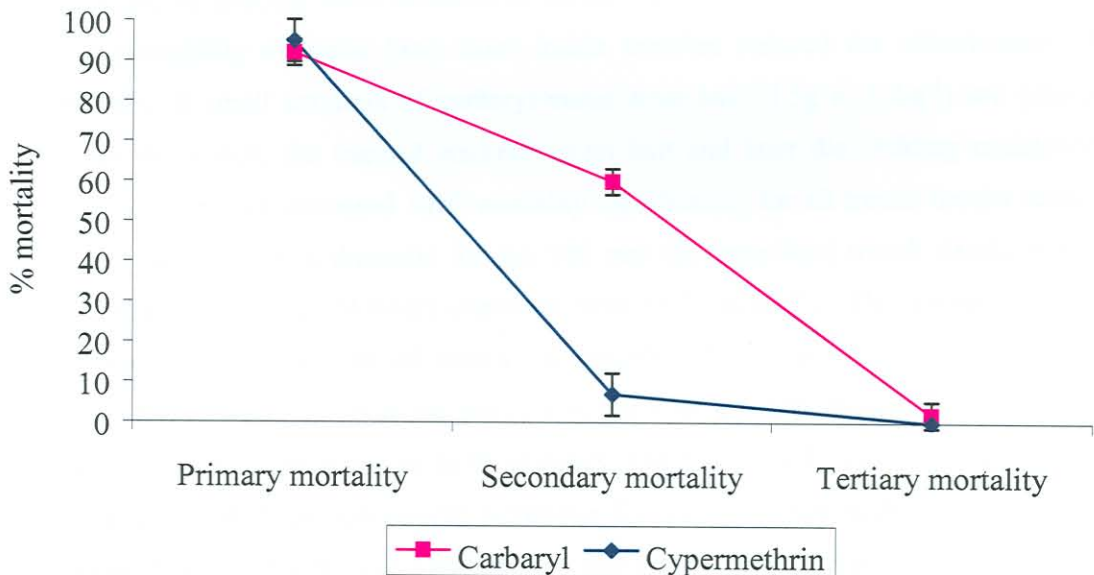


Fig. 2.3. Efficacy of carbaryl (3 g a.i./kg) and cypermethrin (0.09 ga.i./kg) bran baits on ABC; secondary and tertiary mortality due to feeding of ABC on crickets that died of primary poisoning. Bars indicate standard error (SE).