AN ABSTRACT OF THE THESIS OF

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The adult western spotted cucumber beetle (WSCB), *Diabrotica undecimpunctata undecimpunctata* Mannerheim is considered a major pest of snap beans grown for processing in Western Oregon. Control consists of 1 or 2 insecticide sprays prior to harvest. A commercial semiochemical-based insecticide bait, Adios TIC®, 1.3% carbaryl + 5% buffalo gourd root powder + 0.7% TIC mixture (1,2,4-trimethoxybenzene, indole, and *trans*-cinnamaldehyde, 1:1:1), was evaluated for the control of WSCB in commercial snap beans. Adios TIC® did not result in improved WSCB control as reflected by number of beetles and pod damage.

Twenty one plant volatile compounds were evaluated for their attractivity to WSCB in snap bean and squash fields by comparing the number of beetles caught on sticky traps in 1 to 3 day periods. The chemicals beta-ionone, benzyl alcohol, and indole consistently attracted significantly more beetles than unbaited traps. An equal part of these chemicals caught more WSCB than those baited with any of the two component

mixture blends or even individual components. Captures of WSCB increased significantly as doses of the IBb mixture in trap increased. Baited traps placed inside and at the edge of alfalfa fields caught similar numbers of WSCB. More WSCB were caught on traps inside than outside fields. Traps baited with the IBb mixture placed at canopy level or at 10 - 15 cm above canopy caught equal numbers of WSCB. Traps below plant canopy caught fewer WSCB. Most WSCB were caught from 10:00 to 13:00 hrs and from 16:00 to 18:00 hrs. Fresh preparations of IBb were the most attractive to the beetle. The majority of WSCB attracted by the IBb mixture were males (≈ 80%). Its individual components, indole and benzyl alcohol attracted mostly females (≈ 73% and 70% respectively), beta-ionone attracted mostly males (≈ 80%).

Attractivity of Plant Volatiles and a Semiochemical-Based Bait to the Western Spotted Cucumber Beetle, *Diabrotica undecimpunctata undecimpunctata* Mannerheim (Coleoptera: Chrysomelidae)

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Tawatchai Hongtrakul

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1. Introduction

The western spotted cucumber beetle (WSCB), *Diabrotica undecimpunctata undecimpunctata* Mannerheim, is a common pest of the coastal, Western United States. The larvae cause economic damage to potatoes, corn, table beets, and peppermint as well as other crops in Western Oregon. Adult beetles feed on numerous wild and cultivated host plants including ornamentals and bush, snap beans. The adult WSCB is a major pest of snap beans grown for processing in the Willamette Valley, Oregon. It feeds on foliage, flowers and developing bean pods. Pod blemishes caused by beetle bites reduce market quality and result in substantial dockage in prices paid by processors to growers. Because infestations of WSCB in beans are erratic and variable by year or time of season, most fields receive at least one "insurance" insecticide spray near harvest.

Semiochemicals are defined as substances or mixtures of substances emitted by plants or animals that modify the behavior of receptor organisms of like or different species (USEPA, 1982). Many studies document that cucurbitacins arrest or elicit a feeding stimulation in certain species of *Diabrotica* beetles (corn rootworms). Cucurbitacins are bitter substances characteristic of the Cucurbitaceae. They, however, are not volatile. Thus, they are not effective as attractants. However, recent research into the chemical ecology of *Diabrotica* spp. has shown that the adults are attracted to numerous plant volatiles. A practical application of this knowledge is the formulation of these volatiles with cucurbitacins into insecticidal baits for the control of pest species. This is the "attract and kill" concept. Theoretically, effective control could be realized by incorporating very small amounts of an insecticide into a bait. The bait would attract and kill food-seeking beetles prior to crop injury.

Many attractants and arrestants as well as semiochemical-based insecticides have been evaluated for *Diabrotica* spp. in the Midwest. There is a dearth of knowledge in this area for the congeneric WSCB. The purpose of this research is two fold. The first is to evaluate the effectiveness of a commercial semiochemical-based insecticide bait for the control of WSCB. The second is to determine if commercially available plant volatile compounds are attractive to WSCB.

1.1 Literature Review

The western spotted cucumber beetle, *Diabrotica undecimpunctata undecimpunctata* Mannerheim, is a common pest in the Willamette Valley of Western Oregon (Eltoma, 1979; Rockwood and Chamberlin, 1943). Adult beetles feed on numerous wild and cultivated host plants including snap beans. The adult WSCB is considered a major pest of snap beans because it feeds on and injures foliage, flowers and developing bean pods. Beetle bites are unacceptable blemishes on processed snap beans. Pod damage affects the quality of frozen and canned beans. Dockage to the grower and even rejection of beans delivered to the processor is common when beetles are numerous (Weinzierl et al., 1986).

Management of WSCB in snap beans is based on preventive application of insecticide to limit potential damage. Numerous insecticides are registered to control this pest. However, preharvest intervals or restrictions on the use of field and cannery waste as animal feeds result in most applications being carbaryl or diazinon.

Species of the Chrysomelid genus, *Diabrotica*, display close associations with host plants of the Cucurbitacae, particularly *Cucurbita* (Metcalf, 1979). The cucurbitacins (Tetracyclic triterpines) contribute to the bitterness characteristic of the Cucurbitacae and also serve as potent feeding stimulants or arrestants for these beetles (Metcalf et al., 1980, 1982). Cucurbitacins are not volatile, and such, they are not considered effective as attractants.

Recent research on the chemical ecology of rootworms (*D. undecimpunctata howardi* Barber, the southern corn rootworm, SCR; *D. virgifera virgifera* Leconte, the western corn rootworm, WCR; and *D. barberi* Smith and Lawrence, the northern corn rootworm, NCR) has shown that these species orient and move towards numerous plant volatiles associated with certain cucurbits. Indole, isolated from squash blossoms, *Cucurbita maxima* Duchesne, was evaluated by Andersen and Metcalf in 1986. Their field tests showed that indole is a potent attractant for WCR and striped cucumber beetle (*Acalymma* spp). Subsequent literature reports that eugenol attracts NCR (Ladd et al., 1983). Cinnamaldehyde and 1,2,4-trimethoxybenzene were attractants for WCR, and cinnamyl alcohol was found to be an effective attractant for NCR (Lampman and Metcalf, 1987; Metcalf and Lampman, 1989). SCR is apparently attracted to many compounds, including indole, veratrole and phenylacetaldehyde (Lampman et al., 1987).

Various mixtures of these compounds show degrees of olfactory synergism. A mixture of 1,2,4-trimethoxybenzene, indole and *trans*-cinnamaldehyde (TIC) was found to be attractive to a variety of corn rootworms. Apparently these chemicals display an olfactory synergism when blended (Lampman and Metcalf, 1987, 1988; Lampman et al., 1991). Field experiments in cornfields with the TIC mixture as well as its individual components documented their attractivity to *Diabrotica* adults. The blend of TIC displays more than an additive response for attracting WCR, NCR and SCR (Lampman et al., 1991). The TIC mixture was also highly attractive to the striped cucumber beetle, *Acalymma vittatum* (Lewis et al., 1990). The TIC mixture is simply a blend of three cucurbit blossom volatiles that are components in the floral odor of *C. maxima* (Andersen and Metcalf, 1987; Metcalf and Metcalf, 1992).

From a pest management perspective, a bait formulation of volatile attractants, a cucurbitacins arrestant and an insecticide might offer economic control of pests while reducing insecticide load in the environment. By luring beetles to toxicants and inducing them to feed, semiochemicals can substantially increase the efficiency of insecticidal baits (Metcalf et al., 1987; Lance, 1988). Field-cage and small-plot studies with semiochemical-based baits containing carbaryl or methomyl have reduced Midwest adult corn rootworm populations by more than 90%. This results even when toxicants are applied at rates twenty times less than the current label rates. Semiochemical bait technology has the potential of being an effective management tool while effectively reducing the annual amount of soil applied insecticide (Lance and Sutter, 1990).

1.2 Study Objectives

The first objective was to determine if a specific semiochemical-based bait applied to bush snap beans would control WSCB better than a commercial insecticide program. The second objective was to identify and evaluate known volatiles of the cucurbits for atttractivity to the western subspecies, *Diabrotica undecimpunctata undecimpunctata* Mannerheim. Strong attractants to this subspecies could be used in a monitoring or management program in crops where WSCB is a pest.

2. Materials and Methods

2.1 Efficacy of a semiochemical-based insecticide bait for control of WSCB in snap beans

Field experiments were designed to evaluate the efficacy of a semiochemical-based insecticide bait for the control of WSCB in a commercial variety of bush snap beans, OR-91. The commercial bait, Adios TIC®, 1.3% carbaryl + 5% buffalo gourd root powder + 0.7% TIC mixture (1,2,4-trimethoxybenzene, indole, and *trans*-cinnamaldehyde, 1:1:1) was provided by BASF. The carrier of this bait is a corncob granule. The bait was applied either once or twice at two rates. Control was compared to a carbaryl spray at a typical 1.0 lb ai per acre rate.

Tests were conducted at the OSU Vegetable Research Farm in Linn County, Oregon, in 1995. OR-91 snap beans were seeded in rows with a spacing of 30 inches. Four replications were used in a randomized complete block design. Each block varied by both planting date and location on the 160-acre farm to account for seasonal variation of beetle activity and influence of adjacent hosts. Block I consisted of 24 rows each measuring 240 ft long. There were 16 rows, measuring 480 ft long in Blocks II and III. Block IV consisted of 20 rows, measuring 240 ft. Blocks were divided into subplots, consisting of 8 rows by 50 ft in Blocks I and IV, and 8 rows by 65 ft in Blocks II and III, respectively. Average plant density was 6 plants/row foot. Standard irrigation, fertilization, and weed control practices were used.

Treatments consisted of

Adios TIC 0.065 lb ai carbaryl/acre, (5 lbs formulated bait)

Adios TIC 0.125 lb ai carbaryl/acre, (10 lbs formulated bait)

Adios TIC 0.065 lb ai carbaryl/acre, (5 lbs formulated bait), 2 applications

Adios TIC 0.125 lb ai carbaryl/acre, (10 lbs formulated bait), 2 applications

Standard insecticide (Sevin XLR Plus) 1.0 lb ai carbaryl/acre

Untreated Check

Adios treatments were applied using shaker jars to deliver the granules in a uniform band onto the bean plant canopy. A CO_2 pressurized backpack sprayer operating at 45 psi using a 3-nozzle boom, with 8004 nozzles, was used to deliver the reference chemical in an equivalent of 25 gallons of water per acre. Plots were treated during late bloom. Those treatments consisting of two applications received the second five to six days later. Relative adult WSCB populations in the plots were estimated 1 - 3 days before the application of the treatments. The efficacy of the treatments was evaluated by different methods at 1,3 and 7 days after treatments (DAT). Live and dead WSCB were recorded within the interior of each plot by counting beetles from 10 randomly chosen spots in the rows, each measuring 1 meter. At harvest, damage to the bean pods was evaluated using a sample of 500 pods/plot. Any significant differences among treatments were evaluated by analysis of variance. Individual means were separated by Fisher's protected LSD.

2.2 Attractivity of plant volatile compounds for WSCB

Field experiments on attraction of WSCB to various plant volatile compounds were conducted in commercial snap bean, squash and alfalfa fields. Attraction of WSCB to plant volatile compounds was measured by the average number of beetles caught per day on a 10-oz transparent cups (Solo cup), modified from Lampman and Metcalf (1987) which used 1.0 liter cylindrical paper cartons, evenly coated with clear insect adhesive, Tangle Trap® (Tanglefoot Co.). The candidate attractants were applied by capillary micropipettes to cotton wicks (10.0 mm long by 8 mm diameter), modified from Lampman and Metcalf (1987) using cotton dental wicks size 13 mm long by 6 mm diameter, at a dosage of 100 mg per trap. The wicks were previously soaked in mineral oil to prolong volatilization; excess oil was squeezed out prior to treatment of each compound (Lampman and Metcalf, 1987). The cotton wicks, treated and untreated, were attached to the inside of the cups with Tangle Trap®. The cups were inverted and placed on stakes just above the plant height in experimental fields. Tests were conducted in a randomized complete block design with at least 4 replications at numerous times and locations over two seasons. The sticky traps were placed in a line 6 - 10 m apart to minimize possible overlapping of volatile flumes (personal communication, M. Kogan and J. Gaggero). Beetle counts were taken and WSCB as well as other insects were removed from traps everyday during 1 to 3 day-periods, modified from Lampman and Metcalf (1987) in which beetle counts were made at 24 h only. The trap catches were transformed to SQRT (X + 0.1) to satisfy the assumption of equal variance prior to statistical analysis. The significance of attraction was determined by analysis of variance and the individual means were separated by Fisher's protected LSD. The series of steps and experiments outlined below were completed.

2.2.1 Screening of individual plant volatiles

Numerous volatiles of the flowers of the Cucurbitaceae were evaluated for relative attractivity to WSCB. These compounds were indole; *trans*-cinnamaldehyde;

cinnamyl alcohol; 1,2,4-trimethoxybenzene; benzyl alcohol; eugenol; estragole; veratrole; and phenylacetaldehyde. All have been reported to be attractants to at least one *Diabrotica* species (Andersen and Metcalf, 1986; Lampman and Metcalf, 1987; Lampman et al., 1987; Metcalf and Lampman, 1989). Others included 4-methoxybenzyl alcohol; 1,4-dimethoxybenzene; 1,2,3-trimethoxybenzene; beta-ionone; *p*-anisaldehyde; nerolidol; hexyl alcohol; *trans*-3-hexen-1-ol; *cis*-2-hexen-1-ol; linalool; mycene; and limonene. These are chemicals displaying structural similarities to known attractants and deemed worthy for evaluation (Andersen, 1987). Candidate volatiles were applied to cotton wicks, and evaluated as in methods described above. Tests were conducted in snap bean planting at the OSU Vegetable Research Farm and in commercial squash fields at Stahlbush Island Farms, Linn County, Oregon.

2.2.2 Attraction tests of blends of plant volatile compounds

Blends of promising volatiles were investigated for response by WSCB. Initial screening trials identified indole (I), benzyl alcohol (B), and beta-ionone (b) as promising attractants for WSCB. An equal part mixture of these compounds (IBb mixture) as well as its individual components were tested for the attractiveness. Field trials also included the TIC mixture (1,2,4-trimethoxybenzene, indole, and *trans*-cinnamaldehyde, 1:1:1), a highly attractive blend to a variety of *Diabrotica* beetles, (Lampman and Metcalf, 1987, 1988). Trapping techniques described above were used. Tests were conducted in snap bean and squash fields stated above.

2.2.3 Effect of trap color

The IBb mixture shown to be attractive in the previous section was further evaluated using different color traps to determine if color of trap enhanced daily WSCB catches. A 9-oz Solo cup color yellow, red, blue, 10-oz transparent cup, and 10-oz white paper cup were used. Attractants were applied to color traps as the methods described above. Daily mean numbers of WSCB caught on each trap, baited or unbaited were compared. Tests were conducted in a commercial squash field at Stahlbush Island Farms.

2.2.4 Trap location

Numerous investigators have shown differential trap catches of target insects according to field placement, wind direction or adjacent landscape. In this study the effect of trap location at varying distances from the edge of an alfalfa field on the capture of WSCB was investigated. Tests were conducted at the OSU Vegetable Research Farm on weed free fallow upwind of an alfalfa field. Both baited and unbaited traps were placed, in pairs 5 m apart, at distances of 10 m inside the field, at the field edge, as well as 10 m, 20 m, 30 m, 40 m and 50 m on fallow ground, from the edge of the study field. Trap catches at each distance were compared for significant differences.

2.2.5 Dose response of WSCB to attractants

Attractivity of WSCB to doses of the IBb mixture ranging from 0 - 100 mg/trap was evaluated. Each dose was applied and evaluated as described above. Regression analysis was used to determine the correlation of dose and response of WSCB to the attractant.

2.2.6 Trap height

Tests were conducted in squash to determine the optimum height of trap placement in relation to plant canopy for maximum attraction of WSCB to the IBb mixture. Traps were placed at three levels, I: under the leaf canopy and from 20 - 30 cm above ground, II: even with the canopy, and III: 10 - 15 cm above the canopy.

2.2.7 Hourly response of WSCB to attractants

This test was developed to determine if WSCB displayed a periodicity of attractivity, from sunrise to sun set, to the IBb mixture. WSCB caught on traps, baited and unbaited, were recorded and removed from traps every hour from 7:00 AM to 19:00 PM. Temperature in the field as recorded by a hand held thermometer ranged from 60° – 112° F. Day was mostly sunny with wind calm in the morning and light in the afternoon. Trials were conducted only on days conducive to beetle movement (minimal cloud cover, no rain or strong winds and temperatures > 60° F). Mean numbers of WSCB caught on traps from each period were compared. This information will be useful for the monitoring WSCB populations and interpreting trap catches. Tests were conducted in the commercial squash field at Stahlbush Island Farms.

2.2.8 Effect of field exposure on attraction

This trial was designed to determine how long the IBb mixture remains attractive to WSCB upon exposure to field conditions. Beetles captured on both baited and unbaited traps were observed, recorded and removed daily. Mean numbers of beetle captured each day were compared. Tests were conducted in the commercial squash field at Stahlbush Island Farms.

2.2.9 Sex ratio of WSCB attracted to plant volatile compounds

Previous researchers indicated that the sex ratio of the Diabrotica beetles varied with attractant types. Andersen (1986) reported that indole attracted more females WCR in August and early September, but captured more males in late July and late September. Mixtures of attractants the TIC and the VIPAE, a mixture of veratrole (V), indole (I), phenylacetaldehyde (P), *trans*-anethole (A), and eugenol (E), attracted primarily males SCR (Herbert et al., 1996; Lampman and Metcalf, 1987). This study determined sex ratio of WSCB caught on traps baited with the IBb mixture and its individual components. Sex determination of WSCB was begun with the examination of mating pairs for external morphological differences. Males have an extra sclerite at the abdominal apex that is lacking in females. This character was confirmed by dissection. Subsequent literature reviewed indicated that this sexual character was reported by Smith and Allen (1931) for males of *D. duodecimpunctata* Fab., *D. vittata* Lec., *D. balteata* Lec., *D. virgifera* Lec., and *D. longicornis* Say. Also, White (1977) reported that this same sexual character occurs in all 78 species of Central and South American members of *Diabrotica* in the United States National Museum collection.

3. Results

3.1 Efficacy of a semiochemical-based insecticide bait for control of WSCB in snap beans

There were no significant differences in number of live adult WSCB between blocks or treatments for the pre-treatment counts. Additionally, no significant differences in numbers of live adult WSCB were observed between treatments on 1,3 and 7 days after the applications. There were slightly fewer number of live beetles in the plots treated with carbaryl, Sevin XLR Plus (Table 1). Significantly greater numbers of dead WSCB were observed in the carbaryl treated plots at each post treatment observation date (Table 2). Adios TIC® 0.125 lb ai, rate with 2 applications was observed to have significantly more dead beetles than the untreated check at 1 and 7 days following the first application. None of the treatments provided acceptable reduction in the percentage of pods damaged by WSCB feeding activity. In fact, damaged pods were significantly greater in all but one of the Adios TIC® treatments compared to the carbaryl standard (Table 3).

Table 1. Mean numbers of live WSCB ± standard error[a] observed per 1 m of bean row.

Treatment (lb ai carbaryl/acre)	Pre-treatment	1 DAT	3 DAT	7 DAT[c]
Adios TIC 0.065	1.06 ± 0.15	1.21 ± 0.10	1.40 ± 0.23	1.55 ± 0.15 b
Adios TIC 0.065[b]	1.15 ± 0.15	1.17 ± 0.19	1.60 ± 0.35	1.39 ± 0.07 b
Adios TIC 0.125	1.24 ± 0.14	1.17 ± 0.19	1.46 ± 0.30	1.46 ± 0.23 b
Adios TIC 0.125[b]	1.25 ± 0.10	1.40 ± 0.09	1.56 ± 0.24	1.57 ± 0.23 b
Unbaited Control	1.01 ± 0.09	1.20 ± 0.07	1.29 ± 0.18	1.46 ± 0.12 b
Carbaryl 1.0	1.08 ± 0.10	1.04 ± 0.24	1.28 ± 0.44	1.09 ± 0.12 a

[a] n = 4, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).
[b] Two applications.
[c] Replication 3, data taken 9 DAT instead of 7 DAT.

Table 2. Mean numbers of dead WSCB ± standard error[a] observed per 1 m of bean row.

Treatment (lb ai carbaryl/acre)	1 DAT	3 DAT	7 DAT[c]
Adios TIC 0.065	0.16 ± 0.03 bc	0.20 ± 0.07 b	0.08 ± 0.05 c
Adios TIC 0.065[b]	0.29 ± 0.04 bc	0.32 ± 0.04 b	0.24 ± 0.08 bc
Adios TIC 0.125	0.09 ± 0.04 c	0.27 ± 0.07 b	0.21 ± 0.13 c
Adios TIC 0.125[b]	0.49 ± 0.09 b	0.42 ± 0.11 b	0.52 ± 0.07 b
Untreated Control	0.02 ± 0.02 c	0.02 ± 0.02 b	0.03 ± 0.03 c
Carbaryl 1.0	1.56 ± 0.75 a	2.43 ± 1.02 a	1.53 ± 0.40 a

[a] n = 4, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).
[b] Two applications.
[c] Replication 3, data taken 9 DAT instead of 7 DAT.

Table 3. Mean percent pod damage ± standard error[a].

Treatment (lb ai carbaryl/acre)	% Pod Damage
Adios TIC 0.065	12.56 ± 2.63 bc
Adios TIC 0.065 [b]	12.94 ± 1.96 bc
Adios TIC 0.125	12.23 ± 3.03 ab
Adios TIC 0.125 [b]	16.27 ± 3.02 c
Untreated Control	10.75 ± 0.98 ab
Carbaryl 1.0	8.53 ± 1.24 a

[a] n = 4, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).
[b] Two applications.

3.2 Attractivity of plant volatile compounds for WSCB

3.2.1 Screening of individual plant volatiles

The mean numbers of WSCB captured on 10-oz transparent cup traps baited with candidate compounds in a field of OR-91 bush snap beans are presented in Table 4. There were significant differences in trap catches among individual volatile compounds tested. The chemicals, beta-ionone, benzyl alcohol, phenylacetaldehyde, and indole caught significantly more WSCB than the untreated control (clear cup, unbaited). Among these compounds, beta-ionone was the most attractive. The numbers of WSCB caught on traps baited with beta-ionone was significantly greater than those for the other three attractive volatiles and about four times greater than the unbaited clear cups. Trap catches from benzyl alcohol were significantly greater than those caught by indole. In this test, cinnamyl alcohol; *trans*-cinnamaldehyde; 1,2,4-trimethoxybenzene; veratrole; estragole; and eugenol, reported to be attractants for at least one species of *Diabrotica* beetles, were not attractive to WSCB. The numbers of the beetles caught on cups baited with these compounds were not significantly different from the control traps. Other volatiles tested were not attractive to WSCB.

The second tests were conducted in squash. The response of WSCB to sticky traps baited with tested compounds in squash is shown in Table 5. There were highly significant differences on the number of WSCB caught on sticky traps baited with the candidate compounds. Traps baited with benzyl alcohol, 4-methoxybenzyl alcohol, beta-ionone, *p*-anisaldehyde, indole, and estragole, caught significantly more WSCB than control traps. Among these treatments, benzyl alcohol, beta-ionone, and indole were consistently attractive to WSCB as in the previous tests conducted in snap bean. The highest mean numbers of WSCB caught were found on traps baited with benzyl alcohol. However, there was no significant difference among the numbers of WSCB caught in traps baited with benzyl alcohol, beta-ionone, and 4-methoxybenzyl alcohol. Traps baited with these three compounds captured significantly greater beetles than traps baited with indole, estragole, and *p*-anisaldehyde. The mean numbers of WSCB caught on traps baited with benzyl alcohol, 4-methoxybenzyl alcohol, b-ionone were 4.5, 3.5, 2.9, times higher, respectively, than those caught on control traps. Although in these tests, traps baited with *p*-anisaldehyde, and estragole caught significantly more WSCB than control traps, the numbers trapped were not significantly different from many of the less effective chemicals tested. In other words, estragole, and *p*-anisaldehyde were weakly attractive to the beetles. Phenylacetaldehyde was not consistently attractive to WSCB as in the previous tests in snap bean. The TIC mixture is reported to be the most attractive chemical blend for many other *Diabrotica* beetles, and to show olfactory synergism to all species investigated (Lampman and Metcalf, 1987, 1988; Lampman et al., 1991) but did not draw as many WSCB to traps in this experiment. Although the numbers of WSCB caught in traps baited with the TIC mixture was greater than control traps, there was no significant difference between them.

In subsequent screening trials, 9-oz yellow plastic cups were used as the standard sticky trap. It is interesting that the trap catches of candidate compounds were inconsistent over the period of testing. No significant differences were observed among the candidate attractants compared to the yellow control trap (Table 6). Since yellow is quite attractive to *Diabrotica* beetles, using yellow traps may have overcome slight differences in chemical olfactory attractivity.

Table 4. Attraction of WSCB to plant volatile compounds in snap beans, at the OSU Vegetable Research Farm, during 7/10 - 20/1996.

Treatment	Mean numbers of WSCB per sticky trap ± standard error[a]			
	1 DAT	2 DAT	3 DAT	Average
nerolidol	3.50 ± 1.04 ef	1.88 ± 0.83 e	1.00 ± 0.50 e	2.13 ± 0.52 f
1,4-dimethoxybenzene	2.13 ± 0.64 f	2.75 ± 1.24 de	1.63 ± 0.60 cde	2.17 ± 0.74 f
cinnamyl alcohol	3.38 ± 0.82 def	2.00 ± 0.71 de	1.88 ± 0.58 bcde	2.42 ± 0.44 ef
trans-cinnamaldehyde	3.50 ± 1.60 ef	2.38 ± 0.94 de	1.50 ± 0.50 cde	2.46 ± 0.70 ef
1,2,4-trimethoxybenzene	2.88 ± 1.03 ef	1.88 ± 0.69 de	2.75 ± 0.80 abcd	2.50 ± 0.62 ef
1,2,3-trimethoxybenzene	3.88 ± 0.99 def	2.38 ± 0.86 de	1.50 ± 0.60 de	2.58 ± 0.70 ef
cis-2-hexen-1-ol	4.75 ± 1.53 cdef	2.25 ± 0.65 cde	1.38 ± 0.71 de	2.79 ± 0.90 ef
clear cup	3.63 ± 1.07 def	2.50 ± 1.07 de	2.38 ± 1.31 cde	2.83 ± 1.02 ef
p-anisaldehyde	3.75 ± 1.42 def	2.75 ± 1.26 cde	2.00 ± 0.93 bcde	2.83 ± 1.11 ef
veratrole	3.50 ± 0.87 def	2.50 ± 0.91 cde	2.75 ± 1.15 abcde	2.92 ± 0.87 def
estragole	4.25 ± 1.19 def	3.63 ± 1.66 bcde	1.25 ± 0.45 de	3.04 ± 0.94 def
eugenol	3.75 ± 1.24 def	3.13 ± 1.57 de	2.63 ± 1.07 abcde	3.17 ± 1.19 def
hexyl alcohol	4.63 ± 0.91 cde	2.38 ± 0.75 cde	2.75 ± 0.67 abcd	3.25 ± 0.65 cdef
trans-3-hexen-1-ol	5.50 ± 1.84 cde	3.38 ± 1.29 cde	2.00 ± 1.07 bcde	3.63 ± 1.27 cdef
4-methoxybenzyl alcohol	5.13 ± 1.16 cde	5.13 ± 1.60 bc	1.88 ± 1.08 de	4.04 ± 0.91 cde
indole	6.75 ± 1.75 cd	3.75 ± 1.31 bcd	2.75 ± 0.84 abcd	4.42 ± 1.10 cd
phenylacetaldehyde	8.13 ± 1.94 bc	4.25 ± 1.76 bcde	3.88 ± 1.51 abc	5.42 ± 1.51 bc
benzyl alcohol	10.75 ± 3.37 ab	5.63 ± 1.66 b	4.38 ± 1.76 ab	6.92 ± 2.13 b
beta-ionone	14.63 ± 4.33 a	11.38 ± 4.24 a	5.25 ± 2.14 a	10.42 ± 2.92 a

[a] n = 8, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).

Table 5. Attraction of WSCB to plant volatile compounds in squash field, at Stahlbush Island Farms, during 7/22 - 28/1996.

Treatment	Mean numbers of WSCB per sticky trap ± standard error[a]			
	1 DAT	2 DAT	3 DAT	Average
cinnamyl alcohol	3.17 ± 0.79 fg	3.83 ± 1.40 f	2.17 ± 1.05 de	3.06 ± 0.73 f
clear cup	2.83 ± 1.30 g	4.67 ± 1.09 def	2.67 ± 1.09 de	3.39 ± 0.97 f
1,4-dimethoxybenzene	3.17 ± 1.08 g	5.17 ± 1.80 def	2.17 ± 0.79 de	3.50 ± 0.79 f
cis-2-hexen-1-ol	4.00 ± 1.21 efg	4.83 ± 1.01 def	1.67 ± 0.56 e	3.50 ± 0.80 f
trans-3-hexen-1-ol	4.00 ± 1.06 efg	4.17 ± 1.62 f	2.50 ± 0.85 cde	3.56 ± 1.09 f
nerolidol	2.33 ± 0.42 g	4.50 ± 0.96 def	4.00 ± 1.06 bcde	3.61 ± 0.54 ef
trans-cinnamaldehyde	3.00 ± 0.73 fg	5.83 ± 0.75 cdef	3.83 ± 1.47 cde	4.22 ± 0.62 def
TIC	5.00 ± 1.39 defg	4.50 ± 0.99 def	4.17 ± 2.82 cde	4.56 ± 1.51 def
phenylacetaldehydehyde	5.00 ± 1.59 defg	4.00 ± 0.86 ef	4.67 ± 2.49 cde	4.56 ± 1.51 def
veratrole	6.67 ± 1.61 bcdef	4.67 ± 1.36 def	2.33 ± 0.67 de	4.56 ± 0.89 def
1,2,3-trimethoxybenzene	5.17 ± 0.95 defg	3.50 ± 1.18 f	5.83 ± 0.70 abc	4.83 ± 0.54 def
eugenol	4.33 ± 0.84 defg	4.67 ± 1.54 def	6.33 ± 3.40 abcd	5.11 ± 1.70 def
1,2,4-trimethoxybenzene	3.33 ± 0.88 efg	6.17 ± 1.58 cdef	6.00 ± 2.31 abcd	5.17 ± 1.33 def
hexyl alcohol	6.33 ± 1.23 bcdef	6.00 ± 1.57 cdef	3.33 ± 0.84 cde	5.22 ± 0.85 def
estragole	6.00 ± 1.32 cdefg	8.83 ± 2.36 bcd	5.33 ± 1.56 abcd	6.72 ± 1.49 cde
indole	10.50 ± 2.62 abc	6.00 ± 1.93 cdef	4.00 ± 1.24 cde	6.83 ± 1.47 cde
p-anisaldehyde	7.00 ± 1.75 bcde	8.17 ± 2.36 cde	5.33 ± 1.05 abcd	6.83 ± 1.28 bcd
beta-ionone	10.67 ± 1.48 ab	10.00 ± 1.88 bc	9.50 ± 2.62 a	10.06 ± 1.48 abc
4-methoxybenzyl alcohol	9.17 ± 2.97 bcd	15.67 ± 5.23 ab	10.67 ± 5.02 ab	11.83 ± 3.82 ab
benzyl alcohol	16.50 ± 4.47 a	19.33 ± 4.80 a	10.17 ± 2.17 a	15.33 ± 3.57 a

[a] n = 6, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).

Table 6. Capture of WSCB on yellow sticky traps baited with various volatiles.

Treatment	Mean numbers of WSCB per sticky trap ± standard error[a]	
	Snap Bean[b]	Squash[c]
1,2,3-trimethoxybenzene	6.40 ± 0.98[d]	6.17 ± 0.57 bcde
1,2,4-trimethoxybenzene	4.00 ± 0.71	4.17 ± 0.91 e
1,4-dimethoxybenzene	4.60 ± 0.40	5.92 ± 0.96 bcde
4-methoxybenzyl alcohol	4.60 ± 0.93	8.17 ± 0.44 ab
p-anisaldehyde	5.20 ± 1.24	4.00 ± 0.43 e
benzyl alcohol	3.40 ± 1.17	9.33 ± 1.34 a
beta-ionone	3.80 ± 0.37	9.25 ± 0.75 a
trans-cinnamaldehyde	3.60 ± 0.51	5.50 ± 1.48 cde
cinnamyl alcohol	5.00 ± 1.10	4.08 ± 0.69 e
cis-2-hexen-1-ol	4.80 ± 1.07	5.33 ± 0.76 cde
estragole	4.00 ± 1.70	4.67 ± 0.49 cde
eugenol	3.60 ± 1.03	5.50 ± 0.78 bcde
hexyl alcohol	4.60 ± 0.93	5.00 ± 1.19 cde
indole	7.60 ± 1.40	6.92 ± 0.85 abc
limonene	3.20 ± 0.86	4.00 ± 1.05 e
linalool	5.20 ± 0.80	5.17 ± 0.75 cde
mycene	4.40 ± 0.75	6.00 ± 0.95 bcde
nerolidol	2.40 ± 0.60	5.25 ± 1.44 cde
phenylacetaldehyde	6.80 ± 0.80	5.00 ± 0.91 cde
TIC	6.00 ± 1.30	5.08 ± 0.42 cde
trans-3-hexen-1-ol	4.40 ± 0.93	4.50 ± 0.93 de
veratrole	3.20 ± 0.37	4.75 ± 1.24 cde
yellow unbaited	4.60 ± 0.24	6.75 ± 1.54 abcd

[a]Means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).
[b]Mean numbers of trap catches, n = 5, over 1 day period during 8/7 – 8/1996.
[c]Mean numbers of trap catches per day, n = 4, over 3 days period during 8/11 – 14/1996.
[d]No significant differences, $F = 1.58$, $df = 22, 88$, $p = 0.0709$.

3.2.2 Attraction tests of blends of plant volatile compounds

Indole (I), benzyl alcohol (B), and beta-ionone (b) were determined to be effective attractants for WSCB based on the previous trials. Therefore, a mixture of equal parts of these compounds (IBb mixture), as well as its individual components were tested for the attractiveness to WSCB. The tests conducted in snap beans show that only the IBb mixture caught significantly higher numbers of WSCB than the unbaited control, yellow cups (Table 7).

Table 7. Attraction of WSCB to mixtures of plant volatile compounds in snap bean, at the OSU Vegetable Research Farm, during 8/16 - 19/1996.

Treatment	Mean numbers of WSCB ± standard error [a]			
	1 DAT	2 DAT	3 DAT	Average
indole	4.25 ± 1.80 bc	1.75 ± 0.75	2.25 ± 1.11 b	2.75 ± 0.98 b
cocktail mixture [b]	2.75 ± 0.85 c	2.75 ± 0.48	3.00 ± 1.58 b	2.83 ± 0.63 b
benzyl alcohol	4.75 ± 0.48 bc	2.25 ± 0.63	2.25 ± 0.75 b	3.08 ± 0.16 b
TIC	4.25 ± 1.11 bc	2.75 ± 1.25	3.50 ± 0.87 ab	3.50 ± 0.80 b
yellow unbaited	5.75 ± 0.48 ab	3.50 ± 1.32	2.25 ± 0.48 b	3.83 ± 0.65 b
beta-ionone	4.75 ± 1.25 bc	4.00 ± 0.58	4.00 ± 0.58 ab	4.25 ± 0.60 ab
IBb mixture	8.75 ± 1.11 a	3.50 ± 0.96	7.25 ± 0.85 a	6.50 ± 0.40 a

[a] n = 4, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).
[b] mixture of all compounds tested in screening trials (excluded indole, benzyl alcohol, and b-ionone).

The tests were repeated in squash. The mean numbers of WSCB caught per trap were consistently and significantly greater on traps baited with IBb mixture (Table 8). The IBb mixture caught 3.2 times more WSCB than control traps. beta-ionone and indole also caught significantly more WSCB than the control. There were no significant differences among trap catches for the TIC mixture, the cocktail mixture of all compounds tested in screening trails (excluding indole, beta-ionone, and benzyl alcohol), benzylalcohol and the control.

Table 8. Attraction of WSCB to mixtures of plant volatile compounds in squash, at Stahlbush Island Farms, during 8/15 – 18/1996.

Treatment	Mean numbers of WSCB ± standard error [a]			
	1 DAT	2 DAT	3 DAT	Average
yellow unbaited	7.67 ± 1.54 d	1.00 ± 0.37	3.17 ± 0.65	3.94 ± 0.58 d
TIC	10.67 ± 1.41 bcd	0.50 ± 0.22	1.83 ± 0.70	4.33 ± 0.54 cd
benzyl alcohol	9.00 ± 1.46 cd	1.00 ± 0.26	3.50 ± 1.20	4.50 ± 0.84 cd
cocktail mixture [b]	12.17 ± 1.14 bc	1.67 ± 1.48	1.83 ± 0.48	5.22 ± 0.65 bcd
beta-ionone	12.00 ± 3.15 bcd	0.67 ± 0.49	4.00 ± 0.45	5.56 ± 1.19 bc
indole	15.17 ± 1.22 b	1.67 ± 0.42	3.00 ± 0.68	6.61 ± 0.69 b
IBb mixture	30.33 ± 2.51 a	3.83 ± 2.66	4.17 ± 1.01	12.78 ± 0.44 a

[a] n = 6, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).

[b] mixture of all compounds tested in screening trials (excluding indole, benzyl alcohol, and beta-ionone).

To test the interaction of the compounds of the IBb mixture, indole, benzyl alcohol and beta-ionone were mixed into 2-component mixtures as IB mixture (indole + benzyl alcohol), Ib mixture (indole + beta-ionone) and the Bb mixture (benzyl alcohol + beta-ionone). The treatments also included the IBb mixture, its individual components, and the TIC mixture. There were highly significant differences among the treatments. Traps baited with the IBb mixture caught the greatest number of WSCB (Table 9). The IB mixture and the Bb mixture were ranked second in attractiveness to WSCB based on trap catches. The number of WSCB caught on traps baited with IBb mixture, IB mixture, and Bb mixture was significantly greater than that from traps baited with the individual components. Interestingly, the overall response of WSCB to these mixtures was considered to be simply additive. Only the Ib mixture caught similar numbers of beetles to indole, which was one of its individual components. Traps baited with the TIC mixture did not catch significantly greater numbers of WSCB than the control traps.

Table 9. Attractiveness of the IBb mixture and two components blends to WSCB in squash, at Stahlbush Island Farms, during 8/23 - 26/1996.

Treatment	Mean numbers of WSCB ± standard error [a]			
	1 DAT	2 DAT	3 DAT	Average
yellow unbaited	17.00 ± 1.86 g	15.33 ± 3.37 d	5.33 ± 0.92 bc	12.56 ± 1.23 f
TIC	20.33 ± 2.64 fg	16.83 ± 4.16 cd	3.17 ± 0.65 c	13.44 ± 1.80 ef
beta-ionone	24.50 ± 4.60 ef	18.17 ± 3.75 cd	7.33 ± 1.50 ab	16.67 ± 2.61 de
benzyl alcohol	30.33 ± 2.95 de	21.33 ± 2.70 c	7.50 ± 1.20 ab	19.72 ± 1.59 d
indole	38.17 ± 4.38 cd	32.83 ± 5.55 b	8.33 ± 1.80 ab	26.44 ± 2.72 c
Ib mixture	42.17 ± 2.29 c	32.67 ± 5.75 b	8.83 ± 1.58 ab	27.89 ± 2.28 c
Bb mixture	68.33 ± 8.06 ab	36.83 ± 6.27 b	8.33 ± 1.28 ab	37.83 ± 2.65 b
IB mixture	59.00 ± 5.74 b	49.17 ± 10.72 a	9.17 ± 0.83 ab	39.11 ± 4.14 b
IBb mixture	78.83 ± 8.66 a	58.50 ± 10.60 a	11.83 ± 2.96 a	49.72 ± 5.53 a

[a] n = 6, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).

3.2.3 Effect of trap color

The IBb mixtures were tested using different colors of traps. There were highly significant differences among the treatments tested. All 5 color traps baited with the IBb mixture caught significantly greater numbers of WSCB than unbaited traps of the same color (Table 10). The ratio of trap catches between baited and unbaited traps of the same color were, yellow = 2.7 : 1, clear or transparent = 7.4 : 1, blue = 10.5 : 1, red = 16.1 : 1, and white = 8.9 : 1. Among IBb baited traps, yellow traps caught the highest number of the beetles, followed by clear, blue, red, and white traps. However, yellow traps gave the lowest ratio between baited and unbaited.

Table 10. Capture of WSCB on color traps baited with IBb mixture in squash field, at Stahlbush Island Farms, during 8/28 - 31/1996.

Trap Color	Mean numbers of WSCB ± standard error [a]			
	1 DAT	2 DAT	3 DAT	Average
Blue + IBb	48.40 ± 7.31 b	26.40 ± 4.39 bc	15.80 ± 1.80 ab	30.20 ± 2.79 b
Blue	4.20 ± 0.97 d	2.80 ± 1.20 ef	1.60 ± 0.60 e	2.87 ± 0.50 de
Clear + IBb	61.80 ± 11.50 ab	31.00 ± 2.83 ab	16.40 ± 2.25 ab	36.40 ± 4.10 b
Clear	6.20 ± 1.24 d	4.60 ± 1.21 e	4.00 ± 0.84 d	4.93 ± 0.75 d
Red + IBb	30.20 ± 5.25 c	21.00 ± 4.01 c	10.00 ± 1.00 c	20.40 ± 1.71 c
Red	2.00 ± 0.32 d	1.20 ± 0.37 f	0.60 ± 0.40 e	1.27 ± 0.24 e
White + IBb	32.80 ± 4.50 c	10.80 ± 1.20 d	4.60 ± 1.69 d	16.07 ± 1.92 c
White	4.00 ± 0.45 d	1.00 ± 0.45 f	0.40 ± 0.24 e	1.80 ± 0.17 e
Yellow + IBb	79.00 ± 12.14 a	41.80 ± 5.26 a	20.60 ± 1.44 a	47.13 ± 3.94 a
Yellow	29.60 ± 2.46 c	10.00 ± 2.74 d	13.20 ± 1.39 bc	17.60 ± 1.39 c

[a] n = 5, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).

3.2.4 Trap location

Traps baited with the IBb mixture placed at the edge and 10 m inside of the alfalfa field caught similar numbers of WSCB (Table 11). These catches were significantly greater than those caught on traps placed on the fallow ground at the distance of 10, 20, 30, 40, and 50 m from the edge of the field. Baited traps caught significantly more beetles than unbaited traps placed at the same distances.

Table 11. Effect of trap location on the capture of WSCB on traps baited with the IBb mixture in an alfalfa field, at the OSU Vegetable Research Farm, during 9/4 - 9/1996.

Distance in meters	Mean numbers of WSCB ± standard error [a]	
	Baited	Control
-10	54.75 ± 13.02 a	10.00 ± 2.58 a
0	50.25 ± 13.95 a	5.00 ± 2.12 b
10	12.75 ± 4.42 b	1.50 ± 0.29 bc
20	14.50 ± 6.38 b	1.50 ± 1.19 bc
30	11.25 ± 6.37 b	1.50 ± 1.19 bc
40	8.75 ± 4.42 b	0.50 ± 0.50 c
50	11.50 ± 3.57 b	1.25 ± 0.75 bc

[a] n = 4, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).

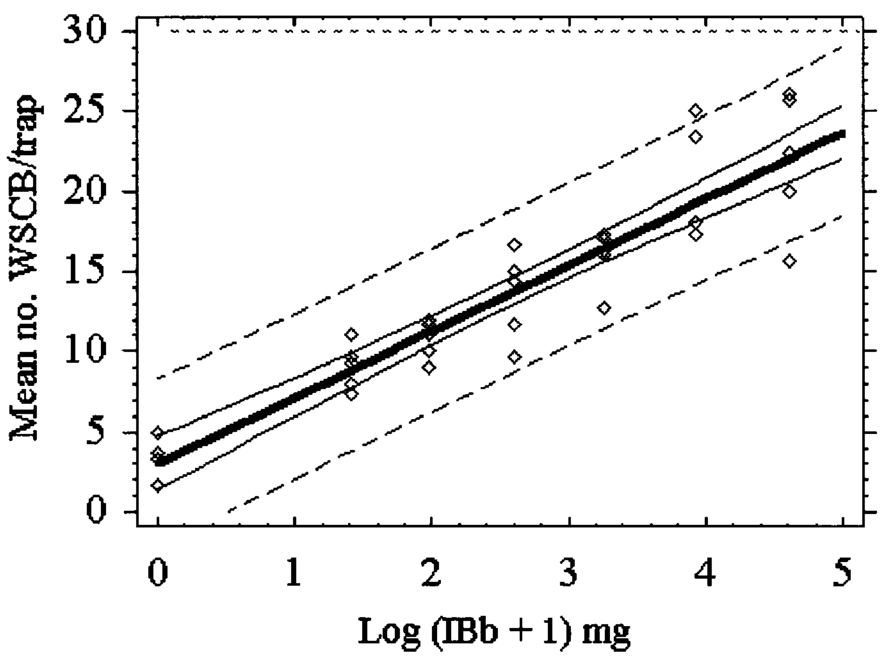
3.2.5 Dose response of WSCB to the IBb mixture

Traps baited with the IBb mixture at doses ranging from 3.125 to 100 mg per trap caught significantly more WSCB than the unbaited traps. The number of beetles captured with doses of 50 and 100 mg per trap were significantly greater than those baited with lower doses. These higher doses caught about six times more beetles than the control (Table 12). The regression analysis showed that the numbers of WSCB caught were correlated with increasing dose (Figure). The equation for the fitted model to describe the relationship between number of WSCB caught on traps and doses of attractant is: $Y = 3.07 + 4.12 * \log (X + 1)$, $R^2 = 86.27$ %, $p < 0.01$. Where Y = predicted mean of WSCB on attractant traps, and X = dosage of the IBb mixture.

Table 12. Dose response of WSCB to the IBb mixture in snap bean, at Hamlin Farm, during 7/30 - 8/2/1997.

Dose (mg/trap)	WSCB [a]	SE	Minimum	Maximum
0.0	3.4 e [b]	0.53	1.66	5.0
3.125	9.06 d	0.64	7.33	11.0
6.25	10.73 cd	0.55	9.0	12.0
12.5	13.46 bc	1.24	9.66	16.66
25.0	15.86 b	0.83	12.66	17.33
50.0	20.33 a	1.59	17.33	25.0
100.0	21.93 a	1.91	15.66	26.0

[a] n = 5, mean numbers of beetles caught per day.
[b] Means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).



Log-dose response curve of WSCB to the IBb mixture. The equation of the fitted model is $Y = 3.07 + 4.12 * \log(X + 1)$, $R^2 = 86.27$ %, $p < 0.01$. Test conducted in commercial snap bean field, at Hamlin Farm, during 7/30 - 8/2/1997.

3.2.6 Trap height

Traps baited with the IBb mixture placed at the same level as the height of the squash plants and at 10 - 15 cm above the plants did not differ significantly in numbers of WSCB caught (Table 13). Traps placed under the leaf canopy, 20 - 30 cm above the ground, caught significantly fewer beetles, than those traps placed at or above the plants. However, the number of WSCB caught on baited traps placed under the leaf canopy was 3.5 times higher than control traps at the same level.

Table 13. Comparison of trap height on the capture of WSCB in squash, at Stahlbush Island Farms, during 8/28 - 31/1996.

Treatment	Mean numbers of WSCB ± standard error [a]			
	1 DAT	2 DAT	3 DAT	Average
Canopy level + IBb	73.50 ± 7.01 a	41.75± 8.23 a	15.75 ± 2.72 a	43.67 ± 4.24 a
Canopy level	27.25 ± 1.03 b	16.50± 3.77 b	7.25 ± 1.25 b	17.00 ± 1.27 b
Taller + IBb	85.25 ± 6.54 a	38.50± 12.28 a	6.00 ± 0.71 b	43.25 ± 2.52 a
Taller Unbaited	34.25 ± 4.99 b	14.50± 5.12 b	4.75 ± 0.85 b	17.83 ± 3.53 b
Under + IBb	4.00 ± 1.29 c	3.25± 1.65 c	1.50 ± 0.29 c	2.92 ± 0.34 c
Under Unbaited	0.75 ± 0.48 d	0.75± 0.25 c	1.00 ± 0.41 c	0.83 ± 0.17 d

[a] n = 4, means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).

3.2.7 Hourly response of WSCB to attractants

According to the number of beetles caught on traps baited with the IBb mixture, flight activity was lower during the 7:00 - 9:00 AM intervals. From 9:00 - 10:00 AM, the activity increased significantly from the earlier period stated above. The numbers of WSCB caught during the period of 10:00 - 11:00 AM, 11:00 - 12:00 AM, 12:00 - 13:00 PM, 14:00 - 15:00 PM, 16:00 - 17:00 PM, and 17:00 - 18:00 PM were the highest and were not significantly different from each other. During the period of 13:00 - 14:00 PM and 15:00 - 16:00 PM, trap catches significantly declined. From 18:00 - 19:00 PM, the number of WSCB caught declined further. No beetles were caught after 19:00 PM (Table 14). Sunrise at 6:28 AM and sunset at 19:50 PM, source: the weather channel.

Table 14. Hourly response of WSCB to the IBb mixture in squash, at Stahlbush Island Farms, during 8/29/1996.

Time	Mean numbers of WSCB ± standard error [a]	
	Baited [b]	Control [b]
7:00	0.40 ± 0.16 f	0.00 ± 0.00 f
8:00	0.50 ± 0.22 f	0.40 ± 0.24 ef
9:00	7.60 ± 1.19 d	3.60 ± 1.03 ab
10:00	16.70 ± 2.06 ab	5.00 ± 1.18 a
11:00	18.80 ± 1.35 a	3.40 ± 0.68 ab
12:00	17.10 ± 1.65 ab	5.00 ± 1.26 a
13:00	14.20 ± 1.53 bc	3.00 ± 1.30 abcd
14:00	18.10 ± 1.17 ab	3.40 ± 1.03 abc
15:00	11.50 ± 1.48 c	1.60 ± 0.81 cde
16:00	17.00 ± 2.76 ab	1.20 ± 0.58 def
17:00	16.20 ± 1.63 ab	1.40 ± 0.40 bcde
18:00	2.70 ± 0.79 e	0.80 ± 0.20 def
19:00	0.00 ± 0.00 f	0.00 ± 0.00 f

[a]Means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).
[b]Baited trap, n = 10; control, n = 5.

3.2.8 Effect of field exposure on attraction

The numbers of WSCB caught on traps baited with the IBb mixture at the dose of 100 mg per trap were observed to decline after the first day of placement. The fresh preparation of the volatile compound mixture was the most attractive to the beetle. It caught about four times more WSCB than those mixtures after six days exposure in the field (Table 15). Since the activity of WSCB changed every day, the ratio of catches between baited and control traps were used to adjust the estimated number of WSCB each day before statistical analysis. The regression analysis showed an inverse relationship between the number of beetles caught and the number of days the chemicals had been in the field. The equation of the fitted model is: $Y = 5.5 - 0.67\ (X)$, $R^2 = 62.85\ \%$, $p < 0.01$. Where Y = the predicted ratio of the number of WSCB caught on traps baited with the IBb mixture, and X = the number of days that traps were in the field.

Table 15. Effect of field exposure on attractiveness of IBb mixture in squash, at Stahlbush Island Farms, during 8/28/1996 - 9/3/1996.

Day	Mean numbers of WSCB ± standard error [a]		
	Baited [b]	Control [b]	Ratio
1	140.80 ± 7.57 a	28.60 ± 3.92 a	4.92 ± 0.26 a
2	52.80 ± 3.43 b	13.00 ± 1.30 b	4.06 ± 0.26 b
3	50.10 ± 3.31 b	14.40 ± 2.73 b	3.48 ± 0.23 b
4	7.80 ± 1.48 e	3.40 ± 0.24 c	2.29 ± 0.44 c
5	12.80 ± 1.70 d	5.60 ± 2.01 c	2.29 ± 0.30 c
6	26.70 ± 2.59 c	18.00 ± 1.64 b	1.48 ± 0.14 d

[a]Means in the same column followed by the same letter are not significantly different ($p \leq 0.05$; Fisher's protected LSD).
[b]Baited trap, n = 10; control, n = 5

3.2.9 Sex ratio of WSCB attracted to plant volatile compounds

The sex ratio of WSCB caught on traps baited with the IBb mixture and its individual components were determined. Indole and benzyl alcohol attracted mostly females (≈ 73% and ≈70% respectively), while beta-ionone attracted mostly males (≈ 80%). The majority of WSCB attracted by the IBb mixture were males (≈ 80%) (Table 16 and 17).

Table 16. Sex ratio of WSCB observed in fields and from traps baited with the IBb mixture.

Crop	Date	Field Population[a]				IBb Mixture			
		n	M	F	Ratio F : M	n	M	F	Ratio F : M
Potato	7/2/97	69	31	38	1.2 : 1	55	48	7	1 : 6.9
Bean	7/3/97	48	24	24	1.0 : 1	74	58	16	1 : 3.6
Bean	7/16/97	56	15	41	2.7 : 1	246	189	57	1 : 3.3
Bean	7/23/97	46	37	9	0.2 : 1	66	63	3	1 : 21.0[b]
Bean	7/25/97	68	27	41	1.5 : 1	46	37	9	1 : 4.1
Bean	7/31/97	44	13	31	2.4 : 1	80	63	17	1 : 3.7
Squash	8/5/97	73	39	34	0.9 : 1	215	145	70	1 : 2.1
Bean	8/13/97	123	45	78	1.7 : 1	106	80	26	1 : 3.1
					Ave = 1.5 : 1				Ave = 1 : 4

[a]Numbers of WSCB observed from sweep net or direct catch.
[b]Not included in average for ratio.

Table 17. Sex ratio of WSCB observed from traps baited with the IBb mixture and its individual components.

Indole		Benzyl alcohol		beta-ionone		IBb Mixture	
n	Ratio F : M	n	Ratio F : M	n	Ratio F : M	n	Ratio F : M
30	2.3 : 1	69	2.8 : 1	113	1: 9.3	55	1: 6.9
303	3.3 : 1	84	2.4 : 1	42	1: 2.5	74	1: 3.6
286	2.7 : 1	174	2.6 : 1	66	1: 2.5	246	1: 3.3
	Ave = 2.8 : 1	164	1.5 : 1	257	1: 1.3	46	1: 4.1
			Ave = 2.3 : 1		Ave = 1: 3.9	80	1: 3.7
						215	1: 2.1
						106	1: 3.1
							Ave = 1: 3.8

4. Discussion

4.1 Efficacy of a semiochemical-based insecticide bait for control of WSCB in snap beans

One or two applications of Adios TIC® did not control WSCB as reflected by both the number of beetles and pod damage at harvest. Lance and Sutter (1990) evaluated baits with these components for control of the western corn rootworm in walk-in field cages in maize. They reported that granules containing carbaryl plus buffalo gourd root powder reduced number of live beetles per cage by 80 % within 24 h. Efficacy of baits was apparently not enhanced by adding 1.5 % TIC. The reasons for this were unclear, however, Lance and Sutter discuss that, given the distribution of granules within the cages, perhaps most beetles encountered the baits by chance during the test period. Barbercheck et al. (1995) tested the TIC commercial bait for control of southern corn rootworm in peanuts. They observed no statistically significant differences in in-shell yield because of treatments. However, they reported that the percentage of undamaged pods was highest in chlorpyrifos treated and lowest in bait treated peanuts. They also mentioned that the number of beetles captured in traps baited with TIC to monitor beetles were low relative to those baited with sex pheromone.

In the current study, one possible explanation for the lack of efficacy of Adios TIC® granular bait is probably that the bait is not attractive to WSCB. Cucurbitacins are effective as feeding stimulants and arrestants only when beetles contact and feed on them. In the absence of an "attractant at distance component", contact with the bait is random. Further, Adios TIC® granules did not stick well on the leaf surfaces and were easily removed by wind and overhead irrigation. Therefore, the chance of beetles encountering the baits was very low.

The TIC mixture was reported to be highly attractive to many *Diabrotica* spp. (Lampman and Metcalf, 1987, 1988; Lampman et al., 1991). However, Hoffman et al. (1996) observed that traps with the TIC mixture captured more WSCB than the control traps in only 1 of 2 trials. In the current study, WSCB did not respond significantly to the TIC mixture in any of the field trials. In subsequent research, 3 volatile compounds, indole (I), benzyl alcohol (B) and beta-ionone (b) were found to be promising attractants for WSCB. An equal part mixture (IBb mixture) of the three compounds consistently attracted a significant number of WSCB in all experiments. Therefore, incorporation of the IBb mixture, or other more attractive compounds, into this type of bait should be investigated.

4.2 Attractivity of plant volatile compounds for WSCB

Twenty one plant volatile compounds were evaluated for their attractivity to WSCB in snap bean and squash fields by comparing the number of beetles caught on sticky traps over 1 to 3 day periods. The chemicals beta-ionone, benzyl alcohol, and indole consistently attracted significantly more beetles than unbaited traps. These three volatiles may have chemical properties that fit into specific receptors of WSCB that stimulate behavior patterns which are a part of the complex process of host plant recognition. Phenylacetaldehyde and 4-methoxybenzyl alcohol, *p*-anisaldehyde and estragole showed some degree of attractiveness but only in one of the two experiments.

Many studies have identified attractants for many *Diabrotica* beetles that belong to the same genus as WSCB. *trans*-cinnamaldehyde; 1,2,4-trimethoxybenzene; and estragole were attractants for WCR. Eugenol and cinnamyl alcohol were found to be the effective attractants for NCR (Ladd et al., 1983; Lampman and Metcalf, 1987; Metcalf and Lampman, 1989). SCR is attracted to many compounds, including veratrole (Lampman et al., 1987). In this test, these compounds were not attractive to WSCB. The number of beetles caught on sticky traps baited with these compounds were not significantly different from the control traps. These results indicate that the response of *Diabrotica* beetles to plant volatile compounds seems to be species specific. Previous studies have also shown that volatile compounds, which were attractive to one species, were not necessarily attractive to other species in the same fields. For example, Ladd et al. (1983) reported that NCR was attracted to eugenol, but WCR was not. Andersen and Metcalf (1986) reported that indole attracted WCR but did not attract SCR.

The interaction of promising compounds was investigated for their attractiveness to WSCB. Equal parts of indole (I), benzyl alcohol (B) and beta-ionone (b) were blended into a three-component IBb mixture, and two-component IB, Bb, and Ib mixtures. All experiments showed that the mean number of WSCB caught was consistently greater on traps baited with the IBb mixture than those baited with any two-component mixtures or their individual components. However, the number of WSCB caught on traps baited with the IBb mixture was only increased additively. Although the TIC mixture was reported to be highly attractive to many *Diabrotica* spp. (Lampman and Metcalf, 1987, 1988; Lampman et al., 1991), traps baited with the TIC mixture caught no more WSCB than the unbaited traps. In 1996, Hoffman et al. observed that traps with the TIC mixture captured more WSCB than the control traps in only 1 of 2 trials. They suggested that the TIC mixture might not be optimal for this species. In the current study, when the trap catches were compared, the TIC mixture caught about three times fewer beetles than the IBb mixture. Results of the current study indicate that only indole was attractive to WSCB. The volatiles with similar molecular structure to *trans*-cinnamaldehyde and 1,2,4-trimethoxybenzene were not attractive. It is not surprising that a mixture was not attractive as well.

To determine if the response of WSCB to plant volatile compounds is enhanced by trap color, the IBb mixture was tested using different colored traps (yellow, transparent, red, blue, and white). Yellow traps caught the most beetles. This is in agreement with the results of Hessler and Sutter (1993) and Hoffmann et al. (1996) studying responses of other *Diabrotica* species to trap color. The IBb mixture caught significantly greater numbers of WSCB than unbaited traps of the same color.

Several factors affecting capture of WSCB on traps baited with the IBb mixture were evaluated to assess its potential use in WSCB management programs. Tests were conducted to compare the effectiveness of the IBb mixture baited traps placed inside an alfalfa field, at the edge, and different distances from the outer edge on fallow ground. Baited traps placed inside and at the edge of alfalfa field caught similar numbers of WSCB. The field and edge traps caught more than those traps placed outside the field.

Captures of WSCB increased significantly as doses of the IBb mixture in the trap increased. Traps baited with the IBb mixture placed at the same level as the height of squash plants and at 10 - 15 cm above the plant height caught the same numbers of WSCB. Hoffman et al. (1996) reported similar results. In pumpkins with tall canopies, traps baited with the TIC mixture placed at canopy and midcanopy height captured the most beetles. This is probably due to a stronger and more uniform scent plume than would be released from near ground, under canopy sources.

The greatest numbers of WSCB were caught during the period of 10:00 AM to 13:00 PM and from 16:00 PM to 18:00 PM. Lance (1990) reported that the most capture of the western corn rootworm and the northern corn rootworm on attractant traps occurred around midday. Similar results on times of the day and trap catches were also reported by Hoffman et al (1996). It was observed that WSCB had more flight activity when it was sunny and warm. Also higher temperatures may result in higher release rate of volatile compounds.

A fresh preparation of the IBb mixture was the most attractive to the beetle. It caught about four times more WSCB than those mixtures exposed six days in the field. However, in all of the observed dates, baited traps still caught significantly more WSCB than the unbaited controls. Although the rate of dissipation of the compound was not quantified, strong odors were still noticeable on wicks when the tests were terminated. It is believed that this mixture could be used to monitor WSCB population in the field for more than 10 days.

Previous studies reported that different attractants or mixtures of attractants lured significantly more males than females, or vice versa within a *Diabrotica* species. In the current studies, the majority of WSCB attracted by the IBb mixture were male beetles ($\approx$ 80%). Its individual components, indole and benzyl alcohol attracted mostly female beetles ($\approx$ 73% and 70% respectively) and beta-ionone attracted mostly male ($\approx$ 80%).

As in previous studies with the adult corn rootworms, there is no apparent explanation for the differential attractiveness of one chemical or mixture of chemicals to the sexes.

The IBb mixture has potential use for monitoring the population density of WSCB in snap bean fields. Further studies will better define environmental influences of temperature, humidity, wind speed and cloud cover on the attraction of WSCB to the IBb mixture.

4.3 Conclusion

The use of semiochemical-based baits for selective insect control using a minimal amount of insecticide is a promising concept. Baits containing attractants and a toxicant offer potential to increase the efficacy of insecticides. The specificity of a bait results in fewer non-target organisms exposure to the toxic compounds. Unfortunately, these field trials indicate that Adios TIC® was not effective in the control of WSCB. The granular baits did not stick well on the foliage. Since adult WSCB feed above the ground surface, the chances that beetles may encounter baits on the soil surface are very low. Also, the TIC mixture was not attractive to and failed to lure WSCB to the bait granules.

This study has confirmed indole (I), benzyl alcohol (B), and beta-ionone as promising attractants for WSCB. An equal part mixture of these compounds, the "IBb mixture" was highly attractive to the male beetle. Several tests were conducted to investigate factors affecting capture of WSCB on traps baited with the IBb mixture. It is believe that the IBb mixture has potential use for monitoring populations of WSCB in commercial snap bean fields and that numbers trapped may correlate to the need for chemical control. Baits employing the IBb mixture and a toxicant should be evaluated against commercial carbaryl or diazinon sprays for WSCB control and management strategies.

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