METHODS OF SELECTION FOR HAIRY CHINCH BUG, <u>BLISSUS</u> LEUCOPTERUS HIRTUS, RESISTANCE IN COOL SEASON TURFGRASSES

> by Paul Benjamin Baker

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## APPROVAL SHEET

Title of Dissertation: Methods of Selection for Hairy Chinch Bug, Blissus leucopterus hirtus, Resistance in Cool Season Turfgrasses

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#### ABSTRACT

Title of Dissertation: Methods of Selection for Hairy Chinch Bug, Blissus leucopterus hirtus, Resistance in Cool Season Turfgrasses

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Laboratory procedures were developed for rearing the hairy chinch bug and evaluating Kentucky bluegrass cultivars for tolerance to adult chinch bug feeding. Progress was made toward development of a technique for measuring adult non-preference in cool season turfgrasses. Chinch bugs were reared in the laboratory on corn sections in 1/2-pt. cardboard cartons. Field and laboratory reared adults were confined in 1/2-gal. cartons on corn sections to provide a source of eggs for laboratory colonies. There was significantly higher nymphal survival and return of adults when eggs were surfacesterilized in 2% sodium hypochlorite solution as compared to those treated with a 1% solution or untreated eggs. There was also a significantly higher return of adults when chinch bugs were reared on corn sections treated with a 2% sodium hypochlorite solution versus stems treated with 2% Arasan or untreated stems. Developmental times for nymphal instars were determined as follows: 1st 12.3 ± 6.0; 2nd 5.4 ± 2.7; 3rd 5.2 ± 1.7; 4th 4.9 ± 1.3; 5th 7.1 ± 0.9 total 35.5 ± 7.4 days. The preoviposition period was determined to be 10.8 ± 4.4 days with nearly 80% of the females tested ovipositing within 24 days.

Cultivars of Kentucky bluegrass were evaluated in the laboratory for tolerance to adult chinch bug feeding when ca. 1 month old. Selections were seeded in 15.2-cm pots in 4 tufts (groups) of seed/ pot. Tufts were thinned to 5 plants each, 7-10 days prior to infestation, and cut to 3.8 cm the day of infestation. During infestation, plants were confined within a plastic cylindrical cage 10 x 20.3 cm high. The cage was divided longitudinally by a flat piece of clear plastic glued between the halves of the cylinder. Adults were placed in one side of the cage; the other side served as uninfested check. After the infestation period the following growth responses were recorded: regrowth, yield, percent dry matter, root development, plant survival, and tillering. In the majority of the cases, chinch bug feeding had a significant effect on plant responses. At infestation rates of 2 adults/plant or higher, plants were severely injured and top and root growth significantly reduced. There were also significant differences in regrowth, yield, percent dry matter, and plant survival among cultivars, indicating that these may be useful criteria for measuring tolerance.

Several approaches were explored to measure adult non-preference. These included exposing adults to single plants, small groups of plants and various sized circular areas of grass ranging from 0.2 to 6.3 cm<sup>2</sup>. Results showed a low response of adults to single plants or small groups of plants, but an inconsistent response to grasses seeded in the various sized circular areas. Adults appeared to prefer the larger areas. Evaluation of 5 cool season grass species planted in 6.3 cm<sup>2</sup> areas, indicated that chinch bug adults may prefer finer leafed turfgrasses. However, this response may be influenced by density of plants in the area, since there were more plants/area of the finer leafed species than the broader leaf species, thus possibly providing better shelter and a more favorable microenvironment.

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## INTRODUCTION

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In recent years, insect problems on turfgrass have become increasingly more evident as a result of the removal of various insecticides for use on turfgrass and the development of insecticidal resistance by many of the major insect pests. In the past, most of the major insect pests, such as chinch bug complex Blissus spp., sod webworm, Crambus spp., billbug Sphenophorus spp., and scarabaeid grubs, were controlled primarily by insecticides. During that time problems related to disease control or agronomic aspects of turfgrass production frequently received more attention because of the relative ease in controlling insects chemically. With the frequent use of insecticides, however, entomologists became aware of other detrimental effects on the turfgrass ecosystem. Streu (1973) noted that the impact of multiple applications over a number of seasons can be cumulative, resulting in pest resurgence, insecticide resistance, and other changes such as plant species succession and plant growth response.

Cost of controlling turfgrass insects in Maryland is presently estimated at 2 - 4 million dollars (L. Hellman, personal communication). Such losses are found in many states. Thus, there is a growing need to develop non-chemical control methods to reduce dependence on insecticides and provide turfgrass growers and users with alternative methods of control at a lower cost. Host plant resistance provides such an alternative for the suppression of insect damage, provided resistant germplasm can be identified and incorporated into

agronomically acceptable cultivars.

The chinch bug complex, Blissus spp., consists of 15 species in the New World, with the most economically important species being the chinch bug, B. leucopterus leucopterus (Say); the hairy chinch bug, B. 1. hirtus Montandon; and the southern chinch bug, B. insularis Barber (Leonard 1968). Members of this complex, particularly B. leucopterus leucopterus, have been reported as pests of the Gramineae family since the 1780's. The first record of damage by B. leucopterus, presently B. 1. leucopterus, was reported on wheat in North Carolina and South Carolina in about 1783 (Leonard 1966). There were similar reports of damage to wheat in North and South Carolina and Virginia in succeeding years. It was not until the 1840's, however, that serious chinch bug infestations were again reported; this time in the Carolinas, Virginia and Illinois. During the period from the 1840's-1880's reports of chinch bug problems in the Midwest coincided with the opening of the tall grass prairies for agricultural crops. The first widespread outbreak in the Midwest occurred in 1864 on wheat, when losses in Illinois alone were estimated at 73 million dollars (Leonard 1966). Since then, many of the corn, small grain and sorghum producing states have reported outbreaks of chinch bugs periodically up to the present (Dahms et al 1936, Snelling et al 1937, Leonard 1966).

The first report of chinch bug damage to a timothy pasture was from New York by Lintner (1883). Howard (1887) reported the first chinch bug damage on turfgrass in a Brooklyn lawn. However, it was not until recently that the hairy chinch bug was considered to be a serious pest of turfgrass due in part to the development of

insecticide resistance (Streu and Cruz 1972). Selection for resistance to chinch bugs has been largely confined to corn and sorghum (Snelling et al 1937, Dahms 1948). Field evaluation for resistance to <u>B. 1. leucopterus</u> in forage grasses was reported by Hayes and Johnson (1925), but resistance in turfgrasses was not studied extensively until the early 1970's when research on southern chinch bug resistance in St. Augustinegrass was begun (Reinart 1974). Cooperative research in Texas and Florida resulted in the development and release of the chinch bug resistant St. Augustinegrass cultivar 'Floratam' (Horn et al 1973). There is a similar need to develop programs to select for hairy chinch bug resistance in cool season turfgrass.

The present study was designed for this purpose and was part of a larger research program to select for insect resistance in cool season grasses. The major objectives were to develop methods for: (1) rearing large numbers of hairy chinch bugs in the laboratory for use in a screening program; (2) screening grass germplasm in the laboratory for resistance to the hairy chinch bug; and (3) to identify the mechanism(s) of resistance. The research was divided into 3 areas: rearing and other biological studies, tolerance, and non-preference to the hairy chinch bug.

#### MATERIALS AND METHODS

## Rearing Studies

Parker and Randolph (1972) reported rearing the chinch bug on corn sections in gallon cardboard containers in the laboratory. I attempted to rear the hairy chinch bug using their method, but found that it was not adequate for rearing large numbers of individuals because of the high mortality of early instars. Mortality appeared to be associated primarily with mold build-up on the corn sections and eggs which trapped young nymphs. To reduce the mold and thus increase survival in the early instars, I conducted 2 studies to investigate the influence of surface sterilization of eggs and corn on reduction of mold (and possibly other organisms). Once adequate methods for rearing the insect were developed, I conducted a series of tests to determine the biology of the insect reared by this procedure.

The following general procedures were used in all rearing studies. Insects were fed on field corn grown in the greenhouse under a 16-18 h photoperiod of natural and artificial light. Corn was harvested when 4-6 weeks old and stems were cut into 7.5-cm sections for feeding. One cut end was coated with paraffin to reduce moisture loss. Adults used for egg sources were maintained in 1/2gal. cartons with cut corn sections and were supplied with 2 rolls of cheesecloth to serve as oviposition sites as described by Parker and Randolph (1972). Eggs were collected and placed on moist filter paper on a moistened sponge in a 9 x 1.9-cm petri dish. After hatch, nymphs crawled from moist filter paper to the corn

sections. Corn sections and 1/2-pt cardboard cartons were changed at least weekly until all nymphs had matured or died. Cartons were placed in clear plastic bags in an environmental chamber maintained at 26°C, 40-75% RH and a 16-h photoperiod.

Egg Treatment: -- Eggs were collected from 1st generation laboratory-reared adults and were treated with either 1 or 2% sodium hypochlorite (SH) solution or distilled water. Eggs were surfacesterilized in SH (Treatments 1 and 2) by immersing them for 20 min in the solution and then in distilled water for 20 min. Unsterilized eggs (Treatment 3) were immersed only in distilled water for 20 min. Following removal from the distilled water 30 eggs per treatment were transferred by brush to a 5.5 x 1.2-cm petri dish with a moist filter paper on a moist sponge. Petri dishes were placed into cartons with 2 corn sections, which provided food for hatching nymphs. Before corn sections were placed in cartons, they were surface-sterilized in 2% SH solution for 20 min, then rinsed in distilled water for 20 min and allowed to air dry on paper towels. A 10  ${\rm cm}^2$  piece of paper towel was forced over the rim of the carton by the top to form a tight seal. Each treatment was replicated 5 times on 3 dates. Observations were made on nymphal development every 3-5 days, and cartons and corn sections were changed weekly.

Corn Treatment:--Thirty eggs were surface-sterilized in 2% SH, rinsed and transferred to petri dishes and placed in cartons with 2 corn sections as described for Study 1. Prior to placement in cartons, corn sections were divided into 3 groups and treated as follows: treatment 1 - surface-sterilized for 20 min in 2% SH solution, rinsed in distilled water for 20 min and air dried on paper

towels; treatment 2 - sections were placed in a 2% Arasan solution for 20 min and air dried on paper towels; treatment 3 - no treatment. A water vial (6.8 x 1.8-cm) with a cotton plug was added to each container to maintain high relative humidity. Nymphal development was checked every 3-5 days at which time corn sections and cartons were changed. Each treatment was replicated 5 times on 4 dates.

Nymphal development:--Eggs were collected, surface-sterilized in 2% SH and handled as described in the general procedure until 5-7 days old. At that time, 75 red eggs (indicating embryo development) were transferred singly to moist filter paper disks (0.6 cm<sup>2</sup>) and placed in cartons. Prior to this, the bottom seam of each carton was sealed with melted wax and the upper 2 cm of the inside surface was coated with talcum powder to reduce loss of nymphs. Two corn sections, surface-sterilized with 2% SH were placed in the carton over a  $3.2 \text{ cm}^2$  section of paper towel. Cartons were then closed and stored as described in the general procedure. Nymphal development was observed daily and corn sections were changed every 3-5 days.

Female Longevity and Oviposition:--Fourth and 5th instars were collected from the field and 1st generation laboratory colonies and maintained in separate 1/2 gal cardboard cartons on corn sections until mature. Newly emerged females were removed daily and placed individually with a male and 2-3 corn sections in 1/2-pt cardboard cartons with a vented top. Cartons were held in a reach-in chamber as in previous studies. Eggs were collected every 2-3 days and corn sections changed weekly. Dead males were replaced at each check. The total number of eggs/day/female was compared for laboratory and field collected adults. Total egg counts included viable and

nonviable (flattened) eggs.

Preoviposition Period:--First-generation laboratory reared 4th and 5th instars were pooled in 1/2-gal cardboard cartons on corn sections. Newly emerged adults were collected for each day and pooled in 1/2-pt cartons on corn sections. After 3-5 days, individual females were confined with at least 1 male and placed with 2-4 corn sections in 1/2-pt cartons with a vented top. Cartons were checked for eggs daily and corn sections were changed weekly.

A standard ANOVA was conducted on the Egg and Corn treatment studies. A standard 't' test was conducted for the Preoviposition and Longevity and Oviposition studies, with means and standard deviation used to express Nymphal Developmental times.

## Tolerance Tests

Four experiments were conducted to evaluate Kentucky bluegrass cultivars for tolerance to adult chinch bug feeding. The following general procedures were used in all tests. Grass cultivars were evaluated for tolerance to feeding when <u>ca</u>. 1 month old. Each selection was seeded in 15.2-cm pots of sand in 4 groups (tufts) of seed per pot and cut to 3.8 cm after 3 weeks. Sand was obtained from the field, screened, and the appropriate nutrients (N, P, K and micro-nutrients) and lime were added (J. Murray, personal communications). Tufts were thinned to 5 plants each 7-10 days prior to infestation, and were cut to 3.8 cm the day of infestation. During infestation, plants were confined within a clear plastic cylindrical cage 10 cm in diam and 20.3 cm high. The cage was divided longitudinally by a flat piece of clear plastic glued between the

halves of the cylinder. A 7.6-cm hole was cut in each side and center section of the cage approximately 4.0 cm from the base and covered with mesh screen for ventilation (Fig. 1). The screen on the sides of the cage was fastened so as to form a flap that could be opened to enable cutting of the grass without removal of the cage. Rubber bands and a piece of wood (10.2 x 2.0 cm) were used to secure the flap. Adults were placed in one side of the cage; the other side served as an uninfested check. Cages were secured with white gravel on the inside and sand on the outside. Talcum powder was placed on the top 2 cm of surface of the infested side to prevent insects from escaping. Following an infestation period of 17-19 days, cages were removed, insects collected and counted, and regrowth, yield, % dry matter, root development, plant survival, and tillering were recorded. To enable root measurements pots with plants were submerged in a bucket of water until plants could be lifted free from the sand. Excess sand was washed free in a 2nd bucket. The root mass of each plant was then measured from the tips of the roots to the plant crown and the measurements for the 5 plants in each tuft were averaged. Root weights were taken for each tuft of 5 plants after the root masses were oven dried at 140°C for 3-5 days. Root organic matter was obtained by ashing the dried roots at 600°C for at least 6 hours and subtracting the remaining weights from the dried weights.

Insects used in these studies were 1st-generation laboratoryreared adults, except in Experiment 3, when field collected adults were used. All experiments were conducted in a rearing room at 21-24°C, 50-80% RH and a 14-h photoperiod. Treatments were

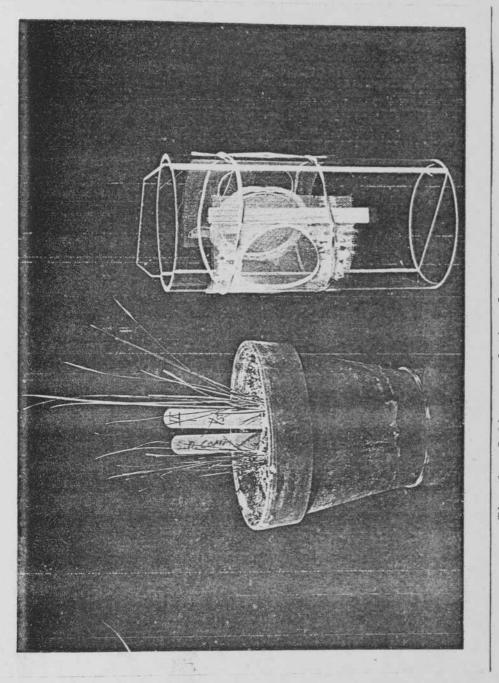


Fig. 1. Split cage design for tolerance tests

replicated 4 times for Experiments 2 and 3 and 5 times for Experiments 1 and 4, using a randomized complete block design.

Experiment 1:--The 5 cultivars, 'Adelphi', 'Baron', 'Fylking', 'Newport', and 'South Dakota Common' (SDC) were chosen because of their diversified field responses to chinch bug feeding (J. Murray, personal communication). An infestation rate of 2 adults/plant was used. Following removal of adults and recording of plant data, plants were allowed to regrow in the greenhouse without insects present (2nd cutting) for 19 days after which the same measurements were again recorded.

Experiment 2:--The same 5 cultivars were evaluated at infestation rates of 1 and 2 adults/plant. Second-cutting data were not recorded.

Experiment 3:--'Fylking' and 'SDC' were evaluated at infestation rates of 1, 1.5, 2 and 2.5 adults/plant. Second cutting data were taken as described for Experiment 1.

Experiment 4:--Twelve cultivars ('A-34', 'Adelphi', 'Bonnieblue', 'Campina', 'Delta', 'Geronimo', 'Kenblue', 'Newport', 'Parade', 'Park', 'Ram I', and 'Troy') with different agronomic characteristics were evaluated at an infestation rate of 1.5 adults/plant.

#### Preference Studies

In initial studies I evaluated the response of adults to a single plant (stem) or a small group of plants of various grasses in an effort to measure preference. Adults were not generally attracted to the plants offered under these conditions even though they were varieties that they normally fed on. As a result, further tests were designed to evaluate the response of adults to various sized

areas of plants in an effort to reduce the possible influence of micro-environment created by plant density. The following general procedures were used in all studies. Grass sections were seeded into various sized areas within a 15.2 cm diam circle in a wooden flat (60.8 x 30 x 7.5-cm) containing soil. Each of the areas was randomized within the circle and each circle constituted a replication. Approximately 3-5 weeks after seeding, plants were cut to ca. 5.1 cm. One week later plants were again cut to 5.1 cm, and a clear plastic cylinder 10.2 cm in diam was placed around each circle. Cylinders were inserted into the soil and secured with white gravel on the inside and soil on the outside. Talcum powder was placed around the top 2 cm of the inside surface to prevent insects from escaping. Sixty 1st-generation laboratory-reared adults were released in the center of each circle. After a specified time period adults were aspirated off and counted. Adults were counted as within an area if they were on/in the grass or within a 0.75-cm circumference of the area. Following removal of adults, the grass from each group was cut at the crown and oven dried at 140°C for 3-5 days. Counts were then calculated as the number of adults per mg of dry weight of plant material for each area. Tests were conducted in an environmental chamber at 24°C with 70-100% RH and a 15-h photoperiod.

Area Study 1:--Perennial ryegrass, <u>Lolium perenne</u> L. var. Manhattan, was seeded in 6 areas within each circle, with 8 circles (replications)/flat. Each flat represents a time interval of 24, 48 and 72 h. Area sizes increased by a factor of 2 from 0.19 - 6.3 cm<sup>2</sup>.

Area Study 2:--Material and methods were identical to Area Study I except there were 10 replications and counts were taken only once

after 24 h.

Species Study:--Five species of cool season turfgrass were seeded or transplanted from the field to areas of 6.3 cm<sup>2</sup> in a circle. The species included: <u>Agrosta</u> spp. (Bentgrass); <u>Festura arundinacea</u> Schreb (Tall Fescue); <u>F. rubra</u> L. (Red Fescue); <u>L. perenne</u> L. (Perennial Ryegrass); <u>Poa pratensis</u> L. (Kentucky Bluegrass) and a check of dried wheat stubble (non-host plant). A number of plugs of the 5 species were obtained from the field and transplanted into pots in the greenhouse. Selections were made among those that survived in the greenhouse and subsequently were transplanted to test flats. When field collected plants showed adequate regrowth, all plants were tested for preference. Plant material was cut to 5.1 cm every 7-10 days until tested.

A standard ANOVA was conducted on data from all Tolerance Tests and Preference Studies, with Duncan's Multiple Range tests used to separate means.

### RESULTS AND DISCUSSION

## Rearing Studies

Egg Treatment:--Results are presented in Table 1. There was no significant effect of treatment on egg hatch or survival of 1st and 2nd instars. However, mortality through the 2nd stadium was high (ca. 70%) for all treatments with over 55% occurring from loss or death of nymphs. Survival of 3rd-5th instars and adults was highest from groups receiving the 2% SH treatment, however, I cannot explain why egg treatment should influence survival of older nymphs or adults. The modification that I made to Parker and Randolph's (1972) technique, such as smaller cartons, more frequent changing of corn sections, and maintaining higher RH, improved nymphal survival over that obtained in the initial trials. However, it was still necessary to reduce the high mortality occurring to the early instars if this method was to be used successfully in rearing the hairy chinch bug.

Corn Treatment:--Results are presented in Table 2. There was no significant effect of corn treatment on egg hatch, however nymphal survival through the 2nd stadium was significantly greater on untreated corn or corn treated with 2% SH than on corn treated with 2% Arasan. There were no significant differences in survival of 3rd-5th instars among treatments and over 80% of the 3rd instars matured to adults. However, the total number of insects reaching maturity was significantly greater on 2% SH treated corn than on either untreated or Arasan-treated corn.

A comparison of the 2 studies showed that increased production of adults in Test 2 resulted primarily from higher survival of 1st and

% sodium hypochlorite	Age interval (x)	Mean no. alive at start of age interval	Mean no. dying during age interval	% mortality during age interval <sup>2</sup>
	Egg			
2 1		30	5.2	17.3 a
1		30	4.7	15.5 a
0		30	6.3	21.1 a
	1-2nd ins	tar		
2		24.8	15.6	62.9 a
2 1 0		25.4	16.7	65.7 a
0		23.6	17.7	75.0 a
	3-5 insta	r		
2		9.2	2.3	25.0 a
1 0		8.7	4.6	52.8 b
0		6.0	2.1	35.0 a
	Adult			
2		6.8 a	.1	1.1
1		4.0 b	.1	3.2
2 1 0		4.6 b	.2	3.4

Table 1. A partial life table of the hairy chinch bug reared on corn stem sections after eggs were treated with distilled water, 1 or 2% sodium hypochlorite.  $^{\rm l}$ 

<sup>1</sup>15 Replications.

<sup>2</sup>Means not followed by the same letter are significantly different (P < 0.05) (Duncan's Multiple range test).

Treatment	Age interval (x)	Mean no. alive at start of age interval	Mean no. dying during age interval	% mortality during age interval <sup>2</sup>
	Egg			
S.H. Arasan Untreated	1-2nd ins	30 30 30 30	5.0 4.7 5.7	16.6 a 15.5 a 19.0 a
S.H.	3-5 insta	25.0	10.8	42.4 a
Arasan		25.4	13.8	54.3 b
Untreated		24.3	11.7	48.1 a
S.H.	Adult	14.2	1.8	12.6 a
Arasan		11.6	1.7	14.6 a
Untreated		12.6	2.5	19.8 a
S.H.		12.4 b	0.0	0.0
Arasan		9.4 a	0.0	0.0
Untreated		10.1 a	0.0	0.0

Table 2. A partial life table of the hairy chinch bug reared on corn stem sections which were treated with 2% sodium hypochlorite (S.H.) Arasan or untreated.  $^{\rm l}$ 

<sup>1</sup>20 Replications.

 $^2Means$  not followed by the same letter are significantly different (P < 0.05) (Duncan's Multiple range test).

2nd instars. Egg hatch and survival of 3rd-5th instars to adults was in most cases very similar (ca. 80%) in both tests. It appears that improved survival in Test 2 resulted primarily from more frequent changing of corn sections rather than from the influence of corn treatments <u>per se</u>, although survival was slightly higher on the SHtreated stems than untreated stems. A comparison of results of the 2 studies shows that there was approximately a 20% increase in survival for 1st-2nd instars, a 10% increase in survival of 3rd-5th instars, and a 50% increase in the production of adults in Test 2 vs Test 1. This in turn, resulted in an overall increase in return of adults of 41% on 2% SH-treated stems in Test 2 vs 22% in Test 1.

There was no significant effect of egg or corn treatment on sex ratio or percentage of short- and long-winged forms of adults produced within each study (Table 3). Under these conditions there was a slightly higher percentage of males produced (53%) and more short-winged forms of both sexes. Leonard (1966) reported B. 1. hirtus as having an overall total of 63.7% brachypterous population. Luginbill (1922) and Chambliss (1895) reported a majority of the spring generation were long-wing forms and summer generation were short-winged forms for <u>B</u>. <u>1</u>. <u>leucopterus</u>. Sweet (1964) in reporting on the biology and ecology of the Rhyparochrominae of New England (Lygaeidae) found a good correlation between the proportion of brachypterous forms and permanency of habitat. Leonard (1966) indicated a similar strong correlation was evident in some species of Blissus. Based on these reports, one would expect a higher percentage brachypterous adults among the hairy chinch bug reared under the conditions of this study.

Treatme	ent	% Males	% Male short wings	% Female short wings	
Eggs <sup>1</sup> -	0	55.7	88.3	60.6	
	1	54.1	83.3	51.2	
	2	49.4	77.7	49.2	
Corn <sup>2</sup> -	untreated	54.9	77.6	78.0	
	Arasan	54.1	83.5	75.7	
	2%	50.5	80.3	69.8	

Table 3. Percent males, males with short wings, and females with short wings in rearing studies of the hairy chinch bug.

<sup>1</sup>15 Replications.

<sup>2</sup>20 Replications.

Development Time: -- The developmental time, in days, for the various instars in these tests was as follows: 1st - 12.3 ± 6.0; 2nd - 5.4 ± 2.7; 3rd - 5.2 ± 1.7; 4th-4.9 ± 1.3; 5th - 7.1 ± 0.9; total 35.5 ± 7.4 days. The developmental time for the first 3 instars was very similar to that reported by Luginbill (1922) for the chinch bug (B. 1. leucopterus) reared on corn leaves in small shell vials. However, Luginbill reported a much longer developmental time of 14.7 and 24.6 days for the 4th and 5th instars, respectively. This resulted in a total development time of 66.4 days to adult, which is almost twice that found for the hairy chinch bug in this study. Chambliss (1895) and Snelling et al (1937) reported B. 1. leucopterus developed in 4-6 weeks on small grains in the Midwest. In Maryland the hairy chinch bug develops in approximately 4-6 weeks in the summer (Hellman, personal communications). Therefore the developmental time reported here is within that range. Luginbill (1922) also reported that the length of the immature stages was somewhat extended in his study because the nymphs were kept under unnatural conditions. This could also result from feeding on excised corn sections since, as tissues degenerate, certain nutrients may become limited in quantity, but over time, the nymphs may have been able to accumulate enough of this factor(s) to enable it eventually to mature. I have also found nymphs that appear to have extended developmental times in other colonies under study. However, regardless of this extended time, the hairy chinch bug can be successfully reared in the laboratory using the above described procedure.

Female Longevity and Oviposition: -- There were no significant differences between laboratory and field collected adults in longevity or oviposition although field adults laid more eggs (170 to 118) and lived longer (102 to 73 days). I could not find reports of similar data on the hairy chinch bug in the literature. However, Janes (1935) and Dahms (1947) reported on the oviposition and longevity of B. 1. leucopterus reared on wheat and sorghum, respectively. Janes (1935) found that field collected adults laid an avg. of 532 eggs and lived for an avg. period of 94 days at 24.5°C on young wheat plants. Dahms (1947) reported B. 1. leucopterus feeding on sorghum seedlings grown in balanced nutrient solutions produced an avg. of 189 eggs and lived an avg. of 45.9 days. The much greater oviposition rate and longer longevity reported by Janes (1935) than Dahms (1947) may result from the fact that Janes (1935) changed material daily, or it may be due to differences in host plants (wheat vs sorghum) on the insects. Similarly, the lower rate I obtained may be due to host plant, the use of excised material or possibly inherent differences between Blissus species.

Preoviposition:--A preoviposition period of  $10.9 \pm 4.4$  days was obtained for 1st-generation laboratory-reared adults. Of the 68 females ovipositing, 51 laid eggs within 30 days. Of these ca. 80% oviposited between 5-14 days. Janes (1935) reported field populations of <u>B. 1. leucopterus</u> oviposited at 9 days with a range of 5-15 days at 24°C.

#### Tolerance Tests

Painter (1951) defines tolerance as a basis of resistance in which the plant shows an ability to grow and reproduce itself or to repair injury to a marked degree in spite of supporting a population approximately equal to that damaging a susceptible host. In this

study tolerance was expressed as a percent difference from the uninfested check; the smaller the difference the more tolerant the cultivar. Differences were expressed as negative (-) a reduction, or positive (+) an increase from the uninfested checks.

The effects of chinch bug feeding on plant responses is summarized in Table 4. Plant responses are shown for 3 of the 4 tests and are an average for all cultivars in the respective tests. Data from Test 3 were not included because the infested value used in the analysis was based on an average of 4 infestation levels, while the value used in the other tests was based on only 1 or 2 infestation levels (Test 2). In all cases, except plant survival in Test 2, chinch bug feeding had a significant effect on plant response. Regrowth, dry weight, yield, root length and plant survival were reduced by adult feeding. Percent dry matter was increased in all cases. Chinch bugs feed primarily in the phloem and xylem tissues of plants, resulting in stunted growth (Painter 1928) and thus could be a major contributor toward moisture depletion in the plant. This could account for the fact that infested plants contained a higher % dry matter than the uninfested checks. The significant reduction in top and root growth resulting from adult feeding in these tests, helps to explain the severe injury sustained in the field from high chinch bug populations. An infestation rate of 1.5 adults/plant as used in Test 4, in the laboratory would correspond to ca. 350 adults/ ft<sup>2</sup> in the field. Populations of this level are not uncommon in Florida (Reinart and Kerr 1973) and were reported from field plots at Beltsville during the summer of 1978 (Ratcliffe, personal communication). The results of these tests demonstrated the usefulness of

			Tests	1		
Plant Responses	I <sup>2</sup>		II3		IV4	
regrowth (cm)						
Infested	4.4	a	7.5	a	2.0	a
Uninfested			17.9		13.7	
dry weight (mg)						
Infested	14.7	a	7.7	a	3.9	a
Uninfested	26.1	b	31.1	b	27.3	b
% dry matter						
Infested	26.1	a	30.0	a	51.7	a
Uninfested			19.9		20.2	
root length (cm)						
Infested	9.3	a	10.6	а	3.9	a
Uninfested	13.7	b	13.1	b	6.8	Ъ
plant survival (%)	1					
Infested		a	96.0	a	41.0	a
Uninfested			99.3		96.2	

Table 4. Effects of feeding by the hairy chinch bug on plant responses for 3 tests.

<sup>1</sup>Means for a given plant response within the same column not followed by same letter are significantly different (P < .05) (Duncan's multiple range test).

<sup>2</sup>Infestation level of 2 adults/plant.

<sup>3</sup>Infestation level averaged for 1 and 2 adults/plant.

<sup>4</sup>Infestation level of 1.5 adults/plant.

the split-cage method employed, both to evaluate tolerance in the laboratory and to enable investigators to correlate plant responses in the laboratory with those in the field. In addition, the cage design would enable one to cut grasses periodically while under infestation, to simulate conditions of insect infestation and cutting practices which occur in the field. By this means it would be possible to study the influence of the interaction of insect feeding and cutting practices on plant growth and survival.

Experiment 1:--The results of this experiment are presented in Table 5. At first cutting there were no significant differences among cultivars for regrowth during infestation, although 'Newport' showed the least percent reduction (more tolerance). When losses in dry matter were compared 'Newport' was significantly different than 'SDC' and 'Adelphi'. At 2nd cutting 'Newport' and 'Baron' had significantly less percent reduction in regrowth and yield than 'Adelphi'.

Experiment 2:--'Newport' and 'Baron' were significantly more tolerant than 'Fylking' and 'SDC' based on percent reduction in regrowth when infested with 1 adult/plant (Table 6). In initial tests some cultivars showed little differences in percent reduction at an infestation level of below 1 adult/plant. At an infestation level of 2 adults/plant there were no significant differences among cultivars for tolerance based on either regrowth or yield. 'Newport' and 'Baron' were significantly more tolerant than 'SDC' based on percent reduction in root weight when infested with 1 adult/plant.

Experiment 3:--In general, first cutting regrowth and yield were reduced at all infestation levels for both cultivars (Table 7). Reductions increased with an increase in infestation rates from 1 to 2

Table 5. Tolerance of Kentucky bluegrass cultivars to feeding by adult hairy chinch bugs expressed as percent difference between uninfested and infested plants<sup>1,2</sup>.

		1st cutting	ng		17	2nd cutting	
Entry	Regrowth cm	Yield mg	% Dry Matter	Regrowth cm	Yield mg	% Dry Matter	Root Length
Newport	-71.0 a	-54.0 a	+75.2 ab	-41.3 b	-39.3 b	+6.0 a	-16.4 b
Fylking	-80.1 a	-74.6 ab	+60.7 a	-56.9 ab	-61.9 ab	+1.9 a	-30.7 ab
Baron	-80.9 a	-73.0 ab	+67.0 ab	-43.6 b	-45.9 b	+4.1 a	-27.3 ab
S.D.C.	-82.0 a	-77.7 b	+71.7 ab	-37,7 b	-53.4 ab	+1.1 a	-31.7 ab
Adelphi	-85.5 a	-77.0 b	+113.7 b	-73.2 a	-82.3 a	+10.1 a	-46.6 a

<sup>1</sup>Density level of 2 adults/plant.

I <sup>2</sup>Means for a given variable followed by the same letter are not significantly different at the 5% level DMR. Table 6. Tolerance of Kentucky bluegrass cultivars to feeding by adult hairy chinch bugs expressed as percent difference between uninfested and infested plants<sup>1,2</sup>.

						KOOT	
Entry	<pre># Adults per plt.</pre>	Regrowth cm	Yield mg	% Dry Matter	Weight	Organic Matter	
Baron	1	-35.3 c	-52.2 b	+3,3 a	-24.4 b	-26.9 c	
Newport	1	-37.6 bc	-62.4 ab	+11.0 a	-23.5 b	-33.7 bc	
Adelphi	1	-54.0 ab	-71.9 ab	+54.5 ab	-53.3 ab	-54.6 abc	
Fylking	1	-56.7 a	-76.1 a	+70.6 ab	-53.6 ab	-51.2 abc	
S.D.C.	1	-59.5 a	-77.5 a	+22.0 a	-65.2 a	-63.1 ab	
Baron	2	-64.3 a	-79.1 a	+25.1 a	-43.3 ab	-67.9 a	
Newport	2	-66.6 a	-82.9 a	+54.1 ab	-56.5 ab	-51.7 abc	
Adelphi	2	-66.6 a	-84.7 a	+131.4 h	-59.5 ab	-60.5 ab	
Fylking	2	-64.2 a	-81.0 a	+46.6 ab	-50.6 ab	-51.2 abc	
S.D.C.	2	-66.2 a	-84.8 a	+60.3 ab	-56.9 ab	-56.6 abc	

1 <sup>2</sup>Means for a given variable followed by the same letter are not significantly different at the 5% level DMR.

Table 7. Tolerance of Kentucky bluegrass cultivars to feeding by adult hairy chinch bugs expressed as percent difference between uninfested and infested plants  $^{1}$ ,  $^{2}$ .

			BIITINN IST	20	SIIT LULLING	CTIIS
Entry	<pre># Adults per plt.</pre>	Regrowth cm	Yield mg	% Dry Matter	Regrowth cm	Yield mg
Fylking	1.0	-65.3 a	-55.9 b	+74.7 ab	-53.7 abc	-81.8 a
Fylking	1.5	-65.2 a	-67.5 ab	+73.9 ab	-67.4 a	-75.7 a
Fylking	2.0	-79.9 a	-74.3 ab	+115.1 b	-71.2 a	-78.9 a
Fylking	2.5	-82.9 a	-80.7 ab	+111.8 b	-82.9 a	-87.0 a
S.D.C.	1.0	-33.9 b	-38.8 b	+21.5 a	-22.3 c	-68.7 a
S.D.C.	1.5	-63.1 a	-67.7 ab	+37.5 a	-31.0 bc	-80.8 a
S.D.C.	2.0	-83.3 a	-89.5 a	+69.7 ab	-61.7 ab	-93.9 a
S.D.C.	2.5	-82.0 a	-85.8 a	+119.4 b	-81.1 a	-88.8 a

1 <sup>2</sup>Means for a given variable followed by the same letter are not significantly different at the 5% level DMR. adults/plant and then leveled off. There was a significant difference in percent reduction in regrowth on 'SDC' between rates of 1 and 1.5 through 2.5 and yield at 1 and 2.0 and 2.5 adults/plant. 'SDC' showed significantly less percent dry matter differences at infestation rates of 1 and 1.5 adults/plant than at 2.5 adults/plant. At 2nd cutting, 'SDC' had significantly less percent reduction in regrowth at infestation rates of 1 and 1.5 than 2.5 adults/plant.

A comparison of 1st and 2nd cutting data for regrowth and yield showed for the most part greater differences in these criteria at 2nd cutting than 1st cutting. This would indicate that even after insects are removed, plant responses are still under stress and recovery may be incomplete or not at all. Painter (1928) reported chinch bug injury is caused mainly by the withdrawal of fluids from the phloem and xylem tubes and stoppage of the conducting tissues by sheath materials, resulting in the starvation of roots for synthesized foods and moisture. Therefore recovery under these or prolonged conditions following feeding may be a good means of evaluating tolerance to the hairy chinch bug.

Experiment 4:--It appeared from Experiment 3, that the greatest differences between uninfested and infested plants occurred between infestation rates of 1 and 2 adults/plant. Thus, it was decided that 1.5 adults/plant would give enough feeding pressure to manifest tolerance without applying so much as to mask any expression of tolerance. On the basis of percent reduction in regrowth, 'Bonnieblue' was significantly more tolerant than 'Kenblue', 'Ram I', 'A-34', 'Geronimo', 'Newport', 'Campina' and 'Adelphi' (Table 8). 'Bonnieblue' expressed significantly less yield loss than 'Campina'.

				Root	
Entry	Regrowth cm	Yield mg	% Dry Matter	Length	Weight
Bonnieblue	-72.6 c	-76.1 b	+116.2 ab	-30.7 ab	-31.0 bc
Delta	-75.6 bc	-78.7 ab	+133.8 abc	-29.7 ab	-20.7 c
Troy	-81.1 abc	-77.8 ab	+45.5 a	-41.5 ab	-36.4 abc
Park	-83.1 abc	-83.2 ab	+141.7 abc	-33.9 ab	-38.9 abc
Parade	-85.2 abc	-87.0 ab	+171.7 abc	-50,9 ab	-46.2 abc
Kenblue	-87.5 ab	-88.2 ab	+122.6 ab	-45.4 ab	-46.7 abc
Ram I	-88.2 ab	-89.5 ab	+219.2 bc	-49.0 ab	-49.8 ab
A-34	-88.4 ab	-86.6 ab	+168.8 abc	-18.7 b	-32.5 abc
Geronimo	-88.6 ab	-87.4 ab	+128.0 ab	-56.3 a	-59.0 a
Newport	-88.7 ab	-82.3 ab	+143.0 abc	-42.7 ab	-45.2 abc
Campina	-90.2 a	-92.2 a	+170.5 abc	-52.4 ab	-49.1 ab
Adelphi	-91.6 a	-88.3 ab	+263.3 c	-43.8 ab	-37.2 abc

<sup>2</sup>Means for a given variable followed by the same letter are not significantly different at the 5% level - DMR.

There was considerable variation in percent dry matter data and little significant differences among cultivars. However, 'Troy' showed the least effect of feeding on changes in percent dry matter and was significantly more tolerant than 'Ram I' and 'Adelphi'. 'Bonnieblue' and 'Delta' showed significantly less percent reduction in root weight than 'Geronimo'.

Results of the 4 tests demonstrated that certain criteria were more useful in determining tolerance than others. There were significant differences in regrowth, yield and percent dry matter in almost all tests, while little or no differences in tillering, root length, and plant survival in any of the tests. A comparison of the more useful criteria for determining tolerance indicated, in general, that the responses of the cultivars from test to test were consistent (Tables 9-11). A comparison of the percent reduction in regrowth criterion for the tests showed no significant differences between cultivars in any of the tests (Table 9). The cultivars, however, responded in a relatively similar manner throughout the tests. A comparison of percent reduction differences in yield showed 'Newport' in the 1st test being significantly different than 'Adelphi' and 'SDC', but in the remaining tests there were no significant differences (Table 10). The cultivars, however, again responded in a relatively similar manner throughout the tests. A comparison of percent differences in dry matter data showed greater variation but 'SDC' and 'Adelphi' showed fairly consistent results (Table 11). 'Fylking' and 'SDC' through the 3 tests, were never significantly different from each other, despite differences in values between tests. Under similar conditions one would expect that these cultivars would respond

	% reduction from uninfested checks in tests				
Entry	I	II	III		
Newport	-71.0 a	-66.6 a			
Fylking	-80.1 a	-64.2 a	-79.9 a		
Baron	-80.9 a	-64.3 a			
S.D.C.	-82.0 a	-66.2 a	-83.3 a		
Adelphi	-85.5 a	-66.6 a			

Table 9. A comparison of data for the criterion 'regrowth' over tests  $^{1,2}$ .

<sup>1</sup>Density level 2 adults/plant.

 $^2\mathrm{Means}$  for a given variable followed by the same letter are not significantly different at the 5% level - DMR.

	% reduction fr	% reduction from uninfested checks in tests				
Entry	I	II	III			
Newport	-54.0 a	-82.9 a				
Baron	-73.0 ab	-79.1 a				
Fylking	-74.6 ab	-81.0 a	-74.3 a			
Adelphi	-77.0 b	-84.7 a				
S.D.C.	-77.7 b	-84.8 a	-89.5 a			

Table 10. A comparison of data for the criterion 'yield' over tests  $^{1,2}$ .

<sup>1</sup>Density level 2 adults/plant.

 $^2{\rm Means}$  for a given variable followed by the same letter are not significantly different at the 5% level - DMR.

	% reduction	% reduction from uninfested checks in tests				
Entry	I	II		III		
Fylking	+60.7	a +46.6	ab	+115.1 b		
Baron	+67.0	ab +25.1	a			
S.D.C.	+71.7	ab +60.3	ab	+69.7 ab		
Newport	+75.2	ab +54.1	ab			
Adelphi	+113.7	b +131.3	b			

Table 11. A comparison of data for the criterion 'percent dry matter' over tests  $^{1,2}$ .

<sup>1</sup>Density levels of 2 adults/plant.

<sup>2</sup>Means for a given variable followed by the same letter are not significantly different at the 5% level - DMR.

in a consistent manner. Thus, it would appear that tolerance to chinch bug feeding might be consistently measured on the basis of reduction in regrowth and yield and possibly increase in percent dry matter.

## Preference Studies

Area Studies 1 and 2:--Results of Area Studies 1 and 2 are presented in Table 12. Results for Study 1 were pooled for 3 days since there were no significant interactions between sampling time and adult response. The results of the 2 tests are somewhat conflicting. In Study 1 adults responded significantly more to the 2 smaller areas, while in Study 2 they preferred the 3 larger areas. This would indicate that more experiments need to be conducted on other possible influential and significant factors such as aggregating pheromones, tuft micro-environment or density factors. However, despite these conflicting results, I decided to use the largest area because it would be easier to establish and handle plugs from the field that were to be screened in the laboratory.

Species Study:--Results are presented in Table 13. There were no significant differences in response of adults to cultivars resulting from growth in the laboratory or field prior to testing. The following discussion will be based on the analysis of combined data from the laboratory and field (column 3, Table 13). Adult response was significantly greater to red fescue than to perennial ryegrass, tall fescue or wheat stubble. Also, the response to red fescue, bentgrass and Kentucky bluegrass was significantly greater than to tall fescue or wheat stubble. These results may indicate,

	<pre># of adults pe wt. of plant m</pre>		
Area (cm <sup>2</sup> )	11	2 <sup>2</sup>	
6.27	0.16 a	0.26 ab	
3.240	0.15 a	0.31 a	
1.620	0.18 a	0.21 ab	
0.790	0.19 a	0.14 bc	
0.384	0.29 b	0.01 c	
0.196	0.32 b	0.06 c	

Table 12. The mean response of adult hairy chinch bugs to six different areas of Lolium perenne var Manhattan.

<sup>1</sup>Mean of 24 replications.

 $^{2}Mean$  of 10 replications.

<pre># of adults per mg of dry wt. of</pre>				
Lab <sup>2</sup>	Field	Combined		
0.124 a	0.105 ab	0.114 a		
0.079 ab	0.129 a	0.104 ab		
0.120 a	0.057 bc	0.088 ab		
0.036 bc	0.090 ab	0.063 bc		
0.073 ab	0.014 c	0.043 c		
0.000 c	0.000 c	0.000 d		
	0.124 a 0.079 ab 0.120 a 0.036 bc 0.073 ab	0.124 a       0.105 ab         0.079 ab       0.129 a         0.120 a       0.057 bc         0.036 bc       0.090 ab         0.073 ab       0.014 c		

Table 13. The mean response of adult hairy chinch bugs to five species of laboratory and field grown cool season turfgrasses.  $^{\rm l}$ 

<sup>1</sup>Area 6.27 cm<sup>2</sup>.

<sup>2</sup>Mean of 8 replications.

in general, that the chinch bug adults preferred the narrower bladed species of grass, i.e., red fescue, bentgrass and Kentucky bluegrass, to the wider bladed species; i.e., perennial ryegrass and tall fescue. In this test the leaf width of perennial ryegrass was intermediate between the narrower leafed species and tall fescue. This may have resulted in the somewhat intermediate response of adults to perennial ryegrass. The density of the grass within an area may play a role in preference. Adults may prefer the narrower bladed species because there are more plants per area thus providing better shelter or a more favorable micro-environment. An aggregating pheromone may also be influencing the preference, especially if density and micro-environment are initially important. The response of the adults to all grasses was significantly higher than to wheat stubble, indicating that adults were responding to the live material. Thus, it appears the adults were responding to the live material and generally toward the narrower bladed turfgrass species.

## CONCLUSION

Hairy chinch bugs reared on corn sections by the procedures described were similar enough in development rates, reproductive capacity and longevity to field collected insects to warrant use of this method for rearing colonies for plant resistance studies. The method proved to be superior to rearing insects on rooted plants, because of the ease of retrieval and access of any given stage for test purposes. The availability of a satisfactory laboratory rearing method enables year-round evaluations of grasses for resistance, with reduced dependence on natural populations as a source of test insects.

Measurement of injury caused by chinch bug feeding by the method described for evaluating plant tolerance demonstrated the significant effect feeding had on plant regrowth, yield and root development. The differences in tolerance expressed by some of the Kentucky bluegrass cultivars may be indicative of resistance present in other lines or species of turfgrass, and emphasizes the need for continuing research to identify germplasm with increased tolerance. A sufficient level of tolerance in grass cultivars could reduce injury sustained by plants from chinch bug feeding without subjecting insects to types or levels of resistance that could ultimately result in selection of resistant insect biotypes. However, the nature of factors contributing to the tolerance expressed by some Kentucky bluegrass cultivars needs to be studied further.

The preference technique described herein indicates many factors may be contributing to the insects' selection of grasses for

shelter, food or oviposition. Among those could be grass morphology, density, and micro-environment, or the influence of chinch bug aggregating pheromones. However, these initial studies may prove valuable as part of a foundation for further research to develop methods for identifying and selecting for nonpreference resistance in cool season grasses.

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