Preferences of Japanese Beetle and Southern Masked Chafer (Coleoptera: Scarabaeidae) Grubs Among Cool-Season Turfgrasses[1]

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ABSTRACT Feeding preferences of Japanese beetle, *Popillia japonica* Newman, and southern masked chafer, *Cyclocephala lurida* Bland, grubs for six common cool-season turfgrasses were evaluated in choice tests in the greenhouse. On the basis of larval distributions, *Popillia japonica* consistently preferred perennial ryegrass, *Lolium perenne* L., over all other turfgrasses tested. In contrast, *C. lurida* showed no consistent pattern of preference. Presence of one grub species did not affect distribution of the other species. Grubs did not discriminate between tall fescue, *Festuca arundinacea* Schreb., infected with the endophyte *Acremonium coenophialum* Morgan-Jones & Gams and endophyte-free tall fescue.

KEY WORDS Insecta, Turfgrass, *Popillia japonica, Cyclocephala lurida.*

Larvae of the Japanese beetle, *Popillia japonica* Newman, feed on roots of a wide variety of garden and field crops, ornamental plants, and grasses (Fleming 1972, Tashiro 1987). Japanese beetle larvae are believed to prefer tender grasses to tougher grass varieties and other plants (Fleming 1972). Larvae of the southern masked chafer, *Cyclocephala lurida* Bland (formerly *C. immaculata* Olivier), also feed on roots of various grasses (Tashiro 1987, Potter et al. 1992). Both species, commonly known as white grubs, are serious pests of cool-season turfgrasses in the northeastern and midwestern United States (Tashiro 1987).

Host preferences of root-feeding scarabs have been evaluated in other plant systems and may be important for pest management. Using 3-month-old pasture plants, Wensler and Dudzinski (1971) found that grubs of *Sericesthis geminata* Boisduval, an Australian species, preferred perennial ryegrass (*Lolium perenne* L.) and orchard grass (*Dactylis glomerata* L.) over a canary grass (*Phalaris tuberosa* L.), and that all three grasses were preferred over white clover (*Trifolium repens* L.). Similar results occurred in comparisons with lyophilized roots. Roots of several pasture legumes were more attractive to the New Zealand grass grub, *Costelytra zealandica* (White), than roots of perennial ryegrass (Sutherland and Hillier 1974, Kain and Atkinson 1977). *Costelytra zealandica* did not discriminate between roots of susceptible and resistant legume species,

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but were highly attracted to alfalfa (*Medicago sativa* L.) and lotus (*Lotus pedunculatus* Cav.), plants previously shown to inhibit larval growth and survival (Farrell and Sweney 1972, Sutherland et al. 1975a, 1975b). Kelsey (1967) reported no differences in numbers of *C. zealandica* among four perennial ryegrass varieties in the field. Resistance of oat (*Avena sativa* L.), wheat (*Triticum aestivum* L.), and barley (*Hordeum vulgare* L.) cultivars to *Phyllophaga congrua* (LeConte) was attributed to non-preference (Crocker et al. 1990).

In Kentucky, populations of *P. japonica* and *C. lurida* grubs often occur together. *Popillia japonica* are highly aggressive, and under crowded experimental conditions may injure or kill one another by biting (Fleming 1963, Régnière et al. 1981). By comparison, *C. lurida* are much less aggressive (Potter, unpublished data). Interactions between the two grub species have not been studied.

All common cool-season turfgrasses are suitable for feeding and growth of *P. japonica* and *C. lurida* grubs (Potter et al. 1992); however, whether the grubs prefer roots of particular grasses is unknown. Below-ground herbivory can cause dramatic changes in composition of mixed-grass pasture swards (King and East 1979), so selective feeding by one or both grub species could affect the composition of mixed-species turfgrass stands.

Infection of tall fescue, *Festuca arundinacea* Schreb., by the endophytic fungus *Acremonium coenophialum* Morgan-Jones & Gams (Clavicipitaceae: Ascomycetes) conveys varying degrees of resistance to many leaf, stem, and phloem-feeding insects (Breen 1994). There is mixed evidence that white grubs may be adversely affected by *A. coenophialum* (Potter et al. 1992, Murphy et al. 1993), but whether grubs discriminate between endophyte-infected (EI) and endophyte-free (EF) tall fescue is unknown. The objectives of this study were to investigate feeding preferences of *P. japonica* and *C. lurida* grubs for different cool-season turfgrasses and for EI versus EF tall fescue, and to determine if the distribution of one grub species is altered by the presence of the other species.

Materials and Methods

Four greenhouse preference trials were conducted from spring 1991 to fall 1992. Aluminum pans (33.0 × 22.9 × 5.1 cm), containing different turfgrass species in various free-choice arrangements, comprised the basic experimental units in each trial. Kentucky bluegrass, *Poa pratensis* L., was used as a standard because it is the most widely planted, general purpose turf and lawn grass in the United States (Tashiro 1987). Turfgrasses were watered as needed and clipped weekly to 6.4 cm height. Field-collected, third instar *P. japonica* and *C. lurida* were used in all trials. Grubs used in Trials 1 and 2 were collected from Kentucky bluegrass; grubs in Trial 3 were from mixed Kentucky bluegrass and tall fescue, while those used in Trial 4 were from tall fescue. Grubs were held in soil for <24 h before being used in experiments.

Trial 1. Paired comparisons with mature turfgrass sod. Sections of mature, established, turfgrass sod (22.9 × 16.5 × 5.1 cm) were obtained from adjacent field plots on a common Maury silt loam soil located at the University of Kentucky's Spindletop Research Farm, near Lexington, KY. The plots had been fertilized twice each fall with 37 kg nitrogen/ha from urea. Two sod sections, each from a different turfgrass species, were installed in each aluminum pan on 11 March 1991. Sheet-metal dividers were inserted between paired sod sections to segregate root growth. Six different comparisons were established. Kentucky bluegrass cultivar 'Kenblue', was paired with each of five other turfgrasses: EI or EF tall fescue, 'Chiefton'; hard fescue, *F. ovina* L. variety duriuscula, 'SR3000'; perennial ryegrass, 'Derby'; and creeping bentgrass, *Agrostis palustris* (Huds.), 'Penncross.' Another two-choice test compared response of grubs to EI and EF tall fescue. Rates of endophyte infection were tested by tissue immunoblot assay (Gwinn et al. 1991), and found to be 4 and 96% in EF and EI tall fescue, respectively.

Metal dividers were removed on 12 April 1991 and *P. japonica* or *C. lurida* were placed separately (20 grubs per pan), or in combination (10 grubs each per pan) along the seam between the sod sections. Grubs that had failed to burrow into the turf within 10 min were replaced with fresh grubs. Treatment combinations (18 total) were arranged in a randomized complete block design across greenhouse benches, from north to south, to account for variation in natural sunlight. There were six replications of each comparison. Greenhouse temperatures ranged from 20 to 30°C, and natural light was supplemented with light from sodium vapor lamps to provide a 16 h photoperiod.

Metal dividers were re-inserted after 1 wk and the number of grubs in each sod section was determined. Dead grubs and those injured by the divider were not included in the analysis.

Trial 2. Paired comparisons with seedling turfgrasses. Pans were filled to 3.8 cm height with a mixture of top soil, peat moss, and sand (4:1:1 ratio by volume). Metal dividers were placed into each pan to make two separate subcompartments of equal area (22.9 × 16.5 cm). Turfgrass seeds (1.2 ml) were applied to the soil surface in each subcompartment and watered daily to ensure germination and development. Four different comparisons were established: Kentucky bluegrass, 'Kenblue', paired with tall fescue, 'Kentucky 31' (EF), hard fescue, 'Aurora', perennial ryegrass, 'Derby', or creeping bentgrass, 'Penncross'. There were six replications for each comparison. Seedling grasses were planted 13 August 1992 and allowed to establish for 6 wk. Metal dividers were then removed, and *P. japonica* or *C. lurida* grubs (20 per pan) were placed into the seam between turfgrasses on 23 September 1992. Treatment combinations (eight total) were arranged and evaluated as in Trial 1.

Trial 3. Response of grubs to paired turfgrass plugs. Plastic pots were filled with soil and seeded with individual turfgrasses (0.6 ml seed per pot), using the same soil mixture, grass species, and cultivars as in Trial 2. Seed were planted on 16 July 1991. After 7 wk of growth, resulting turfgrass plugs (10.3 cm diam, 3.8 cm deep) were removed from pots and transplanted into pans filled with the soil mixture. Plugs of two different turfgrasses were installed 6.4 cm apart in each pan. Four and three comparisons were established for *P. japonica* and *C. lurida,* respectively. For *P. japonica,* perennial ryegrass was paired with Kentucky bluegrass, creeping bentgrass, and tall fescue. We also compared grub preference between Kentucky bluegrass and tall fescue. For *C. lurida,* tall fescue was paired with Kentucky bluegrass, hard fescue, and creeping bentgrass. Each comparison was replicated seven times. Grubs (20 per pan) were placed in a furrow between the two plugs and covered with moist soil on 12 September 1991. Treatment combinations (seven total) were arranged as described for Trial 1. Numbers of larvae within the root zone of each turfgrass plug, or in the remaining soil were determined after 1 wk.

Trial 4. Choice tests with all grass species. Plastic pots were filled with the same soil mixture used in Trial 2 and seeded with individual turfgrass species on 7 March 1992. Resulting turfgrass plugs (5.1 × 5.1 × 3.8 cm) were removed from pots after 5 wk and transplanted into pans filled with the soil mixture. Grasses included Kentucky bluegrass, 'Kenblue', hard fescue, 'Aurora', creeping bentgrass, 'Penncross', perennial ryegrass, 'Derby', and tall fescue, 'Kentucky 31'. We had intended to include both EF and EI tall fescue of the same genotype in the experiment, but discovered later that the seed believed to have been EI was actually EF. Thus, there were two EF tall fescue plugs grown from different seed lots within each pan. The six plugs were randomized and positioned 5.1 m apart and 3.8 cm from the sides of the pan. Pans were infested with either *P. japonica* or *C. lurida* (30 grubs per pan) on 14 April 1992. Grubs were evenly distributed within each pan and allowed to burrow into the soil. There were eight replications (pans) for each grub species. After 2 wk, the soil in each pan was divided into six equal regions (11.2 × 11.2 cm), with plugs occupying the center of each region. Numbers of grubs within the root zone of each turfgrass plug, and within each region (including the plug) were counted.

Data Analyses. In Trials 1-3, numbers of grubs found within each turfgrass were pooled across replicates, and data were then analyzed using a Chi-square goodness of fit test for significant departure from an equal (50:50) distribution. In Trial 4, numbers of grubs within each turfgrass plug or region were pooled within each treatment, and data were analyzed using a Chi-square goodness of fit test (single classification with more than two classes) for significant departure from an equal distribution (Steel and Torrie 1960).

Results

Popillia japonica introduced into pans containing mature turfgrass sod showed significant preference for perennial ryegrass and creeping bentgrass over Kentucky bluegrass (Table 1). *Cyclocephala lurida* preferred Kentucky bluegrass over either EF or EI tall fescue. Neither grub species discriminated between EF and EI tall fescue nor between any other combination of turfgrass species. Similar results were obtained when *P. japonica* and *C. lurida* were evaluated in combination (Table 2), except that significantly more *C. lurida* were found in creeping bentgrass when tested with Kentucky bluegrass.

In choice tests with turfgrass seedlings, *P. japonica* again significantly preferred perennial ryegrass over Kentucky bluegrass (Table 3), but did not discriminate in other turfgrass comparisons. The distribution of *C. lurida* was not significantly different from equality in any comparison between grasses.

Numbers of *P. japonica* were significantly greater in perennial ryegrass seedling plugs than in Kentucky bluegrass or creeping bentgrass (Table 4), and grubs also significantly preferred tall fescue over Kentucky bluegrass. Grubs of *C. lurida* were significantly more abundant in tall fescue than in creeping bentgrass plugs.

Table 1. Distribution of third instar *P. japonica* or *C. lurida* held separately for 1 wk in pans containing two different mature, field-collected, turfgrass sod sections (Trial 1).

Turfgrass comparison*	*P. japonica* Grubs per turfgrass**	*P. japonica* x^2 value†	*C. lurida* Grubs per turfgrass	*C. lurida* x^2 value
KB : TF(EI)	54 : 45	0.82	72 : 36	12.00[b]
KB : TF(EF)	51 : 60	0.73	73 : 27	21.16[b]
KB : HF	55 : 50	0.24	56 : 52	0.15
KB : PR	34 : 66	10.24[b]	48 : 56	0.62
KB : CB	41 : 64	5.04[a]	43 : 60	2.81
TF(EI) : TF(EF)	53 : 60	0.43	51 : 53	0.04

* KB = Kentucky bluegrass, TF(EI) = tall fescue (endophyte-infected), TF(EF) = tall fescue (endophyte-free), HF = hard fescue, PR = perennial ryegrass, CB = creeping bentgrass.
** Total number of grubs per turfgrass pooled from six replications of each comparison.
† x^2 goodness of fit test for departure from an equal distribution ($^aP < 0.05$, $^bP < 0.01$, df = 1).

Table 2. Distribution of third instar *P. japonica* or *C. lurida* held collectively for 1 wk in pans containing two different mature, field-collected, turfgrass sod sections (Trial 1).

Turfgrass comparison*	*P. japonica* Grubs per turfgrass**	*P. japonica* x^2 value†	*C. lurida* Grubs per turfgrass	*C. lurida* x^2 value
KB : TF(EI)	33 : 25	1.10	46 : 8	26.74[b]
KB : TF(EF)	25 : 32	0.86	42 : 11	18.13[b]
KB : HF	22 : 25	0.19	23 : 32	1.47
KB : PR	20 : 35	4.09[a]	28 : 27	0.02
KB : CB	18 : 36	6.00[a]	17 : 32	4.59[a]
TF(EI) : TF(EF)	21 : 31	1.92	21 : 30	1.59

* KB = Kentucky bluegrass, TF(EI) = tall fescue (endophyte-infected), TF(EF) = tall fescue (endophyte-free), HF = hard fescue, PR = perennial ryegrass, CB = creeping bentgrass.
** Total number of grubs per turfgrass pooled from six replications of each comparison.
† x^2 goodness of fit test for departure from an equal distribution ($^aP < 0.05$, $^bP < 0.01$, df = 1).

Table 3. Distribution of third instar *P. japonica* or *C. lurida* held separately for 1 wk in pans containing 6-wk-old seedlings of two different turfgrasses (Trial 2).

	P. japonica		*C. lurida*	
Turfgrass comparison*	Grubs per turfgrass**	x^2 value†	Grubs per turfgrass	x^2 value
KB : TF(EF)	33 : 41	0.87	43 : 40	0.11
KB : HF	34 : 50	3.05	49 : 36	1.99
KB : PR	31 : 58	8.19[b]	35 : 50	2.65
KB : CB	40 : 40	0.00	32 : 41	1.11

* KB = Kentucky bluegrass, TF(EF) = tall fescue (endophyte-free), HF = hard fescue, PR = perennial ryegrass, CB = creeping bentgrass.
** Total number of grubs per turfgrass pooled from six replications of each comparison.
† x^2 goodness of fit test for departure from an equal distribution ($^bP < 0.01$, df = 1).

Table 4. Distribution of third instar *P. japonica* or *C. lurida* after 1 wk in pans containing two different 7-wk-old turfgrass plugs grown from seed (Trial 3).

Grub species	Turfgrass comparison*	Grubs per turfgrass**	x^2 value†
P. japonica	KB : PR	11 : 33	11.00[b]
	KB : TF(EF)	10 : 32	11.52[b]
	PR : TF(EF)	32 : 19	3.31
	PR : CB	35 : 6	20.51[b]
C. lurida	TF(EF) : KB	19 : 17	0.11
	TF(EF) : HF	16 : 22	0.95
	TF(EF) : CB	23 : 5	11.57[b]

* KB = Kentucky bluegrass, TF(EF) = tall fescue (endophyte-free), HF = hard fescue, PR = perennial ryegrass, CB = creeping bentgrass.
** Total number of grubs per turfgrass pooled from six replications of each comparison.
† x^2 goodness of fit test for departure from an equal distribution ($^bP < 0.01$, df = 1).

In comparisons with all six turfgrasses, *P. japonica* were significantly more abundant in perennial ryegrass plugs than in the other turfgrasses tested (Table 5). In addition, numbers of grubs were higher in the region around and including the perennial ryegrass plugs than in all other regions. In contrast, grubs of *C. lurida* were equally distributed among turfgrass plugs or regions. *Popillia japonica* were equally distributed among turfgrass plugs (x^2 = 3.24, df = 4) or regions (x^2 = 5.24, df = 4) when perennial ryegrass was removed from the statistical analysis.

Table 5. Distribution of third instar *P. japonica* or *C. lurida* after 2 wks in pans containing six different 5-wk-old turfgrass plugs grown from seed (Trial 4).

	P. japonica		*C. lurida*	
Turfgrass*	Grubs per plug**	Grubs per region†	Grubs per plug	Grubs per region
KB	12	26	8	22
TF(EF)1	14	33	10	25
TF(EF)2	13	25	9	26
HF	18	35	11	26
PR	29	44	15	36
CR	9	20	14	31
x^2 value‡	15.84[a]	12.11[a]	3.48	4.53

* KB = Kentucky bluegrass, TF(EF)1 = tall fescue (endophyte-free) seed lot 1, TF(EF)2 = tall fescue (endophyte-free) seed lot 2, HF = hard fescue, PR = perennial ryegrass, CB = creeping bentgrass.

** Total number of grubs per turfgrass plug pooled from eight replications.

† Total number of grubs per turfgrass plug and region pooled from eight replications.

‡ x^2 goodness of fit test for departure from an equal distribution ([a]$P < 0.05$, df = 5).

Discussion

Popillia japonica consistently preferred perennial ryegrass over other cool-season turfgrasses. The only possible exception was in Trial 3 (Table 4), where the preference of grubs for perennial ryegrass over tall fescue approached significance ($P < 0.07$). In contrast, *C. lurida* showed no consistent pattern of host preference.

Popillia japonica preferred mature creeping bentgrass sod over mature Kentucky bluegrass. Likewise, grubs of *C. lurida* preferred Kentucky bluegrass to tall fescue sod. These preferences, however, were not apparent in trials with turfgrass seedlings. It is possible that turfgrass age may influence preference by scarabaeid larvae. For example, third-instar *S. geminata* demonstrated a reversal in preference between 3- and 9-month-old pasture plant species (Wensler and Dudzinski 1971).

We had hypothesized that interspecific antagonistic interactions might affect distribution of *C. lurida* more than *P. japonica*. Presence of one white grub species, however, did not appear to alter behavior of the other species. Distributions of both *P. japonica* and *C. lurida* were consistent whether the species were tested individually or in combination. The only exception was in the comparison between Kentucky bluegrass and creeping bentgrass, where *C. lurida* showed no preference when tested alone, but preferred creeping bentgrass when tested with *P. jaonica*. In the latter case, both grub species preferred creeping bentgrass and occupied the same area.

White grubs did not distinguish between EI and EF tall fescue sod. In laboratory studies, endophyte-associated alkaloids deterred feeding by *P. japonica* on agar-based medium (Patterson et al. 1991). However, in no-choice tests, both *P. japonica* and *C. lurida* consumed similar amounts of EI and EF root tissue (Potter et al. 1992).

Use of feeding preferences of root-feeding white grubs may have limited application for pest management because grubs can survive on all cool-season turfgrasses tested (Potter et al. 1992), and because larval distributions are determined mainly by selection of oviposition sites by adult females. However, if white grubs preferentially feed on certain turfgrasses, they could exert strong selection on turfgrass mixtures and affect the long-term composition of turfgrass stands.

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