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MELANOTRICHUS BOYDI (HEMIPTERA: MIRIDAE) IS A SPECIALIST ON THE NICKEL HYPERACCUMULATOR *STREPTANTHUS POLYGALOIDES* (BRASSICACEAE)

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ABSTRACT—Nickel hyperaccumulator plants contain at least 1,000 μg Ni/g dry mass. Their high-Ni tissues present a chemically unique resource for herbivorous insects. A prior survey of insects associated with the Ni hyperaccumulator *Streptanthus polygaloides* in California yielded a new species, *Melanotrichus boydi* Schwartz and Wall (Hemiptera: Miridae). Here we document the host preference of *M. boydi.* Surveys of 10 *S. polygaloides* populations across its geographical and elevational ranges documented the presence of *M. boydi* upon *S. polygaloides* at all sites. A host-choice experiment contrasting *M. boydi* with the polyphagous mirid *Lygus hesperus* (both collected from *S. polygaloides*) showed *M. boydi* preferred *S. polygaloides* but *L. hesperus* did not. Other host-choice studies showed *M. boydi* preferred *S. polygaloides* to 2 other ultramafic soil plant species in the Brassicaceae, including another species of *Streptanthus.* No preference was observed when *M. boydi* was offered samples from several populations of *S. polygaloides* or given a choice between high-Ni and low-Ni *S. polygaloides* plants. These results show that *M. boydi* feeds primarily on *S. polygaloides* but lacks population-level specificity. This is the first report of a North American insect species monophagous on a Ni hyperaccumulator species.

RESUMEN—Las plantas hiperacumuladoras de níquel contienen al menos 1,000 μg Ni/g de peso seco. Sus tejidos con altas concentraciones de Ni representan un recurso químico único para los insectos herbívoros. Un estudio previo de los insectos asociados a la especie hiperacumuladora de Ni *Streptanthus polygaloides* en California condujo al hallazgo de una nueva especie, *Melanotrichus boydi* Schwartz y Wall (Hemiptera: Miridae). En el presente trabajo se documenta la preferencia en cuanto a hospedero de *M. boydi.* La investigación en diez poblaciones de *S. polygaloides* a lo largo de su distribución geográfica y altitudinal documentó la presencia de *M. boydi* en todos los sitios donde *S. polygaloides* crecía. Un experimento de selección de hospedero contrastando *M. boydi* con la chinche ligus *Lygus hesperus* (ambos colectados en *S. polygaloides*) mostró que *M. boydi* prefirió *S. polygaloides* mientras que *L. hesperus* no. Otros estudios de selección evidenciaron que *M. boydi* prefirió *S. polygaloides* antes que otras dos especies de Brassicaceae de suelos ultramáficos, incluyendo otra especie de *Streptanthus.* No se observó preferencia cuando a *M. boydi* se le ofrecieron muestras de varias poblaciones de *S. polygaloides* ni cuando se le dio a escoger entre plantas de *S. polygaloides* con concentraciones altas o bajas de Ni. Estos resultados muestran que *M. boydi* se alimenta primariamente de *S. polygaloides* pero carece de especificidad a nivel poblacional. Este constituye el primer registro de un insecto norteamericano monófago de una especie hiperacumuladora de Ni.

The chemical composition of ultramafic soils creates a unique habitat in which organisms can evolve (Kruckeberg, 1992). Although ultramafic soils comprise only 1% of the California Floristic Province (CFP), almost 10% of the endemic plant species in the CFP are restricted to these soils (Kruckeberg, 1992). The high level of floristic endemism on ultramafic soils often is attributed to the high concentrations of heavy metals and low concentrations of calcium in these soils (Proctor and Woodell, 1975; Nagy and Proctor, 1997). Many authors have hypothesized that a corresponding unique fauna should also occur on serpentine soils (e.g., Proctor and Woodell, 1975; Kruckeberg, 1984; Brooks, 1987), but little research has investigated this question.

Nickel hyperaccumulators are unique plant species associated with ultramafic soils. Tissues of Ni hyperaccumulators contain over 1,000 μg Ni/g on a dry-mass basis (Brooks et al., 1977). Among the hypotheses advanced to explain the evolution of metal hyperaccumulation (Boyd and Martens, 1992), the most widely supported hypothesis proposes that hyperaccumulation serves as an ''elemental defense'' against herbivores (Boyd and Martens, 1998; Boyd, 2004). Because no plant defense is effective against all herbivores (Gatehouse, 2002; Karban and Agrawal, 2002), some herbivores might have evolved counter defenses to high-Ni plant tissues (Boyd, 1998). Pollard (2000) suggested that Ni hyperaccumulator-herbivore systems are ideal for studies of plant-insect coevolution, but, as of yet, little research has been conducted on native herbivorous insects and hyperaccumulator plants.

To our knowledge, the most extensive research on native herbivorous insects of a Ni hyperaccumulator has been conducted in South Africa, where at least 7 species of insects were determined to feed on *Berkheya coddii* (Asteraceae) (Mesjasz-Przybylowicz and Przybylowicz, 2001). Further study of this system found additional insect associates, many of which carry relatively high concentrations of Ni (Boyd et al., 2006). Choice tests using one species, *Chrysolina pardalina* (Coleoptera: Chrysomelidae), showed it was apparently monophagous on *B. coddii* (Augustyniak et al., 2002). To our knowledge, no investigations have been made using herbivores of any other Ni hyperaccumulator species, despite the occurrence of Ni hyperaccumulators in many other parts of the world (Brooks, 1998; Reeves, 2003).

In North America, Wall and Boyd (2002) surveyed arthropods associated with *Streptanthus polygaloides* (Brassicaceae), the only Ni hyperaccumulator species endemic to California ultramafic soils (Kruckeberg and Reeves, 1995). A previously undescribed species, *Melanotrichus boydi* Schwartz and Wall (Hemiptera: Miridae), was discovered that was exceptional in containing over 700 μg Ni/g dry mass on a whole body basis (Schwartz and Wall, 2001). Schwartz and Wall (2001) suggested it was monophagous on *S. polygaloides*. The objective of our current study was to determine the host preference of *M. boydi* by using field surveys and choice experiments. We used a hierarchical design of food-preference experiments that allowed us to distinguish the taxonomic level at which *M. boydi* recognizes potential hosts. First, we compared the feeding preference of *M. boydi* to that of *Lygus hesperus*, a broadly polyphagous mirid (Scott, 1977), by using ultramafic plant species from different families. We then explored feeding preferences of *M. boydi* at successively lower taxonomic levels.

METHODS—*Study Species*—*Streptanthus polygaloides* is a winter annual endemic to ultramafic soils on the western slope of the Sierra Nevada in California, ranging from Butte County in the north to Fresno County in the south (Fig. 1; Table 1) (Kruckeberg, 1984). Throughout its range, *S. polygaloides* is restricted to ultramafic soils and hyperaccumulates Ni (Kruckeberg and Reeves, 1995). Like other western species of *Streptanthus* (Mayer et al., 1994), *S. polygaloides* is phenotypically variable across its range (Table 1). For instance, sites 1, 4, 5, 6, and 9 (Fig. 1 and Table 1) contain morphologically similar plants that produce yellow sepals, whereas sepals of *S. polygaloides* from sites 2, 3, and 8 are purple. *Streptanthus polygaloides* from site 7 is unique; although the flowers are morphologically similar to the above populations, sepals of young unopened flowers are yellow and turn purple upon anthesis. Finally, at site 10, *S. polygaloides* has yellow sepals, but the sepals of each flower have undulate margins and several large warts at the apex.

Melanotrichus boydi is morphologically similar to *M. mistus* and *M. stanleyaea*, differing from them in characteristics of the dorsal setae, antennal color and segmentation, and structure of the male genitalia (Schwartz and Wall, 2001). Species of this Holarctic genus feed primarily on plants in the Asteraceae, Brassicaceae, or Chenopodiaceae (Henry, 1991). In general, mirids use a ''lacerate and flush'' feeding technique, in which the stylets of the insect rip open a series of adjacent cells to suck up cellular material (Miles, 1972). In 3 years of observations, we have seen both nymphs and adults of *M. boydi* feeding on young leaves and flowers of *S. polygaloides*. This choice of host organs for feeding is consistent with our current understanding of the feeding habits of phytophagous Miridae (Schuh and Slater, 1995; Wheeler, 2001).

Distribution of M. boydi *on* S. polygaloides—From mid June to mid July of 1998, we visited 10 ultramafic sites (Fig. 1) in the foothills of the western Sierra Nevada within the reported geographic range of *S. polygaloides*. Ultramafic sites were located using geologic maps supplemented by information in Kruckeberg (1984) and personal communications with other ultramafic ecologists. We selected sampling sites that included most of the geographic range of *S. polygaloides* and sampled populations representing all known phenotypic variation (Table 1).

Sampling for *M. boydi* at each ultramafic site included searches on *S. polygaloides* as well as other plant species. We visually inspected flowering individuals of *S. polygaloides* or sampled stands of *S. polygaloides* for insects by sweep netting. We selected dense monospecific stands of *S. polygaloides* for sweep netting to minimize the chances of capturing insects feeding on other plant species. Other dominant plant species on these sites were sampled separately by sweep netting for *M. boydi*.

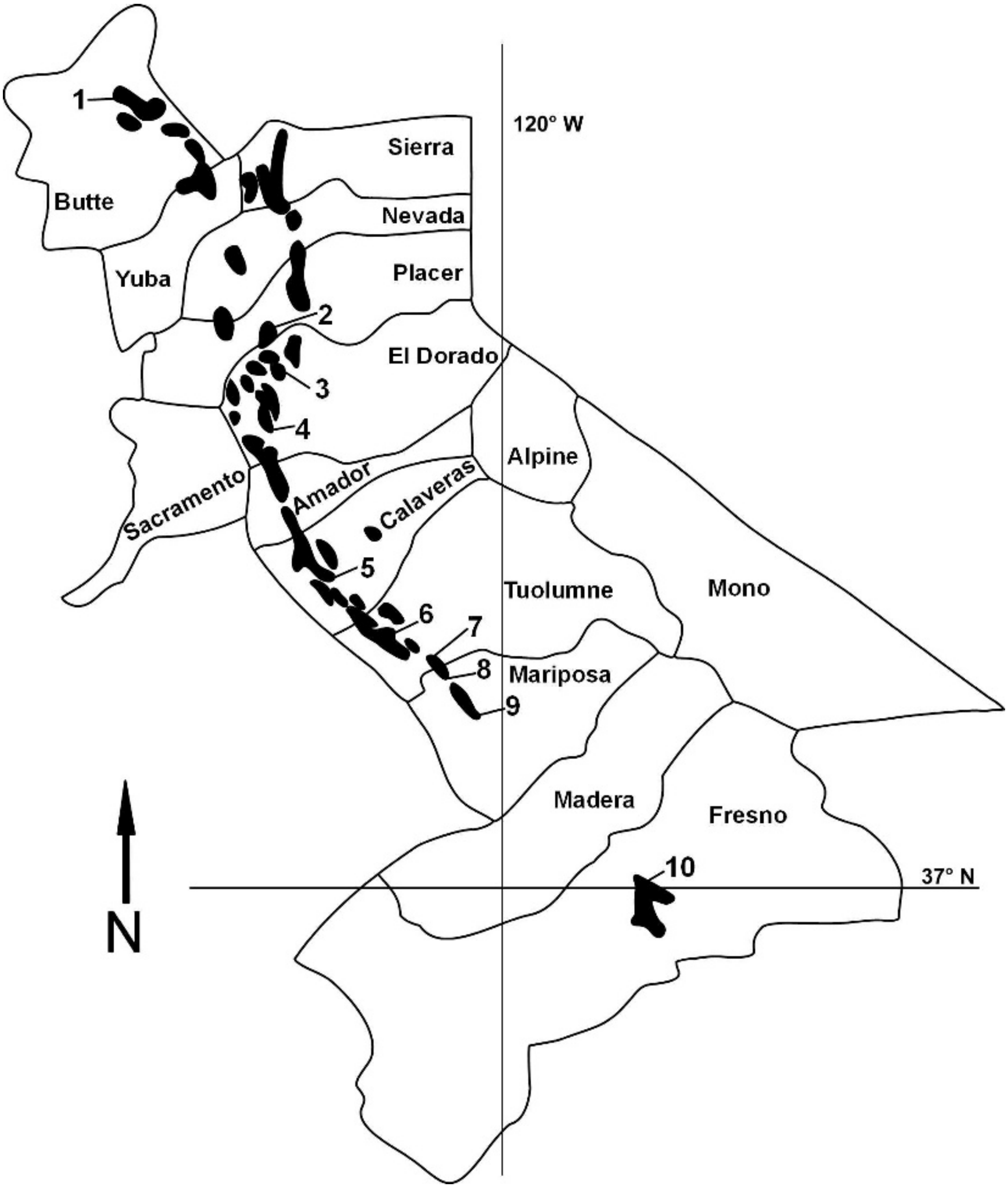

FIG. 1—Distribution of *Melanotrichus boydi* in California (modified from Wall, 1999). Areas filled in black represent distribution of ultramafic soils within the foothills of the Sierra Nevada. *Melanotrichus boydi* was either observed or collected at all numbered sites (numbers correspond to sites listed in Table 1).

Food Preference Experiments—We conducted a series of food preference tests (described below) to determine the discriminatory ability of *M. boydi*. In each experiment, insects were offered choices of food items and their feeding choices observed. For all experiments, feeding was defined as insertion of the stylets into plant tissues, and feeding preference was compared against random expectations by contingency table analysis (Abacus Concepts, 1998). Qualitative colorimetric tests in the field indicated low Ni levels (<100 µg Ni/g) in all plant species used in this study with the exception of *S. polygaloides*.

Preferences of 2 Species of Miridae Collected from S. polygaloides—We compared food preferences of

TABLE 1—California ultramafic sites (numbered on Fig. 1) and *Streptanthus polygaloides* phenotypes sampled for the presence of *Melanotrichus boydi* during June–July 1998.

Site	Location	Elevation (m)	Phenotype
1	N of Magalia, Butte County	800	Yellow sepals
2	S of Washington, Placer County	1,330	Purple sepals
3	S of Sugar Pine Reservoir, El Dorado County	1,270	Purple sepals
4	N of Coloma, El Dorado County	900	Yellow sepals
5	N of San Andreas, Calaveras County	330	Yellow sepals
6	S of Chinese Camp, Tuolumne County	470	Yellow sepals
7	SE of Moccasin, Tuolumne County	700	Young sepals yellow, becoming purple at anthesis
8	NW of Coulterville, Mariposa County	700	Purple sepals
9	Highway 49 bridge at Lake McClure, Mariposa County	900	Yellow sepals
10	NW of Pine Flat Lake, Fresno County	400	Yellow sepals with undulate margin and warty apex

M. boydi and *L. hesperus* (both collected from site 6) by using 4 plant species representing different families. We chose *Clarkia biloba* (Onagraceae), *Calycadenia multiglandulosa* (Asteraceae), *Ceanothus cuneatus* var. *cuneatus* (Rhamnaceae), and *S. polygaloides.* These species were among the most abundant plants at site 6, where the study insects were collected, and *S. polygaloides* and *C. multiglandulosa* represent 2 families (Brassicaceae and Asteraceae) on which other species of *Melanotrichus* feed (Henry, 1991).

In mid-June 1998, 30 adults of *M. boydi* and 16 adults of *L. hesperus* were collected from *S. polygaloides* at site 6. We placed each insect in an individual 10-cm diameter plastic Petri plate containing material from *S. polygaloides*, *C. biloba*, *C. multiglandulosa*, and *C. cuneatus.* Petri plates were divided into numbered quadrants, and a sample of each of the 4 plant species was randomly assigned to each quadrant. Plant material collected the same day as the insects was refrigerated in plastic bags until trials began. Only the young portions (within 1 cm of the apex) of the stems, including developing leaves, of these plants were used in the trials. *Ceanothus cuneatus* was fruiting during the time of the experiment, whereas *S. polygaloides*, *C. biloba*, and *C. multiglandulosa* were all flowering. At 12 h and 24 h, we recorded if each insect was feeding and, if so, the identity of the host.

Food Preferences of M. boydi *Collected from* S. polygaloides—Three experiments were conducted to determine if *M. boydi* had a host preference at the plant genus, species, or population levels. In the first choice experiment, *M. boydi* was offered *S. polygaloides* and *Brassica nigra.* Although it is not a native species, *B. nigra* was chosen because it was the only other annual member of the Brassicaceae in flower at the site where the insects were collected. In late June 1999, 21 adults of *M. boydi* were collected from *S. polygaloides* at site 3. Each individual was placed in the center of a 10-cm diameter plastic Petri plate. Plant material (apical 1 cm of flowering stem) from *S. polygaloides* and *B. nigra* was placed into each half of each Petri plate. Both *S. polygaloides* and *B. nigra* were collected from the same site as the insects. At 12 h and 24 h, we recorded the identity of the host for each feeding insect.

In a second choice experiment to determine the feeding preference of *M. boydi* within the genus *Streptanthus*, we tested *S. tortuosus* against *S. polygaloides.* This species was chosen because it is the only other *Streptanthus* species known to occur at sites at which *M. boydi* is present (Kruckeberg, 1984). In mid-June 1998, 10 adults of *M. boydi* were collected from *S. polygaloides* at site 7 (Table 1). A preference test was conducted in the same manner as with the generic-level experiment above. Both *S. polygaloides* and *S. tortuosus* were collected in flower from the same location (site 7), and the apical 1 cm of flowering stem of each species was used as the plant material. At 12 h and 24 h, we recorded if each insect was feeding and, if so, the identity of the host.

A third set of experiments tested *M. boydi* for population-level food preferences. The test used *S. polygaloides* populations that represented several phenotypes. We collected flowering stems of *S. polygaloides* and 6 *M. boydi* adults from each of sites 8, 9, and 10 (Table 1). Additional young flowering stems of *S. polygaloides* were collected from site 6. These 4 plant populations represented 3 phenotypes (sites 6 and 9 have yellow sepals, site 8 has purple sepals, and site 10 has yellow undulate sepals) from the southern range of *S. polygaloides* (Table 1). The terminal portion of the flowering stem (apical 1 cm) of plants from each population was randomly assigned to quarter sections within 10-cm diameter Petri plates. We then randomly assigned an individual of *M. boydi* from one of the 3 sites (sites 8, 9, and 10) to each plate. At 12 h and 24 h, individuals of *M. boydi* feeding on plants from their home site were scored as ''home site'' and those feeding on plants collected from other sites were scored as ''off site.''

The second population-level test focused on *M. boydi* collected from a single site. We collected 12 *M. boydi* from site 4 and offered them plants from that site and 3 others (sites 1, 6, and 10; Table 1). At 12 and 24 h, we recorded which plant each insect was feeding upon to test if insects from this population demonstrated a preference for plants from their ''home site.''

Effect of S. polygaloides *Ni Concentration on* M. boydi *Host Preference—Streptanthus polygaloides* is the only Ni hyperaccumulator species known from these ultramafic sites on the western slope of the Sierra Nevada (Kruckeberg and Reeves, 1995). To determine if the high levels of Ni found in *S. polygaloides* play a role in host recognition by *M. boydi*, we conducted the following test. In April 1999, seeds of *S. polygaloides* were sown on low-Ni potting soil (ProMix, Premier Horticulture, Red Hill, Pennsylvania) in a greenhouse in Auburn, Alabama. Previous experiments (Boyd and Martens, 1994; Boyd and Moar, 1999) have shown that the resulting plants contain little Ni (no more than 20 μg Ni/g dry mass). In June 1999, flowering stems of these ''low-Ni'' plants were clipped, wrapped in moist paper towels, sealed in plastic bags, and chilled in coolers for transport to California. Upon arrival in California, 21 adults of *M. boydi* and flowering stems of ''high-Ni'' plants of *S. polygaloides* were collected from site 6. Each individual *M. boydi* was placed in the center of a 10-cm diameter plastic Petri plate. Plant material from both ''high-Ni'' and ''low-Ni'' plants was placed in opposite halves of each plate. At 12, 24, and 38 h, we recorded which plant each insect was feeding on to test if insects from this population demonstrated a preference for high-Ni or low-Ni plants. The experiment began less than 48 h after ''low-Ni'' plants had been cut in Auburn.

RESULTS—*Distribution of* M. boydi *on* S. polygaloides—*Melanotrichus boydi* was associated with *S. polygaloides* throughout the range of the plant. Sampling of *S. polygaloides* at all sites yielded adults and sometimes nymphs of *M. boydi*. A taxonomically wide variety of other plant species was also sampled at these sites, including one conifer [*Pinus sabiniana* (Pinaceae)], 3 woody dicots [*Quercus durata* (Fagaceae), *C. cuneatus*, and *Eriodictyon californicum* (Hydrophyllaceae)], one herbaceous monocot (unidentified perennial grass), and 4 herbaceous dicots. The herbaceous dicots were *C. multiglandulosa* and *C. biloba*, and 2 members of the Brassicaceae, *B. nigra* and *S. tortuosus*. The plant species sampled represented a wide taxonomic range (8 families), diverse growth forms, and several of the apparently abundant species during mid June to mid July, when *M. boydi* adults and nymphs were abundant on *S. polygaloides*. Sampling of these other species did not yield *M. boydi*.

Melanotrichus boydi and *S. polygaloides* have fairly wide ecological ranges. Both plant bug and host plant were present at sites ranging in elevation from 330 m to approximately 1,330 m above sea level (Table 1). The sites differed greatly in their surrounding plant communities. Lower-elevation sites were surrounded by chaparral (e.g., site 7) or blue oak savanna (e.g., site 6), whereas higher-elevation sites were surrounded by coniferous forest (e.g., sites 2 and 3). Presence of *S. polygaloides* was key to the occurrence of *M. boydi* across a wide range of elevation and surrounding plant communities.

Food Preference Experiments: Preferences of 2 Species of Miridae Collected from S. polygaloides—*Melanotrichus boydi* and *L. hesperus* differed in host preference. Observed frequencies of *M. boydi* on the offered food plants differed significantly from expected values at both 12 h and 24 h, with almost all individuals feeding on *S. polygaloides* (Table 2). In contrast, *L. hesperus* did not demonstrate a statistically significant preference for any of the food plants offered at either 12 h or 24 h (Table 2). *Lygus hesperus* tended to prefer *C. cuneatus* at 12 h, resulting in a nearly significant chi-square value (Table 2), but this tendency disappeared at 24 h.

Food Preferences of M. boydi *Collected from* S. polygaloides—When given the choice of 2 genera within the Brassicaceae, *M. boydi* significantly preferred *S. polygaloides* to *B. nigra*. At 12 h, all feeding insects were on *S. polygaloides* ($\chi^2 = 7.0$, $df = 1$, $P < 0.008$, $n = 7$). When 2 species of *Streptanthus* were offered as food, *M. boydi* preferred *S. polygaloides* to *S. tortuosus*, although this preference was not demonstrated statistically until 24 h. *Melanotrichus boydi* did not significantly differentiate between *S. polygaloides* and *S. tortuosus* at 12 h, although 63% of feeding insects were probing *S. polygaloides* at that time ($\chi^2 = 0.50$, $df = 1$, $P = 0.48$, $n = 8$). At 24 h, all feeding individuals of *M. boydi* were probing *S. polygaloides* ($\chi^2 = 6.0$, $df = 1$, $P = 0.014$, $n = 6$).

Melanotrichus boydi did not discriminate between *S. polygaloides* at the level of plant populations. In our choice experiment using *M. boydi* from 3 sites, insects did not demonstrate a preference for ''home site'' plants at either 12 h ($\chi^2 = 0.02$, $df = 1$, $P = 0.88$, $n = 15$) or 24 h ($\chi^2 = 0.02$, $df = 1$, $P = 0.88$, $n = 13$). At 12 h, 27% of 15 feeding insects were feeding on material from ''home sites'', and at 24 h, the corresponding percentage for 13 feeding insects was 23%. In the choice experiment using insects from site 4 and plants from sites 1, 4, 6, and 10, *M. boydi* did not significantly prefer plants from site 4 at either 12 h ($\chi^2 = 0.70$, $df = 3$, $P = 0.88$, $n = 12$) or 24 h ($\chi^2 = 0.80$, $df = 3$, $P = 0.85$, $n = 11$). At 12 h, percentages of 12 insects feeding on plant materials from each site were: site 1 (25%), site 4 (33%), site 6 (17%), and site 10 (25%). By 24 h, percentages of 11 feeding insects for materials from each site were: site 1

TABLE 2—Feeding activity of *Melanotrichus boydi* and *Lygus hesperus* collected from *Streptanthus polygaloides* at site 6 in Tuolumne County, California, and offered a choice of 4 host-plant species. Feeding was examined 12 and 24 h after the start of the experiment. Statistical results are contingency table analyses comparing actual insect activity against an expected even distribution of activity across all sample types.

	M. boydi		*L. hesperus*	
Host plant	12 h	24 h	12 h	24 h
S. polygaloides	86%	96%	13%	20%
Clarkia biloba	3%	4%	27%	40%
Ceanothus cuneatus	7%	0%	53%	20%
Calycadenia multiglandulosa	4%	0%	7%	20%
Statistical analyses:				
χ^2, $df = 3$	53	56	7.7	1.2
P-value	<0.001	<0.001	0.053	0.75
Number of insects feeding	29	28	15	10

(18%), site 4 (36%), site 6 (27%), and site 10 (18%).

Effect of S. polygaloides *Ni Concentration on* M. boydi *Host Preference*—Nickel concentration of *S. polygaloides* did not significantly affect host choice by *M. boydi.* Although few insects were observed feeding at any one time, we observed a total of 6 feeding events on high-Ni and 4 feeding events on low-Ni plants over the course of the experiment. Contingency table analysis showed no significant preference for high-Ni or low-Ni plants of *S. polygaloides* ($\chi^2 = 0.4$, $df = 1$, $P = 0.5$, $n = 10$).

DISCUSSION—Our field surveys indicated that *M. boydi* is monophagous on *S. polygaloides.* We never collected *M. boydi* from any plant species other than *S. polygaloides.* Nymphs, which are flightless and, thus, reliable indicators of host-plant suitability for reproduction (Scott, 1977), were encountered only on *S. polygaloides.* Results of the feeding experiments also indicated that *S. polygaloides* is a preferred host for *M. boydi.* The polyphagous *L. hesperus* showed no clear preference for any of the host species used, whereas *M. boydi* significantly preferred *S. polygaloides* (Table 2). This contrast supports our contention that *M. boydi* is monophagous on *S. polygaloides* and also corresponds with published information about these insect genera. *Lygus hesperus* is a polyphagous pest of many crops (Scott, 1977; Schwartz and Foottit, 1998). On the other hand, many species of *Melanotrichus* are considered specialists on plants in the Asteraceae, Brassicaceae, or Chenopodiaceae (Henry, 1991).

The preference of *M. boydi* for *S. polygaloides* was less obvious when individuals were allowed to choose between 2 *Streptanthus* species. It took more than 12 h for *M. boydi* to demonstrate a significant preference for *S. polygaloides* over *S. tortuosus*, perhaps reflecting the relatively similar chemical nature of these species. Members of the Brassicaceae are well known for their production of glucosinolates, defensive secondary chemicals (Feeny, 1977; Rosenthal and Janzen, 1979; Louda and Mole, 1991), that are present in *Streptanthus* in general (Rodman et al., 1981) and *S. polygaloides* in particular (Davis and Boyd, 2000; Jhee et al., 2006). Although glucosinolates deter many generalist herbivores, they act as feeding stimulants to many herbivorous insects that specialize on the Brassicaceae (van Emden, 1972; Louda and Mole, 1991). If glucosinolates serve as a feeding stimulant for *M. boydi,* this could explain the rapid and significant preference for *S. polygaloides* in the comparative experiment using the generalist *L. hesperus.* On the other hand, similarities in glucosinolate composition between *S. polygaloides* and *S. tortuosus* might have caused the delayed preference for *S. polygaloides* in the experiment comparing *M. boydi* preference for these *Streptanthus* species.

Melanotrichus boydi showed no preference when offered plant material from widely varying *S. polygaloides* populations. Although botanists might be unable to discern differences between populations of a species, herbivorous insects often do (Abrahamson and Weis, 1997). However, none of the populations of *M. boydi* used in this study revealed a significant preference for particular populations or phenotypes of *S. polygaloides*, suggesting that a chemical feature common to all *S. polygaloides* populations signaled their acceptability as hosts. Because all populations of *S. polygaloides* hyperaccumulate Ni (Reeves et al., 1981; Kruckeberg and Reeves, 1995), it is tempting to suggest that Ni (or a Ni-containing complex) serves as this signal. However, our choice experiment using high-Ni and low-Ni *S. polygaloides* plants showed that *M. boydi* did not discriminate between them. The lack of discrimination might be explained by the results of Davis and Boyd (2000), who found that glucosinolate concentrations of *S. polygaloides* plants grown on high-Ni soil vs. low-Ni soil did not significantly differ. Thus, we suggest that *M. boydi* uses organic chemicals, such as glucosinolates, to recognize its *S. polygaloides* host rather than plant Ni concentration.

According to Thompson (1994), the majority of phytophagous insects are specialists. Although many insect species are feeding-specialists within a plant genus, our data support the monophagy of *M. boydi* on *S. polygaloides* in nature. *Melanotrichus boydi* is only the third North American mirid reported to specialize on a species of Brassicaceae. The other 2 specialists on North American Brassicaceae also belong to the genus *Melanotrichus* (Henry, 1991). The phylogenetic relationship of these 3 mustard-feeding specialists relative to the rest of the genus would be helpful in understanding the evolution of host specialization in *Melanotrichus*.

Nickel hyperaccumulating plants offer a unique chemical defense against herbivores. Studies of *S. polygaloides* have shown that high-Ni plants are toxic to folivores (Martens and Boyd, 1994; Boyd and Jhee, 2005; Jhee et al., 2005; Jhee et al., 2006), a pathogenic fungus, and a pathogenic bacterium (Boyd et al., 1994). However, Ni is ineffective in protecting against phloem-feeding aphids and spider mites (Boyd and Martens, 1999; Jhee et al., 2005), a virus (Davis et al., 2001), a xylem-feeding spittlebug (Jhee et al., 2005), and *Lygus lineolaris* (Jhee et al., 2005). Boyd (1998) suggested at least 3 ways that herbivores can circumvent metal-based defenses: 1) by avoiding plant parts in which metals are concentrated; 2) through dilution of metal in the diet; and 3) by being physiologically tolerant of high levels of dietary metal. Avoidance seems unlikely in the case of *M. boydi*. In our field studies, we observed both adults and nymphs of *M. boydi* feeding only on young flowers and leaves of *S. polygaloides*. As these tissues contain 2,860 to 16,400 μg Ni/g (Reeves et al., 1981), Ni is not being avoided. Furthermore, dietary dilution is not an option for a monophagous herbivore. Thus, we suggest that *M. boydi* employs physiologic tolerance, similar to that reported for *C. pardalina* feeding on *B. coddii* from South Africa (Mesjasz-Przybylowicz et al., 2004), to circumvent the metal-based defense of *S. polygaloides*. Certainly, the elevated concentration of Ni in *M. boydi* (Schwartz and Wall, 2001) indicates an exceptional degree of Ni tolerance and also refutes avoidance of Ni defenses by this herbivore.

Melanotrichus boydi is the only North American insect known to be monophagous on a Ni hyperaccumulator. Another mirid, *Coquilletia insignis*, feeds on *S. polygaloides* (Boyd et al., 2004), but is found on other hosts (mainly *Lupinus* and other Fabaceae) in its range in western North America (McIver and Stonedahl, 1987) and, thus, is not monophagous. Only 9 Ni hyperaccumulator species have been identified from North America (Kruckeberg and Reeves, 1995). By contrast, Cuba is host to more Ni hyperaccumulator species than any other locality on Earth (Reeves et al., 1996, 1999). It seems likely that surveys of such Ni hyperaccumulator ''hotspots'' will uncover more examples of monophagous Ni-tolerant insect species from North America.

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