Host Range of *Herpetogramma basalis* (Lepidoptera: Crambidae), a Biological Control Agent for the Invasive Weed *Alternanthera philoxeroides* (Centrospermae: Amaranthaceae) in China

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Received 8 July 2019; Editorial decision 24 September 2019

Abstract

*Alternanthera philoxeroides* (Mart.) Griseb. is an invasive herbaceous amphibious weed species in China. A pyralid moth *Herpetogramma basalis* (Walker) was discovered feeding on *A. philoxeroides* through field surveys and may be a potentially useful biocontrol agent. To determine the host range of *H. basalis* and evaluate its potential to control *A. philoxeroides*, no-choice and multiple-choice tests were conducted. *Herpetogramma basalis* fed on target weeds and 29 nontarget plant species. In addition to the target weed *A. philoxeroides*, *H. basalis* developed to adult on eight other nontarget species. *Herpetogramma basalis* survived to adulthood successfully on *A. philoxeroides* and less successfully on several other Amaranthaceae species. In multiple-choice studies, *H. basalis* showed a strong oviposition preference for *A. philoxeroides* over *Amaranthus tricolor* L. (Centrospermae: Amaranthaceae). *Amaranthus tricolor* was the only crop plant that supported the complete development of *H. basalis*. We cautiously recommend *H. basalis* for the biological control of *A. philoxeroides* in China.

Key words: biological weed control, oviposition preference, host specificity

*Alternanthera philoxeroides* (Mart.) Griseb. (Centrospermae: Amaranthaceae, common name alligator weed) is a stoloniferous and rhizomatous perennial that grows rapidly in both terrestrial and aquatic habitats. Native to South America, it has invaded all continents except Africa and Antarctica (Julien et al. 1995, Sosa et al. 2004). *Alternanthera philoxeroides* was introduced into suburban Shanghai from Japan as a forage crop in the late 1930s and spread rapidly to eastern and southern China (Ye et al. 2003). Currently, *A. philoxeroides* is primarily distributed in up to 18 provinces in the warm temperate and subtropical regions of China (Zhang et al. 1993, Wan et al. 2005). As a highly competitive invasive species, *A. philoxeroides* has caused serious problems, such as reducing crop production, blocking drainage and irrigation channels, restricting river traffic, polluting water systems, and reducing biodiversity (Spencer and Coulson 1976, Coulson 1977, Julien et al. 1995, Tao et al. 2009).

Classical biological control is potentially an ecologically and economically sound methodology for controlling invasive plants and their negative impacts (McFadyen 1998, Reeves and Lorch 2012). For example, a flea beetle, *Agasicles hygrophila* (Selman & Vogt) (Coleoptera: Chrysomelidae), from South America was released in the southeastern United States to control *A. philoxeroides* (Hawkes et al. 1967). Introduction of *A. hygrophila* resulted in a positive ecological shift in the aquatic plant community in most of the areas where it was released (Maddox and McCready 1975). This beetle was imported from Florida, and introduced into four areas of China in 1987 (Wang et al. 1998, Ma et al. 2003). Subsequently, *A. hygrophila* established populations in some of the release sites and spread gradually to other places in southern China. It caused the collapse of alligator weed populations from May to June and September to October in aquatic habitats in both Fujian and Hunan provinces (Guo et al. 2012). Thus far, this case represents one of the most successful weed biological control programs in China. However, as a biological control agent, *A. hygrophila* has obvious weaknesses. Previous studies indicated that the beetle’s pupation rate was affected by the morphotypic characteristics of the stem among different ecotypes of *A. philoxeroides*, reducing the biocontrol efficiency in terrestrial stands of *A. philoxeroides* (Vogt et al. 1992, Ma et al. 2004). Currently, a pyralid moth, *Herpetogramma basalis* (Walker) was discovered feeding on *A. philoxeroides* through field surveys and may be a potentially useful biocontrol agent. To determine the host range of *H. basalis* and evaluate its potential to control *A. philoxeroides*, no-choice and multiple-choice tests were conducted. *Herpetogramma basalis* fed on target weeds and 29 nontarget plant species. In addition to the target weed *A. philoxeroides*, *H. basalis* developed to adult on eight other nontarget species. *Herpetogramma basalis* survived to adulthood successfully on *A. philoxeroides* and less successfully on several other Amaranthaceae species. In multiple-choice studies, *H. basalis* showed a strong oviposition preference for *A. philoxeroides* over *Amaranthus tricolor* L. (Centrospermae: Amaranthaceae). *Amaranthus tricolor* was the only crop plant that supported the complete development of *H. basalis*. We cautiously recommend *H. basalis* for the biological control of *A. philoxeroides* in China.
A. philoxeroides plants were collected from minimally disturbed areas such as wastelands and woodlands. They were planted in pots (15-cm diameter, 12-cm depth) containing disinfected soil and often watered to keep the soil moist. Newly hatched larvae were placed on potted host plants (30–40 cm in height) in the artificial climate incubator. Host plants were added as needed until all larvae pupated. Pupae were removed to petri dishes without lids and placed into eclosion cages (25 × 28 × 30 cm). The cages were made of five-sided Plexiglas with the backside covered by 0.2-mm mesh plastic gauze. The left side had a sliding door through which material could be placed in the cage or removed. The front side of the cage was vented with a 14-cm hole lined with cotton sleeve material.

A 250-ml Erlenmeyer flask containing 150-ml water was placed on the bottom of the cage and a bouquet of alligator weed was placed in the flask. After emergence, adult moths were provided with 1% honey in a Petri dish with cotton gauze as the source of nutrition. Adult moths mated in the cage and oviposited on the leaves of the host plants. Newly hatched larvae were used for host range tests and colony maintenance.

No-Choice Larval-Feeding Tests

The objective of this experiment was to determine which species supported larval feeding and development of H. basalis in the laboratory. Plants for the no-choice larval-feeding tests were chosen following a procedure proposed by Waspine (1974, 1989). Under this procedure, potential biological control agents were exposed to a sequence of plant species, ranging from the most closely related to the known host plant, to successively more distant relatives, until the host range of the biocontrol insect was adequately circumscribed. In the present test, the known host plant was A. philoxeroides and the selection of other plant species followed this procedure. The plant species, which included crops, trees, ornamental plants and weeds, were obtained from several different environments such as farms, parks, water areas, and forests in Wuhan, Hubei Province, China. More than 140 plant species belonging to 26 orders, 43 families, and 116 genera were included (Supp Table 1 [online only]). Some of these plants were invasive species (Weber et al. 2008). Healthy plants without any damage were collected and immediately taken back to the laboratory. Generally, bouquets of cut foliage were used in the tests, but occasionally complete plants were used when the plants were smaller. Plants were cultured in Erlenmeyer flasks (250 ml) containing 150 ml water and placed into the eclosion cages (25 × 28 × 30 cm) as described above; these cages reduced the interference between different treatments or repetitions. The cages were placed in incubators (LRH-400-GS, Shaoguan Taihong Medical Instrument Co. Ltd.) at 27 ± 1°C, 70 ± 5% relative humidity, and a photoperiod of 13:11 (L:D) h. Thirty newly emerged, unfed first instars (<12 h old) were transferred onto the plant leaves using a soft goat hairbrush and kept in the incubator as described above. Leaf damage, numbers of surviving larvae and their instars were recorded every 24 h. New plants were added if feeding occurred. Larval development was observed until they perished or successfully pupated. The numbers of pupae and eclosing adults were recorded. Tests were terminated when all larvae had died or completed development to adult moths. Tests were replicated three times for each plant species. Larval-feeding damage was scored from 0 to 5 following the scheme proposed by Ramadan et al. (2011): (0) no damage, (1) probing and superficial feeding on < 5% damage of leaf area, (2) light feeding on 5–20% of leaf area, (3) moderate feeding on 20–40% of leaf area, (4) severe damage on 40–60% of leaf area, and (5) intense feeding on 60% of leaf area and stems, eventually killing the plant.

Materials and Methods

Insect Rearing and Colony Maintenance

An initial culture of A. basalis (Walker, 1866) was obtained from field populations in Wuhan (30.52°N, 114.31°E), Hubei Province, China. The culture was screened for three generations in quarantine to ensure that it was healthy and that no parasitoids were associated with it. The insects were reared continuously in an incubator (LRH-400-GS, Shaoguan Taihong Medical Instrument Co. Ltd.) containing several pots of A. philoxeroides at 27 ± 1°C, 70 ± 5% relative humidity, and a photoperiod of 13:11 (L:D) h. Healthy A. philoxeroides plants were collected from minimally disturbed

Journal of Insect Science, 2019, Vol. 19, No. 6
Multiple-Choice Oviposition Tests

Oviposition tests were carried out on the experimental farm of the Hubei Academy of Agricultural Sciences. They were conducted in several 3.6 × 2.4 × 1.8-m mesh cages (mesh size = 0.5 mm), and the temperature range was about 18–30°C. The front side of each cage was cut with a 1.5-m gap lined with a zipper for entry into the cages. The cages were set on open ground outdoors. The hemoines of the cages that contacted the ground were covered with wet soil to prevent test insects or other creatures from passing through. Plastic film was hung 50 cm above the roof of the cages to avoid direct precipitation on the leaves. The cage design and placement were intended to create conditions that were as near natural as possible.

Two plant species that supported complete development of *H. basalis*, including the target weed *A. philoxeroides* and crop species *A. tricolor*, were tested under these experimental conditions to confirm that *H. basalis* would utilize these plants as suitable hosts for the oviposition choice experiments (Table 2). Several other crops that were more likely to appear in the same habitat with *A. philoxeroides* were also included in the tests because of their economic importance, although feeding damage was not observed on them in the no-choice conditions (Supp Table 2 [online only]). All 35 plant species were cultured in advance to ensure that all plants were used at a stage when they were growing most vigorously at the commencement of the tests. The pots (30-cm diameter, 20-cm depth) containing different plants were arranged in four equally spaced rows. The distance between two adjacent pots was 10 cm. Care was taken to ensure that plants did not touch each other. Plants were watered as needed. The positioning of plants was randomized using a random number table in each cage. Three replications were conducted in three different cages.

Each test commenced with the release of 20 female–male pairs of newly eclosed *H. basalis* adults into the center of each cage. Beginning on the second day after release, eggs were counted on each plant daily with a pocket lens (10× magnification) until all the adult moths had died. Observations continued through egg hatch and larval development. The numbers of larvae were monitored and their development observed until host plants were completely damaged by larval feeding. Larval feeding damage was assessed and recorded. Numbers of pupae were recorded. The plants with observed eggs were analyzed closely during this period, but other plants were also monitored to ensure plants were not omitted owing to the small size of the eggs.

Statistical Analyses

All statistical analyses were performed with SPSS 19.0 for windows (SPSS Inc., Chicago, IL) with a significance level of \( \alpha = 0.05 \). All data for one-way analysis of variance (ANOVA) analyses were first checked for homogeneity of variance using Levene’s test and percentage data were normalized using the arcsine-transformation before comparisons were made. The data of no-choice tests were compared by one-way ANOVA among treatments. LSD test was used for multiple comparisons. The data on the numbers of eggs, larvae and feeding score in multiple-choice tests were compared by \( t \)-test.

Results

No-Choice Larval-Feeding Tests

Tests were conducted with 147 plant species belonging to 26 orders, 43 families, and 116 genera (Supp Table 1 [online only]). Feeding damage occurred on target weeds and 29 nontarget plant species (Supp Table 1 [online only]). However, the feeding marks were extremely slight (feeding score 0–1) on 12 nontarget plant species and the larvae of *H. basalis* reared on these plants did not survive beyond the first instar. Apart from these slightly damaged plant species, target weeds and 17 other plant species supported *H. basalis* development to some degree, and only 9 supported development to the adult stage (Table 1). Seven of the nine species were in the family Amaranthaceae, one in the closely related family Chenopodiaceae, and one in Asteraceae. Heavy feeding damage (feeding score 5) occurred only on Alternanthera and Amaranthus generas of Amaranthaceae (Table 1). Survival to adulthood was the highest on *A. philoxeroides* (84.4 ± 5.9%), *A. sessilis* (74.4 ± 6.2%), *A. viridis* (67.8 ± 4.0%), and *A. tricolor* (65.6 ± 6.2%); intermediate on *A. retroflexus* (57.8 ± 4.0%), *A. hybridus* (54.4 ± 2.9%), *K. scoparia* (30.0 ± 5.1%), and *A. spinosus* (35.6 ± 4.0%); and low on *Helianthus tuberosus* species (4.4 ± 1.1%) (df = 8, 18; \( F = 25.88; P < 0.001 \)). Among all tested plant species, only *A. sessilis* (22.7 ± 2.2), *A. tricolor* (21.0 ± 2.5) and *A. viridis* (21.3 ± 1.7) were the same statistically as *A. philoxeroides* (25.7 ± 1.9) in pupal production (df = 8, 18; \( F = 17.58; P < 0.001 \)) and only *A. sessilis* adult production (22.3 ± 1.9) was statistically the same as *A. philoxeroides* (25.3 ± 1.8; df = 8, 18; \( F = 25.88; P < 0.001 \); Table 1, Fig. 1).

Multiple-Choice Oviposition Tests

Eggs were laid on two species: target weed *A. philoxeroides* and vegetable crop *A. tricolor*. No eggs were recorded on any of the other 33 crop species. Egg numbers and larval numbers on the *A. tricolor* were significantly less than those on *A. philoxeroides* (the number of eggs: \( t = 19.89; df = 2; P = 0.003 \); the number of the first instar larvae: \( t = 24.38; df = 2; P = 0.002 \); the number of the third instar larvae: \( t = 12.61; df = 2; P = 0.006 \); Table 2). Feeding damage on the two species was obviously different. By the time the larvae had reached the third instar, *A. philoxeroides* was almost completely damaged (feeding score 5) with only old stems remaining, whereas feeding score of 1–2 were observed for *A. tricolor* (\( t = 11.000; df = 2; P = 0.008 \); Table 2).

Discussion

The risk of damage to nontarget species by biological control agents can be reduced if the potential for damage can be identified prior to release (McFadyen 1998, Schooler et al. 2006, Manrique et al. 2014). No-choice tests in the laboratory can help identify the range of plants on which the insect can feed and those on which the insect can develop at least partially—which defines the fundamental host range and physiological host range, respectively (Coombs et al. 2004, Sheppard et al. 2005, Fowler et al. 2012). With a sufficiently large and phylogenetic-oriented selection of potential host plants, no-choice tests usually delineate an insect’s host range accurately (Simberloff 2012). In the present study, no-choice tests were used to classify the host suitability of plant species for a given insect into four categories: plants with no feeding damage, plants with tentative feeding, plants supporting insect development to some stage, and plants supporting insect development to the adult stage. Under no-choice conditions, although feeding damage occurred on target weeds and 29 nontarget plant species, only 9 species supported development to the adult stage. The development of *H. basalis* was the most successful on the species belonging to the family Amaranthaceae. Although *H. basalis* could also develop to adults on the a few species in the families, Amaranthaceae, Chenopodiaceae, and Asteraceae, the survival proportion was very low (Table 1). This result demonstrates the usefulness of the centrifugal phylogenetic
Table 1. Larval feeding and development tests with *Herpetogramma basalis* under no-choice conditions

| Family           | Species                               | Status a | Feeding score | No. of larvae in each instar b | No. of pupae | No. of adults |
|------------------|---------------------------------------|----------|---------------|-------------------------------|--------------|--------------|
|                  |                                       |          |               | 1st instar  | 2nd instar  | 3rd instar  | 4th instar  | 5th instar  |               |               |
| Amaranthaceae    | *Alternanthera philoxeroides* (Mart.) Griseb. | I, 5    | 30            | 27.7 ± 1.3 a | 27.3 ± 1.2 a | 27.3 ± 1.2 a | 26.7 ± 1.9 a | 25.7 ± 1.9 a | 25.3 ± 1.8 a |
| Amaranthaceae    | *Amaranthus sessilis* (L.) DC.         | N 5     | 30            | 27.3 ± 1.2 b | 26.7 ± 1.5 a | 23.7 ± 1.2 b | 23.3 ± 1.2 b | 22.3 ± 1.9 b | 22.3 ± 1.9 b |
| Amaranthaceae    | *Amaranthus tricolor* L.               | N, C 5  | 30            | 25.0 ± 1.8 b | 23.3 ± 1.8 b | 23.3 ± 1.8 b | 22.7 ± 2.4 b | 22.0 ± 2.5 b | 19.7 ± 1.9 bc |
| Amaranthaceae    | *Amaranthus viridis* L.                | I 5     | 30            | 24.0 ± 1.5 b | 23.3 ± 1.7 bc | 22.3 ± 1.2 b | 21.3 ± 1.7 bc | 20.3 ± 1.2 bc |               |
| Amaranthaceae    | *Amaranthus retroflexus* L.            | I 5     | 30            | 25.3 ± 1.5 b | 21.7 ± 1.9 bc | 19.3 ± 0.9 bc | 18.7 ± 0.9 bc | 18.3 ± 1.2 bc | 17.3 ± 1.2 cd |
| Amaranthaceae    | *Amaranthus hybrius* L.                | I 5     | 30            | 23.7 ± 1.2 b | 21.7 ± 1.2 bc | 20.7 ± 1.8 d | 18.7 ± 1.8 bc | 17.7 ± 1.8 bc | 16.3 ± 0.9 cd |
| Amaranthaceae    | *Amaranthus spinosus* L.               | I 4-5   | 30            | 22.0 ± 2.1 b | 18.3 ± 1.7 c | 16.0 ± 1.7 bc | 14.3 ± 1.9 bc | 12.3 ± 1.5 d | 10.7 ± 1.2 e |
| Amaranthaceae    | *Achyanthes bidentata* Blume           | N 2     | 30            | 13.0 ± 2.3 cd | 8.3 ± 2.3 de | 3.3 ± 1.5 bc | –              | –              |               |
| Amaranthaceae    | *Celosia argentea* L.                  | N 2     | 30            | 16.3 ± 2.3 f | 12.3 ± 2.0 f | 5.3 ± 1.9 f | 1.0 ± 0.6 f | –              |               |
| Chenopodiaceae   | *Kochia scoparia* (L.) Schrad.         | N 4-5   | 30            | 22.3 ± 1.7 b | 20.7 ± 1.5 ec | 19.3 ± 1.2 bc | 18.7 ± 1.3 bc | 16.7 ± 1.5 cd | 15.0 ± 1.6 d |
| Leguminosae      | *Arachis hypogaea* L.                  | N, C 1  | 30            | 11.3 ± 1.0 ef | 4.7 ± 0.9 gf | 1.3 ± 0.7 h | –              | –              |               |
| Leguminosae      | *Lablab purpureus* (L.) Sweet          | N, C 0-1| 30            | 7.3 ± 1.5 fg | 2.3 ± 0.9 h | –              | –              | –              |               |
| Leguminosae      | *Vigna radiata* (L.) Wilczek           | N, C 0-1| 30            | 6.7 ± 1.2 g | 0.7 ± 0.3 i | –              | –              | –              |               |
| Leguminosae      | *Glycine max* (L.) Merr.               | N, C 0-1| 30            | 4.3 ± 0.9 g | 3.3 ± 0.9 h | 1.0 ± 0.6 f | –              | –              |               |
| Cucurbitaceae    | *Cucurbita moschata* (Duch. ex Lam.)   | N, C 1  | 30            | 11.7 ± 2.0 ef | 1.7 ± 0.9 f | –              | –              | –              |               |
| Convolvulaceae   | *Ipomoea aquatica* Forsk.              | N, C 1  | 30            | 7.3 ± 1.5 fg | 4.0 ± 1.0 f | 1.7 ± 0.9 h | –              | –              |               |
| Asteraceae       | *Helianthus tuberosus* L.              | I 1     | 30            | 8.7 ± 1.2 de | 4.7 ± 1.5 ef | 4.0 ± 1.2 bc | 1.7 ± 0.7 d | 1.3 ± 0.3 f | 1.3 ± 0.3 f |
| Asteraceae       | *Xanthium sibiricum* Patrin ex Widder | N 1     | 30            | 12.3 ± 2.3 d | 8.3 ± 1.2 bc | 3.3 ± 1.2 bc | –              | –              |               |

Tests were replicated three times. Thirty new hatched larvae were used in each test. Feeding score: 0 (no damage), 1 (< 5% damaged), 2 (5–20% damaged), 3 (20–40% damaged), 4 (40–60% damaged), and 5 (> 60% damaged). The plant species which supported phased development were included in this table. Data are presented in means ± SE. And those followed by different lowercase letters within a column are significantly different (one-way ANOVA, LSD tests, P < 0.05).

aC, crop; I, invasive; N, native.

bNo. of the first instar was the number of new hatched larvae transferred onto the plant leaves at the beginning of the tests, and number of larvae in other instars was the maximum count of larvae that survived to this instar.
method used (Wapshere 1974). The Amaranthaceae includes about 70 genera and 900 species worldwide and 15 genera (one introduced) and 44 species (three endemic, 14 introduced) in China (Bao et al. 2003). Most of these species are not economically significant; however, one crop species in this group is a widely cultivated vegetable species, A. tricolor, which supported the development of H. basalis to the adult stage in the no-choice tests. This suggests that H. basalis may pose risks to economically important nontarget species, an issue that will be discussed later.

Given that H. basalis was able to complete its development on several nontarget plant species in the no-choice tests, oviposition tests were subsequently conducted to predict the potential host range of H. basalis. Although H. basalis completed its development on several plant species, only A. philoxeroides and A. tricolor were used in the subsequent oviposition choice tests. In addition to the target species A. philoxeroides, A. tricolor was chosen, because it is a vegetable crop species. The other species that supported the complete development of H. basalis were all weeds and most of them were closely related to these two species. Given the emphasis on understanding oviposition choice between crop plants and weeds in our choice tests, we selected almost all local crops of economic importance for our oviposition choice tests. Of these, only A. tricolor supported oviposition but was not the preferred host.

Many potential insect biological control agents for weeds were selected and tested under restricted cage conditions and have been rejected as unsafe for introduction because they had a wider host range under cage conditions than that observed under natural conditions (Wapshere 1989). The tendency to overestimate host range is possibly owing to limited feeding on plants that are not normally hosts in no-choice specificity tests (Heard 2000, Zachariades et al. 2002, Ramadan et al. 2011). To avoid discarding potentially useful biological control agents as unsafe, tests under conditions as near natural as possible are necessary (Wapshere 1989, Frye et al. 2010). Similarly, in oviposition tests, the restricted test arena and small size of test plants used under quarantine conditions results in indiscriminate oviposition on artificial surfaces in both no-choice and choice trials (Dhileepan et al. 2015).

The problems caused by alternate hosts must be taken seriously when the biocontrol agents were not monophagous (Wapshere 1974). Our research showed that A. sessilis was the only host other than A. philoxeroides that supported development of an equal number of adults. Therefore, A. sessilis had a great possibility to become an alternate host of H. basalis in the field. However, A. sessilis was not included in the multi-choice test. The main reason for this is to consider the status of A. sessilis in the ecological environment compared with other plant species. A. sessilis is a perennial herb and the only native congener of A. philoxeroides in China. It can also spread by clonal growth but mainly by sexual reproduction. A. philoxeroides has wider temperature adaptability, stronger photosynthetic capacity and faster growth ability than A. sessilis. These characteristics make A. philoxeroides more competitive than A. sessilis. As a result, the population of A. sessilis is much smaller than that of A. philoxeroides, although their distribution areas are basically overlapping. (Geng et al. 2006, Zhang et al. 2006, Sun et al. 2010). Therefore, if released in the field, H. basalis would rarely even have a chance to choose between the two species. Of course, it is of great scientific significance to study the host preference of H. basalis to A. philoxeroides and A. sessilis, and new experiments need to be conducted to clarify this issue in the future.

Ideally, risk assessments should include open field studies of host specificity in the weed’s native range, but these tests are difficult to conduct owing to limited funding and the collaboration required conducting studies on large scales (McFadyen 1998). Our multiple-choice trials utilized field mesh cages to observe the oviposition preference of H. basalis. Environmental factors, such as temperature, humidity, and soil in the cages were very similar to natural conditions, with the exception of the mesh and film over the cages. Crops, weeds, and target host weeds were placed in the cages simultaneously, similar to the conditions outside the cages. Thus, conditions for the oviposition choice studies were as authentic as we could achieve on a small scale. There were marked differences between larval feeding fitness and adult oviposition preference. For example, H. basalis had high rates of developmental completion to the adult stage on A. tricolor, but very low proportions of adults selected these

![Fig. 1. Mean survival to adult stage (%) ±SE of Herpetogramma basalis on Alternanthera philoxeroides and eight nontarget plant species, A. sessilis, A. tricolor, A. viridis, A. retroflexus, A. hybridus, A. spinosus, Kochia scoparia, and Helianthus tuberosus during no-choice larval-feeding tests. Multiple comparisons were carried out using LSD tests. Bars topped by the same letters are not significantly different among plant species (P < 0.05). The percentages of survival to the adult stage on all nontarget plant species except A. sessilis were significantly lower than that on A. philoxeroides.](image)

Table 2. Adult oviposition, larval development, and feeding score of Herpetogramma basalis under multiple-choice conditions

| Species               | No. of eggsa | No. of first instar larvaea | No. of third instar larvaeab | Feeding scorec |
|-----------------------|--------------|-----------------------------|-----------------------------|---------------|
| Alternanthera philoxeroides | 477.7 ± 29.0 | 316.3 ± 10.7                | 82.3 ± 5.2                  | 5.0 ± 0       |
| Amaranthus tricolor    | 39.3 ± 15.0  | 21.0 ± 5.7                  | 14.7 ± 3.8                  | 1.3 ± 0.3     |

Tests were replicated three times. Feeding score: 0 (no damage), 1 (< 5 % damaged), 2(5–20% damaged), 3 (20–40% damaged), 4 (40–60% damaged) and 5 (> 60% damaged). Data are presented in means ±SE.

aMeans of the indexes between different plants are significantly different (t-tests, P < 0.05).

bObservation of larval numbers terminated at the third instar larval stage when the target weeds A. philoxeroides were almost completed damaged.
species for oviposition. Host suitability of the insect is determined by both oviposition and larval development. For a holometabolous insect, the survival of offspring larvae depends on whether they lay eggs on suitable host plants, so their oviposition preference is very important. Thus, use of multiple-choice field experiments simulating natural conditions as applied in our studies warrants further attention.

Although *H. basalis* was able to complete its development on *A. tricolor*, the observation that *A. tricolor* was not a preferred ovipositional host could explain why we found *H. basalis* feeding on *A. philoxeroides* but not *A. tricolor* in field surveys. Whether *H. basalis* can be used as a candidate biological agent given that *A. tricolor* is an important crop species needs to be evaluated. In our opinion, the moth could be a valuable potential biological agent. First, *H. basalis* has been present in China for a long time and has never been recorded as a pest of any crop in China. The current regulatory policies of China on biological control are all related to invasive or quarantine organisms (PRCME 2002), and the use of existing organisms such as *H. basalis* for biological control in China is not regulated by current laws and policies. Nevertheless, adequate assessments and tracking observation are needed before and after large-scale release to ensure that there are no risks. Second, when given a choice for oviposition, *H. basalis* significantly preferred *A. philoxeroides* and the proportion of eggs laid on *A. tricolor* was very low. Although monocultures approach the no-choice situation and alter the population dynamics of insects introduced for biological control (McConachie 2015), *H. basalis* would possibly always have *A. philoxeroides* available under field conditions. The population of *A. philoxeroides* and other closely related weed species, such as *A. retroflexus*, far exceed the populations of *A. tricolor*. Finally, *A. tricolor* would possibly not be at risk from feeding by *H. basalis* stems since *A. tricolor* is an annual crop with a short growth period and is cultivated from April to September. This is the same period that *A. philoxeroides* grows most vigorously. *A. philoxeroides* is a perennial weed with robust vegetative growth and ecological adaptability. The growth period of *A. philoxeroides* extends far beyond the growth period of *A. tricolor* unless the latter is grown in a greenhouse. Moreover, *A. tricolor* as a crop, would be well managed, whether in the field or in a greenhouse owing to its economic value. If *H. basalis* occurred on *A. tricolor* occasionally, it could be controlled as long as with other insect pests. In short, considering the spatial and temporal distribution of weedy host plants and *A. tricolor*, and the oviposition and feeding preferences of *H. basalis*, there is little risk of *H. basalis* becoming a significant pest of *A. tricolor*.

In conclusion, the results presented here indicate that *H. basalis* is an oligophagous herbivore with a preference for the target weed *A. philoxeroides* and inferior preference for closely related species that are mostly restricted to the family Amaranthaceae. The only crop species that supported complete development of *H. basalis* was *A. tricolor*. Based on the arguments presented above, *H. basalis* could be a valuable potential biological agent in China, nevertheless, it is not suitable for release in any of the leading weed biological control countries where *A. philoxeroides* is invasive, due to its host range being too broad. Whether it can be applied also depends on its control effect, population dynamics and ecological adaptability in the field.

**Supplementary Data**

Supplementary data are available at *Journal of Insect Science* online.

**Acknowledgments**

We thank Professor Fanghao Wan and Dr. Peng Wan for offering useful suggestions. This research was funded by the Agricultural Science and Technology Innovation Center of Hubei Province, China (No. 2011-620-003-03-04).

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