

Resistance of *Glycine* Species and Various Cultivated Legumes to the Soybean Aphid (Homoptera: Aphididae)

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ABSTRACT The soybean aphid, *Aphis glycines* Matsumura, is a new pest of soybean, *Glycine max* (L.) Merr., in North America. It has become widespread on soybean in North America since it was first identified in the Midwest in 2000. Species of *Rhamnus* L. (buckthorn) are the primary hosts of *A. glycines*, and soybean is known as a secondary host. There is limited information about the secondary host range of *A. glycines*. Aphid colonization on various legume hosts was compared in choice experiments. Aphid colonization occurred on species in the genus *Glycine* Wild. No colonization occurred on *Lablab purpureus* (L.) Sweet, *Lens culinaris* Medik, *Phaseolus vulgaris* L., *Pisum sativum* L., or species of *Vicia* L. and *Vigna* Savi. Colonization was limited or aphids were transient on species of *Medicago* L., *Phaseolus* L., and *Trifolium* L. There were significant differences in aphid colonization among *Medicago truncatula* accessions with numbers ranging from 7 to 97 aphids per plant. Six *Glycine soja* Sieb. & Zucc. accessions were as resistant as *G. max* accessions to *A. glycines*; these may represent novel sources of *A. glycines* resistance not found in *G. max*. Antibiosis was found to play a large role in the expression of resistance in three of the *G. soja* accessions. Results of this study indicated that *G. max* and *G. soja* were the best secondary hosts of *A. glycines*; however, its secondary host range may include other leguminous species. Therefore, *A. glycines* did not seem to have a highly restricted monophagous secondary host range.

KEY WORDS *Aphis glycines*, aphid, *Glycine*, resistance, soybean

A NATIVE OF ASIA, *Aphis glycines* Matsumura was first identified in the Midwest in 2000 (Hartman et al. 2001). It rapidly spread throughout the region and into other parts of North America (Patterson and Ragsdale 2002). High aphid populations reduce crop production directly when their feeding causes severe stunting and leaf distortion (Sun et al. 1990, Patterson and Ragsdale 2002, Hill et al. 2004). According to an online document from the University of Minnesota Extension Service (Ostlie 2002), in fields where there were high aphid populations, yield losses attributed to the aphid were estimated at 13% in replicated plots in Wisconsin in 2000 and >50% in experimental plots in Minnesota in 2001. Soybean aphids have reduced yields by 58% (Wang et al. 1994) and plant height by ≈ 21 cm (Wang et al. 1996) in China. An additional threat posed by the aphid is its ability to transmit certain plant viruses to soybean, such as *Soybean mosaic virus* (SMV) (Hartman et al. 2001).

A. glycines and a close relative *Aphis gossypii* Glover, the cotton or melon aphid, are the only aphid species found colonizing soybean in the United States. In other parts of the world *Aphis craccivora* Koch, *Aulacorthum solani* (Kaltenbach), and other species have been found colonizing soybean (D. Voegtlin, personal communication).

A. glycines has a heteroecious holocyclic life cycle pattern (Guang-xue and Tie-sen 1982, Hartman et al. 2001). *Rhamnus* L. spp. (buckthorn) are the primary hosts of *A. glycines*, and soybean is a secondary host. In autumn when the soybean crop matures, the aphid moves to *Rhamnus*, where mating and egg deposition occurs. The egg stage overwinters on *Rhamnus*. During the following spring, the eggs hatch and a few wingless generations are produced before alates (winged females) migrate to soybean fields.

There is little information about other secondary hosts of *A. glycines* besides soybean. Host range information is important in developing an integrated control approach and for assessing the potential spread of aphid-transmitted viruses. About 10% of all aphids are heteroecious and are classified as polyphagous because they can colonize different hosts (Eastop 1973). However, most aphids colonize one plant species at a time and are therefore regarded as sequentially monophagous (Dixon 1987).

The objectives of this study were to test the ability of *A. glycines* to colonize and damage other cultivated

Trade and manufacturers' names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by USDA implies no approval of the product to the exclusion of others that may also be suitable.

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legume species besides *G. max* and to compare colonization on different *Glycine* species, including wild soybean, *Glycine soja* Sieb. & Zucc., a potential source of new genetic traits for soybean improvement (Singh and Hymowitz 1999).

Materials and Methods

Culture of *A. glycines*. A single clone of virus-free aphids was propagated from an individual first instar from a population of *A. glycines* collected on soybean in Urbana, IL, in 2000 (Hill et al. 2004). David Voegtlin (Illinois Natural History Survey, Urbana, IL) confirmed the aphid identification. Aphids were reared and maintained on V_c-V₂ stage (Fehr and Caviness 1977) virus-free plants of soybean 'Williams 82', grown inside a growth chamber (model E-54U, Percival Scientific, Inc., Boone, IA) at 22°C, the optimum temperature for population development (Hirano et al. 1996), under continuous 200 $\mu\text{mol m}^{-2} \text{s}^{-1}$ PAR illumination. The Urbana clone was used in all experiments described below.

Plant Culture. Seed of *G. max* and other legume species, collected from various sources, was directly sown into soil-less medium (Sunshine Mix, LC1, Sun Gro Horticulture Inc., Bellevue, WA) and covered with course grade vermiculite (Hummert International, Earth City, MO). Plants were grown in plastic multipots (#D812, Hummert International) with pot sizes ranging from 30 by 40 by 60 mm to 60 by 60 by 60 mm, depending on the experiment, and placed in plastic trays without holes (#F1020, Hummert International). Immediately after planting, five to 10 pellets of a slow release fertilizer (Nutricote, 18:6:6) were added over the surface of the medium in each pot.

Seed of noncultivated *Glycine* species, provided by T. Hymowitz (Department of Crop Sciences, University of Illinois, Urbana, IL), were scarified by nicking the side of each seed opposite from the hilum with a razor blade to enhance germination. Scarified seeds were placed on moist filter paper (90-mm Whatman No. 1) within 100 by 15-mm plastic petri dishes under continuous 20 $\mu\text{mol m}^{-2} \text{s}^{-1}$ PAR illumination at 25°C for 5–7 d. Germlings were transplanted into soil-less medium in multipots placed in trays without holes and were covered with a clear plastic dome (#CW221, Hummert International) after transplanting. The covered trays were placed in the shade below a bench in the greenhouse to acclimatize the seedlings.

Aphid Choice Tests. Five choice tests were conducted to study host preferences and possible antixenosis. In these tests, aphid movement was not restricted, allowing them to seek and accumulate on susceptible hosts. All experiments were conducted in an air-conditioned greenhouse maintained at 22–25°C with supplemental continuous illumination (200 $\mu\text{mol m}^{-2} \text{s}^{-1}$ PAR at night) (Hill et al. 2004). The greenhouse was dedicated to soybean aphid work exclusively. No pesticides were used and entry into the greenhouse was restricted to avoid introduction of aphid predators and parasitoids. Plants were bottom

watered to avoid disturbing the aphids by filling the trays containing the plants with water as needed.

Aphid colonization on a range of various legume species, soybean cultivars, and *G. soja* accessions were compared in experiment 1. Rows of two plants of 48 entries collected from various sources were arranged in a randomized complete block (RCB) design with four replications. Two resistant *G. max* cultivars ('Jackson' and PI71506) (Hill et al. 2004), and several susceptible *G. max* cultivars were included in the experiment. Soon after plant emergence, seedlings were directly infested with aphids by placing leaves from 'Williams 82' plants containing dense colonies of aphids of all stages on top of each seedling. Within a day, aphids moved from the infested leaves to the test seedlings and the transferred leaves were removed and discarded. The trays were grouped together on a single greenhouse bench, allowing apterae (wingless) and alatae (winged) forms to wander within and between trays. Randomization of test entries within the flats minimized the effect of potential variation in numbers of aphids on infested leaves transferred to the test plants. Within a week after aphid transfer, aphids had moved and began accumulating on susceptible host plants. Aphid colonization on each row of two plants was rated 17 d after infestation, giving more than adequate time for populous aphid colonies to develop and begin to cause observable plant damage on susceptible plants. Aphid colonization indices (Hill et al. 2004) were calculated by taking the product of the estimates of aphid population density with plant damage. Aphid population density was estimated using a 0–3 scale, where 0 represents no aphids observed; 1, low population density; 2, medium population density; and 3, dense population (usually >100 aphids per plant). A 0–3 scale was also used to estimate plant damage, where 0 represents no perceptible damage; 1, mild leaf discoloration or distortion; 2, moderate leaf discoloration or distortion; and 3, severe leaf distortion, stunting, or plant death. The product of the two estimates gave a broader range of index values, from 0 to 9, to maximize potential differences among test entries.

In experiment 2, a direct count of numbers of aphids on plants of 12 accessions of various legume species was recorded. Rows of four plants of each accession were arranged in a RCB design with three replications. Resistant 'Jackson' and susceptible 'Williams 82' *G. max* were included in the experiment. Infested leaves were placed on 7-d-old seedlings. The total number of aphids on each plant was counted 13 d later.

In experiment 3, numbers of aphids on plants of 14 accessions of different *Glycine* species, obtained from the USDA soybean germplasm collection housed at the University of Illinois, Urbana, IL, were counted. As in experiment 2, resistant 'Jackson' and susceptible 'Williams 82' *G. max* were included in the experiment. Rows of two plants of each accession were arranged in a RCB design with four replications. Seedlings were indirectly infested with aphids by exposing them to alates that migrated from plants surrounding the ex-

periment. Total numbers of aphids on each test plant were counted 21 d later.

Seedlings of 24 accessions of *M. truncatula*, Plant Introduction (PI) numbers 2203, 2204, 2218, 2252, 2729, 2748, 2806, 2820, 2826, 2831, 2840, 2841, 3047, 3054, 3115, 3116, 3308, 3536, 3537, 3562, 3569, 3573, 3648, and 3653, obtained from the South Australian Research and Development Institute, were directly infested in experiment 4. In this experiment, three plants of each accession were arranged in a RCB design with two replications. Total numbers of aphids on each test plant were counted 21 d later.

Numbers of aphids on several accessions of *G. soja* were compared with susceptible and resistant *G. max* accessions (Hill et al. 2004) in experiment 5. Two *G. max* accessions, PI87059 and PI88508, obtained from the USDA soybean germplasm collection, that had low aphid colonization indices in a preliminary germplasm screen, were included in the experiment. *G. soja* accessions with PI numbers were obtained from the USDA soybean germplasm collection, whereas the other *G. soja* accessions included in the experiment were obtained from B. Diers (Department of Crop Sciences, University of Illinois, Urbana, IL). The accessions were selected because they were identified as putatively resistant or susceptible after *A. glycines* inadvertently attacked several *G. soja* accessions inside a field cage in Urbana, IL, in 2001. Rows of two plants of each accession were arranged in a RCB design with four replications. Infested leaves were placed on 7-d-old seedlings and 14 d later, the total number of aphids on each plant was counted.

Aphid Nonchoice Test. A nonchoice test was conducted to study the role of antibiosis in the resistance of five *G. soja* and two *G. max* accessions, 'Dowling' (resistant) and 'Loda' (susceptible) (Hill et al. 2004). Four of the five *G. soja* accessions included were tested in experiment 5 and the fifth one, 'HAS', from B. Diers' collection, was found to be susceptible in a preliminary test and was included as a susceptible check. Four plants of each accession were arranged in a completely randomized design. The experiment was conducted in a Conviron plant growth chamber (model #CMP4030, Controlled Environments Ltd., Winnipeg, MB, Canada) at 22°C under continuous 300 $\mu\text{mol m}^{-2} \text{s}^{-1}$ PAR irradiation and 70% RH. A single alate, 1 or 2 d old, was placed on the abaxial side of the lamina of the center leaflet of a new, fully expanded true leaf of individual V1-V2 stage plants (Fehr and Caviness 1977) with the aid of a moist camel's-hair brush. Aphids were isolated on the leaves by attaching leaf cages over the aphids to restrict their movement. The cages were made with 1-mm-thick plastic tubing with a 10-mm-internal diameter, cut 12 mm in length, and covered with plastic mesh with 100- μm openings (Sterling Net Co., Montclair, NJ) glued on one end. On the opposite end of the cage tubing, a 4 mm in width by 4 mm in thickness foam ring with a 8-mm internal and 12-mm outer diameter was centered and glued on to provide a seal between the cage and leaf surface when attached to the leaf. Cages were placed over the aphids with the foam end down on the leaf

surface and were fastened to the leaf with a metal clip held closed by spring tension. Alates were placed on four individual plants of each test entry. Nymphs produced from each alate placed on each plant were counted and removed daily to avoid overcrowding of the cages. After 12 d, the cumulative number of aphid offspring on each plant was determined.

Statistical Analyses. All statistical data analyses were performed with the aid of JMP version five (SAS Institute 2002). Aphid counts were first transformed to $\log_{10}(\text{count} + 1)$ before performing analysis of variance (ANOVA) and least squared means were de-transformed before presenting them in the tables. Mean separation was done by calculating the least significant difference (LSD) at $P = 0.05$ when treatment means were significantly different ($P < 0.05$) in the ANOVA.

Results

Aphid Colonization on Different Legume and *Glycine* species. There were significant differences ($P < 0.01$) among the entries in experiment 1 (Table 1). Plants of many of the legume species were free of aphids 17 d after infestation, including *Lablab purpureus* (L.), *Lens culinaris* Medik, *Vigna* spp., *Vicia* spp., one *Phaseolus vulgaris* L. accession, and *Pisum sativum* L. Observations indicated that aphids avoided those plants.

A few live aphids were observed on some accessions of *Lotus corniculatis* L., *Onobrychis vicifolia* Scop., *Phaseolus lunatus* L., and *P. vulgaris*. Most of the live aphids were probably the aphids originally transferred to the plants and were transient on those hosts, passing over them while seeking a better host, sometimes stopping to feed on them for a time. Dead aphids were commonly found on *P. vulgaris*, and a few were also found on some of the other legume species, indicating that aphids tried to feed on those species but could not survive on them. No plant damage caused by aphids was observed on those species.

Larger numbers of aphids were found on *Medicago sativa* L. and *Trifolium* spp., up to ≈ 40 aphids with a few small aphid colonies on some plants. Numbers on the *Phaseolus coccineus* L. accession exceeded 40 aphids per plant and its mean aphid colonization index was not significantly different from the resistant soybean accession 'Jackson'. Aphid feeding seemed to cause minor distortion and crinkling of *P. coccineus* leaves.

Aphid colonization on *G. soja* varied among the accessions. One *G. soja* accession, PI518282, had an index significantly lower than 'Jackson', indicating it had strong resistance. Most of the other *G. soja* accessions had intermediate indices and were less resistant than 'Jackson'. *G. soja* accession PI423993 had a high index, indicating that it had high aphid numbers and severe plant damage, similar to susceptible *G. max* test entries.

Although, numbers of aphids on 'Jackson' and PI71506 were higher (up to ≈ 50 aphids per plant) than on non-*Glycine* species, they were thinly scattered and

Table 1. Soybean aphid colonization on seedlings of various legume and soybean germplasm accessions in a choice test 17 d after aphid infestation

Species	Name	Source/Description	Aphid colonization index (0–9)
<i>Lablab purpureus</i>	PI288467	India	0.0a ^a
<i>Vigna angularis</i>	Erimo Shozu	Adzuki bean	0.0a
<i>Vicia villosa</i>	Hairy Vetch		0.0a
<i>Vigna aureus</i>	Kiloga	Mung bean	0.0a
<i>Vicia sativa</i>	PI170474	Turkey	0.0a
<i>Pisum sativum</i>	PI206832	Pea	0.0a
<i>Vicia faba</i>	PI469199	United Kingdom	0.0a
<i>Lens culinaris</i>	PI508091	Lentil	0.0a
<i>Vigna angularis</i>	PI93815	China	0.0a
<i>Phaseolus vulgaris</i>	SEA-10 Mulatinho	Common bean	0.0a
<i>Vigna angularis</i>	Takara Shozu	Adzuki bean	0.0a
<i>Phaseolus vulgaris</i>	A176 Jalinho	Common bean	0.2a
<i>Lotus corniculatus</i>	Maitland	Birdsfoot trefoil	0.2a
<i>Phaseolus vulgaris</i>	XAN309	Common bean	0.2ab
<i>Onobrychis vicifolia</i>	PI110400	Sainfoin	0.3abc
<i>Phaseolus vulgaris</i>	Miss Kelly Amendoin	Common bean	0.4abcd
<i>Trifolium repens</i>	Common	White clover	0.6abcd
<i>Phaseolus lunatus</i>	PI549453		0.7abcde
<i>Trifolium pratense</i>	Marathon	Red clover	0.9bcde
<i>Phaseolus vulgaris</i>	Pompadour B	Common bean	0.9bcde
<i>Phaseolus vulgaris</i>	Top Crop	Common bean	0.9bcde
<i>Medicago sativa</i>	PI536532	Alfalfa	1.0cdef
<i>Trifolium pratense</i>	C11	Red clover	1.2defg
<i>Trifolium subterraneum</i>	Woogenelcup		1.2defg
<i>Glycine soja</i>	PI518282	Soybean ally	1.8efgh
<i>Phaseolus coccineus</i>	Scarlet Runner Bean		2.0efgh
<i>Glycine max</i>	Jackson (PI548657)	Aphid resistant	2.1fghi
<i>Glycine soja</i>	PI424006A	Soybean ally	2.4fghij
<i>Glycine max</i>	PI71506	Aphid resistant	2.7hijk
<i>Glycine soja</i>	PI447003A	Soybean ally	3.0hijkl
<i>Glycine soja</i>	PI424006B	Soybean ally	3.3hijkl
<i>Glycine soja</i>	PI468396B	Soybean ally	3.3hijkl
<i>Glycine soja</i>	PI518281	Soybean ally	4.1jklm
<i>Glycine max</i>	Essex	Soybean cultivar	4.3jklm
<i>Glycine max</i>	L93P-941	Dense pubescence	4.3jklm
<i>Glycine soja</i>	PI468918	Soybean ally	4.3jklm
<i>Glycine max</i>	Soden-daizu	PI229358	4.6klm
<i>Glycine max</i>	Williams 82	Soybean cultivar	4.6klm
<i>Glycine soja</i>	PI522212B	Soybean ally	5.1lm
<i>Glycine max</i>	Clark	Soybean cultivar	5.4lm
<i>Glycine max</i>	Fiskeby V	PI360955A	6.0m
<i>Glycine max</i>	L62-1579	Dense pubescence	6.0m
<i>Glycine max</i>	L95P-65	Dense pubescence	6.0m
<i>Glycine soja</i>	PI423993	Soybean ally	7.0m
Mean			1.4

Means followed by the same letters are not significantly different by the least significant difference test ($P = 0.05$).

^a Mean of four replications. Aphid colonization index: the product of the estimates of aphid population density with plant damage. Aphid population density was estimated using a 0–3 scale, where 0 represents no aphids observed; 1, low population density; 2, medium population density; and 3, dense population (usually >100 aphids per plant). A 0–3 scale was also used to estimate plant damage, where 0 represents no perceptible damage; 1, mild leaf discoloration or distortion; 2, moderate leaf discoloration or distortion; and 3, severe leaf distortion, stunting, or plant death.

did not seem to form colonies, whereas significant colonization, with >100 aphids per plant, densely packed together, combined with moderate-to-severe damage occurred on susceptible *G. max* test entries.

Dense pubescence in *G. max* did not reduce aphid colonization on the three accessions possessing the trait (Table 1). All other *G. max* accessions had normal pubescence densities, including the resistant accessions 'Jackson' and PI 71506.

Aphid numbers were significantly different among the entries in experiment 2 (Table 2). *P. sativum* plants had no aphids present on them, supporting the results of experiment 1 that indicated it was a poor host of the soybean aphid, whereas the other legume species had at least a few aphids per plant. As in experiment 1, the

aphids were transient on some species and those observed were probably the aphids originally transferred to the plants. *G. max* 'Jackson' plants had significantly more aphids than the other legumes, but significantly less than 'Williams 82' plants.

In experiment 3, there were significant differences in aphid numbers among the *Glycine* species (Table 3). *Glycine clandestina* Wendl., accession PI440958, had significantly fewer aphids than 'Jackson', whereas the majority of the other *Glycine* species had aphid numbers not significantly different from 'Jackson'. Two *G. soja* accessions and the *Glycine latifolia* (Benth.) C. Newell & Hymowitz accession had numbers significantly higher than on 'Jackson', but they were significantly lower than numbers on 'Williams

Table 2. Number of soybean aphids on various legumes 13 days after aphid-infested leaves were placed on 7-d-old seedlings

Entry	Species	Mean no. of aphids/plant
PI206832	<i>Pisum sativum</i>	0.0a ^a
PI469199	<i>Vicia faba</i>	0.1ab
PI508091	<i>Lens culinaris</i>	0.3abc
PI170474	<i>Vicia sativa</i>	0.3abc
Kiloga	<i>Vigna aureus</i>	0.9abcd
SA1316	<i>Medicago truncatula</i>	1.4bcd
Common	<i>Trifolium repens</i>	1.8cd
SA1306	<i>Medicago truncatula</i>	1.8cd
Top Crop	<i>Phaseolus vulgaris</i>	2.9d
Marathon	<i>Trifolium pratense</i>	3.0d
Jackson	<i>Glycine max</i>	21.6e
Williams82	<i>Glycine max</i>	184.9f
Mean		2.6

Means followed by the same letters are not significantly different by the least significant difference test ($P = 0.05$).

^a Mean of three replications of four plants each.

82'. Considerable variability in aphid counts occurred on plants of *G. argyrea*. There was no significant aphid mortality observed on any of the *Glycine* species.

Stocks of *M. truncatula* had significantly different aphid numbers, indicating differences in susceptibility among the stocks. Numbers of aphids ranged from seven on PI3054 to 97 aphids per plant on PI3115. The overall mean number of aphids on the *M. truncatula* stocks was 23, and the standard error was about three aphids per plant.

Numbers of Aphids on *G. soja* Accessions. Four *G. soja* accessions had aphid numbers not significantly different from the most resistant *G. max* accessions 'Sato', 'Sugao Zarai', and 'Jackson', in experiment 5 (Table 4). Numbers on *G. max* accessions PI87059 and PI88508 and *G. soja* accessions 'G3' and 'Z9' were not significantly different from resistant checks 'Palmetto', 'Dowling', and 'CNS', indicating that they had equal levels of resistance. *G. soja* accession PI522212B

Table 3. Number of aphids on plants of accessions of different *Glycine* species 21 d after exposure to alates during the V_c stage

Entry	<i>Glycine</i> species	Mean no. of aphids/plant
PI233139	<i>falcata</i>	35ab ^a
PI440958	<i>clandestina</i>	36a
PI440963	<i>cyrtoloba</i>	67bc
Jackson	<i>max</i>	68bc
PI505151	<i>argyrea</i>	72abcd
PI440928	<i>canescens</i>	73bc
PI440956	<i>microphylla</i>	82cd
PI505166	<i>curvata</i>	86cd
PI373990	<i>tabacina</i>	87cd
PI483218	<i>tomentella</i>	89cd
PI447003A	<i>soja</i>	143de
PI378709	<i>latifolia</i>	178ef
PI424006A	<i>soja</i>	316f
Williams 82	<i>max</i>	636g
Mean		108

Means followed by the same letters are not significantly different by the least significant difference test ($P = 0.05$).

^a Mean of four two-plant replications.

Table 4. Number of soybean aphids on seedlings of resistant and susceptible *G. max* and *G. soja* germplasm accessions 14 d after aphid infestation

Entry	<i>Glycine</i> species	No. of aphids
S12	<i>soja</i>	7a ^a
Sato (PI548409)	<i>max</i>	15ab
L4	<i>soja</i>	16ab
Taichung 38 (PI518282)	<i>soja</i>	19abcd
Sugao Zarai (PI200538)	<i>max</i>	20ab
Jackson (PI548657)	<i>max</i>	20ab
JS1	<i>soja</i>	23b
Palmetto (PI548480)	<i>max</i>	26bc
CNS (PI548445)	<i>max</i>	26bc
Dowling (PI548663)	<i>max</i>	28bcde
G3	<i>soja</i>	60cdef
Moyashimame (PI87059)	<i>max</i>	61cdef
Z9	<i>soja</i>	66defg
Showa No. 1-4 (PI88508)	<i>max</i>	67efg
Taichung 37 (PI518281)	<i>soja</i>	70fg
PI423993	<i>soja</i>	92fg
PI424006B	<i>soja</i>	96fgh
PI424006A	<i>soja</i>	117fghi
PI4683396B	<i>soja</i>	128fghi
PI447003A	<i>soja</i>	139fghij
PI468918	<i>soja</i>	145fghijk
Ina	<i>max</i>	230hijkl
Williams 82	<i>max</i>	269ijkl
Pioneer 93B01	<i>max</i>	309jkl
Pana	<i>max</i>	311jkl
PI522212B	<i>soja</i>	337kl
Loda	<i>max</i>	437l
Mean		105

Means followed by the same letters are not significantly different by the least significant difference test ($P = 0.05$).

^a Mean of four replications of two plants each.

had aphid numbers similar to 'Loda', the most susceptible *G. max* cultivar.

Results in nonchoice test experiment 6 indicated that *G. soja* accessions 'JS1', 'L4', and 'S12' had a similar antibiosis effect against the soybean aphid as the resistant check 'Dowling' (Table 5). Although the number of offspring produced on accession 'Z9' was lower, it was not significantly different from the susceptible check 'Loda', suggesting that antibiosis had a lesser role in resistance expression in 'Z9' compared with 'JS1', 'L4', and 'S12'. Although alates of uniform age were not used in this study, the magnitude of differences between resistant and susceptible accessions

Table 5. Cumulative number of nymphs produced in 12 d by single *A. glycyines* alates caged on seedlings of *G. max* and *G. soja* germplasm

Entry	<i>Glycine</i> species	No. of aphids
JS1	<i>soja</i>	2a ^a
Dowling	<i>max</i>	2a
L4	<i>soja</i>	3a
S12	<i>soja</i>	8a
Z9	<i>soja</i>	64b
Loda	<i>max</i>	105b
HAS	<i>soja</i>	121b
Mean		15

Means followed by the same letters are not significantly different by the least significant difference test ($P = 0.05$).

^a Mean of four plants.

was great enough to limit the importance of variability in population development due to potential bias of the age of adult used to initiate colonies.

Discussion

Results in this study indicated that *A. glycines* readily colonized *Glycine* species, in particular, *G. max* and *G. soja*, along with a couple of the perennial species such as *G. latifolia*, identifying those species as good hosts of the soybean aphid.

Poor hosts were also identified, including *P. sativum*, species of *Vicia*, and the other species in this study that did not support soybean aphid colonization by *A. glycines*. However, a comprehensive sample of germplasm of these species was not tested; therefore, definitive conclusions about the ability of *A. glycines* to colonize those species cannot be made.

There was limited *A. glycines* colonization on *P. coccineus* and species of *Trifolium* and *Medicago* in these experiments; however, numbers of aphids were generally lower on those species than on the resistant *G. max* accessions. Under field conditions, colonization of these legumes may be less likely to occur because environmental conditions are more variable and aphid pressure would probably be lower than in the greenhouse tests. Paik (1972) listed *P. coccineus*, the scarlet runner bean, as a host of *A. glycines* in Korea. There are no reports of *A. glycines* colonization on legume crops other than soybean in North America.

On the other hand, *A. glycines* was transient on many species in the experiments, temporarily stopping to feed while seeking more susceptible hosts to colonize. Although colonization on those species may not occur in nature, *A. glycines* may still be able to probe or feed on them and acquire viruses, such as *Alfalfa mosaic virus* (Hill et al. 2001), for transmission to virus-susceptible crops.

Results in this study suggested that *A. glycines* might not have a highly restricted secondary host range, although it may still be considered sequentially monophagous compared with highly polyphagous species such as *Myzus persicae* (Sulzer). Most aphids, including heteroecious species, show a high degree of host specificity (Dixon 1987).

Dead aphids, frequently observed on *P. vulgaris* and some of the other legume species, indicated a high level of aphid mortality, possibly due to an antibiotic factor. It is known that *P. vulgaris* leaves contain an alpha amylase inhibitor that inhibits insects' digestive enzymes (Moreno and Chrispeels 1989, Grossi de Sa et al. 1997, Ishimoto et al. 1999). Another possible mechanism for the antibiosis could be the presence of sharp, hooked trichomes on the leaf surface of many *P. vulgaris* cultivars that may impale the aphids (Simmonds and Blaney 1989). Hill et al. (2004) established that antibiosis was an important resistance factor in resistant *G. max* accessions; however, the exact effects on aphid biology and mechanisms of action were not characterized.

Results in this study regarding resistance to *A. glycines* in *Glycine* species were in general agreement with an earlier report from China (Zhuang et al. 1996), except that the accessions of *Glycine canescens* F.J. Herm. & *G. tabacina* (Labill.) Benth. tested in this study were resistant to *A. glycines*, whereas the accessions tested in China were classified as susceptible. Variability for resistance to *A. glycines* may exist in those species. Another explanation could be the existence of variability in host specialization among *A. glycines* populations; however, there are no reports of host specialization or biotypes in *A. glycines*.

Significant differences in aphid colonization on different *M. truncatula* genetic stocks (VandenBosch and Frugoli 2001, Thoquet et al. 2002) could be the basis for genetic, biochemical, and physiological studies of host factors involved in susceptibility or resistance to *A. glycines*. Information discovered in such studies might lead to novel aphid control approaches that could be applicable to soybean.

An earlier report identified three *A. glycines*-resistant *G. soja* accessions after screening ≈ 1000 accessions (Sun et al. 1990). Six new resistant *G. soja* accessions are reported here. Three of them had a strong antibiotic effect on *A. glycines*. They may be novel sources for *A. glycines* resistance, unrelated to those discovered in *G. max*, and could be useful in breeding programs to develop *A. glycines*-resistant soybean cultivars because they can be successfully crossed with cultivated soybean (Singh and Hymowitz 1999).

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