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ABUNDANCE OF RICE ROOT APHID AMONG SELECTED PLANT SPECIES AND ON PLANTS GROWN WITH DIFFERENT SOIL-SURFACE MEDIA

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ABSTRACT

The rice root aphid, *Rhopalosiphum rufiabdominalis* (Sasaki), is distributed worldwide and colonizes a wide range of plants. However, relatively little is known about the suitability of different host plants, optimal rearing techniques, and the aphid's impact on plant fitness. To improve understanding of these factors, laboratory experiments were conducted to compare the abundance of rice root aphid on plants grown using three different soil-surface media and among selected monocotyledonous and dicotyledonous plants. Rice root aphid was more abundant on plants grown with a sandy soil surface than a surface with fine wood chips or only bare non-sandy soil. Rice root aphid was more abundant on 'Elbon' rye than on 'Bart 38,' 'Dart,' 'Fletcher' and 'Ramona 50' wheat. More winged rice root aphids were produced on Elbon rye than on Dart wheat, but the number of winged aphids on Elbon rye did not differ from that on other wheat lines. Rice root aphid was more abundant on Elbon rye and 'TAM 110' wheat than on 'Marmin,' 'Marshall' and 'Sharp' wheat. Additional observations with monocotyledonous plants showed that abundance of rice root aphid on 'Kivu 85' triticale was comparable to that on Elbon rye. Rice root aphid did not reproduce on potato or soybean, although winged adults persisted up to 24 days on caged potato plants. The implications of differential abundance of rice root aphid on plants are discussed in regard to colony rearing, future experiments and possible pest management considerations.

The rice root aphid, *Rhopalosiphum rufiabdominalis* (Sasaki) (Hemiptera: Sternorrhyncha: Aphididae), is a cosmopolitan species that colonizes many kinds of plants (Doncaster 1956, Blackman and Eastop 1994, 2000). Its primary hosts are species of *Prunus* trees (Blackman and Eastop 1994). The rice root aphid colonizes leaves of *Prunus* spp. in the fall and overwinters as eggs on stems. However, rice root aphid typically survives throughout the year in milder climates by continuous parthenogenesis on secondary hosts, which include rice and many other species of Graminae, Cyperaceae, and some dicotyledonous plants (Patch 1938, Etzel and Pettit 1992, Halbert 1996, Tsai and Liu 1998, Blackman and Eastop 2000). The rice root aphid generally infests the roots of its secondary hosts (Etzel and Pettit 1992, Blackman and Eastop 2000), but may also occur on shoots in the field, greenhouse, and laboratory (Kieckhefer and Gustin 1967, Paliwal 1980, Kindler et al. 2004).

Rice root aphid is an indirect pest of many crop plants because of it ability to vector disease agents such as barley yellow dwarf virus (BYDV) (Paliwal 1980, Jedlinski et al. 1981) and sugarcane yellow leaf virus (Schenck and Lehrer 2000). Rice root aphid is often one of the most abundant aphids infesting wheat and other small-grain crops in North America (Palmer 1939, Kieckhefer and Gustin 1967, Paliwal 1980, Chapin et al. 2001). Despite its lengthy history

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and abundance on small-grain crops, tests of direct impact by rice root aphid on plant fitness are lacking.

A critical first step in evaluating the effect of a potential pest aphid on plant fitness is to determine the range of host plants that are capable of supporting large populations of the aphid. Once identified, these plants may then be used as rearing hosts and as test plants for evaluating any impacts of aphid infestations (Blackman 1990). The identification of suitable rearing plants is important, as pretest conditions in which aphids are held may directly affect experimental outcomes (Smith et al. 1994). For instance, the use of a particular plant species or line for rearing and subsequent impact testing may predispose test aphids to feed on it and lead to exaggerated estimates of impact.

It is also important to determine particular conditions that support suitable numbers of aphids on rearing plants or experimental plants or that facilitate the execution of experiments (Blackman 1990, Tsai and Liu 1998). For instance in laboratory experiments with the bird cherry-oat aphid, *Rhopalosiphum padi* (L.), a thin layer of light-colored quartzite sand serves as a useful contrasting surface for tracking this generally dark colored aphid during infestation and evaluative counting, when it may become dislodged from test plants and fall onto the soil surface of experimental arenas (Hesler and Tharp 2005). As bird cherry-oat aphid and rice root aphid closely resemble one another morphologically (Richards 1960, Pike et al. 1990), light-colored sand may also be a useful soil surface in experiments with rice root aphid, even though a dark soil mix is suitable for rearing and for experiments (Paliwal 1980, Kindler et al. 2004). Anecdotal observations have also indicated that population growth of rice root aphid may be facilitated by the placement of a thin layer of fine wood chips on the soil surface around the base of rearing plants (Kindler et al. 2004), but a layer of wood chips may also hinder tracking the aphids during infestation and evaluative counting. Thus, the population growth of rice root aphid needs comparison among different soil-surface treatments, such as sand and wood chips, to determine their utility in future experiments. The objective of this research is to determine the suitability of selected plants and soil media for rice root aphid by measuring the abundance of rice root aphid over time when placed on selected monocotyledonous and dicotyledonous plants.

MATERIALS AND METHODS

Aphids. Rice root aphids used in the experiments were obtained from a virus-free, multiclonal stock colony maintained on 'Elbon' rye plants (Kindler et al. 2004) growing in a 15.2-cm diameter pot covered with a cylindrical cellulose nitrate cage (Hesler and Tharp 2005). The colony was maintained in a growth chamber (Controlled Environments Inc., Pembina, ND, USA) under constant conditions (13 h light at 19°C, 11 h dark at 18°C) at the USDA North Central Agricultural Research Laboratory (NCARL), Brookings, South Dakota. A non-viruliferous colony of rice root aphid was established by collecting numerous individuals from a winter wheat field near Brookings in autumn 1999, placing them on sachets of Parafilm® (American National Can Co., Greenwich, CT, U.S.A.) containing 20% sucrose solution, removing neonate offspring, and transferring them to noninfested rye plants (Hesler and Tharp 2005). This procedure was repeated about once per year with colony aphids, and occasionally leaf tissue was tested serologically (Agdia, Elkhart, IN, U.S.A.) to ensure that colony plants were free of BYDV. The integrity of the colony was also checked weekly by examining a few hundred individuals to ensure no contamination by morphologically similar species such as bird cherry-oat aphid. Winged aphids became present in cages 3 weeks after initial infestation and aggregated on the inner surface of each cylindrical cage. The colony was perpetuated by regularly infesting one-week-old rye plants with winged rice

root aphids obtained from caged plants infested 3 to 4 weeks earlier (Kindler et al. 2004). Voucher specimens of the aphids are deposited at NCARL.

Abundance of rice root aphid on Elbon rye using different soil-surface media. Experimental plants were prepared by germinating seeds between layers of moist paper towels held in plastic containers in the dark at 20°C (Hesler and Tharp 2005). After 24 to 48 h, 25 to 40 individual seedlings exhibiting uniform root and coleoptile growth were planted into a 2:1:1 mixture of Vienna soil (fine-loamy, mixed Calcic Hapludolls), perlite, and coarsely ground coconut shells (Coir, J. R. Johnson Supply Inc., Roseville, MN, U.S.A.). Pots were thinned to 20 seedlings 5 or 6 days later.

Three types of soil-surface media treatments were tested, and they were applied to pots when rye plants were 7-d old. Treatments consisted of adding volumes of 5 oz. of soil mixture, 8 oz. of fine cedar-wood chips, or 5 oz. of light-colored quartzite sand to each test pot and spreading each soil treatment over the soil surface and around the base of test plants. After treatments were applied, the modified soil surface in each pot was sprayed lightly with water. Then, each pot of 20 seedlings was infested with 28 winged rice root aphids selected randomly from the cylindrical cages that had covered colony plants infested 3 to 4 weeks earlier. Infested plants were immediately caged and placed into a growth chamber (13 h light at 19°C, 11 h dark at 18°C) as a randomized complete block design with four replications.

Abundance of rice root aphids was measured 24 d later when plants were about 40-cm tall with 3 to 4 leaves and had dense aphid populations around their basal half. Aphids were counted immediately from plants clipped at the soil surface of each quadrant in an experimental pot (4 plants total). Winged aphids on the cut plants were more likely to escape and were counted first. The numbers of all aphids on each set of 4 plants was summed for each treatment replicate. Additional plants from the pots were gently dug to confirm that rice root aphids had not infested roots, consistent with our previous, unpublished observations. As a second measure of abundance, the number of winged aphids on the inner surface of each treatment cage was also counted. The four-plant counts and counts of winged aphids from cages were subjected to separate analyses of variance (PROC ANOVA; SAS Institute 2002), and treatment means were separated by Tukey's HSD method. A significance level of $\alpha = 0.05$ was used for statistical tests.

Abundance of rice root aphid on selected monocotyledonous plants. The abundance of rice root aphid on monocot plants was evaluated in two separate, quantitative experiments and a third experiment in which abundance was observed but not quantified. The first experiment compared rice root aphid abundance on Elbon rye to that on wheat lines 'Fletcher,' 'Dart,' 'Baart 38,' and 'Ramona 50.' These lines express symptoms of BYDV infection (Oswald and Houston 1953; NGRP 2007a, b, c) and were evaluated as possible hosts of rice root aphid for studies on virus transmission and yield effects. A second experiment compared abundance of rice root aphid on Elbon rye to that on two spring-wheat lines, 'Marshall' and 'Sharp,' and two winter- wheat lines, 'Marmin' and 'TAM 110.' Marshall, Sharp, Marmin, and TAM 110 are lines that are regionally adapted to the northern Great Plains. Each of these two experiments was conducted identically to the soil-media experiment. In addition to these quantitative tests, abundance of rice root aphids was also evaluated qualitatively on 'Kivu 85' triticale, which in preliminary tests appeared to support large populations of rice root aphids. Sets of triticale plants were infested and maintained in the same manner as the quantitative experiments with wheat and rye. A set of Elbon colony plants was infested at the same time and placed into a separate chamber (13 h light at 19°C, 11 h dark at 18°C). After 4 weeks, aphid abundance on triticale and rye was observed *in situ*.

Abundance of rice root aphid on selected dicotyledonous plants. Potato and soybean were evaluated as hosts of rice root aphid in separate experiments. In the first experiment, three potato plants per pot were used. To achieve this, three cuttings of potato tuber (each from a different tuber) were planted about 2 inches deep in a 15.2-cm diameter pot filled with a modified soil mix. The mix was modified to enhance potato growth by adding an equal volume of sand to the original soil mix used with monocots. One of three common, locally available lines of potato ('Irish Cobbler,' 'Norkotah Russet,' or 'Red Pontiac') was planted per pot. A separate control treatment of Elbon rye (20 plants per pot) was also included. Potato plants were three-weeks old and rye plants were one-week old when they were infested on the same date with 28 winged rice root aphids. Infested treatment plants were placed into a growth chamber (13 h light at 19°C, 11 h dark at 18°C) according to a randomized complete block design with four replications. After 24 days, aphids were counted on the potato plants in each pot and from 4 randomly selected rye plants per pot. In a second experiment, seeds of soybean line '91B91' were planted in 15.2-cm diameter pot with soil mix and thinned to two to three plants two weeks later. Each of three pots of soybean was infested with 28 winged rice root aphids, immediately caged, and placed into a growth chamber (16 h light at 22°C, 8 h dark at 19°C). A set of three Elbon colony plants was infested at the same time and placed into a separate chamber (13 h light at 19°C, 11 h dark at 18°C). After 2 weeks, aphid abundance on soybean and rye were observed *in situ*.

RESULTS

Abundance of rice root aphid on Elbon rye using different soil-surface media. Abundance of rice root aphid on Elbon rye varied with soil-surface media ($F = 1.28$, df = 2, 14, $P = 0.0012$). Plants grown with a sandy soil surface ($\bar{x} \pm$ SE = 900.1 ± 92.8 per 4 plants) had more aphids than a surface with fine wood chips ($\bar{x} \pm$ SE = 494.8 ± 54.2 per 4 plants) or only soil ($\bar{x} \pm$ SE = 453.8 ± 53.9 per 4 plants). In addition, more winged aphids ($F = 37.59$, df = 2, 14, $P < 0.001$) were collected from cages in the sand treatment ($\bar{x} \pm$ SE = 240.6 ± 23.9) than with treatments of wood chips ($\bar{x} \pm$ SE = 60.4 ± 8.9) or only soil ($\bar{x} \pm$ SE = 52.1 ± 11.7).

Abundance of rice root aphid on selected monocotyledonous plants. In the first experiment (Table 1), rice root aphid was more abundant on Elbon rye than on Bart 38, Dart, Fletcher and Ramona 50 wheat ($F = 8.10$, df = 4, 12, $P = 0.002$). More winged rice root aphids were collected from cages of Elbon rye than from those of Dart wheat, but the number of winged aphids on Elbon rye did not differ from that of other wheat lines ($F = 3.70$, df = 4, 12, $P = 0.035$). In the second experiment (Table 1), rice root aphid was more abundant on Elbon rye and TAM 110 wheat than on Marmin, Marshall and Sharp wheat ($F = 28.13$, df = 4, 12, $P < 0.001$), and more winged aphids were collected from cages of Elbon rye and TAM 110 wheat than from cages of other wheat plants ($F = 18.67$, df = 4, 12, $P < 0.001$). Additional observations showed that rice root aphid became highly abundant on Kivu 85 triticale, with abundance appearing comparable to that on Elbon rye infested for the same length of time. Stems and lower leaves of rye and triticale were virtually covered with dense colonies of rice root aphids, and winged aphids were abundant on the inner surfaces of cages.

Abundance of rice root aphid on selected dicotyledonous plants. Winged rice root aphids were found on potato and soybean plants within a few hours after being introduced into test cages, and they remained active in test arenas for at least several days. In potato tests, alates were generally observed on plants rather than on the inner surface of cages. After two weeks, however, dead alates began to appear increasingly on the soil surface of the potato test arenas, but several alates survived on potato throughout the 4-week test period. However, no offspring were found on the shoots, roots, or tubers of potato plants

Table 1. Abundance of rice root aphid ($\bar{x}$ ± SE) after 24 d of infestation on rye and wheat.

Plant line	Number per 4 plants	Number of winged per cage surface[1]
Experiment 1		
Elbon rye	1669.0 ± 261.7a	162.8 ± 35.7a
Baart 38 wheat	825.5 ± 168.3b	108.5 ± 17.3ab
Dart wheat	331.5 ± 132.6b	50.8 ± 12.7b
Fletcher wheat	544.0 ± 126.9b	83.3 ± 22.3ab
Ramona 50 wheat	749.8 ± 95.4b	76.8 ± 8.7ab
Experiment 2		
Elbon rye	1064.5 ± 50.9a	291.3 ± 42.8a
Marmin wheat	457.5 ± 116.4b	84.3 ± 14.1b
Marshall wheat	433.3 ± 20.9b	83.5 ± 18.9b
Sharp wheat	249.8 ± 20.9b	53.0 ± 12.1b
TAM 110 wheat	1019.5 ± 97.3a	190.5 ± 16.7a

For each column within an experiment, means ± SE followed by different letters are significantly different (Tukey's HSD test, $\alpha = 0.05$).
[1] Twenty plants per cage.

after 24 days. After 24 days in the potato test, Elbon rye had 1816.8 ± 466.5 ($\bar{x}$ ± SE) aphids per 4 plants and 290.8 ± 170.8 ($\bar{x}$ ± SE) winged aphids per cage. Most of the winged aphids on soybean died in about one week and no offspring were found on soybean plants, but after 4 weeks Elbon rye was heavily infested with hundreds of rice root aphids per stem and many alates were found on the inner surface of cages.

DISCUSSION

The abundance of rice root aphid was greater on plants growing above a sandy soil surface than with surfaces of wood chips or soil mix. Our test was not designed to determine why rice root aphids were more abundant in the sand treatment, and this may be addressed in future studies. Nonetheless, the use of light colored sand could facilitate tracking aphids during infestation and counting, and the use of a sandy surface is recommended especially for laboratory and greenhouse experiments involving infestations of shoots with rice root aphids. The sandy surface could also enhance colony production of rice root aphids on Elbon rye.

Rice root aphid had differential population growth among various plant species and no population growth on potato, which had been reported previously as a host (Essig 1944). The population of rice root aphids in this study was collected from a field of 'Roughrider' wheat, and results showed that it was well adapted to rye, wheat and triticale, but not potato or soybean. Previous results have shown that this population of rice root aphid also becomes only moderately abundant on barley and oats and is poorly adapted to rice and other grasses (Kindler et al. 2004), but other North American populations of rice root aphid have been well adapted to barley and oats (Paliwal 1980). Collectively, these results suggest that the population of rice root aphid in this study may represent a biotype based on its differential survival and development on particular host plants (Eastop 1973, Diehl and Bush 1984, Drés and Mallet 2002).

The rice root aphid did not reproduce on potato and soybean, but alates had prolonged survival on potato and limited survival on soybean. The survival of

alates on potato and soybean raises the question of whether rice root aphid may be a vector of plant-disease viruses in these crops, but its ability to transmit viruses to potato and soybean is unknown. Several congeneric aphid species do not colonize potato or soybean, but they are a vector of stylet-borne plant-disease viruses to these crops. For instance, both corn leaf aphid, *Rhopalosiphum maidis* (Fitch), and bird cherry-oat aphid transmit potato leafroll virus to potato (Halbert et al. 2003) and soybean mosaic virus to soybean (Halbert et al. 1981), and *Rhopalosiphum insertum* (Walker) is capable of transmitting potato virus Y to potato (van Hoof 1980).

Rice root aphid may be prevalent in regions of North America where wheat is grown near soybean and potato (Kieckhefer and Gustin 1967, Al-Raeesi et al. 1992, Chapin et al. 2001, Kindler et al. 2004), but it has not been considered significant in the epidemiology of aphid-borne viruses in potato and soybean. There is no record of rice root aphid colonizing soybean, and it composes <<1% of alates captured in traps within soybean fields (Halbert et al. 1981). Alate rice root aphids have not been trapped in Minnesota and North Dakota potato fields (DiFonzo et al. 1997). However, as rice root aphid (as *Cerosipha californica*) has been recorded on potato in California (Essig 1944), and given the prolonged survival of alates on potato in the present study, tests of its ability to transmit viruses to potato may be warranted.

Rice root aphids in this study differed in abundance among wheat lines, with greatest numbers on TAM 110 wheat and with decreased abundance of winged aphids on Dart wheat. The differential abundance of rice root aphid among wheat lines has some implications for rearing and experimentation. First, TAM 110 was the only wheat line with an abundance of rice root aphid comparable to that on Elbon rye. Abundance of rice root aphid on Kivu 85 triticale, a wheat × rye cross, was also comparable to that of Elbon rye. Thus, TAM 110 and Kivu 85 may be equally suitable to Elbon rye as rearing hosts. However, the differential abundance of rice root aphid among wheat and triticale lines suggests that experiments to test for an impact of rice root aphid on the growth and grain yield need to be designed to maintain an equal number of aphids across test plants over time (Lamb and MacKay 1995). From a pest management standpoint, the differential abundance of rice root aphids among wheat lines suggests inherent variation in wheat that could be exploited to develop and eventually deploy lines that limit aphid infestations (Webster 1991).

One objective of this study was to identify wheat lines that support large numbers of rice root aphid and also readily express symptoms of BYDV infection for possible use in studies on virus transmission and yield effects. The wheat lines Baart 38, Dart, Fletcher and Ramona 50 are moderately to extremely susceptible to BYDV (Oswald and Houston 1953; NGRP 2007a, b, c), and rice root aphid was fairly abundant on these lines, although relatively low numbers of aphids were produced on Dart. Thus, Baart 38, Fletcher and Ramona 50 may be useful as colony plants for maintaining rice root aphids and BYDV, and in future experiments to determine the effects of rice root aphid and BYDV on wheat growth and yield.

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