Beat Sampling Accuracy in Estimating Spruce Spider Mite (Acari: Tetranychidae) Populations and Injury on Juniper

PAULA M. SHREWSBURY¹ AND MARK R. HARDIN^{2, 3}

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ABSTRACT The use of a standardized beat sampling method for estimating spruce spider mite, Oligonychus ununguis (Jacobi) (Acari: Tetranychidae), densities on a widely used evergreen ornamental plant species, Juniperus chinensis variety 'Sargentii' A. Henry (Cupressaceae), was examined. There was a significant positive relationship between total spruce spider mite densities and spider mite densities from beat sampling on juniper. The slope and intercept of the relationship may be used by pest managers to predict total spider mite densities on plants from beat sample counts. Beat sampling dramatically underestimates the total number of spider mites on a foliage sample. The relationships between spruce spider mite feeding injury and spider mite density estimates from beat sampling juniper foliage and total spider mite counts on foliage were also examined. There was a significant positive relationship between spruce spider mite density as estimated from beat sampling and injury to the plants. There was a similar positive relationship between the total number of spruce spider mites and injury to the plants, suggesting that a pest manager could use beat sampling counts to estimate plant injury and related thresholds. These findings have important implications to decision-making for spruce spider mite control, especially as it relates to threshold levels and determining rates of predator releases. Further assessment of the effectiveness of beat and other sampling methods across multiple spider mite-host plant associations needs to be examined to enable pest managers to select sampling plans that are feasible and reliable.

KEY WORDS biological control, integrated pest management, monitoring, injury, threshold

SPIDER MITES ARE KEY PESTS in many cropping systems (van de Vrie et al. 1972), including ornamental plant systems where they attack a variety of woody and herbaceous plants (Raupp and Hoitink 1996, Skirvin and De Courcy Williams 1999, Shrewsbury and Skinner 2002). In most agricultural crops, yield determines the economic value of the crop, whereas with ornamental plants it is the aesthetic appearance of the plant that directly relates to its economic value (Sadof and Raupp 1996). Because of this, tolerance to plant injury or action thresholds for spider mites are often lower in ornamental systems than traditional agricultural systems (van de Vrie 1985, Sadof and Raupp 1996, Skirvin and De Courcy Williams 1999).

Accurately estimating spider mite population densities is critical to successful management of spider mites. Accurate population estimates are vital for establishing the relationship between spider mite densities and plant injury, which in turn form decisionmaking guidelines that determine action thresholds for chemical, biological, and other control measures (Binns and Nyrop 1992). Accurate population estimates are also important in determining appropriate release rates of biological control agents to suppress spider mite populations (Opit et al. 2003, Shrewsbury and Hardin 2003). These considerations have become especially important in recent years with increasing emphasis on reducing pesticide inputs and implementing alternative management tactics such as biological controls (Garber et al. 1996, Skirvin and De Courcy Williams 1999).

Sampling or monitoring spider mites is challenging due to their small size, rapid reproduction, and patchy distribution (Wilson and Morton 1993). Two methods commonly used to sample spider mites include a binomial (presence/absence) sampling method and a beat sampling method. Binomial sampling entails counting the proportion of infested leaves rather than the total number of spider mites on the sampling unit (Binns and Nyrop 1992, Opit et al. 2003). Binomial sampling methods have been developed to monitor populations of spider mites for several cropping systems such as peanuts (Margolies et al. 1984), strawberries (Raworth 1986), cotton (Wilson and Morton 1993), and apples (Nyrop et al. 1994). This method has only been examined for a few ornamental plant systems, field-grown roses (Karlik et al. 1995) and greenhouse-grown geraniums (Opit et al. 2003).

The most frequently recommended method of sampling spider mite populations on ornamental plants is

¹Department of Entomology, University of Maryland, College Park, MD 20742-4454.

² Horticultural Services Division, Smithsonian Institution, Arts & Industry Bldg., Washington DC 20560–0420.

³ Current address: Office of Safety and Regulatory Affairs, Howard County Public School System, 10920 Rte. 108, Ellicott City, MD 21042.

the beat sampling technique (Davidson et al. 1988, Nielsen 1989, Studebaker 1992, Lehman 1998, Dreistadt 2001, and numerous extension fact sheets and Web sites; P.S., personal observation). Beat sampling typically consists of placing a sheet of white graph paper (10 by 14 cm) on a clipboard beneath the foliage of a plant and striking the foliage, usually with a wooden dowel or something similar. The foliage is beaten a standardized number of times, and the number of active spruce spider mites (and sometimes natural enemies) on the paper is counted. This is often repeated multiple times per plant to obtain an average spider mite density per plant. To our knowledge, there are no studies examining the accuracy of this monitoring method for estimating total spider mite populations or the relationship between beat sampling mite counts and the total number of mites on a plant. Additionally, few studies have examined the relationship between spider mite densities from beat sampling and plant injury from spider mites on ornamentals (Sadof and Alexander 1993).

Recent unsuccessful efforts in spruce spider mite, Oligonychus ununguis (Jacobi) (Acari: Tetranychidae), control by using acaricides and augmentative release of predatory mites (Shrewsbury and Hardin 2003) raised questions as to the accuracy of beat sampling in estimating spider mite population densities. Shrewsbury and Hardin (2003) found that augmentative release of predatory mites was less effective than conventional miticides in suppressing spruce spider mite densities on junipers. Moreover, both biological and chemical control measures resulted in levels of plant injury that were likely unacceptable to the consumer. Beat sample counts used to estimate predator release rates may have underestimated total spider mite densities. Underestimation of spider mite densities resulted in release of too few predatory mites, likely reducing the effectiveness of predator releases. In addition, pesticide treatments were applied when spider mite populations were high and injury to the plants had already occurred (Shrewsbury and Hardin 2003).

This study examined the usefulness of the beat sampling method for estimating spruce spider mite densities on a widely used evergreen ornamental, Juniperus chinensis variety 'Sargentii' A. Henry (Cupressaceae). The specific objectives of this study were to 1) elucidate the relationship between the number of spruce spider mites removed by a standard beat sampling method and the total abundance of spruce spider mites across a range of spider mite densities; and 2) examine the relationships between spider mite feeding injury and spruce spider mite density estimates from beat sampling and total spider mite counts.

Materials and Methods

Mite Brush Machine Accuracy. A mite brush machine (Leedom Engineering, Twain Harte, CA) was used to determine the number of spider mites present on foliage samples from juniper. Twenty-eight plants were randomly selected from landscape-grown junipers with an active infestation of spruce spider mite. One sample $(15.24 \text{ by } 15.24 \text{ by } 15.24 \text{ cm } [3,539.6 \text{ cm}^3])$ was collected from each plant. Samples were placed in plastic bags, put in a cooler, and returned to the laboratory where spruce spider mites were counted. Each sample was run through the mite brush machine for 1.5 min. Dislodged mites were collected on a 12.7cm-diameter glass plate covered with a thin film of dishwashing soap (to reduce movement of mites). The glass plate was examined under a dissecting microscope $(25\times)$, and active stages of mites were counted and recorded. The foliage sample was then thoroughly examined under the dissecting microscope, and any remaining active stages of spruce spider mites were counted and recorded.

Beat Sampling Accuracy in Predicting Total Mite Abundance. To determine the accuracy of the beat sampling method in estimating total spruce spider mite densities, spruce spider mite counts from beat sampling and mite brush machine sampling were taken from infested junipers. Total spider mite densities were estimated as the sum of beat and brush machine mite counts. Junipers were maintained in the outdoor nursery yard under conventional cultural practices at the University of Maryland Greenhouse Facility in College Park, MD. Junipers were grown in #3 containers (10.4 liters, 24.5 cm in diameter), \approx 3 yr old, and of equal size (0.42 m³ of foliage). Fifty-four junipers were sampled on 23 May, 1 June, 20 June, 8 September, and 3 November 2000. To reduce possible bias in sampling method, the same individual "beat" all plants, and samples were always selected from a similar location on each plant and of uniform size. The volume of foliage sampled was $\approx 3,539.6 \text{ cm}^3$ (15.24 by 15.24 by 15.24-cm section) and located on the lower, outer portion of the plant. Beat sampling was conducted using a standardized method by placing a sheet of white graph paper (10 by 14 cm) on a clipboard under a sampling unit of foliage (same size as described above). The foliage was beat 10 times with a 0.5-m dowel, and the number of active spruce spider mites on the graph paper was counted. Mite brush machine counts were taken from the same unit of foliage sampled by beat sampling. The area of foliage used in the beat sample was removed, placed in a plastic bag, put in a cooler, and transported to the laboratory. Samples were run through the mite brush machine, and spruce spider mites were counted as described above.

Spider Mite Density and Plant Injury. To examine the relationships between spruce spider mite density as estimated by beat sampling and total spider mite density, and spider mite feeding injury, plant injury levels were estimated for 27 junipers. Plant injury was assessed on 28 July 2000 when spider mites were no longer active and maximum feeding injury had accrued. Four individuals with entomological training conducted plant injury ratings. Each individual visually assessed the percentage of leaf area showing visible injury (discoloration) by spider mites by using an incremental scale, where 0 is no spider mite injury, 1 is 1–10%, 2 is 11–20%, 3 is 21–30%, 4 is 31–40%, 5 is 41–50%, 6 is 51–60%, 7 is 61–70%, 8 is 71–80%, 9 is 81–90%, and 10 is 91–100% leaf injury (Sclar et al. 1998, Shrewsbury and Hardin 2003). An average plant injury level was determined for each plant. Spruce spider mite densities were estimated from beat sampling and total mite counts (=beat + brush sampling) for the same 27 plants used for the injury ratings.

Statistical Analysis. To examine the effectiveness of the mite brush machine for removing active stages of spruce spider mites, the average percentage of spruce spider mites brushed from the plant was determined (PROC MEANS, SAS Institute 1999). A Pearson's correlation (PROC CORR, SAS Institute 1999) was used to determine whether spruce spider mite density influenced the effectiveness of the mite brush machine. To determine the relationship between spruce spider mite density, as estimated from beat sampling, and total spruce spider mite density, as estimated from the sum of mite brush machine and beat sampling counts, data were first log₁₀ transformed to meet the assumptions of normality and homogeneity of variances. Data were then analyzed by an analysis of covariance (ANCOVA) (PROC MIXED, SAS Institute 1999) to determine whether sampling date influenced the relationship between beat sampling and the total density of spruce spider mites. Because there was no significant date by total spider mite density interaction, data were pooled across dates and analyzed with a regression analysis (PROC REG, SAS Institute 1999), where total spider mite density was the independent variable (x) and spider mite density as estimated by beat sampling was the dependent variable (y). This is a part-whole relationship where spider mite density from beat sampling is part of the whole, total spider mite density and therefore must be correlated at some level. Consequently, a P value in not meaningful and is not reported. Although beat sample mite counts are truly a response to total mite counts, pest managers beat sample plants and then need to predict total mite densities from beat sample counts. Therefore, an inverse prediction equation was used to predict total spruce spider mite abundance (independent variable) from spruce spider mite abundance as estimated from beat sampling (dependent variable) (Zar 1999). The inverse prediction equation x = (y)(-a)/b is an algebraic rearrangement of the linear regression equation (y = a + bx), where y is the dependent variable (beat sampling mite density), x is the independent variable (total mite density), b is slope of the relationship, and *a* is the *y* intercept (Zar 1999). A regression analysis (PROC REG, SAS Institute 1999) was also used to examine the relationships between spruce spider mite density as estimated by beat sampling and total spider mite density, and spider mite feeding injury to plants.

Results

Mite Brush Machine Accuracy. The mite brush machine removed $95.5 \pm 1.0\%$ (mean \pm SEM) of all active stages of spruce spider mite on the juniper samples. Effectiveness ranged from 81.8 to 100%. There was a significant correlation between the number of spruce spider mites removed by the mite brush machine and the number of mites missed (r = 0.78, df = 27, P = 0.0001). However, there was not a significant correlation between the number of spruce spider mite brushed and the percentage of mites missed (r = 0.12, df = 27, P = 0.55), indicating there was no density bias in the accuracy of the mite brush machine.

Beat Sampling Accuracy in Predicting Total Mite Abundance. The total density of spruce spider mites as estimated by the sum of mite brush machine and beat samples on juniper ranged from 0 to 7,928 and beat sampling counts ranged from 0 to 1,230 spruce spider mites per sampling unit (3,539.6 cm³) of foliage. There was a positive relationship between total spruce spider mite density and beat sampling spider mite density on juniper ($R^2 = 0.789$; df = 1, 264; y = -0.12 + 0.798x) (Fig. 1a). Nontransformed data are presented for ease of interpretation (Fig. 1b).

Predicting the total abundance of spruce spider mites based on beat sample counts can be done using the inverse prediction equation (Zar 1999) and the relationship between total mite and beat sample mite counts (Fig. 1a). The inverse prediction equation [x = (y - a)/b] for this study system is $x = (\log_{10} \text{ of no. of}$ mites beat + 0.12)/0.798. Because the regression equation was calculated from \log_{10} transformed data, the number of spider mites beat must be \log_{10} transformed when used in the inverse prediction equation. In addition, once x is calculated, the anti-log of x should be determined to get the total number of spider mites.

Spider Mite Density and Plant Injury. For the spider mite densities examined in this study, plant injury ratings on juniper ranged from 5.25 to 7.25 (53–73% feeding injury, respectively). There was a significant positive linear relationship between spruce spider mite density (*x*) as estimated from beat sampling and injury to the plants (*y*) ($R^2 = 0.43$; df = 1, 25; P = 0.0002; y = 5.37 + 0.0007x) (Fig. 2a). There was a similar relationship between the total number of spruce spider mites (*x*) and injury to the plants (*y*) ($R^2 = 0.54$; df = 1, 24; P = 0.0001; y = 5.24 + 0.0002x) (Fig. 2b).

Discussion

Several studies have demonstrated the importance of accurately estimating spider mite densities to achieve successful control of spider mites (Hamlen and Lindquist 1981, Hamlen and Poole 1982, Strong and Croft 1996, Skirvin and De Courcy Williams 1999, Shrewsbury and Hardin 2003). In greenhouse crops, Hamlen and Lindquist (1981) demonstrated it was important to introduce predators at low densities of spider mites to prevent plant damage as predators took from 1 to 3 wk to obtain control. Shrewsbury and Hardin (2003) suggested that underestimation of spider mite populations resulted in failure of predator releases to control spruce spider mites. Hamlen and Poole (1982) examined different predator: prev ratios to control twospotted spider mite by using Phytoseiulus macropilis (Banks). Predator: prey ratios of 1:5 and



Fig. 1. (a and b) Relationship between the total number of spruce spider mites on the foliage of *J. chinensis* variety Sargentii (x) and the number of spruce spider mites beat sampled (y) (regression equation y = -0.12 + 0.798x). Log₁₀-transformed (a) and nontransformed (b) data presented for ease of interpretation.

1:10 suppressed spider mite populations below damaging levels, whereas 1:20 and controls (no predators) resulted in significant damage to plants. These studies demonstrated the need to accurately estimate spider mite populations so predators can be released at rates known to suppress spider mite populations.

The data presented in this article demonstrated the beat sampling method could be used in estimating total spider mite densities. Pest managers need to be aware that beat sampling underestimates the total density of spider mites on a sample of foliage. We were able to determine the relationship between total spruce spider mite densities and spruce spider mite densities from beat sampling on juniper. Knowing this relationship allows pest managers to use an inverse prediction equation to determine total spider mite densities from beat sampling counts. For example, in our study system, a beat sample of 100 spruce spider mites actually means 453.6 mites were present on the sampled foliage. This underestimation has important implications for management decisions such as the release rates of predators. The relationship between beat sampling and total spider mite densities found in this study are likely applicable to other mite-host plant systems, especially plants with architectures similar to junipers.

Another important component of pest management decision-making is the relationship between pest densities and plant injury levels. A pest manager needs to be able to sample spider mites and determine whether densities are near action thresholds for implementing control measures to prevent pests from reaching economic injury levels. This is especially true for control measures, such as augmentative biological control, where there may be a lag time between control implementation and pest suppression (Hamlen and Lindquist 1981). The studies presented here identified a positive linear relationship between spruce spider mite counts based on beat sampling and plant injury. We also determined that this relationship was similar to that of total spider mite counts and plant injury. Using another spider mite-host plant system, Sadof



Fig.2. (a and b) Relationships between the number of spruce spider mites beat sampled (*x*) and percentage of plant injury (*y*) (regression equation y = 5.37 + 0.0007x) (a) and between the total number of spruce spider mites on the foliage (*x*) and percentage of plant injury (*y*) (regression equation y = 5.24 + 0.0002x) (b). Note x-axes differ.

and Alexander (1993) examined the relationship between twospotted spider mite densities, as estimated from beat sampling, and plant injury on burning bush, *Euonymus alatus* (Thun.) Sieb., and found that spider mite densities were also positively correlated to levels of plant injury. These studies suggest that a pest manager could use beat sampling to estimate plant injury and related thresholds.

Binomial sampling has been demonstrated to be an economically feasible and accurate method of sampling spider mite populations in several crop systems (Margolies et al. 1984, Raworth 1986, Wilson and Morton 1993, Nyrop et al. 1994, Karlik et al. 1995, Opit et al. 2003). To improve management of spider mites by using biological and other control measure, future research should examine relationships between standardized beat sampling and total mite densities across several mite and host plant associations. In addition, the use of binomial sampling plans to estimate spider mite densities on ornamental plants should be explored to allow an objective assessment of the accuracy of different sampling methods and enable pest managers to select sampling plans that are feasible and reliable.

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