

# *Matsucoccus macrocitrices* (Hemiptera: Matsucoccidae): First Report, Distribution, and Association With Symptomatic Eastern White Pine in the Southeastern United States

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J. Econ. Entomol. 106(6): 2391–2398 (2013); DOI: <http://dx.doi.org/10.1603/EC13251>

**ABSTRACT** We provide the first report of *Matsucoccus macrocitrices* Richards (Hemiptera: Matsucoccidae) feeding and reproducing on eastern white pine, *Pinus strobus* L., in the southeastern United States. Until now, *M. macrocitrices* had been reported only from the Canadian Atlantic Maritimes, New Hampshire, and Massachusetts. Entomological holdings of 27 major museums in eastern North America have no historical records for *M. macrocitrices* from the southeastern region. However, our field surveys and molecular analyses (DNA barcoding) have resulted in the collection and positive identification of *M. macrocitrices* in Georgia, North Carolina, South Carolina, Tennessee, Virginia, and West Virginia. In addition to the new geographic range, *M. macrocitrices* is also being associated with dieback and mortality of all diameter classes of *P. strobus* leading to concern about a potential shift from its historically nonpestiferous presence on the host tree. On *P. strobus*, *M. macrocitrices* was found embedded in cankers or present on top of the bark with necrotic tissue under their feeding area, indicating that they may be creating wounds for opportunistic pathogenic fungi to infect. Further, we found *M. macrocitrices* living outside of the epiphytic mats of its symbiotic fungus, *Septobasidium pinicola* Snell. This study shows that *M. macrocitrices* is now widespread in the southeastern United States, with implications for the future survival and regeneration of *P. strobus* in eastern North America.

**KEY WORDS** eastern white pine, Matsucoccidae, *Matsucoccus macrocitrices*, *Pinus strobus*, southeastern United States

The genus *Matsucoccus* Cockerell, commonly known as the pine bast scales or matsucoccids (Hemiptera: Coccoidea: Matsucoccidae), is a group of widely distributed scale insects in temperate, tropical, and subtropical areas of the world. There are 32 extant *Matsucoccus* species worldwide, with 19 in North America that all feed exclusively on trees in the Pinaceae family (Ben-Dov 2005, 2012). Eastern North American forests have five species of *Matsucoccus*: 1) *Matsucoccus alabamiae* Morrison; 2) *Matsucoccus banksianae* Ray and Williams; 3) *Matsucoccus gallicolus* Morrison; 4) *Matsucoccus macrocitrices* Richards; and 5) *Matsucoccus matsumurae* (Kuwana), a nonnative species for which *Matsucoccus resinosa* Bean and Godwin has

recently been synonymized (Morrison 1939, Parr 1939, Richards 1960, Watson et al. 1960, Ray and Williams 1991, Koszta 1996, Booth and Gullan 2006; Table 1). Among these scale species, *M. gallicolus* has the widest native host and distribution range in eastern North America. *M. alabamiae* is reported only from pine trees in Alabama; *M. banksianae* from jack pine (*Pinus banksiana* Lambert) in Minnesota; *M. macrocitrices* from eastern white pine (*Pinus strobus* L.) in the Canadian Maritime Provinces, New Hampshire, and Massachusetts; and *M. matsumurae* from red pine (*Pinus resinosa* Aiton) in Connecticut, Massachusetts, New Hampshire, New Jersey, New York, Pennsylvania, and Rhode Island (Table 1).

Several *Matsucoccus* species are known to be pestiferous and economically important, as their feeding activities have been implicated in dieback and mortality of both mature and young pine trees. For example, in the southwestern United States, the pinyon needle scale, *Matsucoccus acalyptus* Herbert, can cause defoliation and mortality of pinyon pine (*Pinus edulis* Engelm.) after repeated feeding (McCambridge and Pierce 1964). The nonnative *M. matsumurae* was responsible for foliage discoloration (Bean and Godwin 1955) and mortality of plantation-grown *P. res-*

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**Table 1.** Pine hosts and collection localities of *Matsucoccus* scale species documented in eastern North America

Scale species	Pine hosts in eastern North America	Collection locality in eastern North America	Citations
<i>M. alabamiae</i> Morrison	<i>Pinus</i> spp.	Alabama	Morrison (1939), Drooz (1985)
<i>M. banksianae</i> Ray & Williams	<i>P. banksiana</i> Lambert	Minnesota	Ray and Williams (1991)
<i>M. gallicolus</i> Morrison	<i>P. echinata</i> Miller, <i>P. elliotii</i> Engelm., <i>P. glabra</i> Walter, <i>P. ponderosa</i> Douglas ex Lawson, <i>P. pungens</i> Lambert, <i>P. resinosa</i> Aiton, <i>P. rigida</i> Miller, <i>P. serotina</i> Michaux, <i>P. taeda</i> L., <i>P. virginiana</i> Miller	Connecticut, District of Columbia, Florida, Georgia, Kentucky, Massachusetts, Maryland, Maine, Missouri, North Carolina, New Hampshire, New Jersey, New York, Ohio, Pennsylvania, Rhode Island, Tennessee, Virginia	Morrison (1939), Parr (1939), Drooz (1985), Kosztarab (1996)
<i>M. macrocitrices</i> Richards	<i>P. strobus</i> L.	Massachusetts, New Brunswick, New Hampshire, Nova Scotia, Ontario, Quebec	Richards (1960), Watson et al. (1960), Martineau (1964), Drooz (1985), Kosztarab (1996), Foldi (2004)
<i>M. matsumurae</i> (Kuwana)	<i>P. densiflora</i> Siebold & Zuccarini <sup>a</sup> , <i>P. resinosa</i> , <i>P. rigida</i> , <i>P. tabulaeformis</i> Carrière <sup>a</sup> , <i>P. thunbergii</i> Parlatores <sup>a</sup>	Connecticut, Massachusetts, New Hampshire, New Jersey, New York, Pennsylvania, Rhode Island	Bean and Godwin (1955, 1971), Drooz (1985), Kosztarab (1996), Li and Zhao (1997), New Hampshire Division of Forests and Lands (2012), USDA Forest Service, Forest Health Protection (2013)

<sup>a</sup> Asian pine species.

*inosa* in Connecticut (McClure 1983). *M. gallicolus* was responsible for killing terminal leaders on >63% of pitch pine (*Pinus rigida* Miller) trees in Pennsylvania, and caused >50% mortality in pitch pine stands in Massachusetts (Parr 1939).

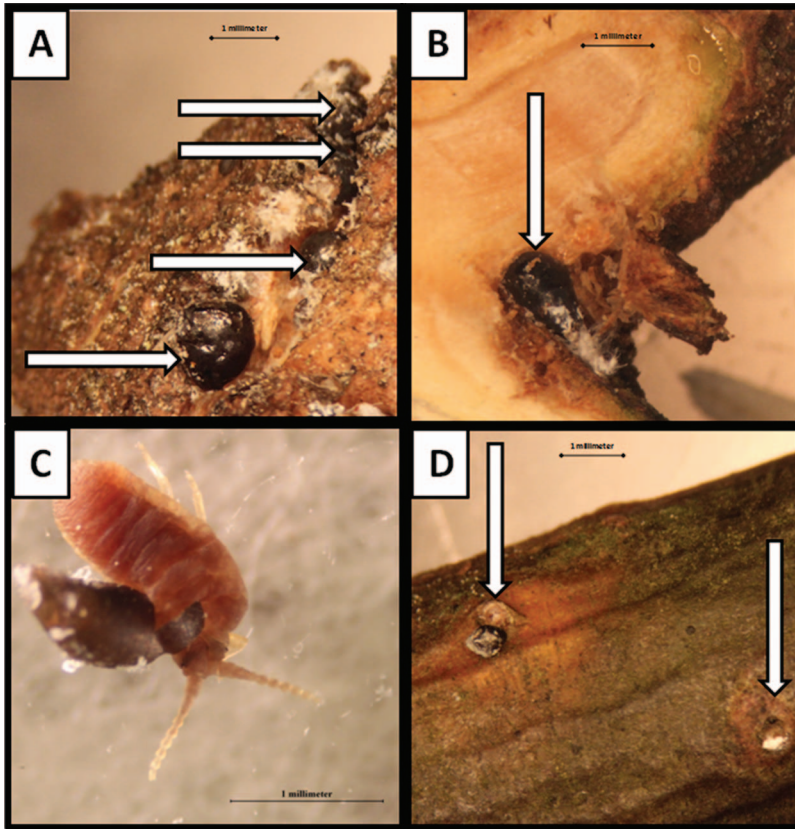
Recently, eastern white pine (*P. strobus*) trees in mixed and pure stands started showing dieback in the Appalachian Mountains of Virginia (2006) and Georgia (2011). Symptoms included branch flagging, crown thinning, resinosis at branch crotches and on the main stem, and cankers on all diameter classes of trees. Follow-up visits to several locations revealed an increase in the number of symptomatic trees as well as tree mortality. On close inspection of branches and main stems, small black immature stages of a scale insect were found embedded in the majority of the cankers, under lichen, and in branch crotches (Fig. 1A and B). In 2007, scale specimens from Bath County, VA, were sent to the Florida Department of Agriculture and Consumer Services where they were tentatively identified as an immature instar of a *Matsucoccus* species based on morphological characteristics (G. Hodges, personal communication). The taxonomy of *Matsucoccus* is based on adult females and immature cyst-stage specimens cannot be identified to species based on morphology. The only known *Matsucoccus* scale to use *P. strobus* as a host is *M. macrocitrices*, but this species has never been documented as being associated with severe dieback or mortality of the host tree, nor has it been reported south of Massachusetts.

*P. strobus* is a long-lived, ecologically and economically important conifer species in North America with a native range encompassing 31 states and provinces (Fig. 2). It is a dominant or codominant tree in 30 Society of American Foresters forest cover types, has been planted to aid in soil reclamation, and is used by

a number of vertebrate wildlife species for food, protection, and shelter (Wendel and Smith 1990). In addition, *P. strobus* is commercially valuable as both a timber and Christmas tree species in the United States (Wendel and Smith 1990). In the northeastern region of the United States, *P. strobus* is one of the most harvested conifer species and is listed as having the highest volume of sawtimber in four states (Maine, Massachusetts, New Hampshire, and Rhode Island; Butler et al. 2012a,b; McCaskill and McWilliams 2012; Morin and Woodall 2012).

Similar to Georgia and Virginia, dieback and mortality of *P. strobus* has also been reported in Maine, New Hampshire, Ontario, and West Virginia, and has been attributed largely to pathogens such as *Caliciopsis pinea* Peck (Lombard 2003, Maine Forest Service 2008, Rose 2011, Llewellyn 2013). Examination of symptomatic *P. strobus* samples from New Hampshire and West Virginia revealed immature insects of a *Matsucoccus* species embedded in the majority of the cankers, including the *C. pinea* cankers. A cursory examination of the fungi associated with cankers and feeding scales of *Matsucoccus* on *P. strobus* in Virginia revealed the presence of a number of common, opportunistic, typically nonaggressive fungal pathogens including *C. pinea*, *Diplodia scrobiculata* de Wet, Slippers & Wingfield, *Fusarium chlamydosporium* Wollenw. & Reinking, and *Fusarium acuminatum* Ellis & Everh. (Cram et al. 2009).

Our research objectives were to: 1) identify the *Matsucoccus* species present on symptomatic *P. strobus*; 2) determine the geographic distribution of this *Matsucoccus* species in the southeastern United States; and 3) conduct a cursory examination of the fungi associated with symptomatic trees of *P. strobus* in



**Fig. 1.** *M. macrocitrices* (from Georgia): (A and B) Overwintering cyst stage embedded in cankers. (C) Emerging adult. (D) Necrotic plant tissue under feeding sites. Arrows indicate *M. macrocitrices* individuals (A and B) or feeding sites (D). (Online figure in color.)

Georgia, including the tissue found with feeding *Matsucoccus* scales.

### Materials and Methods

***Matsucoccus* Identification.** In January, 2012, collections of immature scale insects from the Chattahoochee National Forest (Habersham County), GA, were stored in 90% EtOH and sent to The Australian National University (ANU) and The University of Queensland (UQ), Australia, for identification using DNA sequence analysis. DNA was extracted non-destructively from individual specimens and amplified for nuclear rDNA genes (18S and 28S), which have been shown to be useful in identifying species of *Matsucoccus* (Booth and Gullan 2006). An  $\approx 600$  bp fragment of 18S was amplified using primers 2880 and B- (Von Dohlen and Moran 1995), and the D2–D3 domains of 28S ( $\approx 750$  bp) were amplified with S3360 (Dowton and Austin 1998) and A335 (Whiting et al. 1997). Polymerase chain reaction (PCR) was carried out in 25  $\mu$ l volumes that included 13  $\mu$ l H<sub>2</sub>O, 5  $\mu$ l MangoTaq 5 $\times$  PCR buffer (Bioline, Sydney, Australia), 2  $\mu$ l dNTP (2 mM), 1.5  $\mu$ l MgCl<sub>2</sub> (50 mM), 0.5  $\mu$ l of each of the forward and reverse primers (each at 10  $\mu$ M), 1 U of MangoTaq (Bioline), and 2  $\mu$ l of DNA

template. PCR of both 18S and 28S used an initial step of denaturation at 94°C for 4 min, followed by 35 cycles of denaturation at 94°C for 30 s, primer annealing at 55°C for 30 s and primer extension at 72°C for 1 min, and completed with a final elongation step of 72°C for 3 min. PCR products were treated with Exonuclease I and Antarctic Phosphatase (New England Biolabs, Queensland, Australia) before sending to Macrogen Inc. (Republic of Korea).

Sequences were edited using Geneious R6 (Biomatters 2012) and aligned manually in Se-Al (Rambaut 2002). Nexus files were imported into Geneious R6 and analyzed using Neighbor-Joining clustering method with the Tamura Nei algorithm implemented in Geneious R6. One thousand bootstrap pseudo-replicates were conducted using the same program to assess node support.

In May 2012, adult females were found in Murray County, GA, and were sent to the ANU for species identification based on adult female morphology. These specimens are housed in the Australian National Insect Collection, Commonwealth Scientific and Industrial Research Organization Ecosystem Sciences, Canberra, Australia.

***Matsucoccus* Distribution.** In late 2012 and early 2013, sampling for *Matsucoccus* on *P. strobus* was ex-



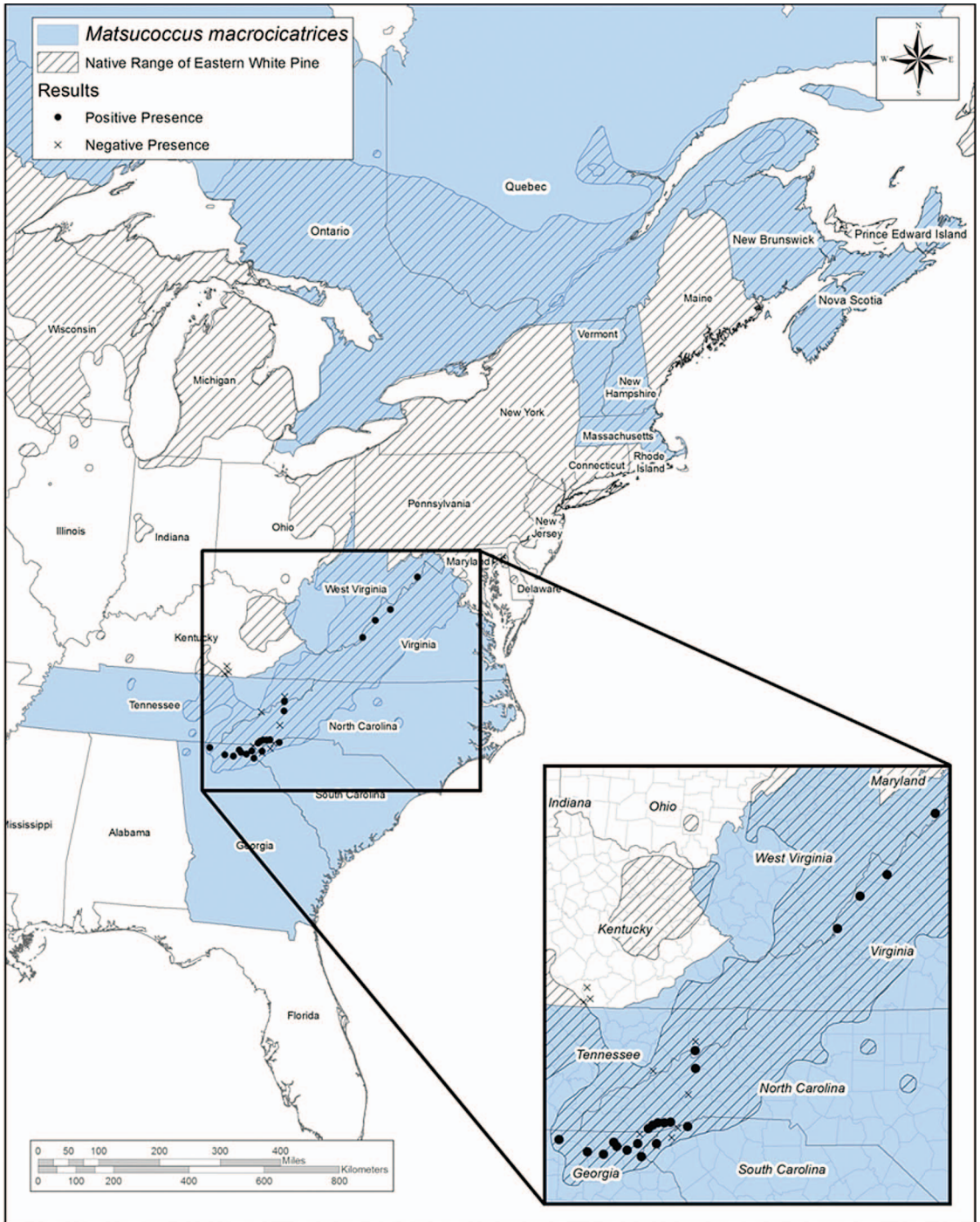


Fig. 2. *M. macrocitrices* distribution map by state and province in eastern North America. Inset map shows collection locations in the southeastern United States as of January 2013. Note that the entire states or provinces were colored to indicate the presence of *M. macrocitrices*, and that this does not indicate that *M. macrocitrices* is present everywhere in that particular state or province. (Online figure in color.)

panded to other states in the southeastern region of the United States. Branches of *P. strobus* from Georgia, Virginia, and West Virginia were collected from

known symptomatic stands, whereas *P. strobus* samples from Kentucky, North Carolina, South Carolina, and Tennessee were randomly collected based on

**Table 2.** Collection information for *M. macrocitrices* on *P. strobus* in the southeastern United States (locations refer to Fig. 2)

State	County	Latitude and longitude	Collection date	
Georgia	Gilmer	34°43'21.179", -84°12'16.308"	20 Nov. 2012	
	Habersham	34°45'24.048", -83°35'01.967"	18 Dec. 2011	
	Lumpkin	34°41'44.448", -83°57'01.511"	28 Sept. 2012	
	Murray	34°52'20.711", -84°39'00.144"	18 April 2012	
	Rabun	34°50'31.415", -83°25'25.752"	4 Jan. 2013	
	Stephens	34°40'17.652", -83°21'36.072"	18 Nov. 2012	
	Townes	34°51'04.212", -83°47'42.648"	28 Sept. 2012	
	White	34°47'56.471", -83°44'42.251"	28 Sept. 2012	
	North Carolina	Jackson	35°07'02.532", -83°00'43.487"	4 Jan. 2013
		Jackson	35°06'47.052", -83°06'44.171"	4 Jan. 2013
Macon		35°02'14.460", -83°15'26.928"	4 Jan. 2013	
Macon		35°04'58.908", -83°11'19.212"	4 Jan. 2013	
Madison		35°49'18.768", -82°31'42.672"	9 Jan. 2013	
Transylvania		35°07'34.175", -82°54.56.123"	4 Jan. 2013	
South Carolina	Greenville	35°04'15.312", -82°38'41.531"	11 Dec. 2012	
	Oconee	34°50'34.907", -83°07'50.627"	11 Dec. 2012	
Tennessee	Unicoi	36°03'11.951", -82°31'51.671"	9 Jan. 2013	
Virginia	Bath	38°01'53.652", -79°51'27.216"	20 July 2011	
	Highland	38°17'59.999", -79°24'29.998"	28 Nov. 2012	
West Virginia	Hardy	39°04'14.268", -78°34'41.051"	22 Oct. 2012	
	Monroe	37°37'07.895", -80°14'19.464"	15 Oct. 2012	

road surveying for trees. In total, 31 locations in 25 counties were sampled with two or more counties per state. Branches were examined for the presence of the immature cyst stages that were collected ( $n = \approx 15$  per location), and sent to UQ for identification. All immature cyst stages were stored in 90% EtOH and DNA-barcoded as described previously for species determination. A reference collection of remaining immature cyst stages and adults not sent to UQ are deposited at the Georgia Museum of Natural History, University of Georgia, Athens. In addition to field surveys, entomological holdings of 27 major museums in eastern North America were searched for *Matsucoccus* specimens to determine whether any historical records of this insect existed that were not recorded in the literature.

**Fungus Identification.** From March to August of 2012, in total, 120 branches (10 branches collected biweekly) were collected from individual symptomatic *P. strobus* trees in Habersham County, GA, to be examined for the occurrence of *C. pinea* or of *Septobasidium pinicola* Snell, an epiphytic fungus known to be mutualistic with some sap-sucking insects, including *Matsucoccus* (Couch 1938, Watson et al. 1960). Identifications of these fungi were based on the spermatogonia and ascocarp fruiting structures of *C. pinea* (Benny et al. 1985) and the diagnostic fungal mat of *S. pinicola* (Couch 1938).

Additional branches from individual *P. strobus* trees in Habersham County, GA, were collected to identify fungi occurring in the necrotic wood tissue surrounding individuals of *Matsucoccus* not associated with cankers. In total, 72 scales (maximum six per branch) were removed from the host and a 5 by 5 mm<sup>2</sup> area of the *P. strobus* tissue that surrounded each scale was placed in 10% bleach at 6% active ingredient for 40 s. The surface-sterilized tissue was then washed in sterile water for 1 min and blotted dry with sterile paper towels. Samples were divided evenly on three types of media: modified Nash-Snyder medium (Nelson et al.

1983), pine needle agar (PNA; Blodgett et al. 2003), and potato dextrose agar with streptomycin and tergitol (PDA+S+T) medium (Steiner and Watson 1965). Plated samples were incubated at 20°C for 4 wk with weekly observations for identification or transfer of isolates to fresh media. Unidentifiable mycelium isolates that did not produce spores on the initial medium used were transferred to carnation-leaf water agar medium (Nelson et al. 1983) or to PNA in an attempt to induce spore production for identification. These second transfers were also observed weekly for 4 wk.

## Results

***Matsucoccus* Identification.** Molecular analysis matched the specimens collected in 2012 and 2013 to *M. macrocitrices*. The sequences of the 18S and 28S D2–D3 gene regions from all collections were identical to those of *M. macrocitrices* previously collected and identified from Massachusetts, with strong bootstrap support (92% for 18S and 100% for the 28S D2–D3 region). Genbank accession numbers for the 28S D2–D3 and 18S regions are KF040554–KF040572 and KF053072–KF053091, respectively. Furthermore, the identity of adult females from Murray County, GA, was confirmed as *M. macrocitrices* based on published adult morphology (Richards 1960).

***Matsucoccus* Distribution.** Of the 25 counties sampled, 19 (76%) contained *P. strobus* with *M. macrocitrices* (Fig. 2; Table 2). The six county samples lacking *M. macrocitrices* were all collected from nonsymptomatic, randomly selected *P. strobus*. Results indicate that *M. macrocitrices* is now present on *P. strobus* in Georgia, North Carolina, South Carolina, Tennessee, Virginia, and West Virginia (Fig. 2; Table 2). The survey of entomological holdings of 27 museums in eastern North America resulted in no historical records of *M. macrocitrices* from the southeastern

Table 3. Reports of *M. macrocitrices* in museum collections in eastern North America

State or province	Institution	Holdings
Alabama	Auburn University	No <i>M. macrocitrices</i> in collection
Connecticut	University of Connecticut	No <i>M. macrocitrices</i> in collection
Florida	Florida Department of Agriculture	No <i>M. macrocitrices</i> in collection
Georgia	University of Georgia	No <i>M. macrocitrices</i> in collection
Indiana	Purdue University	No <i>M. macrocitrices</i> in collection
Kentucky	University of Kentucky	No <i>M. macrocitrices</i> in collection
Maine	Maine Forest Service	No <i>M. macrocitrices</i> in collection
Maryland	Smithsonian National Museum of Natural History	No <i>M. macrocitrices</i> in collection
Massachusetts	University of Massachusetts	No <i>M. macrocitrices</i> in collection
Michigan	Michigan State University	No <i>M. macrocitrices</i> in collection
Mississippi	Mississippi State University	No <i>M. macrocitrices</i> in collection
Newfoundland	Memorial University	No <i>M. macrocitrices</i> in collection
New Hampshire	University of New Hampshire	No <i>M. macrocitrices</i> in collection
New York	Cornell University	No <i>M. macrocitrices</i> in collection
North Carolina	North Carolina State University	No <i>M. macrocitrices</i> in collection
Ohio	Ohio State University	No <i>M. macrocitrices</i> in collection
Ontario	Canadian National Collection	<i>M. macrocitrices</i> type series as described by Richards (1960) coll. from Lynedoch, Denbigh, and Griffith (Ontario), and Fredericton (New Brunswick)—all on <i>P. strobus</i>
Pennsylvania	Penn State University	No <i>M. macrocitrices</i> in collection
Quebec	McGill University	No <i>M. macrocitrices</i> in collection
Rhode Island	RI Museum of Natural History	No <i>M. macrocitrices</i> in collection
Rhode Island	University of Rhode Island	No <i>M. macrocitrices</i> in collection
South Carolina	Clemson	No <i>M. macrocitrices</i> in collection
Tennessee	University of Tennessee	No <i>M. macrocitrices</i> in collection
Vermont	Vermont Department of Forests, Parks, and Recreation	<i>M. macrocitrices</i> . VT <sup>a</sup> coll. from Montpelier (Washington Co.) and St. Johnsbury (Caledonia Co.) associated with <i>S. pinicola</i> , and from Dummerston (Windham Co.)—all on <i>P. strobus</i>
Virginia	Virginia Tech University	No <i>M. macrocitrices</i> in collection
West Virginia	West Virginia University	No <i>M. macrocitrices</i> in collection
Wisconsin	University of Wisconsin	No <i>M. macrocitrices</i> in collection

<sup>a</sup> New state record for *M. macrocitrices*.

United States, but did result in a new state record for this species in Vermont (Table 3).

**Fungus Identification.** Of the 120 symptomatic *P. strobus* branches collected from Georgia, 91.7% had *M. macrocitrices*, 15.8% had *C. pinea* cankers, the pathogen implicated in *P. strobus* dieback in Maine, New Hampshire, and West Virginia, and only 2.5% had *S. pinicola* fungal mats. Fungi isolated from the wood surrounding *M. macrocitrices* included potentially pathogenic species in the genera *Pestalotiopsis* and *Phomopsis* along with a number of saprophytic fungi including *Aspergillus niger* Van Tieghem, *Chaetophoma* spp., and *Peyronellaea* spp. Of the different media used, 12.5% of the PNA plates contained *Pestalotiopsis*, 4.2% of the Nash-Snyder plates had *Pestalotiopsis* and 8.3% with *Phomopsis*, and 12.5% of the PDA+S+T plates contained *Pestalotiopsis* and 4.2% had *Phomopsis*. No cultures of *Fusarium*, *Diplodia*, or *Caliciopsis*, the fungi previously found associated with *P. strobus* cankers in Virginia, were isolated. However, in the case of *Caliciopsis*, there is no known selective medium for this genus, and the media used may not have been optimal for isolation.

### Discussion

We provide the first documentation of the presence of *M. macrocitrices* in six states in the southeastern United States. These results are important because 1)

this species has never been reported or collected from the southeastern United States. It was previously known only from Massachusetts, New Brunswick, New Hampshire, Nova Scotia, Ontario, and Quebec. The museum survey added Vermont to this list; 2) this scale species has never been reported with *P. strobus* showing signs of dieback or mortality; 3) *M. macrocitrices* has not been associated previously with fungal pathogens; and 4) *M. macrocitrices* has never been documented living outside the epiphytic mats of its symbiotic fungus, *S. pinicola*.

The most recent detailed work regarding *M. macrocitrices* was conducted by Watson et al. (1960) in the Canadian Maritimes where *M. macrocitrices* was found to be closely associated with *S. pinicola*, an epiphytic, nonpathogenic, and mutualistic basidiomycete fungus (Couch 1938). Adult females of *M. macrocitrices* emerge in late spring to early summer and move away from the fungal mats to produce their ovisacs in cryptic locations, such as bark crevices or under lichens (Watson et al. 1960). In Georgia, adults emerged in late winter to early spring and the few ovisacs found were in cankers and twig beetle (Coleoptera: Curculionidae: Scolytinae) galleries. After hatching, the mobile yellow-brown crawlers (0.6–1.5 mm) move back to the *S. pinicola* fungal mats where they insert their stylets into the bark and become stationary to form the intermediate cyst stage. The high proportion of branches with *M. macrocitrices*



lacking *S. pinicola* (97.3%) in our study suggests that, although *S. pinicola* requires an insect for nutrition (Couch 1938), *M. macrocitrices* is likely able to survive without the presence of fungal mats.

Watson et al. (1960) observed that the cyst stage remained within the fungus for two winters, suggesting that *M. macrocitrices* has a 2-yr life cycle. Although males were not found initially, suggesting that this species may be parthenogenetic (Watson et al. 1960), males have been described (Richards 1960). Adult females are 3.6–4.0 mm, have fully developed antennae and legs when they emerge from the cyst stage (Fig. 1C), and are wingless. Adult males of *M. macrocitrices* emerge from a “cocoon” made of waxy threads secreted by the prepupa and are winged (Richards 1960). The phenology of *M. macrocitrices* in the southeastern United States is currently under investigation. However, the lack of overlapping generations and synchronous adult emergences in early 2012 and again in early 2013 suggests that *M. macrocitrices* may have a 1-yr life cycle in Georgia.

At present, we are unsure whether *M. macrocitrices* has always been present in the southeastern United States, but has been overlooked because of its small size and cryptic nature, or whether our findings reflect a relatively recent range expansion or introduction into these states. The absence of *M. macrocitrices* in museum holdings from the southeastern United States supports the latter hypothesis (Table 3). Conversely, *S. pinicola* was collected in North Carolina in 1931 (Couch 1938) supporting the hypothesis that *M. macrocitrices* may have always been in the southeastern United States. More surveys in the native range of *P. strobus* will provide a better understanding of the distribution of *M. macrocitrices* in North America.

Unlike some of the other pine bast scales, *M. macrocitrices* has not historically been associated with tree dieback or mortality, which raises interesting questions about its presence on symptomatic *P. strobus*, both in the southeastern United States and in its previously documented range (New Hampshire). The variability of fungi found with cankers and tissue associated with feeding individuals of *M. macrocitrices* in this and other investigations (Cram et al. 2009) indicates that the insect is most likely not associated with a single pathogen. Because scales are found either deeply embedded in the cankers or present on top of the bark with clear necrotic tissue under their feeding area (Fig. 1B and D), we hypothesize that they may be creating wounds that are then infested by opportunistic fungi such as *C. pinea*. The relative contributions of the scale insect–fungal pathogen complex to canker formation and tree dieback, and their interactions with each other, are also being investigated. Overall, our study suggests that a closer examination of *M. macrocitrices* and associated pathogens may be warranted for the future health of natural and planted *P. strobus* stands in North America.

## Acknowledgments

We thank B. Barnes, C. Brissey, and K. Smoot (University of Georgia) for field and laboratory support. J. Sullivan (Georgia Forestry Commission); M. Bohne and I. Munck (U.S. Department of Agriculture Forest Service); K. Lombard (New Hampshire Division of Forests and Lands); B. Heath (North Carolina Forest Service); and K. Carrington, B. Kesecker, and J. Rose (West Virginia Department of Agriculture) kindly sent us *P. strobus* samples. We are also thankful to G. Stanosz and D. Smith (University of Wisconsin), G. Hodges (Florida Department of Agriculture and Consumer Sciences), and D. Henk (Imperial College, United Kingdom) for their assistance with identifications. Thanks also to T. Edgerton (Virginia Department of Forestry) for producing the scale distribution map. Many researchers at various museums assisted with specimen retrievals including S. Alm (University of Rhode Island), B. Blinn (North Carolina State University), C. Bartlett (University of Delaware), S. Boucher (McGill University), D. Chandler (University of New Hampshire), T. Chapman (Memorial University), S. Clutts (University of Kentucky), E. Day (Virginia Tech University), A. Deans (Penn State University), J. Dombroskie (Cornell University), C. Donahue (Maine Forest Service), F. Drummond (University of Maine), R. Footitt (Canadian National Collection), H. Ginsberg (Rhode Island Museum of Natural History), T. Hanson (Vermont Department of Forests, Parks, and Recreation), R. Hoebeke (University of Georgia), S. Krauth (University of Wisconsin), P. Lambdin (University of Tennessee), D. Miller (Smithsonian), J. Morse (Clemson University), L. Musetti (Ohio State University), B. Normark (University of Massachusetts), J. O'Donnell (University of Connecticut), G. Parsons (Michigan State University), A. Provonsha (Purdue University), C. Ray, Jr. (Auburn University), T. Schiefer (Mississippi State University), I. Stocks (Florida Department of Agriculture), and J. Strazanac (West Virginia University). We thank the editor and two anonymous reviewers for their constructive comments on this manuscript. Funding was provided by the Daniel B. Warnell School of Forestry and Natural Resources, University of Georgia; U.S. Department of Agriculture–Forest Service, Forest Health Protection; and Virginia Department of Forestry.

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Received 27 May 2013; accepted 11 August 2013.