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Advertisement Call and Distribution of the Treefrogs *Hyla chrysoscelis* and *Hyla versicolor* in Virginia

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ABSTRACT

The gray treefrog complex consists of two sibling species that are indistinguishable morphologically, the diploid *Hyla chrysoscelis* and the tetraploid *Hyla versicolor*. Identification is possible in the field only by audio recognition of male advertisement call trill rates (pulses/second). During 1979-1983 we evaluated taped calls of these two species taken from 89 populations from throughout Virginia to map their respective ranges and to evaluate differences in call parameters. *Hyla chrysoscelis* occurs in the Coastal Plain, eastern and southern Piedmont, and in southwestern Virginia. *Hyla versicolor* occurs in the Piedmont, Blue Ridge, and Ridge and Valley regions south to Wythe and Tazewell counties. Sympatric sites occur in several locations in the Piedmont and both species are syntopic in several of them. Male trill rates are significantly related to ambient and body temperatures. Rates produced by male *H. chrysoscelis* ($\geq 31/s$) are twice as fast as that for *H. versicolor* ($\leq 30/s$); they did not overlap in our samples at any temperature. Trill rates and call duration in southwestern Virginia populations of *H. chrysoscelis* differed significantly from those in eastern populations when adjusted for ambient temperature. Adjusted trill rate and duration in *H. chrysoscelis* populations in sympatry with *H. versicolor* were not significantly different from allopatric populations but were for *H. versicolor*.

Keywords: Anura, Cope's Gray Treefrog, ecology, Gray Treefrog, *Hyla chrysoscelis*, *Hyla versicolor*, vocalizations, distribution, Virginia

INTRODUCTION

The diploid *Hyla chrysoscelis* (Cope's Gray Treefrog) and the tetraploid *H. versicolor* (Gray Treefrog) differ in the pulse rate of the trills produced by calling males (Johnson 1959, 1966), their karyotypes (Wasserman 1970, Bogart and Wasserman 1972), and in some cytological parameters, such as size of toe pad cells

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(Green 1980), cell size (Bogart and Wasserman 1972), amount of DNA in the nuclei (Bachmann and Bogart 1975), and size of nuclei and number of nucleoli (Cash and Bogart 1975). Ptacek et al. (1994) recognized five sibling species in the Gray Treefrog complex, two of which were diploid and three tetraploid. Espinoza and Noor (2002) examined evidence for gene flow among *H. versicolor* lineages at various locations using PCR-product cloning techniques. These authors verified distinct mitochondrial lineages in *H. versicolor*, but stated that these lineages hybridize when they exist in sympatry. Halloway et al. (2006) determined that *H. versicolor* originated repeatedly from three diploid ancestors, including *H. chrysoscelis*, and merged through inbreeding to result in a single species. Both of these gray treefrogs occur widely in Virginia.

At present, recognition of male mating trill pulse rates is the only method available to distinguish *H. chrysoscelis* and *H. versicolor* in the field (Elliott et al. 2009). They are indistinguishable morphologically. However, because pulse rates are related to ambient temperature, pulse rate alone may not distinguish a warm *H. versicolor* from a cool *H. chrysoscelis*. Thus, elucidation of their respective distributions has been slow to accumulate. They have been mapped out in several states, including Texas (Johnson 1966), parts of Illinois (Brown and Brown 1972), Wisconsin (Jaslow and Vogt 1977), Michigan (Bogart and Jaslow 1979), and Maryland (Otto et al. 2007). Zweifel (1970) examined the distribution of these species in northeastern Virginia, eastern Maryland, Delmarva and southern New Jersey. Based on these studies and their own work, Ralin (1977) and Gerhardt (1999) extrapolated the range limits of *H. chrysoscelis* and *H. versicolor* for North America and their areas of sympatry. For Virginia, Ralin (1977) hypothesized that *H. chrysoscelis* occurs in the southeastern half of the state and *H. versicolor* in the northwestern half. Gerhardt (1999) illustrated the generally accurate distribution patterns for both species and their known areas of sympatry based, in part, on information supplied by us. Range-wide maps of the two Gray Treefrogs are in Cline (2005a, b) and Halloway et al. (2006).

Our study sought to determine the distributions of these two species in Virginia and to investigate differences in their vocalizations. The atlas of amphibians and reptiles published by Mitchell and Reay (1999) used the information we present here, in addition to museum specimens and other sources to create maps of both gray treefrogs in Virginia. This paper presents the basis upon which those maps were delineated and presents results of our analyses of call parameters in allopatry and sympatry.

MATERIALS AND METHODS

During March to August 1979-1983, we drove approximately 71,000 km throughout Virginia to locate calling gray treefrogs. We found choruses at 224 localities. Tentative identifications made by ear in the field were later verified and analyzed at 89 of these sites, the results of which are reported here. We recorded vocalizations on portable standard Panasonic® and General Electric® cassette recorders using fresh batteries. When possible, we took cloacal temperatures (to 0.1°C) with Schultheis quick-reading thermometers. Ambient and water temperatures were taken with Schultheis and Webster thermometers placed as close to the calling frog as possible. In the analyses of call parameters, water temperature was used for the ambient measurement instead of air temperature if the frog was sitting in water when calling.

Sonograms of mating calls were produced with a Kay Electronic Sound Spectrograph Model 6061B. From these sonograms we determined trill rates (number

of pulses per second) and duration of calls (in seconds) of 326 individual males from the 89 localities. Trill rates were determined directly from the sonograms. Scatterplots of trill rate versus body and ambient temperatures were used to illustrate the relationships between temperature and call parameters. Analysis of covariance, with ambient and body temperatures as covariates, was used to compare differences between species and populations. Statistical tests were performed with SysStat 11[®] using a Type I error rate of $\alpha = 0.05$. In this paper, means are reported with $\pm$ one sd.

RESULTS

Hyla chrysoscelis occurs in Virginia in two allopatric areas, the Coastal Plain, including the Eastern Shore, and in the Valley and Ridge and Cumberland Plateau regions south of the New River in the southwestern corner of the state (Figure 1). *Hyla versicolor* occurs allopatrically in the western Piedmont, Blue Ridge Mountains, and Valley and Ridge regions south to eastern Wythe County. The two species are sympatric over a wide portion of the eastern and lower Piedmont region and are syntopic in many locations in central and south-central Virginia (Figure 1). We found no evidence that these two species occur in sympatry in southwestern Virginia and could not determine whether the range of *H. versicolor* extends into North Carolina along the Blue Ridge Mountains. Current locality data suggest that eastern and southwestern *H. chrysoscelis* populations are separated by populations of *H. versicolor*. Recent locality records of *H. chrysoscelis* along the Blue Ridge Parkway in Floyd County (Mitchell and Reay, 1999) where they were not known historically (Hoffman, 1996) suggest that this pattern may be changing.

Comparisons of call parameters between species (Table 1) demonstrate that differences in trill rates are readily apparent before adjustment is made for the effects of temperature. Populations of *H. chrysoscelis* from southwestern Virginia have slightly higher average unadjusted trill rates than populations in eastern Virginia and lower average unadjusted call duration. Unadjusted trill rates of *H. chrysoscelis* populations sympatric with *H. versicolor* average higher than that in allopatric populations, but unadjusted call duration is shorter. Sympatric *H. versicolor* populations, however, exhibit similar unadjusted trill rates and shorter unadjusted call durations than allopatric populations (Table 1).

Trill rates of Virginia Gray Treefrogs are significantly related to ambient temperature and body temperature (Figure 2, 3). Note that trill rates for these two species do not overlap at any ambient or body temperature. Between-species comparisons of the ambient temperature range in Figures 2 and 3 suggest that *H. versicolor* is active over a broader range of temperature conditions than *H. chrysoscelis*.

Southwestern Virginia populations of *H. chrysoscelis* differed significantly from eastern populations in trill rate ($F = 10.24$, $P = 0.002$) and call duration ($F = 4.86$, $P = 0.029$) when adjusted for ambient temperature. Adjusted trill rate and duration in *H. chrysoscelis* populations sympatric with *H. versicolor* were not significantly different from allopatric populations ($F = 3.38$, $P = 0.068$, $F = 1.084$, $P = 0.300$, respectively). Sympatric *H. versicolor* had a significantly lower adjusted trill rate than allopatric populations ($F = 4.93$, $P = 0.029$). There was no significant difference in adjusted call duration between these two groups ($F = 1.25$, $P = 0.269$).

Ambient temperature does not affect dominant frequency (KHz) in Virginia gray treefrogs ($F = 0.177$, $P = 0.675$). Mean dominant frequency of *H. chrysoscelis* and *H.*

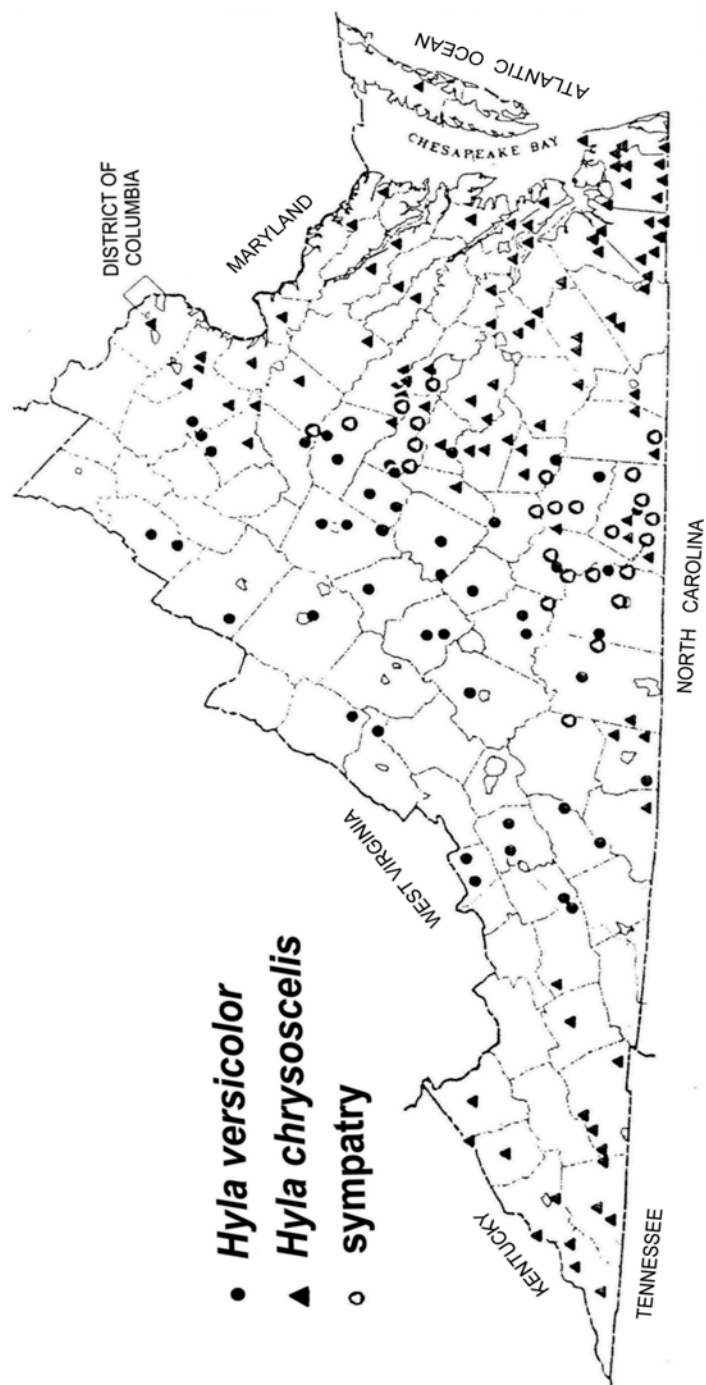


FIGURE 1. Distribution patterns for *Hyla chrysoscelis* and *H. versicolor* in Virginia based on analysis of trill rates in breeding choruses.

TABLE 1. Unadjusted call character and temperature statistics for *Hyla chrysoscelis* and *H. versicolor* samples in allopatric and sympatric regions in Virginia. Eastern Virginia samples of *H. chrysoscelis* include allopatric and sympatric data from sites east of the Blue Ridge Mountains. Sympatric samples for both species are from the eastern Piedmont. The first number in each column is the sample size followed by the mean $\pm$ one SD.

	Body Temp (°C)	Ambient Temp (°C)	Trill Rate	Duration (s) (pulse/s)	Dominant Frequency
<i>H. chrysoscelis</i>					
E. VA	40 22.65 $\pm$ 1.99	194 21.91 $\pm$ 2.06	197 45.63 $\pm$ 4.75	119 0.841 $\pm$ 0.272	46 1882.39 $\pm$ 625.42
SW VA	– –	14 21.55 $\pm$ 2.06	14 48.29 $\pm$ 6.17	9 0.679 $\pm$ 0.159	7 1441.43 $\pm$ 484.85
Allopatric	14 22.79 $\pm$ 1.39	100 21.6 $\pm$ 1.96	101 44.95 $\pm$ 4.64	65 0.855 $\pm$ 0.306	28 1913.93 $\pm$ 636.62
Sympatric	24 22.46 $\pm$ 2.25	103 22.11 $\pm$ 2.18	104 46.62 $\pm$ 4.99	59 0.789 $\pm$ 0.199	24 1748.33 $\pm$ 604.51
All samples	38 22.65 $\pm$ 1.99	217 21.88 $\pm$ 2.06	209 45.81 $\pm$ 4.89	133 0.830 $\pm$ 0.268	53 1824.15 $\pm$ 633.16
<i>H. versicolor</i>					
Allopatric	11 24.51 $\pm$ 1.80	34 21.96 $\pm$ 2.48	34 24.85 $\pm$ 3.29	21 0.809 $\pm$ 0.379	10 1793.40 $\pm$ 438.59
Sympatric	9 22.21 $\pm$ 3.28	71 22.67 $\pm$ 3.15	73 24.12 $\pm$ 3.51	40 0.722 $\pm$ 0.319	21 1914.67 $\pm$ 500.25
All samples	20 23.71 $\pm$ 2.53	105 22.5 $\pm$ 2.90	107 24.54 $\pm$ 3.42	61 0.729 $\pm$ 0.327	31 1785.55 $\pm$ 477.35

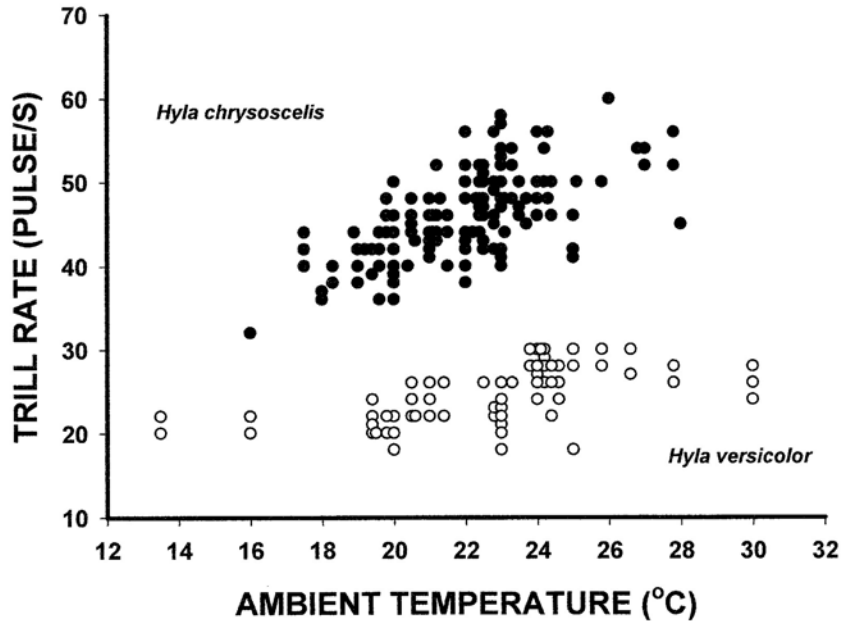


FIGURE 2. Relationship of trill rate to ambient temperature for *Hyla chrysoscelis* and *H. versicolor* in Virginia.

versicolor calls (Table 1) do not differ significantly ($t = 0.424$, $P = 0.673$). The dominant frequency for *H. chrysoscelis* calls in southwestern Virginia populations are not significantly different from those in eastern populations ($t = 1.780$, $P = 0.081$).

DISCUSSION

Twenty years before it was determined that gray treefrogs were, in fact, two separate species, Hoffman (1946), following differences in calls defined by Noble and Hassler (1936) and Walker (1946), delineated their ranges in Virginia with considerable accuracy. The patterns he found, based only on the relative locations of gray treefrogs with "harsh" and "mellow" voices, are essentially the same as those we report here. He noted a population in the central Virginia Piedmont (an area of sympatry) that apparently had calls somewhat intermediate between the two voice types and with longer durations than those he had noted in southeastern Virginia (*H. chrysoscelis* only). We suspect that this was a function of temperature because most of his southeastern records were obtained in summer (based on museum records); his Piedmont record was taken in late September. Hoffman's paper has apparently been overlooked in all the previous literature on the calls and biogeography of these species except Zweifel (1970).

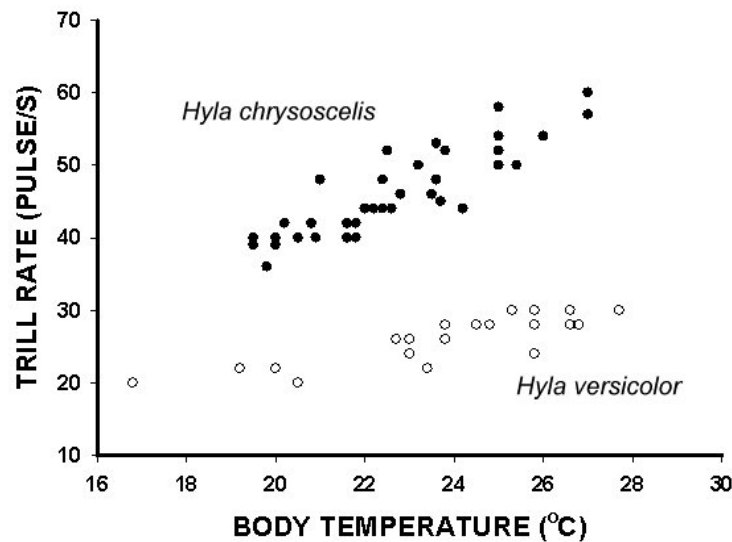


FIGURE 3. Relationship of trill rate to body temperature for *Hyla chrysoscelis* and *H. versicolor* in Virginia.

The distribution pattern hypothesized by Ralin and Sealander (1979) for *Hyla chrysoscelis* and *H. versicolor* in Virginia was essentially correct for these two species east of the Blue Ridge Mountains, although the broad zone of overlap was not predicted. The primary modification of their pattern occurs in southwestern Virginia where only *H. chrysoscelis* is found; they had predicted only *H. versicolor* in that area. Hoffman and Kleinpeter (1948) described what appeared to be a sympatric population of the two species near Burkes Garden, Tazewell County, although R.L. Hoffman and J.A. Fowler heard only *H. versicolor* there three years later (R.L. Hoffman, pers. comm.). This unvouchered record, along with the one for eastern Wythe County, may delineate the range boundary of this species in southwestern Virginia. *Hyla chrysoscelis*, which occurs in southwestern Virginia, also ranges northward into West Virginia (Green and Pauley 1987, M. Little, pers. comm.) and Kentucky (J. MacGregor, pers. comm.) and southwestward throughout Tennessee (Redmond and Scott 1996, Niemiller and Reynolds 2011). The eastern form of *H. chrysoscelis* occurs throughout North Carolina (A. Braswell, pers. comm.) and extends northward into eastern Maryland and New Jersey in the Coastal Plain (Zweifel 1970). In Maryland, *Hyla chrysoscelis* occurs throughout most of the eastern half of the state and upper Delmarva, whereas *H. versicolor* occurs primarily in central and western Maryland; sympatry occurs in several locations (Otto et al. 2007, D. Forester and R. Miller, pers. comm.).

Comparisons of adjusted trill rate and duration for *H. chrysoscelis* populations revealed significant differences between eastern populations and those in southwestern Virginia but not in populations in sympatry with *H. versicolor*. The regional differences may be due to a combination of the large difference in sample sizes for these two areas and the narrow range of ambient temperatures recorded in southwestern counties (20–24°C) compared to the much wider range in eastern samples (17.5–28°C). Additional sampling over a broader range of temperatures in southwestern counties may provide a different result. Lack of a difference in adjusted trill rate and duration for *H. chrysoscelis* and *H. versicolor* in sympatry suggests that there is no species effect. The significant difference in adjusted trill rate between allopatric and sympatric populations in *H. versicolor* may have been influenced by several factors. Differences in trill rate between sympatric populations of these two species in Missouri are greatest at high temperatures (Gerhardt 1982) suggesting that higher temperatures in the Piedmont compared to those in higher elevations contributed to this result.

Despite the fact that trill rates are temperature-dependent, complete lack of overlap at most body and ambient temperatures in Virginia populations suggests that this parameter may be used as an identifying field character once the differences become recognized through experience and training. Trill rates ≥ 31 pulses per second are *H. chrysoscelis* and rates ≤ 30 pulses per second are *H. versicolor*. Johnson (1966) used trill rate differences to construct a key to these species, although there was a 2-pulse overlap in the minimum and maximum values in his study. Other studies (e.g., Zweifel 1970, Ralin 1977, Gerhardt 1982) found a small range overlap in trill rates between cool *H. chrysoscelis* and warm *H. versicolor*. Because individuals have been found to exhibit trill rates intermediate between the two species (Zweifel 1970, Gerhardt 1982), we caution the use of this parameter rate for final species identification without correction for temperature affects. Trill rates are non-overlapping at 20°C (diploid species mean = $35.7 \pm 1.4^\circ\text{C}$, tetraploid species mean = $22.9 \pm 1.5^\circ\text{C}$) (Gerhardt 2005). Mean breeding body temperatures in Virginia (22.7°C for *H. chrysoscelis*, 23.7°C for *H. versicolor*) are higher than the 20°C. Holloway et al. (2006) concur that this is the middle of the range of breeding temperatures for these species range-wide.

Comparisons of the call parameters for Virginia populations of *H. chrysoscelis* with values of these parameters in Ralin (1977) indicate that the populations in eastern and southwestern Virginia correspond with his eastern form. Virginia *H. versicolor* trill rates correspond more closely with Ralin's northern *H. versicolor*, but call duration is considerably longer than those noted for either northern or southern *H. versicolor*. Jaslow and Bogart (1979) found a similar result for gray treefrogs in Michigan. Using our data, it is not possible to determine whether the eastern Virginia populations of *H. chrysoscelis* differ genetically from the southwestern populations. Electrophoretic (e.g., Ralin and Sealander 1979), immunological (e.g., Maxson, et al. 1977), and genetic analyses (Espinoza and Noor 2002, Holloway et al. 2006) have elucidated the historical biogeographical patterns and origins of this diploid-tetraploid species pair in North America. Distribution patterns illustrated in Figure 1a in Holloway et al. (2006) for the two species show sympatry in a continuous range from southwestern Virginia (Wythe County to about Giles County) to about Mecklenburg County in the southern Piedmont and along the western edge of the Piedmont. Our results do not show sympatry west of the Blue Ridge Escarpment and places the overlap zone of these two species in the eastern Piedmont. Their distribution of *H. versicolor* is well to the west of the range in

Virginia that we elucidated. Additional records of these two species in southwestern Virginia and along the eastern side of the Blue Ridge Mountains are needed to clarify this discrepancy.

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LITERATURE CITED

- Bachmann, K., and J.P. Bogart. 1975. Comparative cytochemical measurements in the diploid-tetraploid species pair of hylid frogs *Hyla chrysoscelis* and *Hyla versicolor*. *Cytogenetics and Cell Genetics* 15:186-194.
- Bogart, J.P., and A.O. Wasserman. 1972. Diploid-polyploid species pairs: A possible clue to evolution by polyploidization in anuran amphibians. *Cytogenetics* 11:7-24.
- Brown, L.E., and J.R. Brown. 1972. Mating calls and distributional records of treefrogs of the *Hyla versicolor* complex in Illinois. *Journal of Herpetology* 6:233-234.
- Cash, M.N., and J.P. Bogart. 1975. Cytological differentiation of the diploid--tetraploid species pair of North American treefrogs (Amphibia, Anura, Hylidae). *Journal of Herpetology* 12:555-558.
- Cline, G.R. 2005a. *Hyla chrysoscelis* Cope, 1880, Cope's Gray Treefrog. Pp. 449-452 In M. Lannoo (ed.), *Amphibian Declines, The Conservation Status of United States Species*. University of California Press, Berkeley, CA.
- Cline, G.R. 2005b. *Hyla versicolor* LeConte, 1825, Eastern Gray Treefrog. Pp. 458-461 In M. Lannoo (ed.), *Amphibian Declines, The Conservation Status of United States Species*. University of California Press, Berkeley, CA.
- Elliott, L., Gerhardt, H.C. & Carlos Davidson. 2009. *Frogs and Toads of North America: A Comprehensive Guide to their Identification, Behavior and Calls*. Houghton-Mifflin, NY. 340 pp. with CD.
- Espinoza, N.R., and M.A. Noor. 2002. Population genetics of a polyploid: is there hybridization between lineages of *Hyla versicolor*? *Journal of Heredity* 93:81-85.
- Gerhardt, H.C. 1982. Sound pattern recognition in some North American treefrogs (Anura: Hylidae): Implications for mate choice. *American Zoologist* 22:581-595.
- Gerhardt, H.C. 1999. Reproductive character displacement and other sources of selection on acoustic communication systems. Pp. 515-534 in M.D. Hauser and M. Konishi, eds., *The Design of Animal Communication*. MIT Press, Cambridge, MA.
- Gerhardt H.C. 2005. Advertisement-call preferences in diploid-tetraploid treefrogs (*Hyla chrysoscelis* and *Hyla versicolor*): implications for mate choice and the evolution of communication systems. *Evolution* 59:395-408.

- Green, D.M. 1980. Size differences in adhesive toe-pad cells of treefrogs of the diploid-polyploid *Hyla versicolor* complex. *Journal of Herpetology* 14:15-19.
- Green, N.B., and T.K. Pauley. 1987. *Amphibians and Reptiles in West Virginia*. University of Pittsburgh Press, Pittsburgh, PA. 241 pp.
- Hoffman, R.L. 1946. The voice of *Hyla versicolor* in Virginia. *Herpetologica* 3:141-142.
- Hoffman, R.L. and H.L. Kleinpeter. 1948. Amphibians from Burkes Garden, Virginia. *American Midland Naturalist* 319:602-607.
- Hoffman, R.L. 1996. *Hyla chrysoscelis* also crosses the Blue Ridge: *sic juvat transcendere montes*. *Catesbeiana* 16:3-8.
- Holloway, A.K., Cannatella, D.C., Gerhardt, H.C., and D.M. Hillis. 2006. Polyploids with different origins and ancestors form a single sexual polyploidy species. *American Naturalist* 167: E88-E101.
- Jaslow, A.P., and R.C. Vogt. 1977. Identification and distribution of *Hyla versicolor* and *Hyla chrysoscelis* in Wisconsin. *Herpetologica* 33:201-205.
- Jaslow, A.P., and J.P. Bogart. 1979. Distribution and call parameters of *Hyla chrysoscelis* and *Hyla versicolor* in Michigan. *Life Science Contribution Royal Ontario Museum* 117:1-13.
- Johnson, C. 1959. Genetic incompatibility of the call races of *Hyla versicolor* LeConte in Texas. *Copeia* 1959: 327-335.
- Johnson, C. 1966. Species recognition in the *Hyla versicolor* complex. *Texas Journal of Science* 18:361-364.
- Maxson, L., E. Pepper, and R.D. Maxson. 1977. Immunological resolution of a diploid-tetraploid species complex of tree frogs. *Science* 197:1012-1013.
- Mitchell, J.C., and K.K. Reay. 1999. *Atlas of Amphibians and Reptiles in Virginia*. Special Publication No. 1, Virginia Department of Game and Inland Fisheries, Richmond, VA. 122 pp.
- Niemiller, M.L., and R.G. Reynolds. 2011. *The Amphibians of Tennessee*. University of Tennessee Press, Knoxville, TN. 369 pp.
- Noble, G.K., and W.G. Hassler. 1936. Three salientia of geographic interest from southern Maryland. *Copeia* 1936:63-64.
- Otto, C.R., J.W. Snodgrass, D.C. Forester, J.C. Mitchell, and R.W. Miller. 2007. Climatic variation and the distribution of an amphibian polyploid complex. *Journal of Animal Ecology* 76:1053-1061.
- Ptacek, M.B., H.C. Gerhardt, and R.D. Sage. 1994. Speciation by polyploidy in treefrogs: multiple origins of the tetraploid *Hyla versicolor*. *Evolution* 48:898-908.
- Ralin, D.B. 1977. Evolutionary aspects of mating call variation in a diploid- tetraploid species complex of treefrogs (Anura). *Evolution* 31:721-736.
- Ralin, D.B. and R. K. Sealander. 1979. Evolutionary genetics of diploid-tetraploid species of treefrogs of the genus *Hyla*. *Evolution* 33:595-608.
- Redmond, W.H., and A.F. Scott. 1996. *Atlas of amphibians of Tennessee*. Center for Field Biology, Miscellaneous Publication No. 12:1-94.
- Walker, C.F. 1946. The amphibians of Ohio, Part I. Frogs and toads. *Ohio State Museum Science Bulletin* 1:1-109.
- Wasserman, A.O. 1970. Polyploidy in the common tree toad *Hyla versicolor* Le Conte. *Science* 167:385-386.
- Zweifel, R.G. 1970. Distribution and mating call of the treefrog, *Hyla chrysoscelis* at the northeastern edge of its range. *Chesapeake Science* 11:94-97.

Distributions and Abundances of *Microstegium vimineum* along Forest Roadsides at the Grassy Hill Natural Area, Franklin County, Virginia

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ABSTRACT

In summer 2005, plots were surveyed along roads passing through forest habitats at the Grassy Hill Natural Area Preserve in Franklin County, Virginia to assess the distributions and abundances of *Microstegium vimineum* in transects located at increasing distances away from roadsides into forest interiors. Across plots, *Microstegium* was encountered almost exclusively in roadside transects, where abundances were relatively high. While forest composition and topographic features were similar across plots, percent canopy cover and leaf litter depth were greater in interior compared to roadside transects due to undisturbed tree canopies and ground cover located in interior plot areas. Results imply that *Microstegium* was restricted to forest roadsides at Grassy Hill at the time of the study, likely due to factors that differ between forest edges and interiors.

Key words: canopy cover, edge, Grassy Hill Natural Area, leaf litter, *Microstegium vimineum*

INTRODUCTION

Microstegium vimineum (Trin.) A. Camus (i.e. Japanese stiltgrass) is an Asian endemic widespread in the eastern United States, including Virginia (Gibson et al. 2002, VDCR 2009), where it is invasive (VDCR 2009, Miller and Matlack 2010) and among the most targeted of exotics for management (Heffernan et al. 2001). It occurs in mountain habitats in the state (Heffernan et al. 2001), including those at Grassy Hill where it grows along roads and in limited areas of undisturbed forest (Turner and Demkó 2007). This is not surprising given that it thrives in both open forests and disturbed sites such as roadside habitats (Redman 1995), as well as in less disturbed forest interiors (Oswalt et al. 2007, Warren et al. 2011) where its presence is alarming since it can spread and outcompete native species (Barden 1987, Adams et al. 2009).

Microstegium's success in invading forests results from a high invasive potential related to a large seed set (Gibson et al. 2002, Bauer and Flory 2010), multiple seed dispersal modes (Christen and Matlack 2009), and dense growth (DeMeester and Richter 2010), which give it competitive advantages over other plants, reducing their growth and survival (Bauer and Flory 2010). However, *Microstegium* is not always invasive. While shade tolerant, it is inhibited by dense shade (Miller and Matlack 2010) and grows optimally under moderately high light conditions (Cole and Weltzin 2004, Glasgow and Matlack 2007). It also germinates and grows best on bare mesic soil

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compared to soil covered by plants and/or leaf litter (Barden 1987, Oswalt and Oswalt 2007). Together, these and other factors can inhibit *Microstegium* recruitment, which benefits management efforts at natural areas like Grassy Hill.

The main purpose of this study was to quantify *Microstegium* distributions and abundances along forest roads at Grassy Hill to determine if the species is found in both roadside and adjacent forest interiors, and if so, at what frequencies. Because *Microstegium* was previously observed almost exclusively along roadsides (Greg Turner, personal observations), it was predicted that it would decline in abundance with road distance. The study also assessed canopy and leaf litter cover to determine if these factors differ with road distance, given that they can inhibit *Microstegium* recruitment.

METHODS

I conducted this study at the Grassy Hill Natural Area, a 524 ha state preserve located northwest of Rocky Mount (36°59' 60"N, 79°53' 23"W). The preserve lies in the Piedmont physiographic province (Roberts & Bailey 2000) and contains magnesium-rich bedrock overlain with mafic soils (VDCR 2003). It is mountainous, with northwest-oriented slopes reaching 535 m ASL (USGS and VDMR 1985), and is dominated by hickory (*Carya*), oak (*Quercus*), and pine (*Pinus*) species. A few roads and other corridors cross the preserve, but at the time of the study there were no records of fire, logging, or other major disturbances since the mid-twentieth century (John Ebbert, VA Department of Forestry, personal communication).

In summer 2005, I placed nine 50 x 50 m plots along three roads (i.e. three per road) passing through relatively even-aged forest: a paved two-lane road, a gravel access road, and a dirt access road. Plot locations were determined using a random numbers table and a preserve map to choose start points for each plot, none of which was located within 500 m of another. Roads were chosen because they were contiguous and, thus, not independent from one another, and because they are conduits for exotic plant recruitment. Within each plot, five 4 x 50 m belt transects were established using the methods of Brothers and Spingarn (1992) to form a road proximity gradient in which transects were arrayed parallel to roadsides. Transects ran (-2)-2, 2-6, 10-14, 20-24 and 45-49 meters away from roadsides into interiors and were labeled T(-2), T(2), T(10), T(20), and T(45), respectively. T(-2) transects included road shoulder areas located 2 meters outside of canopy edges (i.e. -2 m from edges). *Microstegium* was sampled in June and July by noting its presence and by counting culms in those transects where it was found to determine its general distributions across plot transects, total abundances per transect in each plot, and mean abundances per transect across roads. Percent canopy cover and leaf litter depth were measured on three consecutive days in July, at 10 equidistant points (i.e. every 2.5 m) within each transect, to yield mean values per transect across roads for each measure. Canopy cover was measured with a handheld spherical densiometer (Forest Densiometers, Bartlesville, OK) and leaf litter depth with a meter stick as the distance between the bottom duff and top leaf layers. Because *Microstegium* was found almost exclusively in T(-2) transects, statistical tests for road distance effects on *Microstegium* abundance, and for associations between abundance and canopy cover or leaf litter depth, could not be conducted since the assumptions of regression and correlation tests could not be met. Thus, only analyses of observational results were attempted.

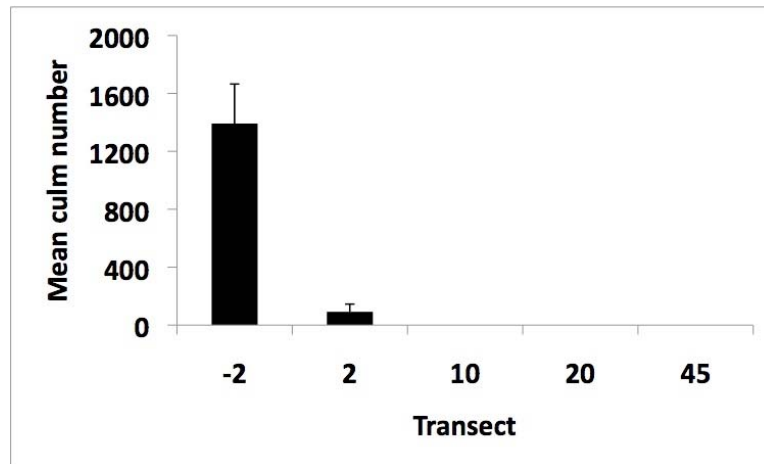


FIGURE 1. *Microstegium vimineum* abundance, measured as the mean number of culms per transect across road plots (error bars = + 1 standard error).

RESULTS

Microstegium vimineum was encountered in every T(-2) transect of each road plot, and in one T(2) transect. Abundance was relatively similar across plots, with relatively high frequencies recorded in T(-2) transects, low numbers in T2 transects, and none beyond any T2 transect (Figure 1). These findings imply that there was virtually no *Microstegium* recruitment into forest interiors away from roadsides at the time of the study. Measures of percent canopy cover showed a pattern of increasing percent cover with distance away from roadsides across plots, with mean percent canopy cover increasing from 82% in T(-2) transects, to 91% in T2 transects, to 98% and higher in T20 and T45 transects across plots (Table 1). Similarly, leaf litter depth generally increased with distance away from roadsides, as mean depth increased from a low of 2.3 cm in T(-2) transects, to 9.9 cm in T2 transects, and to 21.6 cm in T45 transects across plots (Table 1).

DISCUSSION

Microstegium vimineum was found in every study plot, suggesting that it was widespread at Grassy Hill at the time of the study. This is not surprising given that forest structure and topography, and human activities that facilitate *Microstegium* recruitment, did not differ greatly between plots before or during this study (Greg Turner, personal observations). Though widely distributed, *Microstegium* was largely restricted to T(-2) transects, which is important from a management perspective at this preserve, since other regional studies report its presence in less disturbed forest interior habitats. These studies report that *Microstegium* prefers to grow along roadsides and

TABLE 1: Mean canopy cover (%) and leaf litter depth (cm) measured across road transects. The numbers -2, 2, 10, 20, and 45 represent transect distances (m) from forest edges.

Transect Variable	-2	2	10	20	45
Mean canopy cover (%)	82.2	91.0	94.6	98.0	98.4
Mean leaf litter depth (cm)	2.3	9.9	9.1	10.4	21.6

in semi-open habitats, but does grow in closed-canopy interiors (Redman 1995, Huebner 2003, Cole and Weltzin 2004), likely due to shade tolerance (Leicht et al. 2005, DeMeester and Richter 2010). Which leads to the question of why *Microstegium* was absent from plot interiors in my study.

While *Microstegium* grows under high shade (Miller and Matlack 2010), it is light sensitive (Glasgow and Matlack 2007), so much so that germination and seedling growth by the species are negatively correlated with shade (Schramm and Ehrenfeld 2010). Thus, relegation of *Microstegium* to T(-2) transects, where canopy cover was lowest, was not surprising. However, I did expect to find *Microstegium* in some interior transects, given its shade tolerance and past accounts of it growing in interior habitats at Grassy Hill (Greg Turner, personal observations), but I did not. Thus, the high canopy cover conditions that I measured in interior transects may have had some influence on the *Microstegium* distributions I found. Likewise, leaf litter may have been influential, as it has been reported to inhibit *Microstegium* seedling growth, survival, and recruitment in other regional forests (Oswalt and Oswalt 2007, Miller and Matlack 2010). Since litter depth generally increased away from roadsides, due to uniform canopy cover and lack of ground disturbances from humans, large animals, or wind, it is reasonable to infer that it too may have had some influence on the *Microstegium* distributions I found.

Distributions may also have been affected by seed dispersal, which is facilitated by animal and human activities, and by water (Barden 1987, Oswalt and Oswalt 2007). Given the abrupt falloff of *Microstegium* beyond roadsides, seed dispersal may have been lower in forest interiors than along roadsides. Lack of animal dispersal is unlikely, given that granivores common at Grassy Hill (e.g. birds and mice) move in both edge and interior habitats. Similarly, human activities, such as vehicles carrying seed on tires or roadwork that disturbs soil and ground cover (Schmidt 1989, Tyser and Worley 1992), were relegated to roadsides before and during study time. A more likely influential dispersal mode was water, given that no streams or erosion scours were found in any interior transect in any plot, while scours were seen in most T(-2) transects. Thus, *Microstegium* absence from interior transects may reflect a lack of

water borne seed dispersion in them. At this point, it is important to note that any assertions made about any factor that may have influenced *Microstegium* distributions and abundances were only speculative, given that the absence of *Microstegium* beyond most all T(2) transects negated statistical testing. Further, it is also likely that unknown factors, or interactions among factors, influenced *Microstegium* distributions and abundances.

In conclusion, *Microstegium* was restricted to roadsides at Grassy Hill at the time of the study. Given its high invasive potential, its absence from interiors was welcome news. However, periodic new surveys of *Microstegium* along preserve roads are suggested, as well as are studies examining potentially causative factors for *Microstegium* distributions and abundance. Meanwhile, efforts to maintain intact canopies and minimize leaf litter disruption in forest habitats fragmented by roads might be considered as a potentially pragmatic way to restrict *Microstegium* from forest interiors at the preserve.

ACKNOWLEDGMENTS

I thank Mike Leahy, former steward of Grassy Hill, for suggesting that I assess *Microstegium vimineum* at the preserve. I also thank Marianne Demkó, VA Western Community College, for her field assistance, and the VDCR, Division of Natural Heritage, for giving me access to the preserve. Finally, I thank the West Chester University, College of Arts and Sciences, Support and Development Award Committee for funding this project.

LITERATURE CITED

- Adams, S. N., A. M. Katharina, and A. M. Engelhardt. 2009. Diversity declines in *Microstegium vimineum* (Japanese stiltgrass) patches. *Biological Conservation* 142:1002-1010.
- Barden, L. S. 1987. Invasion of *Microstegium vimineum* (Poaceae), an exotic, annual, shade tolerant C4 grass, into a North Carolina floodplain. *American Midland Naturalist* 118:40-45.
- Bauer, J. T. and S. L. Flory. 2010. Suppression of the Woodland Herb *Senna hebecarpa* by the Invasive Grass *Microstegium vimineum*. *American Midland Naturalist* 165:105-115.
- Brothers, T. S. and A. Spingarn. 1992. Forest fragmentation and alien plant invasion of central Indiana old growth forests. *Conservation Biology* 6:91-100.
- Christen, D. C. and G. R. Matlack. 2009. The habitat and conduit functions of roads in the spread of three invasive plant species. *Biological Invasions* 11:453-465.
- Cole, D. N. and J. F. Weltzin. 2004. Environmental correlates of the distribution and abundance of *Microstegium vimineum*, in east Tennessee. *Southeastern Naturalist* 3:545-562.
- DeMeester, J. E. and D. deB Richter. 2010. Differences in wetland nitrogen cycling between the invasive grass *Microstegium vimineum* and a diverse plant community. *Ecological Applications* 20:609-619.
- Gibson, D. J., G. Spyreas, and J. Benedict. 2002. Life history of *Microstegium vimineum* (Poaceae), an invasive grass in southern Illinois. *Journal of the Torrey Botanical Society* 129:207-219.
- Glasgow, L. S. and G. R. Matlack. 2007. The effects of prescribed burning and canopy

- openness on establishment of two non-native plant species in a deciduous forest, southeast Ohio, USA. *Forest Ecology Management* 238:319–329.
- Heffernan, K. E., P. P. Coulling, J. F. Townsend, and C. J. Hutto. 2001. Ranking exotic invasive plant species in Virginia. Richmond, Virginia: Report no. 01-13. VDCR, Division of Natural Heritage, Richmond, Virginia.
- Huebner, C. A. 2003. Vulnerability of oak-dominated forests in West Virginia to invasive exotic plants: spatial and temporal patterns of nine exotic species using herbarium records and land classification data. *Castanea* 68:1-14.
- Leicht, S. A., J. A. Silander, Jr., and K. Greenwood. 2005. Assessing the competitive ability of Japanese Stiltgrass, *Microstegium vimineum* (Trin.) A. Camus. *Journal of the Torrey Botanical Society* 132:573–580.
- Miller, N. P. and G. R. Matlack. 2010. Population expansion in an exotic grass, *Microstegium vimineum*: a test of the channeled diffusion model. *Diversity and Distributions* 16:816-826.
- Oswalt, C. M. and S. N. Oswalt. 2007. Winter litter disturbance facilitates the spread of the non-native invasive grass *Microstegium vimineum* Trin A. Camus. *Forest Ecology and Management* 249:199–203.
- Oswalt, C. M., S. N. Oswalt, and W. K. Clatterbuck. 2007. Effects of *Microstegium vimineum* (Trin.) A. Camus on native woody species diversity in a productive mixed-hardwood forest in Tennessee. *Forest Ecology and Management* 242:727–732.
- Redman, D. E. 1995. Distribution and habitat types for Nepal *Microstegium* [*Microstegium vimineum* (Trin.) Camus] in Maryland and the District of Columbia. *Castanea* 60:270-275.
- Roberts, C. and C. M. Bailey. 2000. Physiographic map of Virginia counties. Virginia Division of Mineral Resources and the USGS of Mineral Producing Localities, Charlottesville, Virginia.
<http://web.wm.edu/geology/virginia/provinces/pdf/va_counties_phys.pdf?svr=www> (December 1, 2012).
- Schmidt, W. 1989. Plant dispersal by motorcars. *Vegetatio* 80:147-152.
- Schramm, J. W. and J. G. Ehrenfeld. 2010. Leaf litter and understory canopy shade limit the establishment, growth and reproduction of *Microstegium vimineum*. *Biological Invasions* 12:3195-3204.
- Turner, G. D. and M. Demkó. 2007. Exotic plant distributions along disturbance corridors at the Grassy Hill Natural Area Preserve, Franklin County, Virginia. *Banisteria* 30:19-26.
- Tyser, R. W. and C. A. Worley. 1992. Alien flora in grasslands adjacent to road and trail corridors in Glacier National Park, Montana (USA). *Conservation Biology* 6:253-262.
- USGS and VA Division of Mineral Resources. 1985. Rocky Mount Quadrangle, Virginia – Franklin County, 7.5-minute series, 36079-H8-TF-024. Richmond, Virginia.
- VDCR. 2003. Grassy Hill Natural Area Preserve. Richmond, Virginia.
<http://www.dcr.virginia.gov/natural_heritage/natural_area_preserves/grassyhill.shtml> (December 1, 2012).

- VDCR. 2009. Invasive alien plant species of Virginia.
<http://www.dcr.virginia.gov/natural_heritage/documents/invlist.pdf>
(December 1, 2012).
- Warren II, R. J., J. P. Wright, and M. A. Bradford. 2011. The putative niche requirements and landscape of *Microstegium vimineum*: an Asian invasive grass. *Biological Invasions* 13:471-483.

**Minutes of the Virginia Academy of Science
Council meeting May 27, 2011
Science Museum of Virginia**

The meeting was called to order at 9:35 am

In attendance: Mike Renfroe; Elsa Falls; Richard Brant; James O'Brien; Deborah L. Neely-Fisher; George C. Grant; Art Conway; Carolyn Conway; David Hagen; Richard Groover; Darcy Mays; Arum Verma; Weiner Wieland; Jim Martin; Susan Booth; David Crosby

President Mike Renfroe presiding:

President Renfroe started the May Council meeting by introducing Dr. Rich Conti, the 3rd director of the Science Museum of Virginia. Dr. Conti gave a delightful presentation on the future activities and directions of the SMV. Dr. Conti encouraged everyone at the council meeting to see the exhibits and especially the upcoming one called "Great Balls of Fire"

Officers' reports:

President:

President Renfroe gave the past president's written report (Dr. Verma). VAS has a MOU with Norfolk State University for hosting the 2012 Annual meeting Virginia. Tech will be the Host University in 2013. Dr. O'Brien was re-nominated to the board of Trustees. The 2012 slate of officers were approved. An invitation to attend the VAS Annual meeting was sent to the Governors' office.

President Elect:

Ralph Eckerlin, There was an increase in the number of VAS presentations.

Vice President:

No report

Secretary:

No report

Treasurer:

Treasury funds ok

Executive Officer:

Art Conway gave an up to date report on the Sitz awards. \$780 was awarded in 2010 and \$250 in 2011.

Dr. James O'Brien recommended that Deborah L. Neely-Fisher take over the Newsletter. The motion was referred to the Publication Committee.

VJAS Director:

VJAS Report Susan Booth

The JVAS had over 700 in attendance at UR. There were a few glitches. Some rooms didn't have sheets or towels, but these problems were corrected quickly. VJAS needs more readers for papers. VJAS would like to setup onsite registration for guest and student observers.

Committee Reports:**Archives:**

No report

Awards:

No awards at this time

Constitution and Bylaws:

No report

Environment:

No Report

Finance and Endowment:

No report

Fundraising:

No report

Junior Academy of Science:

No report

Long Range Planning:

No report

Membership: Richard Brandt

Working on increasing new membership. Gave special Thanks to Carolyn Conway. Asked all members to assist in increasing membership of the Academy.

Trust:

Quarterly reports on the Endowment Funds showed a loss ~10,000. The Trust Committee wants to reallocate the sub fund. The Trust wants to diversify funds. In response the Trust Committee wants to reallocate trust funds, the president plans to constitute a Finance Planning Task Force for the Future (FPTFF).

Rosemary Barra to be reappointed to the Trust Committee.

Research:

Written report submitted.

Horsley Award: Five papers were submitted.

The Horsley Award paper for 2011:

Eswar Prasad R. Iyer, Srividya Chandramouli Iyer, Ramakrishna Meduri, dennis Wang, and Daniel N. Cox

Department of Molecular and Microbiology, Krasnow Institute for Advance Study, George Mason University, Manassas, VA.

Class-specific profiling and *in vivo* RNAi screen reveal complex transcriptional regulatory networks mediating dendritic architecture.

Andrews Grant: Two proposals were submitted and are being reviewed.

Small Project Grants: Six proposals were submitted and are out for review.

Flora:

“Flora of Virginia” publication is slated for October 12, 2012.

Section Reports**Aeronautical & Aerospace Sciences:**

No report

Agriculture, Forestry and Aquaculture:

The section had a good turnout with lots of student presentations. Awarded 1st place and honorable mention awards.

Astronomy, Mathematics, and Physics with Materials Science:

No report

Biology with Microbiology:

No report

Biomedical and General Engineering:

No report

Botany:

Fifteen papers presented and 4 posters. Had 32 in attendance.

Chemistry:

Fourteen papers presented in this section with five posters. A special seminar was given.

Computer Science:

No report

Education:

Five papers given and one poster. Had 34 in attendance.

Environmental Science:

Twelve presentations with three no shows for this section.

Medical Sciences:

No Report

Natural History and Biodiversity:

Seventeen papers were given. Five posters presentations were set up. Twelve students gave presentations. Presentations came from six institutions. One first place award given with two honorable mentions.

Psychology:

Five paper presentations given as well as six posters. This section had a slight decline in number of presentations.

Statistics:

Ten presentations were given. Four were given by students. One first place award was given and one honorable mention. Section had up to 37 in attendance.

Structural Biology, Biochemistry and Biophysics:

Eighteen scheduled talks with several posters.

Old Business

VJAS is concerned about how the Trust for the Future is generating funds.

Dr. O'Brien reported on the Fellow Challenge is up by \$4000.

Dr. Brant reported on "The Thomas F. and Kate Miller Jeffress Memorial Trust"

New Business

Discussion was initiated concerning the forgiveness of the debt to VJAS. A motion was made to move this to the FPTF for further study and report to the Council.

Motions from the Trust Committee:

1. That the President appoints a Task Force to look at Academy financial

planning for the future.

2. That Rosemary Barra be re-elected by council to the Trust Committee for a 3-year term.

Both motions passed.

President Renfoe appointed Art Burke, Elsa Falls, Rosemary Barra, and Darcy Mays to the Financial Planning Task Force (FPTF)

From Susan Booth:

Motion: Give the Administrative Assistant \$1000 from the Dominion Funds for special expense as a bonus. Second and passed.

From Dr. O'Brien:

Motion: That the Council appoint Deborah Neely-Fisher as editor of the Virginia Scientists to begin September 1st, 2011. Second and passed.

President Renfoe directed that the record show that the Academy greatly appreciates and recognizes the years of service by Dr. James O'Brien as editor of the Virginia Scientist.

Motion for Adjournment was called and second. Council meeting was adjourned.

Respectfully submitted,
David Crosby, Secretary, VAS

NOTES

NOTES

Instructions to Authors

All manuscripts and correspondence should be addressed to the Editor. The Virginia Journal of Science welcomes for consideration original articles and short notes in the various disciplines of engineering and science. Cross-disciplinary papers dealing with advancements in science and technology and the impact of these on man and society are particularly welcome. Submission of an article implies that the article has not been published elsewhere while under consideration by the Journal.

Three complete copies of each manuscript and figures are required. Original figures need not be sent at this time. **OR**, a file of the manuscript in an acceptable word processing format, i.e., Word or WordPerfect. Authors should submit names of three potential reviewers. All manuscripts must be double-spaced. **Do not** use special effects such as bold or large print.

The title, author's name, affiliation, address and e-mail should be placed on a cover page. An abstract (not to exceed 200 words) summarizing the text, particularly the results and conclusions, is required. The text should follow the general format used by professional journals in the author's discipline and the Virginia Journal of Science has an on-line style manual. Literature cited in the text should follow the name-year format: (McCaffrey and Dueser, 1990) or (Williams et al., 1990). In the Literature Cited section at the end of the article, each reference should include the full name of the author(s), year, title of article, title of journal (using standard abbreviations), volume number and first and last page of the article. For a book, include author(s), year, title, pages or number of pages, publisher and city of publication. Examples:

McCaffrey, Cheryl A. and Raymond D. Dueser. 1990. Plant associations of the Virginia barrier islands. *Va. J. Sci.* 41:282-299.

Spry, A. 1969. *Metamorphic Textures*. Pergamon Press, New York. 350 pp.

Each figure and table should be mentioned specifically in the text. All tables, figures and figure legends should be on a separate pages at the end of the text.

Multiple author papers are required to have a statement in the acknowledgments indicating the participation and contribution of each author.

After revision and final acceptance of an article, the author will be required to furnish two error-free copies of the manuscript: 1) printed copy, single spaced, with tables and figure captions at the end of the document, and one set of original figures, each identified on the back by figure number and author's name; 2) a PC file in acceptable format containing the text file, tables and figure legends.

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