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117

Distribution and Call Parameters
of *Hyla chrysoscelis* and
Hyla versicolor in Michigan

James P. Bogart
Alan P. Jaslow

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LIFE SCIENCES CONTRIBUTIONS
ROYAL ONTARIO MUSEUM
NUMBER 117

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Distribution and Call Parameters of *Hyla chrysoscelis* and *Hyla versicolor* in Michigan

Abstract

Information from chromosomes, nucleolar determinations from preserved specimens, and mating call analyses revealed the presence of *Hyla chrysoscelis* populations in seven Michigan counties. Call analyses indicate that Michigan *H. chrysoscelis* are more similar to Wisconsin and "western" populations than to "eastern" populations of *H. chrysoscelis*. Most Michigan *H. chrysoscelis* occur sympatrically with *H. versicolor* which is widespread throughout the state.

Introduction

The present distribution of *Hyla chrysoscelis* and its morphological, tetraploid counterpart, *H. versicolor* has been estimated from pulse rate analyses of calling males involving relatively few populations (Blair, 1958; Johnson, 1966; Ralin, 1968, 1977; Zweifel, 1970; Gerhardt, 1974; Jaslow and Vogt, 1977).

Identification, and therefore determination of distribution, is complicated by the fact that pulse rate is temperature dependent. In some populations, a "hot" *H. versicolor* male recorded on a warm night might be confused with a "cold" *H. chrysoscelis* male recorded on a cool night (Zweifel, 1970; Gerhardt, 1978). Call differences may easily be perceived by ear without bioacoustic analysis if the species occur in sympatry and are both calling together at similar temperatures. The two call types were heard on June 14, 1974 during an overnight stay in Portage Lake State Park Campground, Jackson County, Michigan. Tape recordings were not obtained at that time but voucher specimens of each call type were captured and karyotyped. From the chromosomal information, it was evident that *H. chrysoscelis* occurs sympatrically with *H. versicolor* at that locality.

Finding *H. chrysoscelis* in Michigan was unexpected in that the closest published record of *H. chrysoscelis* to the west is northern Illinois (Brown and Brown, 1972) and the tentative map provided by Ralin (1968) indicated that *H. chrysoscelis* should not occur north of central Ohio and Indiana. Blair (1958), and more recently Gerhardt (1974, 1978), Maxson et al. (1977), and Ralin (1977) discuss a possible partitioning of *H. chrysoscelis*. This study was undertaken to determine the distribution and possible interpopulational affinities of the Michigan treefrogs.

Materials and Methods

Trips were made through some of the southern counties of Michigan from May through July, 1975. Calls were recorded using a Uher 4000S recorder at a tape speed of 19 cm/sec and a Uher M517 microphone. The tapes were analysed with a Kay Electric Sound Spectrograph (7030A) using an effective band width filter of 300 Hz at 1/2 playback speed for pulse rate analysis. Sections were run from calls played back at normal speed using the narrow band width filter (45 Hz) for frequency determinations. Call duration was determined by measuring sonograms using the middle three calls for each individual.

Esophageal temperatures of recorded individuals were taken using a Schultheis thermometer except for two *H. versicolor* males which were calling from the water, in which case the temperature of the water was used for regression analyses. Ten *H. chrysoscelis* males were recorded from five localities and nine *H. versicolor* males were recorded from five localities. Least squares regression analyses were used to calculate the regression of pulse repetition rate vs temperature, call duration vs temperature, and dominant frequency vs temperature.

Chromosomes were obtained from three *H. chrysoscelis* males and from six *H. versicolor* males at one sympatric locality (Portage Lake State Park). The methods used for obtaining chromosomes were the same as in Bogart (1968). Preserved specimens were borrowed from the Museum of Zoology at the University of Michigan and the University of Kansas Museum of Natural History for nucleolar number determinations using the procedure of Fernandez-Gomez et al. (1969) as outlined by Cash and Bogart (1978).

Results

The collected specimens of each call type, distinguished by call, at the Portage Lake State Park all possessed the expected chromosome complement of 48 chromosomes for *H. versicolor* and 24 for *H. chrysoscelis*. The karyotypes are basically similar to those published by Bogart and Wasserman (1972). The accumulated locality information for Michigan *H. chrysoscelis* and *H. versicolor* is included in Table 1 and on the map (Fig. 1). Most of the localities for *H. versicolor* were determined entirely from nucleolar counts from preserved frogs. These do not represent the entire preserved collection examined because some of the specimens did not reveal sufficient cellular detail to determine accurately the number of nucleoli. The relationship of pulse rate to temperature proved to be the only highly significant ($P < 0.005$) correlation for calling males of both species (Fig. 2). The regression formulae for Michigan *H. chrysoscelis* is: $Y = 3.78X - 33.93$ ($r^2 = .81$) and for *H. versicolor* is: $Y = 1.32X - 7.50$ ($r^2 = .90$). Determinations of call parameters for both species in Michigan is included in Table 2.

Table 1 Locality information for *Hyla chrysoscelis* and *H. versicolor* in Michigan. UM and KU numbers refer to catalogued specimens used for nucleolar determinations from the Museum of Zoology at the University of Michigan (UM) or the University of Kansas Museum of Natural History (KU).

County	<i>H. versicolor</i>	Locality	<i>H. chrysoscelis</i>
Alcona:	Sprinkler Lake (UM128633)		
Allegan:	Wayland (UM84198)		
Barry:	T3N/R9W/S32 ^a ; T1N/R9W/S32 ^a ; T2N/R8W/S1 ^a ; T3N/R8W/S36 ^a	T2N/R8W/S1 ^a ; T3N/R8W/S36 ^b T3N/R10W/S29 ^a ; T1N/R9W/S2 ^a T2N/R9W/S22 ^c	
Berrien:	Harbert (UM51186-7); Warrens dunes (UM54374)	Harbert (UM51188); Sawyer (UM53235)	
Calhoun:	T2S/R4W ^a ; Marshall (UM83266)		
Cheboygan:	Douglas Lake (KU55510)		
Clare:	Budd Lake (UM84726)		
Clinton:	Rose Lake (UM96164)		
Eaton:	T2N/R5W/S3 ^a ; Olivet (UM90743); Charlotte (UM83792)		
Gratiot:	Alma (UM56317)	Alma (UM56316)	
Huron:	Caseville (UM37898; UM37904) Sand Point (UM37902-3; UM37899)		
Ingham:	Whiteoak Twp. (UM56633; UM56728; UM56732); Leslie (UM83791)		
Ionia:	Boston Twp. (UM89637)		
Jackson:	Portage Lake State Park ^d ; T3S/R2E/S22 ^a ; T2S/R2E/S4 ^c ; T1S/R2E/S34 ^a ; T1S/R2E/S28 ^a Manchester (UM56571); Grass Lake (UM83793); (UM118458)	Portage Lake State Park ^d ; T2S/R2E/S4 ^c ; T1S/R2E/S34 ^a	
Kalkaska:	(UM65070); (UM69546)	(UM69340)	
Kalamazoo:	Agusta (UM83813); (UM77598)		
Keweenaw:	Lake Bailey (UM83777)		
Livingston:	T1N/R4E ^a ; T1N/R4E/S29 ^a ; Brighton (UM30656); Whitmore Lake (KU68852; KU68855-6)	T1N/R2E/S34 ^a	
Marquette:	Marquette (UM83890); (UM88440); Moccasin Lake (UM100193-4)		
Mecosta:	Paris (UM63136); Remus (UM11712)		
Newaygo:	(UM63457)		
Ogemaw:	Sage Lake (UM89648)		
Otsego:	Pigeon River (UM110529)		
Roscommon:	(UM122550)		
Sanilac:	Melvin (UM96167)		
Shiawassee:	Hopkins Lake (UM74539)		
Van Buren:	South Haven (UM103667); Wolf Lake (UM116309)		

Table 1 continued

County	<i>H. versicolor</i>	Locality	<i>H. chrysoscelis</i>
Washtenaw:	T3S/R4E/S23 ^b ; T1S/R4E/S4&9 ^a ; T2S/R3E/S6 ^a ; T2S/R3E/S5 ^a ; T1S/R3E/S28 ^d ; T1S/R3D/S20 ^a ; T2S/R5E ^a ; Ann Arbor (UM30805; UM30707; UM30654-5; UM99999; UM99408); Dixboro (KU68845-6; KU68848)	T1S/R3E/S28 ^c ; T2S/R3E/S5 ^a	
Wayne:	Dearborn (UM51920-1; Denton (UM31600)		

* Township/Range/Section number; ^a Determined by call only; ^b One specimen recorded; ^c Two specimens recorded; ^d Three specimens recorded.

Table 2 Call parameters in Michigan *Hyla chrysoscelis* and *Hyla versicolor* populations $\pm$ 95% confidence limits. Ranges are included in parentheses.

Species	N	Mean temperature (C)	Mean pulse repetition rate (pulses/sec)	Mean call duration (sec)	Mean dominant frequency (KHz)
<i>Hyla chrysoscelis</i>	10	23.2 (21.0 – 26.0)	53.2 $\pm$ 1.0 (40.7 – 67.5)	0.46 $\pm$ 0.06 (0.34 – 0.59)	2.52 $\pm$ 0.07 (2.17 – 3.00)
<i>Hyla versicolor</i>	9	22.5 (20.2 – 26.0)	23.3 $\pm$ 0.9 (18.7 – 26.0)	0.57 $\pm$ 0.08 (0.34 – 0.73)	2.29 $\pm$ 0.03 (2.13 – 2.55)

Discussion

Populations of *H. chrysoscelis* in Michigan appear to be concentrated in the southern counties and at all the localities but one (south of Cloverdale in Barry county) *H. chrysoscelis* occurs sympatrically with *H. versicolor*. The distribution of *H. chrysoscelis* is probably more widespread than indicated here as our sampling was concentrated, for field collecting and recording specimens, in a few areas in southern Michigan. The museum specimens were from scattered localities and, in most cases, the populations were represented by only one or a few individuals. Even if the two species occurred sympatrically, *H. chrysoscelis* might not be represented in a small sample. At the one allopatric locality, only two individuals of *H. chrysoscelis* were heard and under more favourable conditions, it is possible that *H. versicolor* would be encountered. Since our study concentrated on locating populations of *H. chrysoscelis*, many allopatric populations of *H. versicolor* were not examined in detail and most of the *H. versicolor* recorded for this study were, for convenience, from sympatric localities.

Personal observations from extensive collecting in other localities in various parts of the range of these two species, reveal the calling male *H. chrysoscelis* in Michigan to be similar to those from Wisconsin and different from those of Texas, Louisiana, and North Carolina. In Michigan, they are smaller and usually all green with few, if any, dorsal markings. The green colour is brighter and more intense than any of the green *H. versicolor* or the more southern *H. chrysoscelis* in their green colour phase and extends to the throat where it may appear yellow-green. The colour of Michigan calling *H. chrysoscelis* is reminiscent of the colour of the green treefrog, *H. cinerea*. Vocalizing *H. versicolor* in Michigan appear comparatively larger than the Michigan *H. chrysoscelis*, have dorsal markings, and are usually moss and kelly-green although some brown and grey individuals were calling.

The small number of *H. chrysoscelis* examined in our study does not provide an adequate sample size to draw any significance from mean snout-vent lengths nor is the colour difference apparent in preserved specimens. It is interesting to note, however, that Wisconsin *H. chrysoscelis*, which appear most similar to Michigan *H. chrysoscelis* were found to be smaller (mean size 35.2 mm) (Jaslow and Vogt, 1977) than *H. chrysoscelis* in Texas, with a mean size of approximately 40.5 mm (extrapolated from fig. 1 in Ralin, 1968). *H. versicolor* does not seem to possess this latitudinal difference in size and from these same studies, has a mean size of 42.6 mm (Wisconsin) and approximately 42.5 mm (Texas).

Gerhardt (1974) found that Georgia and South Carolina populations of *H. chrysoscelis* differed from Texas populations in two call parameters; pulse repetition rate and mean call duration. In Gerhardt's discrimination experiments, females preferentially chose males from their own region. Ralin (1977) also indicated that *H. chrysoscelis* could be divided, east and west, by pulse rate, call duration, and electrophoresis. He differentiated two populations of *H. versicolor*, north and south, which could be distinguished by call and electrophoresis. Southern populations had a faster pulse repetition rate. Microcomplement fixation studies by Maxson et al. (1977) have also indicated separation of *H. chrysoscelis* into two differentiated populations. Considering the possible differentiation between the various populations, it should be instructive to compare the Michigan populations with "eastern" and "western" *H. chrysoscelis* and "northern" and "southern" *H. versicolor*.

Call Duration

Both Gerhardt (1974) and Ralin (1977) provide evidence that the “eastern” *H. chrysoscelis* has a much longer estimated mean call than “western” *H. chrysoscelis*. In our analysis of call duration for Michigan *H. chrysoscelis* and *H. versicolor*, we found that regression equations (call duration vs temperature) were not statistically valid and correction for temperature was, therefore, not possible over our temperature range for either species. Our mean call duration (0.46 ± 0.06) compares most favourably with Gerhardt’s value of 0.45 ± 0.02 for Texas frogs (opposed to 0.75 ± 0.05 for Georgia and South Carolina *H. chrysoscelis*). Regression equations for call duration have only been provided by Ralin (1977) for his “eastern” ($Y = -0.04X + 1.73$) and “western” ($Y = -0.02X + 1.23$) populations of *H. chrysoscelis*. Substituting our mean temperature of 23.3 for X in Ralin’s equations, we obtain a duration of 0.80 (“eastern”) and 0.76 (“western”). Neither of these values would be considered close to our mean call duration for Michigan *H. chrysoscelis*. It is evident that regression lines, using Ralin’s equations for call duration, would intersect when $X = 25$ C and $Y = 0.73$ sec. Any possible discrimination with respect to call duration for Ralin’s populations would appear to be irrelevant at temperatures around 25 C.

Ralin (1977) indicated that the call duration for *H. versicolor* tended to be intermediate between the duration of “eastern” and “western” *H. chrysoscelis*. Our mean call duration for Michigan *H. versicolor* would be considered of intermediate duration when compared with Gerhardt’s values for *H. chrysoscelis* (above). Ralin’s regression equations for “southern” ($Y = -0.04X + 1.62$) and “northern” ($Y = -0.03X + 1.44$) *H. versicolor* would also produce intersecting regression lines when $X = 18$ C and $Y = 0.9$ sec. Substituting our average temperature of 22.5 C for X in Ralin’s equations, we obtain values of 0.72 (“southern”) and 0.86 (“northern”). Neither of these values would be considered close to our mean call duration for Michigan *H. versicolor*.

Pulse Repetition Rate

Pulse repetition rates were positively correlated with temperature for both *H. chrysoscelis* and *H. versicolor* in Michigan (Fig. 2). The regression lines in Fig. 2 compare the Michigan populations with *H. versicolor* from Ralin (1977). Published regression equations and pulse repetition rates are compared with Michigan populations in Table 3. *Hyla chrysoscelis* in Michigan has a pulse repetition rate which is most similar to “western” populations of *H. chrysoscelis* while Michigan *H. versicolor* would be most similar to “northern” *H. versicolor* populations. Ralin (1968, 1977) speculated that a greater pulse rate differential between *H. chrysoscelis* and *H. versicolor* was evident in sympatric populations and this could be the result of call reinforcement for pre-mating isolation. Most of the populations of *H. chrysoscelis* in Wisconsin and Michigan do occur sympatrically with *H. versicolor* and the greater distinction in the north might be attributed to reinforcement, but analyses of large allopatric populations would be necessary to test this hypothesis. It is evident that the slopes of the regression equations for *H. chrysoscelis* are all steeper than those for *H. versicolor* (Fig. 2; Table 3). In Michigan, *H. chrysoscelis* demonstrated a larger variance from the mean than did *H. versicolor* but, despite the

Fig. 2 Pulse repetition rate for *Hyla versicolor* and *H. chrysoscelis*. Determinations are plotted from the calls of Michigan male *H. versicolor* (dots) and *H. chrysoscelis* (circles). The regression lines for Michigan *H. versicolor* and *H. chrysoscelis* (solid lines) are compared with "northern" and "southern" *H. versicolor* and "eastern" and "western" *H. chrysoscelis* (dashed lines) using the regression equations provided by Ralin (1977).

variance, the probability that the slope for Michigan *H. chrysoscelis* is equal to the slope for Michigan *H. versicolor* is very low ($P = .0025$).

Some of the variation shown in the regression analyses (Table 3) could be an artifact produced by sample size used in the various studies or by the methods of recording temperature. Larger sample sizes should result in smaller standard deviations, confidence limit estimations and better correlations. Temperatures recorded from the esophagus resulted in slightly less variance and slightly greater correlations than did temperatures recorded from calling sites in Wisconsin (Jaslow and Vogt, 1977).

Mean Dominant Frequency

The mean dominant frequency for Michigan *H. chrysoscelis* was higher than that for *H. versicolor* (Table 2). Our regression equations (frequency vs temperature) were not statistically valid and the ranges were wide. The actual measurement of dominant frequency is complicated by the fact that these species have frequency modulated (FM) pulses.

Table 3 Comparison of pulse repetition rate in *Hyla chrysoscelis* and *Hyla versicolor* populations. Pulse rate (Y) at 24 C* is determined by substituting 24 for the temperature (X) in the regression equations.

Populations	Regression equation	Mean pulse repetition rate (pulses/sec @ 24 C*)	References
<i>Hyla chrysoscelis</i>			
Michigan	$Y = 3.78X - 33.93$	56.8	This study
Wisconsin	$Y = 2.59X - 2.90$	59.3	Jaslow and Vogt (1977)
Texas	$Y = 2.36X - 3.40$	53.2	Ralin (1977)
Texas and Missouri	$Y = 3.05X - 15.80$	57.4	Gerhardt (1978)
Texas		55.1	Gerhardt (1974)
Georgia, South Carolina		44.6	Gerhardt (1974)
Oklahoma, Mississippi, Georgia, South Carolina, and North Carolina	$Y = 2.88X - 21.75$	47.4	Ralin (1977)
New Jersey, Delaware and Virginia	$Y = 2.41X - 8.46$	49.4	Zweifel (1970)
<i>Hyla versicolor</i>			
Michigan	$Y = 1.32X - 7.50$	24.2	This study
Wisconsin	$Y = 1.30X - 4.95$	26.2	Jaslow and Vogt (1977)
New Jersey, Delaware	$Y = 1.43X - 7.16$	27.2	Zweifel (1970)
New Jersey, New York	$Y = 1.22X - 4.30$	25.0	Ralin (1977)
Texas	$Y = 1.19X - 0.51$	29.1	Ralin (1977)
Missouri	$Y = 1.15X - 2.30$	25.3	Gerhardt (1978)

* 24 C was chosen for comparative purposes in order to incorporate Gerhardt's (1974) values which were corrected to 24 C.

Conclusion

The range map for *H. chrysoscelis* produced by Ralin (1968, 1977) must now be amended to include Wisconsin (Blair, 1958; Jaslow and Vogt, 1977), Missouri (Gerhardt, 1978), and Michigan. From the available evidence, Michigan *H. chrysoscelis* has call characteristics which are most similar to "western" populations and Michigan *H. versicolor* could be aligned with "northern" populations. A close examination of the specimens and localities previously used for the distinction of "eastern" and "western" *H. chrysoscelis* or "northern" and "southern" *H. versicolor* has, for the most part, relied on geographically similar populations (perhaps, in some cases, the same recordings?) (See localities and acknowledgements in Gerhardt, 1974, 1978; Ralin, 1968, 1977; Zweifel, 1970.) Previously, "western" populations were essentially central Texas populations. In the north, the "western" *H. chrysoscelis* ranges farther east than the Texas or Mississippi "eastern" *H. chrysoscelis*.

The information from call duration and frequency indicates that there must be considerable variation within and between populations and these parameters are probably not as important as pulse repetition rate for discrimination. There appears to be a greater divergence in pulse repetition rate between sympatric *H. versicolor* and *H. chrysoscelis* in the north which may be the result of character displacement (Ralin, 1968, 1977). Analyses of additional populations, and especially allopatric northern populations of *H. chrysoscelis* are needed before we accept Ralin's tenet. Perhaps populations from intermediate geographical areas will demonstrate clinal relationships with respect to divergence in pulse repetition rate or slope when compared with *H. versicolor*. Experimentally produced triploid hybrids are viable and do produce adult males (Johnson, 1966; Bogart, unpublished) but, considering all the sympatric populations of *H. chrysoscelis* and *H. versicolor* which have been studied, only one "intermediate call" of a putative hybrid has ever been reported (Zweifel, 1970). Discrimination, therefore, does not seem to be a problem for the frogs. Future studies from a larger number of populations in various regions will help to alleviate the present problems concerning distribution and the call parameters which are actually utilized by the populations for isolation, and possible intraspecific partitioning.

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