

Microgeographical variation in song repertoire and structure between the leks of Green Violetears *Colibri thalassinus* in central Mexico

Carlos LARA^{1*}, Vanessa MARTÍNEZ-GARCÍA¹ & Juan Francisco ORNELAS²

¹Centro de Investigación en Ciencias Biológicas, Univ. Autónoma de Tlaxcala, Km 10.5 Autopista Tlaxcala-San Martín Texmelucan, San Felipe Ixtacuixtla, Tlaxcala 90120, MÉXICO

²Departamento de Biología Evolutiva, Instituto de Ecología, A.C., Carretera Antigua a Coatepec 351, El Haya, Xalapa, Veracruz 91070, MÉXICO

*Corresponding author, e-mail: laracar@posgradouatx.com.mx

Lara C., Martínez-García V., Ornelas J. F. 2015. Microgeographical variation in song repertoire and structure between the leks of Green Violetears *Colibri thalassinus* in central Mexico. *Acta Ornithol.* 50: 23–32. DOI 10.3161/00016454AO2015.50.1.004

Abstract. Hummingbird vocalizations are usually displayed in two contexts. When foraging or defending territories, males and females usually display relative simple calls as territorial advertisement, while during courtship males perform individually elaborate songs to mate attraction or through singing assemblies (leks). About 15% of all described hummingbird species form leks or singing assemblies during the breeding season. However, the vocal repertoire displayed by males in these arenas has been rarely described. Here we studied the songs of Green Violetear *Colibri thalassinus* in a temperate forest from central Tlaxcala, Mexico, to document structure and variation within and between singing assemblies established over 3 years. We found 41 well differentiated and structurally complex syllable types across eleven recorded singing groups of Green Violetears (≈ 12 syllable types per singing group). The songs of most birds were repeats of the same 3–4 syllables, but the syllables types emitted can vary. Birds from the same lek tend to have similar song. Comparison of syllable sharing within-groups showed a Jaccard's similarity coefficient averaging 0.51 ± 0.09 (mean \pm SD), and 0.24 ± 0.01 for among groups comparisons. These differences were corroborated by bootstrapping tests finding no significant similarity among males from different leks. However, significant similarity in syllable sharing was found between males from the same lek. No significant correlation was found between song similarity and geographic distances between leks. We have documented that static songs of lekking Green Violetears are more complex than had previously suggested. The variation of vocal repertoire among leks indicates the apparent presence of "vocal dialects" that are variable across time and space. The song variation in the context of discrimination ability in vocal neighborhoods requires further research.

Key words: *Colibri thalassinus*, hummingbirds, leks, song structure, vocal variation, vocalization, song variability

Received — July 2014, accepted — June 2015

INTRODUCTION

Historically, the study of bird vocalizations has been focused mainly on oscines, rarely on parrots and hummingbirds (Catchpole & Slater 1995, Kroodsma et al. 1996). However, hummingbirds Trochilidae, typically considered as nearly voiceless, produce various calls and songs and, in some species, songs composed of syllables with a highly variable and complex acoustic structure (Baptista 1996, Ficken et al. 2000, Ornelas et al. 2002, Ferreira et al. 2006, González et al. 2011). Like parrots and songbirds, hummingbirds have also developed the trait of vocal learning (Jarvis et al. 2000), and although most hummingbird species sing simple one- or two-syllable songs (Atwood et al. 1991, Kroodsma et al. 1996, Araya-Salas &

Wright 2013), there are species intermediate in vocal complexity (Baptista & Schuchman 1990, Gaunt et al. 1994, Kroodsma et al. 1996), and species with intricate and extraordinarily complex and versatile vocal signals (Kroodsma et al. 1996, Ficken et al. 2000, Jarvis et al. 2000, Ornelas et al. 2002, González & Ornelas 2005, 2009, Ferreira et al. 2006). Therefore, hummingbirds offer many opportunities for studying the function and variation in song advertisement. Males, and in some species, females vocalize in several different contexts (Wiley 1971, Gaunt et al. 1994, Ficken et al. 2000), but the acoustic characteristics, structure and organization, function, and the evolution of vocal display repertoires in hummingbirds are largely unknown (Kroodsma et al. 1996). Knowledge of the context in which birds sing is

important because it can provide valuable clues as to the possible functions of song (Catchpole & Slater 1995).

Hummingbird vocalizations (songs and calls) are usually displayed in two contexts. When foraging, hummingbirds (both sexes) usually display relative simple calls as territorial advertisement. During courtship males may, individually or through song-assemblies (leks), perform elaborate songs to attract a mate (Snow 1968, Wiley 1971, Ficken et al. 2000, Ornelas et al. 2002, Williams & Houtman 2008). Some studies have shown that songs displayed in this sexual context can be very flexible, even reaching differentiation of vocal neighborhoods (Gaunt et al. 1994, Yang et al. 2007, González & Ornelas 2009). Despite this, the behavioral variation in vocal displays among hummingbirds has been infrequently studied. In a recent review, Martínez-García et al. (2013) found that up to 49 hummingbird species had been reported to form leks. Males congregate in leks during the breeding season and individually occupy and defend the same singing territory on the mating arena, typically from an exposed perch, broadcasting vocalizations into the environment. Males sing continuously throughout the day to get the attention of the visiting females that have nearly completed the nest (Wagner 1945, González & Ornelas 2009). Surprisingly, the way females choose males and the vocal repertoire displayed by males in these arenas has not been described for most of the lekking species.

Green Violetear *Colibri thalassinus* is a medium-sized, sexually monochromatic species. Both sexes are iridescent green in color, with glittering violet auriculars and a bluish-violet patch on chest. The tail-feathers are bronzy blue-green with a broad blackish subterminal band (Howell & Webb 1995, Johnsgard 1997). Its geographic distribution is widespread, comprising central Mexico to Guatemala, El Salvador, Costa Rica, Colombia, Ecuador, and Peru. The species is found at altitudes from 1200 to 3000 m a.s.l. in cypress and pine-oak forest during the breeding season and between breeding seasons migrate up to fir forest from 2900 to 3500 m a.s.l. (Wagner 1945, Howell & Webb 1995, Johnsgard 1997). The Green Violetear is a lekking hummingbird with a song composed of several notes (Gaunt et al. 1994, Barrantes et al. 2008, Martínez-García et al. 2013). Leks are typically composed of three to five males that occupy a limited area, usually only 8 to 15 meters apart, so that they can hear and see each other. At these sites, the males sing continuously on exposed

perches to attract the attention of the females during the breeding season (Wagner 1945, Martínez-García et al. 2013). Males apparently do not pay attention to each other and no agonistic behaviors between them occur at the arena. Males sing continuously throughout the day to get the attention of the visiting females that have nearly completed the nest (Wagner 1945). The males advertise their presence and gain the attention of the female by their loud vocalizations and followed by a race to the nesting territory accompanied with display undulating flights (Wagner 1945). Although it is possible to distinguish different leks during a breeding season (Martínez-García et al. 2013), acoustic variation among males displaying at leks has not yet been analyzed.

The objectives of our study were two-fold: (1) To compare acoustic variation and syllable sharing within and among eleven singing assemblies of Green Violetears in a temperate forest in central Mexico, and (2) to test for significant correlation between song similarity and geographic distances among leks.

METHODS

Study site

Green Violetears were studied from 5 June to 6 October in a 3 year study (2009–2011), at the La Malinche National Park (LMNP), Tlaxcala, Mexico (19°14'N, 98°14'W, with elevation ranging from 2000 to 4461 m a.s.l.). The vegetation in the study area is mainly a mosaic of pine forest and second growth vegetation (Villers et al. 2006). At the LMNP, leks of Green Violetears are established under varying spatial and temporal conditions, mainly in secondary vegetation zones with individual singing territories typically established on pine trees separated 8–15 m (Lara 2006, Martínez-García et al. 2013). Green Violetear leks were established in the edges of the LMNP, in areas close to the perimeter of the LMNP. These sites are composed of recovering pine forest (*Pinus montezuma*, *P. pseudostrobus*, *P. leiophylla*, and *P. hartwegii*) with open spaces. Studied leks were distributed into seven different sites of the LMNP: Site 1 — Caseta, 2 — km 78, 3 — km 69, 4 — Albergue, 5 — Ixtenco, 6 — Cabañas, and 7 — Km 73 (Fig. 1). In 2009, only one lek was established (Caseta), three leks (km 78, km 69, and Albergue) in 2010, and seven leks in 2011 in the aforementioned sites, all of which varied in the number of members that belong to the lek. These sites

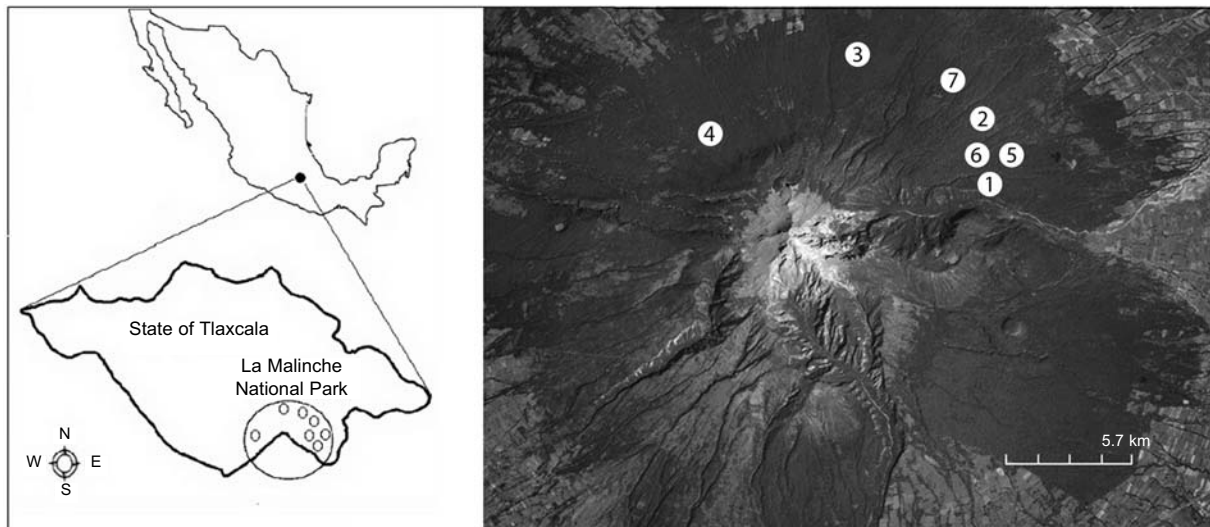


Fig. 1. Geographical location of the La Malinche National Park (LMNP) in the state of Tlaxcala, Mexico. White circles indicate the location of the leks studied: Sites: 1 — Caseta, 2 — km 78, 3 — km 69, 4 — Albergue, 5 — Ixtenco, 6 — Cabañas and, 7 — km 73. Image from Google Earth version 7.1.2.2041.

typically contained flowering patches of firecracker bushes *Bouvardia ternifolia* (Rubiaceae) and beardtongues *Penstemon roseus* (Scrophulariaceae) and were the feeding territories with the greatest abundance of hummingbirds (Martínez-García et al. 2013). The leks were established in an altitudinal strip ranging from 2060–2800 m a.s.l. From the time of its formation, the average (\pm SE) lek lasted 30 active days (\pm 9.52), ranging from 10–50 days, with a constant turnover of individuals in each lek (Martínez-García et al. 2013). The average distance between neighboring leks was 2.39 km (\pm 0.9 km; range = 0.54–9.65 km), while the average distance between perches of singing individuals within a given lek was 99.72 m (\pm 4.95 m; range = 28.75–715.98 m). These data indicate that individuals were quite dispersed and too far out of sight to maintain visual contact, but they were able to maintain vocal contact among each other. Furthermore, leks of this species can be established in previously occupied sites, including the use of the same perch during successive years (Martínez-García et al. 2013). If the same individual visits different leks or kept singing only in one of them is unknown.

Previous results at the study site showed that individuals forming part of a lek begun singing on their perches at sunrise and continued doing so during the whole day with a decrease in activity in the afternoon. The individuals who formed the leks sang from three to five different notes, accumulating a vocal repertoire of 40 notes inside the leks (static singing) and one note in the singing

displayed when foraging (Martínez-García et al. 2013). Three forms of short and monotonous calls that birds repeat continually can be differentiated. The first is emitted when standing on an exposed branch, *huit ti titatia*; *huit tita*, *tetahuit tetahuit*, and *teta tetuit tetahuit*, continually and with little intensity. The second, with greater intensity and louder, *ahuit ahuit ta ta* and *huit ti ta huit ti ta*, is done moving the head from one side to another. And the third type, *huita huita*, is repeated continually while turning the head from one side to another and at the same time raising the feathers of the head and the back (Wagner 1945).

Capturing and color marking of hummingbirds

To evaluate the song structure of lekking Green Violetears, we captured and color marked the majority of individuals of this species once the reproductive season began in the study area. Several weeks prior to and during the initial stages of lek formation, 134 hummingbirds were captured with mist nets in different areas of the LMNP, most typically in those where abundant patches of flowering firecracker bushes and beardtongues were observed. Out of those mist-netted, 114 individuals were color-marked following González & Ornelas (2009). Briefly, marks consisted of small colored circles made of foam sheets, attached with nontoxic fast drying hard glue to the back of the Green Violetears (González & Ornelas 2009). These markings last 2–3 months until the back feathers are replaced by molt

(Martínez-García et al. 2013). Marked individuals were observed defending territories over several weeks after manipulation; none of these birds disappeared or was found dead in the course of this study. Each individual carried an exclusive color combination for further identification. We were able to locate, identify, and record the songs of color-marked lekking males. Because individual identification of recorded males was not possible, and color marks are not permanent, it was not possible to know if the same individuals occupied different leks within a season or the same leks and singing territories in consecutive seasons.

Leks and song recordings

In addition to the capturing and marking of individuals, field observations were conducted in different areas of the LMNP to detect the presence of leks. We identified a potential lek area when a group of individuals were vocalizing for 10–20 consecutive minutes on permanent perches at a constant rate, and once identified these areas the lek activity was daily monitored for about a week. The observations ceased for a given area if individuals were not vocalizing constantly and the establishment of a lek was then not confirmed in these areas. If the establishment of a lek was settled (i.e. lek activity for more than one week), the spatial distribution of each singing individual within the arena was recorded using a GPS (Garmin model Oregon 300). In order to confirm the temporal persistence of the lekking area, subsequent observations at 3-days intervals were conducted for the whole breeding season (June to October) and lek establishment was confirmed, and during this time every new individual joining the lek was recorded.

Vocalizations were recorded across all day approximately twice a week with two digital SONY PCM-M10 recorders, a Sennheiser MKH-70 shotgun microphone, and headphones. Spectrograms and acoustic measurements of recordings of all individuals were generated automatically with Raven Pro 1.3 (Charif et al. 2008). Recordings of marked males were digitized at a sampling rate of 44.1 kHz and stored as 16-bit samples. Each male was recorded once as to get 4 full songs per individual for a day. Thus, after several days, all members within a lek were recorded. Spectrograms were produced with a 349.7-Hz filter bandwidth and a frame length of 512 points (= 11.6 ms). The temporal and frequency-grid resolution of the spectrograms were 64 points (= 1.45 ms) and 2.69 Hz, respectively.

Acoustic and statistical analysis

We used 352 song recordings of 88 individuals from all lek areas for analyses. Each individual song was displayed on the computer screen to identify visually syllable types as units. Syllables were letter-coded and categorized following González & Ornelas (2009). The vocal lek repertoire was defined as continuous series of vocalizations composed of different number or types of syllables emitted by individual birds attending singing assemblies. To document song structure and acoustic variation within and between singing groups, we measured and estimated seven variables on each spectrogram and averaged values for each lek (Fig. 2, Table 1).

Acoustic variation was assessed within and between Green Violetear songs from the studied leks. We conducted a principal components analysis (PCA) with averaged values for each bird ($n = 88$; Table 2) of all acoustic measures using SPSS version 11.5.2.1 (SPSS Inc., Chicago, IL). Data were $\log(x + 1)$ (duration of syllables, maximum and minimum frequency, bandwidth and duration of silent intervals) or square-root transformed (number of syllables and number of syllable types) prior to analysis to achieve normality, but untransformed data are reported in tables and figures. Furthermore, data were subjected to the Kaiser-Meyer-Olkin (KMO) measure of sampling adequacy, which recorded a substantial value of 0.750. The KMO and Bartlett's measure are used to measure sampling adequacy in the use of PCA (Jackson 2003). The KMO statistic varies between 0 and 1; a value of zero indicates that the sum of partial correlations is large relative to the sum of correlations, indicating diffusion of pattern of the correlations and hence factor analysis is likely to be inappropriate (Jackson 2003). After factor

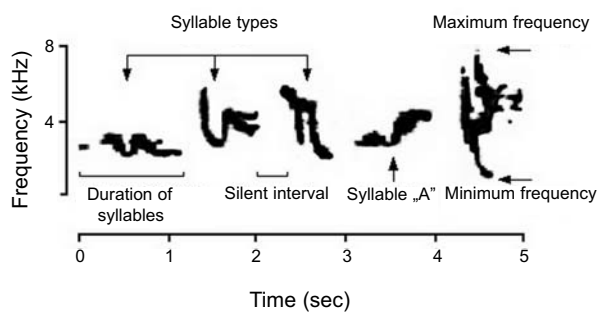


Fig. 2. Acoustic analysis of Green Violetear song and the parameters calculated. Spectrogram illustrates an example of one song produced by a hummingbird male in a lek.

Table 1. Variables measured in Green Violetears songs. Data are means \pm SE across individuals within each lek. Numbers in parenthesis are individual birds estimated per lek. Leks were studied in 2009, 2010, and 2011 — suffixes respectively -09, -10, -11.

| Variable | Singing group (lek) | | | | | | | | | | |
|----------------------------------|----------------------|----------------------|----------------------|----------------------|-----------------------|----------------------|----------------------|----------------------|-----------------------|-----------------------|----------------------|
| | Site 1-09 (N = 8) | Site 2-10 (N = 6) | Site 3-10 (N = 8) | Site 4-10 (N = 4) | Site 1-11 (N = 10) | Site 2-11 (N = 7) | Site 3-11 (N = 8) | Site 4-11 (N = 7) | Site 5-11 (N = 13) | Site 6-11 (N = 10) | Site 7-11 (N = 7) |
| Duration of syllables (s) | 0.05 \pm 0.0 | 0.03 \pm 0.0 | 0.06 \pm 0.0 | 0.04 \pm 0.0 | 0.04 \pm 0.0 | 0.03 \pm 0.0 | 0.04 \pm 0.03 | 0.03 \pm 0.0 | 0.05 \pm 0.0 | 0.05 \pm 0.0 | 0.03 \pm 0.03 |
| Number of syllables | 3.0 \pm 0.0 | 3.3 \pm 0.3 | 4.2 \pm 0.3 | 3.2 \pm 0.1 | 3.1 \pm 0.2 | 3.7 \pm 0.3 | 3.7 \pm 0.0 | 3.3 \pm 0.1 | 3.0 \pm 0.0 | 3.7 \pm 0.1 | 3.1 \pm 0.2 |
| Number of syllable types | 12.3 \pm 0.3 | 3.0 \pm 0.0 | 16.7 \pm 0.9 | 10.4 \pm 0.3 | 14.0 \pm 0.3 | 12.2 \pm 0.0 | 14.2 \pm 0.1 | 15.0 \pm 0.0 | 10.7 \pm 0.4 | 11.1 \pm 0.2 | 13.2 \pm 0.4 |
| Maximum frequency (kHz) | 9.4 \pm 0.2 | 7.9 \pm 0.1 | 9.6 \pm 0.1 | 10.1 \pm 0.2 | 9.7 \pm 0.3 | 8.9 \pm 0.1 | 9.6 \pm 0.4 | 10.1 \pm 0.2 | 9.5 \pm 0.3 | 9.9 \pm 0.1 | 9.3 \pm 0.1 |
| Minimum frequency (kHz) | 3.7 \pm 0.1 | 4.5 \pm 0.5 | 3.4 \pm 0.2 | 3.2 \pm 0.1 | 3.9 \pm 0.1 | 3.7 \pm 7.3 | 3.7 \pm 0.0 | 3.5 \pm 0.1 | 3.5 \pm 0.0 | 3.6 \pm 0.2 | 3.7 \pm 0.3 |
| Bandwidth (kHz) | 6.1 \pm 0.2 | 3.8 \pm 0.9 | 5.9 \pm 0.3 | 6.2 \pm 0.6 | 4.5 \pm 0.4 | 4.4 \pm 0.6 | 5.6 \pm 0.4 | 6.3 \pm 0.4 | 5.3 \pm 0.6 | 5.2 \pm 0.5 | 4.4 \pm 0.5 |
| Duration of silent intervals (s) | 0.31 \pm 0.1 | 0.44 \pm 0.0 | 0.40 \pm 0.1 | 0.28 \pm 0.0 | 0.29 \pm 0.0 | 0.28 \pm 0.0 | 0.27 \pm 0.0 | 0.31 \pm 0.0 | 0.35 \pm 0.1 | 0.29 \pm 0.0 | 0.29 \pm 0.0 |

scores were calculated, these were used as dependent variables to evaluate variation among groups using a series of nonparametric, one-way analyses of variance (Kruskal-Wallis ANOVA) with STATVIEW (Abacus Concepts, Berkeley, CA). Besides comparing acoustic variables among singing groups, with the aid of ESTIMATES (Colwell 2000), we compared syllable sharing among and within leks by using Jaccard's similarity coefficient (S_j). We quantified pairwise similarity among songs of individual birds from all leks on the basis of the presence/absence of syllables in their vocal repertoires. Thus, $S_j = a/(a + b + c)$, where a is the number of syllables shared by two singing leks (A and B), b is the number of syllables present in the repertoire of the A lek but absent in the repertoire of the B lek but absent in the repertoire of the A lek. The range of the coefficient of Jaccard is supposed to be 0 (no similarity) to 1 (complete similarity). To validate similarity in repertoire within and between leks, we compared similarity between randomly selected males from the same and different leks by using a bootstrap method (Efron & Tibshirani 1998). Differences were considered statistically significant if the 95% bootstrap confidence interval, calculated with 1000 iterations, did not bracket zero. Finally, the Mantel correlation statistic (Mantel 1967) was computing using PAST version 3.01 (Hammer et al. 2001) to test for significant correlation between song similarity among leks (the obtained Jaccard's similarity coefficients) and geographical distance (distance between leks obtained from GPS measurements).

RESULTS

Lek and singing behavior

Green Violetear males sang constantly throughout the day and perched typically on the same branch. They seem to prefer singing loudly while perching on small dry twigs usually high up in pine trees. Males were singing on their territorial perches for ca. 25 minutes, producing repeatedly the same short phrases of 2 to five short duration syllables (0.048 ± 0.005 seconds, CV = 0.25) separated by silent intervals (0.311 ± 0.013 , CV = 0.17) producing 80–230 notes per minute.

Song structure

Eleven leks composed of a variable number of members were recorded into seven different

areas of the LMNP throughout the study (Fig. 1). Most leks were separated from one to another by more than 2.5 km. A total of 88 color-marked individuals were identified as members of these singing assemblies, obtaining a total of 352 recordings for analysis (4 full songs per individual). Green Violetear songs are rhythmic metallic chippings (ca. 4 different syllable types sung by individual at all, $CV = 0.11$), punctuated irregularly and often prolonged for several minutes (15–25 min). We found 41 distinct syllable types (coded from units A to AN) across the eleven leks (ca. 12 syllable types per lek; $CV = 0.29$; Fig. 3). Syllables usually lasted < 2 s, and were sung at a high rate (up to 6 syllables per second). No introductory syllable emission was recorded for the song. The frequency ranged from a minimum of 3.7 ($CV = 0.09$) to 9.6 kHz ($CV = 0.06$) as the maximum (see Table 1).

Microgeographic song variation

The PCA analysis of the seven acoustic variables yielded three components (eigenvalues > 0.50), which accounted for most (86%) of the variation recorded in Green Violetear songs (Table 2). The KMO measure achieved a high value (0.757) suggesting the adequacy of the sample size for the PCA. The Bartlett's test of sphericity was also significant suggesting that the population was not an identity matrix. The conventional rule about communality values is that extraction values (eigenvalues) of more than 0.50 at the initial iteration indicates that the variable is significant; and should be included in the data for further analysis or otherwise removed. The eigenvalue and factor loadings were set at conventional high values of 1.00 and 0.50, respectively (Jackson 2003). Applying the latent root criterion on the number of principal components to be extracted suggests that 3 components should

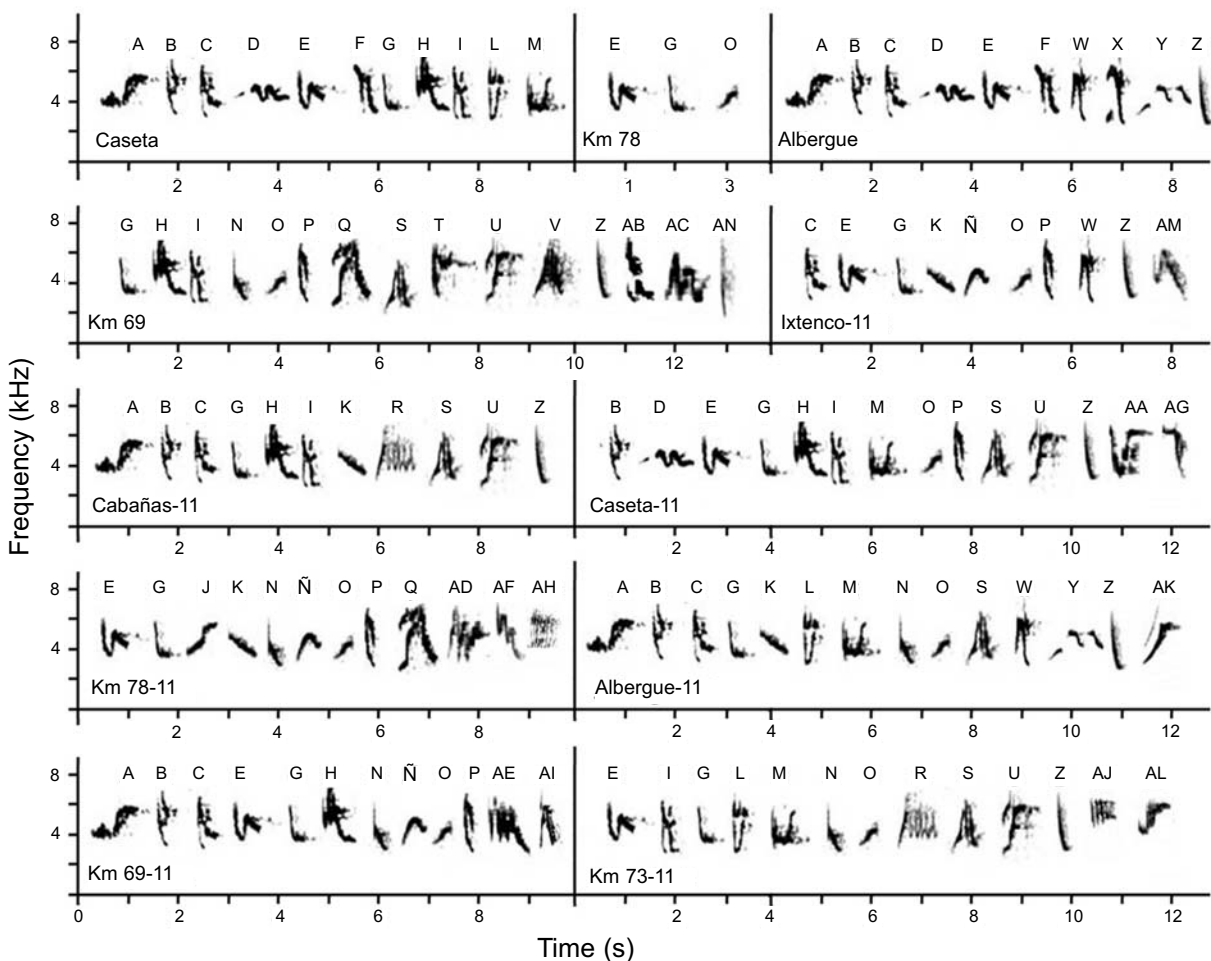


Fig. 3. Composite spectrograms showing all syllable types (alphabetical codes) recorded in each lek throughout the study.

Table 2. Principal components analysis of the correlation matrix among seven acoustic variables in 11 singing groups (leks) of Green Violetear at La Malinche National Park, Tlaxcala, Mexico. Correlations loadings above 0.60 are in bold face.

| Variables | PC1 | PC2 | PC3 |
|----------------------------------|--------------|-------------|-------------|
| Duration of syllables (s) | 0.50 | 0.67 | -0.33 |
| Number of syllables | 0.22 | 0.69 | 0.63 |
| Number of syllable types | 0.81 | 0.02 | 0.37 |
| Maximum frequency (kHz) | 0.93 | -0.20 | -0.08 |
| Minimum frequency (kHz) | -0.92 | 0.03 | 0.08 |
| Bandwidth (kHz) | 0.83 | 0.09 | -0.34 |
| Duration of silent intervals (s) | -0.53 | 0.71 | -0.29 |
| Variance (%) | 52.37 | 21.33 | 12.41 |

be extracted, as their respective eigenvalues are greater than one. Principal component 1 (PC1, 52.37% of the variance) exhibited positive loadings for the number of syllable types, maximum frequency and bandwidth, and negative for minimum frequency. Principal components 2 and 3 (PC2 and PC3, 21.33% and 12% of the variance, respectively) were mainly explained by the duration of silent intervals, and the duration and number of syllables. The apparent lack of group differentiation in the PC1 versus PC2 scatterplot suggests that intragroup variability accounted for much of the observed acoustic variation (Fig. 4). We found no significant differences among groups for the PC components obtained (Kruskal-Wallis ANOVA; PC1 — $\chi^2 = 13.45$, $df = 352$, $p = 0.21$; PC2 — $\chi^2 = 9.33$, $df = 352$, $p = 0.11$; PC3 — $\chi^2 = 10.19$, $df = 352$, $p = 0.28$).

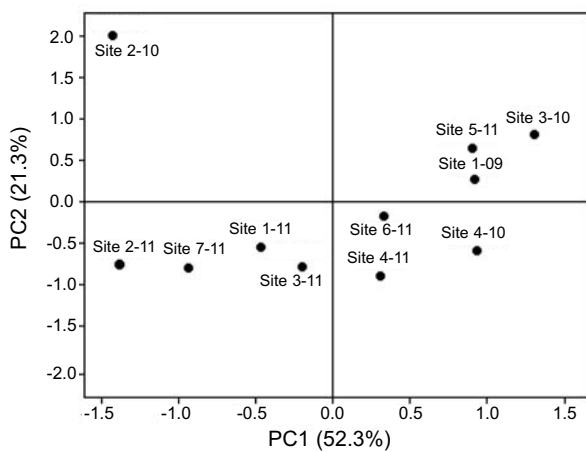


Fig. 4. Bivariate plot of PC factor scores (73.7% of the total variance in the data) for seven acoustic variables of Green Violetear songs from 11 leks. We show the plane of axes 1 and 2. Variation explained by each axis is given in parenthesis.

Comparison of syllable sharing among groups ($n = 55$) showed a Jaccard's similarity coefficient averaging of 0.24 ± 0.01 (mean \pm SD) and 0.51 ± 0.09 for within-group comparisons ($n = 4004$). After resampling, these were corroborated finding no significant similarity among males from different leks (0–0.31, 95% confidence interval of the mean). However, significant similarity in syllable sharing was found between males from the same lek (0.47–0.63, 95% confidence interval of the mean). No significant correlation was found between song similarity and geographic distances between leks (Mantel test: $r = 0.204$, $p = 0.23$).

DISCUSSION

We have documented that static songs of lekking Green Violetears are complex. Our results have shown that at the microgeographic scale there is great variation in song structure and vocal repertoire in leks from this species, which is not primarily explained by the geographical distance between the singing groups. Although songs in most individuals from a lek are a repetition of the same syllables, these can be sung in a different order. Also, although the number of syllables displayed in each lek was similar, its wide diversity of types is intriguing.

Although the pioneer onomatopoeic descriptions for the song of the Green Violetear in Mexico (Wagner 1945), Guatemala (Skutch 1967), and Costa Rica (Stiles & Skutch 1989), showed a large variation in the species, studies on the apparent complexity of its song are scarce. In the first study on the singing pattern for the species, Gaunt et al. (1994) mentioned that in the Cordillera de Talamanca between Cerro Vueltas and Cerro de la Muerte (Costa Rica), males gathered in a lek (7 individuals) sang the same song with respect to individuals from another lek (11 individuals). Also, they mention that the songs displayed in both leks were different to songs of solitary males (11 individuals). Although this work showed that birds in neighboring populations share song types and similarity decreases with geographical distance, acoustic variation among males displaying at leks were not analyzed. Fourteen years later, in the same study area, Barrantes et al. (2008) analyzed the acoustic variation of 19 males at one lek. Their results showed that the songs consisted of two elements (syllables), which can be sung repeatedly alone

or combined. Here we focused on lekking males during the breeding season, in which the use of songs from marked individuals gives us greater certainty of variation within and between the leks studied.

At LMNP the song of the Green Violetear is sung in phrases composed of 2–4 short duration syllables (< 2 s), which can be emitted in different combinations, as was previously shown by Gaunt et al. (1994). Structurally, the song is emitted in a frequency range (range 3–10 kHz) resulting in a broad bandwidth (range 5–7 kHz), which agrees with previous results for the species (Barrantes et al. 2008). In bioacoustics, short vocal phrases emitted with a wide frequency are generally associated with an efficient transmission of acoustic signals that allow an easy location of the singer and the possible masking of other singers producing songs at a lower amplitude frequency (Richards & Wiley 1980, Barrantes et al. 2008). The variation of vocal repertoire among leks analyzed in our study indicates the apparent presence of “vocal dialects” that are variable across time and space, as previously suggested (Gaunt et al. 1994, Barrantes et al. 2008). However, in our study 4 leks were studied in repeated years and recorded songs showed variation between years. This result may suggest the absence of territorial fidelity by individuals, and that males that form leks are learning their songs and incorporating new elements in their vocal repertoire after dispersal. Both hypotheses have been tested in other hummingbird species. For example, González & Ornelas (2009) showed that song neighborhoods within a Wedge-tailed Sabrewing *Campylopterus curvipennis* lek from central Veracruz, Mexico, persist over 4 years. Dispersal patterns of Wedge-tailed Sabrewings are not known, however, based on the observed geographic structure of song among lek members, the authors suggest that this species learn their vocalizations after dispersal, probably to facilitate territory establishment and/or the access to social groups (leks). Also, Araya-Salas et al. (2013) studied vocalizations at four leks of the Long-billed Hermit *Phaethornis longirostris* during 4 years at La Selva, Costa Rica. They found that 18% of males changed their songs over time. Thus, males that changed their songs never again produce the songs previously emitted. However, it is not known how and how long can they learn the songs of a lek because banding hummingbirds was not feasible and color marks were not permanent. Both hypotheses need to be further assessed in the Green Violetear.

It is widely accepted that in most species of hummingbirds that form leks, males emit long songs composed of repeated short-duration notes or syllables (Stiles & Wolf 1979). However, hummingbird songs with repetitive phrases can be versatile in the number of syllables and very complex as shown in the Sparkling Violetear *Colibri coruscans* (Gaunt et al. 1994), the Charming Hummingbird *Amazilia decora* (Kroodsma 1996), the Magnificent Hummingbird *Eugenes fulgens* (Ornelas et al. 2002), and the Anna’s Hummingbird *Calypte anna* (Yang et al. 2007). To our knowledge, our results confirmed for the first time that the static song of Green Violetears is very complex. Despite that, the possible functions of this structured complexity is unknown. As previously suggested (Ornelas et al. 2002), versatile songs can be an easy and efficient way to communicate territory ownership at a low energy cost and without the need of more expensive and potentially harmful chasing and agonistic behaviors. Further studies are needed to evaluate this possibility.

In short, our results showed no significant variation in spectral and temporal song characteristics but the variation in the repertoire was significant. The lack of variation in spectral and temporal song characteristics could be explained by the apparently similar habitats occupied by the leks in which birds should produce signals with similar temporal and spectral characteristics, while the variation in the repertoire could be a result of song learning (i.e. time of song acquisition, dispersion of individuals). Thus, the absence of microgeographic variation with respect to the acoustic characteristics evaluated suggest strong similarities between the LMNP leks, which is independent of the geographic distance between them. Nevertheless, our results also show the great difference between leks in song structure and syllable sharing. This evidence seems to suggest the presence of vocal dialects (song neighborhoods) at a lek level, as previously shown in the Wedge-tailed Sabrewing (Ornelas et al. 2009). The fact that Green Violetear males from the same lek tend to have similar songs and that these differences persist over time, seem to suggest this phenomenon. Nonetheless, it is necessary to confirm whether individuals with similar songs within a lek (song neighborhood) recognize its singing type and discriminate against different songs from different neighborhoods within and outside a lek.

ACKNOWLEDGEMENTS

Field study and experimental procedures were revised and authorized by the Graduate Studies Committee of the Universidad Autónoma de Tlaxcala. We are grateful to I. Morales, L. Gómez, H. Martínez, M. J. Pérez and G. Pérez for field assistance. We thank an anonymous reviewer for helpful comments on a previous version. VMG was supported by a doctoral grant (ESDEPED scholarship) from the Universidad Autónoma de Tlaxcala.

REFERENCES

- Abacus Concepts 1996. Statview Reference. Abacus Concepts, Inc., Berkeley, CA.
- Araya-Salas M., Wright T. 2013. Open-ended song learning in a hummingbird. *Biol. Lett.* 9: 1–5.
- Atwood J. L., Fitz V. L., Bamesberger J. E. 1991. Temporal patterns of singing activity at leks of the White-bellied Emerald. *Wilson Bull.* 103: 373–386.
- Baptista L. F. 1996. Nature and its nurturing in avian vocal development. In: Kroodsma D. E., Miller E. H. (eds). *Ecology and evolution of acoustic communication in birds. Animal Personalities: Behavior, Physiology, and Evolution.* Cornell Univ. Press, Ithaca, New York, pp. 36–90.
- Baptista L. F., Schuchmann K.-L. 1990. Song learning in the Anna's Hummingbird (*Calypte anna*). *Ethology* 84: 15–26.
- Barrantes G., Sánchez C., Hilje B., Jaffé R. 2008. Male song variation of Green Violet-ear (*Colibri thalassinus*) in the Talamanca Mountain Range, Costa Rica. *Wilson J. Ornithol.* 120: 519–524.
- Catchpole C. J., Slater P. J. 1995. *Bird song: biological themes and variations.* Cambridge University Press, Cambridge.
- Charif R. A., Waack A. M., Strickman L. M. 2008. *Raven Pro 1.3 User's Manual.* Cornell Laboratory of Ornithology, Ithaca, NY.
- Colwell R. K. 2000. ESTIMATES: Statistical estimation of species richness and shared species from samples, version 6.0b1. User's Guide and application. Available at <http://www.viceroy.eeb.uconn.edu/estimates>.
- Efron B., Tibshirani R. J. 1998. *An introduction to the Bootstrap.* CRC Press. Boca Raton, FL.
- Ferreira A. R. J., Smulders T. V., Sameshima K., Mello C. V., Jarvis E. D. 2006. Vocalization and associated behaviors of the Sombre Hummingbird (*Aphantochroa cirrhochloris*) and the Rufous-breasted hermit (*Glaucis hirsutus*). *Auk* 123: 1129–1148.
- Ficken M. S., Rusch K. M., Taylor S. J., Powers D. R. 2000. Blue-throated Hummingbird song: A pinnacle of nonoscine vocalizations. *Auk* 117: 120–128.
- Gaunt S. L. L., Baptista L. F., Sánchez J. E., Hernandez D. 1994. Song learning as evidenced from song sharing in two hummingbird species (*Colibri coruscans* and *C. thalassinus*). *Auk* 111: 87–103.
- González C., Ornelas J. F. 2005. Song structure and microgeographic song variation in Wedge-tailed Sabrewings (*Campylopterus curvipennis*) in Veracruz, Mexico. *Auk* 122: 593–607.
- González C., Ornelas J. F. 2009. Song variation and persistence of song neighborhoods in a lekking hummingbird. *Condor* 111: 633–640.
- González C., Ornelas J. F., Gutiérrez-Rodríguez C. 2011. Selection and geographic isolation influence hummingbird speciation: genetic, acoustic and morphological divergence in the wedge-tailed sabrewing (*Campylopterus curvipennis*). *BMC Evol. Biol.* 11: 38.
- Hammer O., Harper D. A. T., Ryan P. D. 2001. PAST: Paleontological statistics software package for education and data analysis. *Paleontologica Electronica* 4: 1–9.
- Howell S. N. G., Webb S. 1995. *A Guide to the birds of Mexico and northern Central America.* Oxford Univ. Press.
- Jackson J. E. 2003. *A user's guide to principal components.* Wiley-Interscience.
- Jarvis E. D., Ribeiro S., Da Silva M. L., Ventura D., Vielliard J., Mello C. V. 2000. Behaviourally driven gene expression reveals song nuclei in hummingbird brain. *Nature* 406: 628–632.
- Johnsgard P. A. 1997. *The hummingbirds of North America.* Smithsonian Institution Press.
- Kroodsma D. E. 1996. Ecology of passerine song development. In: Kroodsma D. E., Miller E. H. (eds). *Ecology and evolution of acoustic communication in birds. Animal Personalities: Behavior, Physiology, and Evolution.* Cornell Univ. Press, Ithaca, New York, pp. 3–19.
- Kroodsma D. E., Vielliard J. M. E., Stiles F. G. 1996. Study of bird sounds in the Neotropics: urgency and opportunity. In: Kroodsma D. E., Miller E. H. (eds). *Ecology and evolution of acoustic communication in birds. Animal Personalities: Behavior, Physiology, and Evolution.* Cornell Univ. Press, Ithaca, New York, pp. 269–281.
- Lara C. 2006. Temporal dynamics of flower use by hummingbirds in a highland temperate forest in México. *Ecoscience* 13: 23–29.
- Mantel N. 1967. The detection of disease clustering and a generalized regression approach. *Cancer Research* 27: 209–220.
- Martínez-García V., Lara C., Ornelas J. F. 2013. Lek characteristics and the static male song of the green violet-ear (*Colibri thalassinus*) during a 3-year study in a temperate forest of central Mexico. *Ornitologia Neotropical* 24: 183–200.
- Ornelas J. F., González C., Uribe J. 2002. Complex vocalizations and aerial displays of the Amethyst-throated Hummingbird (*Lampornis amethystinus*). *Auk* 119: 1141–1149.
- Richards D. G., Wiley R. H. 1980. Reverberations and amplitude fluctuations in the propagation of sound in a forest: implications for animal communication. *Am. Nat.* 115: 381–399.
- Skutch A. F. 1967. Life histories of Central American highland birds. *Nuttall Ornithol. Soc. Publ.* 7.
- Snow D. W. 1968. The singing assemblies of Little Hermits. *Living Bird* 7: 47–55.
- Stiles F. G., Skutch A. F. 1989. *A guide to the birds of Costa Rica.* Comstock Publishing Associates.
- Stiles F. G., Wolf L. L. 1979. Ecology and evolution of lek mating behavior in the Long tailed hermit hummingbird. *Ornithol. Monogr.* 27: 1–78.
- Villers R. L., Rojas G. F., Tenorio L. P. 2006. [Botanic guide to the La Malinche National Park Tlaxcala-Puebla]. Universidad Nacional Autónoma de México. México. D.F.
- Wagner H. O. 1945. Notes on the life history of the Mexican Violet-ear. *Wilson Bull.* 57: 165–187.
- Wiley R. H. 1971. Song groups in a singing assembly of Little Hermits. *Condor* 73: 28–35.
- Williams R. B., Houtman M. A. 2008. Song of Costa's Hummingbird (*Calypte costae*). *Auk* 125: 663–669.
- Yang X., Lei F., Wang G., Jesse A. J. 2007. Syllable sharing and inter-individual syllable variation in Anna's hummingbird, *Calypte anna* songs, in San Francisco, California. *Folia Zool.* 56: 307–318.

STRESZCZENIE

[Zmienność repertuaru i struktury śpiewu pomiędzy tokowiskami uszatka zielonego w centralnym Meksyku]

Głosy wydawane przez kolibry można podzielić na dwie grupy. Podczas żerowania lub obrony terytoriów, samce i samice zwykle wydają relatywnie proste dźwięki dla oznaczenia terytorium, natomiast w czasie toków, samce wykonują dość złożone śpiewy, albo samodzielnie, albo na zbiorowych tokowiskach. Takie tokowiska zostały stwierdzone dla 15% wszystkich opisanych gatunków kolibrów, jednak charakterystyka śpiewów poszczególnych osobników na tokowisku była rzadko badana.

W pracy opisano śpiew osobników uszatka zielonego na 7 oddalonych od siebie o 0,5–9,6 km powierzchniach z tokowiskami rozmieszczonych w lasach sosnowych klimatu umiarkowanego w centralnym Meksyku, w stanie Tlaxcala (Fig. 1). Osobniki były odławiane i znakowane, następnie wyszukiwano tworzące się tokowiska i nagrywano głosy poszczególnych ptaków. Łącznie na tokowiskach stwierdzono 88 znakowanych samców. Średni czas, gdy tokowisko było aktywne, wynosił 30 dni (zakres: 10–50 dni). Na każdym z tokowisk następowała wymiana obecnych na nim samców. Poszczególne śpiewające samce znajdowały się na tokowisku średnio ok. 100 m od siebie. Jako, że badania prowadzono przez 3 lata (2009–2011), różne tokowiska były użytkowane w poszczególnych latach (2009 – 1 tokowisko, 2010 – 3 tokowiska, 2011 – 7 tokowisk), a zastosowane znakowanie osobników nie było trwałe, wyniki z przebadanych 11 tokowisk

traktowane były jako niezależne. W analizach uwzględniono cechy śpiewu takie jak długość trwania sylaby, długość okresu ciszy pomiędzy sylabami, minimalna i maksymalna częstotliwość, a także zidentyfikowano poszczególne sylaby (Fig. 2). Zmienne dotyczące śpiewu analizowano przy pomocy analizy głównych składowych (Tab. 2), zaś podobieństwo piosenek (wykorzystanie tych samych sylab) określano współczynnikiem podobieństwa Jaccarda.

Autorzy zidentyfikowali 41 wyraźnie zróżnicowanych i złożonych sylab dla wszystkich jedenastu badanych tokowisk (Fig. 3), na jedno tokowisko przypadało ok. 12 sylab. Śpiew badanych samców zawierał frazy złożone z 2–4 krótkich sylab (< 2 s), choć skład sylab różnił się między osobnikami (Tab. 1). Większe podobieństwo piosenek stwierdzono dla osobników pochodzących z tych samych niż różnych tokowisk (współczynnik podobieństwa Jaccarda, odpowiednio 0,51 i 0,24). Nie stwierdzono istotnej korelacji pomiędzy podobieństwem piosenek a odległością pomiędzy tokowiskami. Większość obserwowanej zmienności w charakterystyce śpiewu u tego gatunku może wynikać ze zmienności na poziomie tokowiska (Fig. 4).

Różnice w strukturze piosenki oraz podobieństwie sylab sugerują występowanie u tego gatunku dialektów na poziomie tokowisk. Konieczne są dalsze badania w celu potwierdzenia, czy osobniki o podobnych piosenkach, występujące na tych samych tokowiskach, rozpoznają typ piosenki oraz czy są w stanie odróżnić ją od innych piosenek zarówno w obrębie danego tokowiska, jak i poza nim.