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Frequency Modulation During Song in a Suboscine Does Not Require Vocal Muscles

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Amador A, Goller F, Mindlin GB. Frequency modulation during song in a suboscine does not require vocal muscles. *J Neurophysiol* 99: 2383–2389, 2008. First published February 20, 2008; doi:10.1152/jn.01002.2007. The physiology of sound production in suboscines is poorly investigated. Suboscines are thought to develop song innately unlike the closely related oscines. Comparing phonatory mechanisms might therefore provide interesting insight into the evolution of vocal learning. Here we investigate sound production and control of sound frequency in the Great Kiskadee (*Pitangus sulfuratus*) by recording air sac pressure and vocalizations during spontaneously generated song. In all the songs and calls recorded, the modulations of the fundamental frequency are highly correlated to air sac pressure. To test whether this relationship reflects frequency control by changing respiratory activity or indicates synchronized vocal control, we denervated the syringeal muscles by bilateral resection of the tracheosyringeal nerve. After denervation, the strong correlation between fundamental frequency and air sac pressure patterns remained unchanged. A single linear regression relates sound frequency to air sac pressure in the intact and denervated birds. This surprising lack of control by syringeal muscles of frequency in Kiskadees, in strong contrast to songbirds, poses the question of how air sac pressure regulates sound frequency. To explore this question theoretically, we assume a nonlinear restitution force for the oscillating membrane folds in a two mass model of sound production. This nonlinear restitution force is essential to reproduce the frequency modulations of the observed vocalizations.

INTRODUCTION

Songbirds have been widely studied as animal models for vocal learning (e.g., Doupe and Kuhl 1999). Among the aspects studied in detail is the motor control of the vocal organ, the syrinx (reviewed in Suthers and Goller 1997; Suthers and Zollinger 2004; Suthers et al. 1999). In contrast to oscines, song production mechanisms have not been studied in the diverse groups of suboscines. In the *Tyrannidae*, song appears to be innate (Kroodsma 1984; Kroodsma and Konishi 1991), whereas in some other groups, vocal learning may have evolved recently (Kroodsma 2004).

The bipartite vocal organ in Kiskadees contains two sound generators like the songbird syrinx, but only three to four pairs of syringeal muscles (Ames 1971; Miskimen 1963), compared with the six pairs in oscines. In songbirds, song production involves active control of airflow and acoustic parameters (e.g., Goller and Suthers 1996a,b; Suthers et al. 1999), but it is unknown to what degree neural control of vocal muscles is involved in song production in suboscines.

In songbirds, one of the major vocal control tasks is to adjust syringeal tension, which determines the fundamental frequency of sound. The ventral syringeal muscles play an important role in tension control. Electromyographic (EMG) activity in this muscle is exponentially related to fundamental frequency of the generated sounds and predicts FM very closely (Goller and Suthers 1996b). This role was confirmed by theoretical studies, where EMG activity was used to set the parameters for labial tension in a dynamical model of the syrinx (Mindlin et al. 2003), which reproduced the basic frequency characteristics of song.

Frequency control in other bird groups is less well understood but may not involve such direct action by vocal muscles (e.g., Gaunt and Gaunt 1977, 1985; Gaunt et al. 1982; Larsen and Goller 2002; Suthers 2001; Youngren et al. 1974). In the ring dove (*Streptopelia risoria*), FM is closely correlated with fluctuations in pressure in the interclavicular air sac (Beckers et al. 2003), but direct or indirect effects of the tracheolateral muscles cannot be excluded (Beckers et al. 2003; Elemans et al. 2006). It is unknown whether vocal muscles contribute to frequency control in suboscines.

At the level of central motor control of song production, there appear to be strong differences between oscines and suboscines (Farries 2004; Jarvis 2004; Kroodsma and Konishi 1991; Wild 2004). Whereas the motor control circuitry involves several forebrain nuclei in songbirds, in at least two species of suboscines, these nuclei are absent (Kroodsma and Konishi 1991; Nottebohm 1980). The oscine forebrain circuitry of song control consists of a motor pathway and a pathway involved in song learning (e.g., Bottjer et al. 1984; Farries 2004; Scharff and Nottebohm 1991). It is unknown whether this apparent lack of forebrain song motor control circuitry in suboscines is correlated with less sophisticated vocal motor control.

Here we provide the first data on peripheral aspects of song production in a tyrannid, the Great Kiskadee (*Pitangus sulfuratus*). Its song is a sequence of three syllables that are repeated in a stereotyped manner. Subsyringeal air sac pressure and fundamental frequency of sound are positively correlated, and this correlation remains after denervation of the syringeal musculature. With a computational model of the syrinx, we explore how this direct control of sound frequency through respiratory pressure might be effected and show that a restitution constant, nonlinearly related to the membrane fold displacement, can account for the observed frequency control.

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METHODS

Animals and surgery

Experiments were performed on four wild-caught Great Kiskadees. Simultaneous recordings of subsyringeal air sac pressure and sound were performed. For insertion of an air sac cannula, we anesthetized birds with intramuscular injections of ketamine/xylazine. We then inserted a flexible cannula (Silastic laboratory tubing, 1.65 mm OD and 0.76 mm ID) through the abdominal wall just posterior to the last rib, so that it extended a few millimeters into a thoracic air sac. The free end of the Silastic tubing was connected to a miniature piezoresistive pressure transducer (Fujikura model FPM-02PG). To record the pressure signal (low-frequency signal) with a PC sound card (MAYA 1010), we modulated the signal using an electronic device that multiplies it with a sinusoidal signal of 1 kHz. The recorded signal was then demodulated back for analysis of the pressure signal. Sound was recorded on a second channel of the sound card with a TAKSTAR SGC 568 microphone.

As soon as representative sample songs were recorded, we denervated the syringeal muscles by cutting the tracheosyringeal branch of the hypoglossal nerve bilaterally. An incision was made in the skin along the ventrolateral surface of the neck and the trachea was exposed. A 2-mm-long segment of both nerves was removed. The tracheosyringeal nerve is the only known motor supply to the syringeal muscles, and its resection therefore removes all active vocal control. In one additional bird, we ascertained that the tracheosyringeal nerve provides the only motor innervation to the syringeal muscles. We first stimulated the nerve and observed the resulting contractions of the syringeal muscles under the microscope. Step stimulation currents (400 μA, 5 ms), elicited the contraction of all three syringeal muscles. In addition, we investigated the region of the syrinx for nerves running from lateral and dorsal areas to the syrinx. A presumed vagal, afferent nerve branch was reported in some birds (e.g., Nottebohm 2004). We identified one possible candidate visually, but its stimulation did not produce any contractions in the syringeal muscles even at high stimulation currents. It is therefore highly unlikely that another neural pathway for motor innervation of the syringeal muscles exists in the Kiskadee.

Birds typically recovered rapidly from the surgery and started singing on the same day. Song and air sac pressure were recorded for several days after denervation.

Data analysis

For quantification of the air sac pressure-fundamental frequency relationship, we measured both variables throughout the course of each syllable ($\geq$10 points at constant time intervals) using Praat software (Boersma and Weenik 2005). Air sac pressure values were sometimes affected by fluid build-up in the cannula. To avoid biases from different absolute pressure values, we normalized pressure data such that the minimum value during song was zero and the highest value was 1. As the points were clustered along straight lines, regression analyses were performed (SigmaPlot 8.0 software) to obtain the slopes and intercepts of the fitting curves (see Table 1). The significance of the changes of these parameters before and after the nerve cuts were estimated using Student's *t*-test (Kleinbaum and Kupper 1978).

Mathematical model

To generate synthetic song, we implemented mathematically a physical model of the syrinx by adapting a previously described model (Steinecke and Herzel 1995). We used the air sac pressure measurements as input to the model. The output of the model is a synthetic song that we can compare with the recorded one. The similarity between real and synthetic song builds confidence on the proposed model and the hypothesis used to formulate it. A similar strategy was followed in Mindlin et al. (2003), where songbird tonal sounds were synthesized with a one mass model for the labial dynamics.

The Great Kiskadee has a tracheobronchial syrinx (Fig. 1*A*) in which airflow is modulated by two pairs of membrane folds. Each fold is assumed to have internal structure (Steinecke and Herzel 1995; Zaccarelli et al. 2006), and therefore two masses are used to model it; m_1 and m_2 for the lower and upper part (Fig. 1*B*). Each mass is connected to a wall and to the other mass through springs. Their motion is described by means of variables $x_{1,i}$ and $x_{2,i}$ that account for the departures of the lower and upper masses from their equilibrium positions (with the index *i* denoting either right or left side). Therefore the motion equations ruling the dynamics of each membrane fold in one sound source (either right or left), read as

$$m_1\ddot{x}_{1,i} + B\dot{x}_{1,i} + F_{\mathrm{rest},1,i}(x_{1,i}) + K_c(x_{1,i} - x_{2,i}) - G_{1,i} = H_1(t)$$

$$m_2\ddot{x}_{2,i} + B\dot{x}_{2,i} + F_{\mathrm{rest},2,i}(x_{2,i}) + K_c(x_{2,i} - x_{1,i}) - G_{2,i} = 0 \qquad (1)$$

with the index *i* denoting either right or left. *B* stands for a dissipation constant and $G_{1,i}$ and $G_{2,i}$ are restoring forces that emulate collision

$$G_{\alpha,i} = -\theta\left(-l\left(\frac{a_{0\alpha,i}}{1 + x_{\alpha,i}}\right)\right)\frac{a_{0\alpha,i}}{1 + x_{\alpha,i}}\, C_{\alpha,i}$$

with $\alpha = 1, 2$ and $i = \mathrm{r}, \mathrm{l}$ (right or left). The length of the masses along a direction perpendicular to the air flow circulation one is represented by *l*, the area $a_{0\alpha,i} = l\, x_{0\alpha,i}$ with $x_{0\alpha,i}$ the rest position of the respective mass, and $c_{\alpha,i}$ is a parameter that quantifies the forces during collisions. The function $\theta(x)$ is defined as $\theta(x) = 0$ if $x < 0$ and $\theta(x) = 1$ if $x > 0$ and allows introducing a nonzero restoring effect only during collisions. K_c accounts for the elastic coupling between the masses. We define the lumen areas $a_\alpha = a_{\alpha \mathrm{l}} + a_{\alpha \mathrm{r}}$, with $a_{\alpha,i} = l(x_{0\alpha,i} + x_{\alpha,i})$,

TABLE 1. *Analysis of the relationship between air sac pressure and fundamental frequency*

	Pre-Nerve Cut				Post-Nerve Cut							
Bird	*r*	*P*	Slope	Intercept	*r*	*P*	Slope	Intercept	t_{slope}	p_{slope}	$t_{\mathrm{intercept}}$	$p_{\mathrm{intercept}}$
1	0.953	<0.0001	791 ± 46	1,133 ± 31	0.937	<0.0001	872 ± 58	1,275 ± 32	1.11	NS	3.2	SD
2	0.985	<0.0001	737 ± 37	1,060 ± 37	0.987	<0.0001	781 ± 38	927 ± 23	0.83	NS	4.17	SD
3	0.986	<0.0001	711 ± 35	1,414 ± 20	0.97	<0.0001	885 ± 90	1,334 ± 50	1.81	NS	1.77	NS
4	0.991	<0.0001	398 ± 18	1,367 ± 12	0.984	<0.0001	448 ± 22	1,316 ± 14	1.77	NS	2.72	SD
Model[1]	—	—	—		0.981	<0.0001	650 ± 20	1,409 ± 10	—		—	

For all the cases analyzed (4 birds, before and after the nerve cut), the data points were well clustered as shown in Fig. 3. To quantify this, we performed a linear fit to obtain the slope and intercept. To evaluate the goodness of the fit, we use the correlation coefficient (*r*) and the *P* value (probability that $r = 0$). A Student's *t*-test was performed for the slopes and intercepts before and after the nerve cut, leading to conclude that in all the cases the difference between slopes was not significant (NS). The intercepts were significatively different (±SD) in 3 cases but in a nonsystematic manner (the intercept was larger after the cut in *birds 1* and *4* and smaller in *bird 2*), whereas in *bird 3*, the intercepts where not significantly different (NS). [1]based on air sac pressure data from *bird 1*.

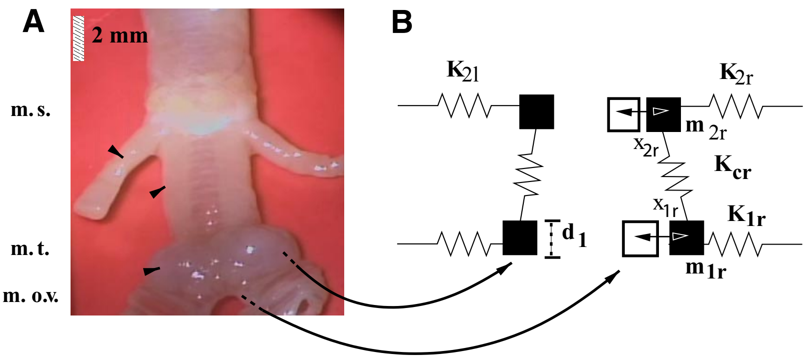

FIG. 1. Syrinx of the Great Kiskadee. *A*: ventral view of the syrinx showing the 3 muscles: m.sternotrachealis (m.s.), m.tracheolateralis (m.t.), and m.obliquus ventralis (m.o.v.). The curved arrows indicate the approximate position of the membrane folds (not visible in the external view), which are the sound source. *B*: schematic diagram of the 2 mass model used to simulate the oscillating folds (see METHODS). *Right*: arrows show the allowed movements of the masses.

and $a_{\min} = \max[0, \min(a_{1l},a_{2l}) + \min(a_{1r},a_{2r})]$ with $\alpha = 1,2$ and $I =$ r,l. The force H_1 is proportional to the pressures acting on the masses

$$H_1 = Pld_1[1 - \theta(a_{\min})(a_{\min}/a_1)^2]\theta(a_1)$$

with P the air sac pressure, d_1 the vertical length of the lower mass, and term in brackets reflects the functional form of the pressure obtained using Bernoulli′s equations when the labia present a convergent profile (i.e., $a_1 > a_2$). Once the dynamics of the masses is obtained, the synthetic sound is proportional to the syringeal airflow $U = \sqrt{2P/\rho}\,a_{\min}\theta(a_{\min})$. The left and right masses are assumed to be equal (both for the upper and lower masses) as well as the coupling constant between upper and lower masses K_c.

We allow the restitution forces $F_{\text{rest},\alpha,i}$ be either linear or nonlinear functions of the folds' midpoint positions $x_{\alpha,i}$.

For air sac pressure, P, we use the recorded values during song to compare the synthetic sound generated by the model with the actual songs.

RESULTS

Syringeal morphology varies considerably among the species within the suboscine groups. The vocal organ of the Great Kiskadee is a tracheobronchial syrinx with two independently controlled sound generators (Fig. 1*A*). Each semi-syrinx contains a set of membrane folds, which are the presumed sound generators and act as valves controlling airflow. Three pairs of muscles control the motion and position of the syringeal cartilaginous framework: the extrinsic muscles m. sternotrachealis and m. tracheolateralis and the intrinsic muscle, m. obliquus ventralis. M. obliquus ventralis is strongly developed and covers the ventral and lateral surface of the syringeal tympanum. The deep fibers of this muscle are oriented in an anteroposterior direction and the superficial ones more dorsoventrally (see also Ames 1971).

The song of the Great Kiskadee consists of three stereotyped syllables (Fig. 2*A, middle*), which are very similar in different individuals and are produced by both sexes. Each syllable is generated by an expiratory pressure pulse, and the intersyllable intervals correspond to short inspirations (as indicated by subatmospheric air sac pressure; Fig. 2*A, top*). The frequency pattern of each syllable contains up- and downmodulation, and the modulation range is from 0.5 to 1 kHz, approximately. The time course of the fundamental frequency of each syllable follows closely that of the subsyringeal air sac pressure pattern (Fig. 2*A*). Quantitative analysis of the relationship for the song syllables shows a highly significant linear relationship between fundamental frequency and air sac pressure (Fig. 3*A*; Table 1).

We tested the role of the syringeal muscles in song production by transecting both tracheosyringeal nerves. The songs

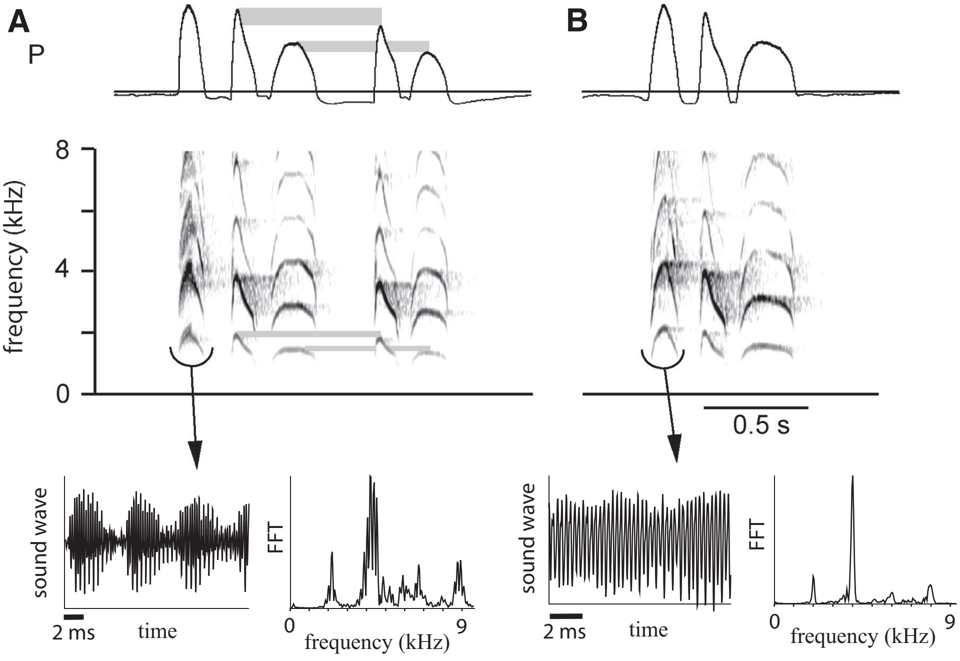

FIG. 2. Simultaneous measurements of sound and air sac pressure during spontaneous singing. The air sac pressure pattern (*P*, horizontal line indicates ambient pressure) of song linearly correlates with the fundamental frequency of sound frequency (shown spectrographically) in the intact bird (*A*) and after a bilateral denervation of the syringeal muscles (*B*). The bird (*1* in Table 1) repeated the 2nd and 3rd syllable (*A*) but produced the 2nd set with slightly lower absolute air sac pressure. This difference in pressure is reflected in slightly lower fundamental frequency of sound (gray areas). The high degree of similarity between syllables before and after the nerve cut suggests that the syringeal muscles do not play a major role in the control of the FM during the song. *Bottom*: corresponds to the sound wave of a segment of the 1st syllable and its corresponding fast Fourier transform (FFT). This acoustic change is the only measurable difference between pre and post nerve cut songs.

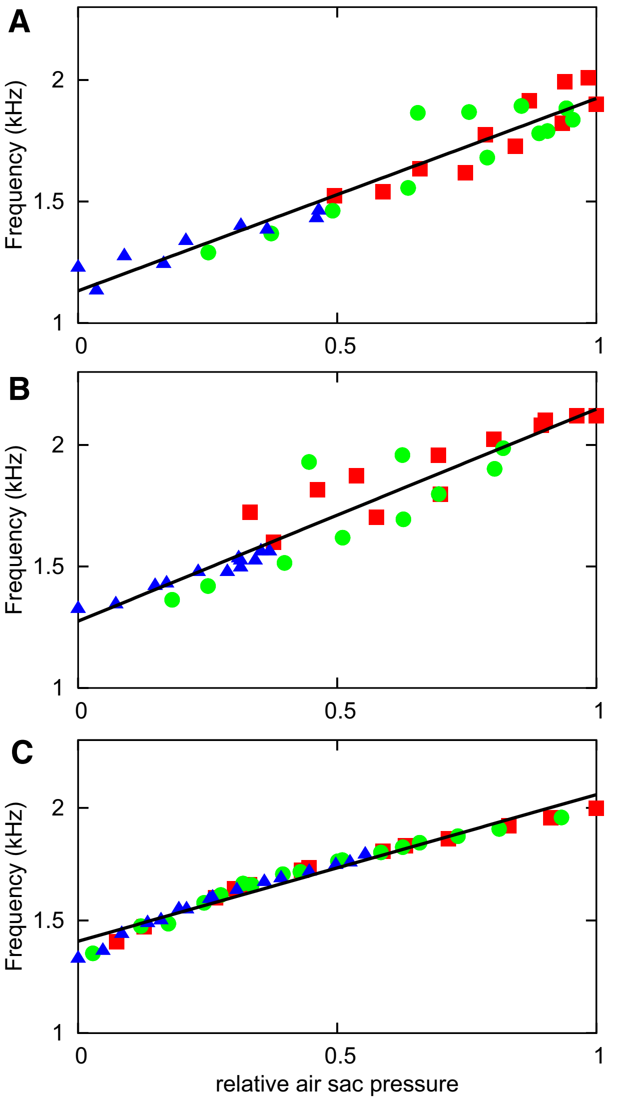

FIG. 3. Linear correlation between relative air sac pressure and sound frequency during song. Each point type refers to a different syllable (circles for the 1st syllable, squares for the 2nd one, and triangles for the 3rd one). Remarkably, the 3 syllables can be fitted by the same linear function, in an intact bird (*A*), the same bird after denervation of the syringeal muscles (*B*), or in the simulations of the model (*C*). In this example, *A* and *B* correspond to *bird 1* in Table 1. The parameters of the linear functions shown in this figure are the following: in *A* slope = 791 ± 46, intercept = 1,133 ± 30, in *B* slope = 872 ± 58, intercept = 1,275 ± 32, and in *C* slope = 650 ± 20, intercept = 1,408 ± 10. The fitting and regression analyses were performed with SigmaPlot 8.0 software.

and air sac pressure patterns are remarkably unaffected by denervation of the syringeal muscles (Fig. 2*B*). The close correlation between air sac pressure and fundamental frequency of syllables remains intact (Figs. 2*B* and 3*B;* Table 1). We found no significant differences in the slopes of the linear regression analyses between pre- and postnerve cut datasets, indicating that the relationship between air sac pressure and sound frequency was not affected by denervation of the syringeal muscles (Table 1). We did find changes of the intercepts after denervation (see Table 1); yet, they were not systematic.

Whereas the basic acoustic structure is similar, we observed smaller acoustic differences between the songs before and after the nerve cuts. The rich harmonic content in the first syllable was lost after the cut (Fig. 2, *bottom*). The observed difference suggests that syringeal muscles play a role in the control of this acoustic feature.

One bird in this study produced more variable call series in addition to its song after denervation. These vocalizations also illustrate the close relationship between frequency and air sac pressure modulation without muscle activity (Fig. 4).

The lack of an obvious role for the syringeal muscles in the control of FM of sound is surprising, taking into account previous studies performed on oscine birds. Previous physical models for sound production in oscines associated the fundamental frequencies of the uttered sounds to the restitution strength of the oscillating labia (Laje et al. 2002; Mindlin et al. 2003), which was assumed to be proportional to the activity of the tension-controlling syringeal muscles.

Because in the Great Kiskadee the syringeal muscles do not contribute to the control the fundamental frequency of the vocalizations, it is necessary to explore new mechanisms for how respiratory pressure may be transduced into frequency. Abandoning the simplifying hypothesis of a linear restitution force for the oscillating folds, it is possible to account for the observed relationship between pressure and sound frequency. The air sac pressure not only allows the onset of folds oscillation, but it also shifts the average midpoint fold position (Gardner et al. 2001). In this way, restitution force, depending nonlinearly on the membrane folds displacements, transduces higher pressure values into higher oscillation frequencies.

We implemented a mathematical model for this physical mechanism (see METHODS). The numerical integration of the equations of motion for the masses allows generating the synthetic sound that would be produced as airflow is modulated by motion of the membrane folds. Figure 5*A* illustrates the synthetically generated song if the restitution forces are assumed to be linear. As expected, there are basically no modulations of the fundamental frequencies during the vocalizations.

Figure 5*B* displays the result of the nonlinear case, where we assumed a restitution force including a cubic dependence on the midpoint position of the membrane fold. Under this assumption, the time course of the fundamental frequency resembles that of the recorded song. In Fig. 3*C*, we show the quantification of the relationship between fundamental frequency and air sac pressure of the synthetic sound. As in the experiment, the points cluster around a straight line. In this way we show that, assuming a nonlinear restitution force for the membrane folds, it is possible to transduce air sac pressure into frequency modulations without active muscle control.

DISCUSSION

The two main tasks of active vocal control are regulating airflow (gating) through the sound generating valves and control of sound frequency (tension) (e.g., Suthers et al. 1999). In songbirds, syringeal muscles actively control the valves and sound frequency, but similar information on what role vocal control plays in sound production in suboscines is missing. Here we show that in the Kiskadee song production is

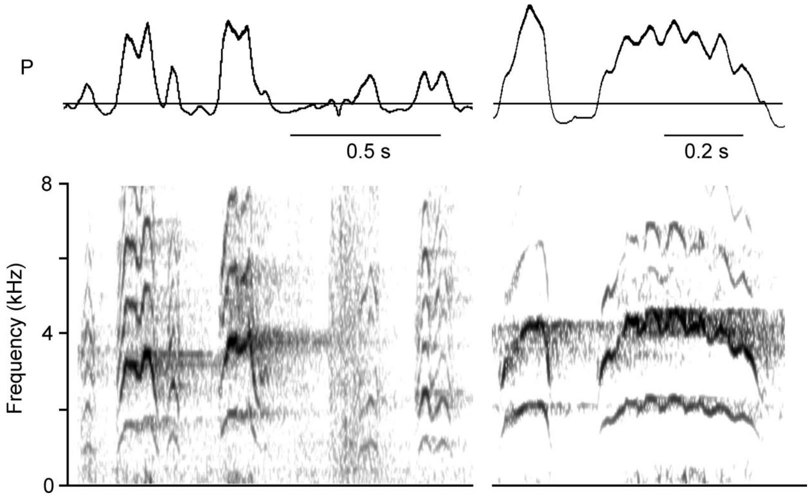

FIG. 4. The close relationship between air sac pressure (*P*) and fundamental frequency was also present in more variable vocalizations. Here we show 2 examples of calls uttered by 1 bird (*3* in Table 1) after denervation.

largely independent of vocal muscles despite the presence of three pairs of well-developed muscles. In a theoretical approach, we show how respiratory activity may be transduced into tension control of the syringeal membrane folds. It is unclear whether the lack of direct vocal control in Kiskadees reflects the difference in song development between *Tyrannidae* and songbirds. More comparative data can address this important issue and shed light on the evolution of vocal learning.

In songbirds, gating and frequency control are effected by syringeal muscles (Goller and Suthers 1996a,b; Suthers et al. 1999). Tracheobronchial muscles are the main gating muscles; the dorsal and ventral tracheobronchial muscles are the adductors and abductors of the lateral labium, respectively. How the positioning of the medial labium is controlled is less well understood, but, most likely, it involves the dorsal syringeal muscle (Larsen and Goller 2002). The activity of the ventral syringeal muscle is most closely correlated with the fundamental frequency of sound. The gating muscles may also contribute to control of tension in a more indirect way, but this role has not been analyzed quantitatively for any species (Goller and Suthers 1996a,b; Vicario 1991).

Song production in the Great Kiskadee does not rely as strongly on vocal muscles as observed in the songbirds. Here we mainly focus on frequency control because the only intrinsic muscle, m. obliquus, of the Kiskadee syrinx is located on the ventral part of the tympanum, similar to the ventral syringeal muscle of the songbird syrinx. However, our experiment also indirectly shows that active syringeal gating of airflow is not required to generate the main features of normal song.

The song of the Kiskadee consists of three syllables, the fundamental frequency of which is within a narrow range of 1–2 kHz and the FM of which is limited to maximally 1 kHz. In many songbirds, the frequency of song syllables can span a much larger frequency range, and FM depths can be much greater than those observed in the Kiskadee. Despite this smaller range, it is surprising that denervation of the syringeal muscles does not affect the main frequency characteristics of song in the Kiskadee.

In songbirds on the other hand, denervation produces striking changes in song production. In some species, nerve cuts result in loss of phonation on the denervated side(s) (e.g., Nottebohm 1971; Nottebohm and Nottebohm 1976; Suthers and Zollinger 2004). Other species after denervation produce song syllables with lower fundamental frequency and rich upper harmonic content (e.g., Daley and Goller 2004; Peek 1972; Seller 1979; Simpson and Vicario 1990; Smith 1976; Suthers 2001). Bilateral cuts can cause respiratory distress or wheezing during deep quiet breathing, suggesting that syringeal muscles are important for abduction of the labial valve (e.g., Nottebohm 1971; Seller 1979; Smith 1976). The different effect of denervation on breathing in different species may indicate different labial biomechanics and therefore different need for active abduction. All these results confirm that active gating plays a role in quiet respiration and that both main roles of syringeal muscles, gating and tension control, are required for normal sound production in songbirds. In contrast, production of song syllables is not equally dependent on syringeal control in the Kiskadee.

It has been hypothesized that intrinsic musculature makes more complex vocal behavior possible by allowing more precise control over individual components of the syrinx than can be achieved through the more indirectly and therefore more globally operating extrinsic muscles (Gaunt 1983). The lack of a clear role of the intrinsic syringeal muscle in vocal control during song generation in the Kiskadee is puzzling. The most consistent change we observed in Kiskadee song after denervation was a reduction in harmonic complexity of the first syllable. This may indicate that the interaction of the two sound sources was disrupted by the denervation, possibly by its effect on syringeal gating of airflow. It is also possible that syringeal muscles are more involved in production of other calls of the vocal repertoire. For example, some of the harsh-sounding calls (Brush and Fitzpatrick 2002) may require activity in the syringeal muscles. Such sound characteristics are accompanied by strong muscle activation in brown thrashers (*Toxostoma rufum*) and starlings (*Sturnus vulgaris*) (Goller and Suthers 1996b; F. Goller and B. G. Cooper, unpublished results). Unfortunately, the birds did not produce these calls in the experimental situation. It is interesting in this context that in parrots activity of intrinsic syringeal muscles does not show a clear correlation with fundamental frequency of sound (Gaunt and Gaunt 1985), and tension control therefore appears to be more indirect through the gating activity of these muscles (Larsen and Goller 2002).

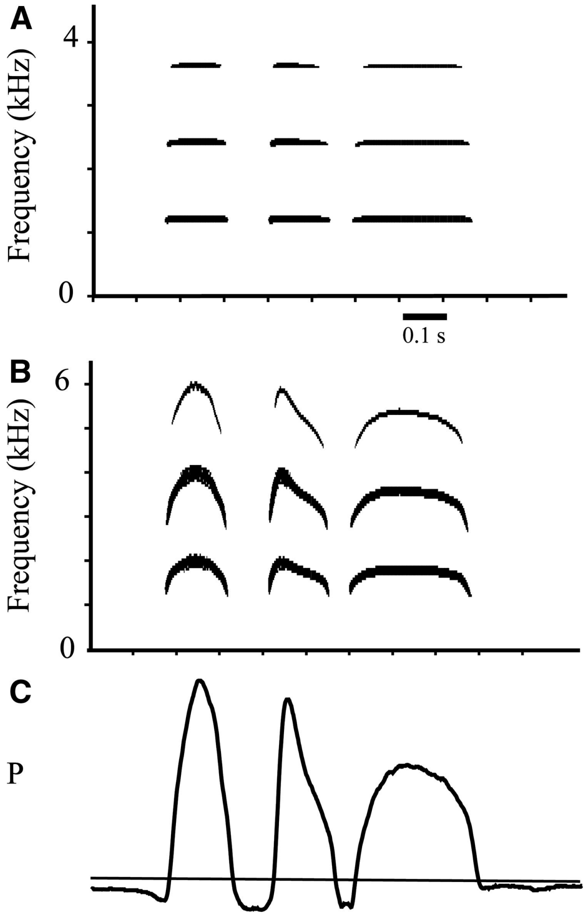

FIG. 5. Different synthetic sounds generated by the 2 mass model under different assumptions on the restitution force acting on a labium. For a linear restitution force, the synthetic sound presents a constant frequency (*A*). Assuming a non linear restitution force, the fundamental frequency follows the air sac pressure. In this case, we use $F_{\text{rest},1,i} = 0.14x_{1,i} + 140x^3_{1,i}$, $F_{\text{rest},2,i} = 0.045x_{2,i} + 140x^3_{2,i}$, with the index *i* denoting either right or left (*B*). The air sac pressure used to drive the model is shown in (*C*). The parameters used in the simulations are $m_1 = 0.018$ *g*, $m_2 = 0.018$ *g*, $l = 0.1$ cm, $d_1 = 0.25$ cm, $B = 1$ *g/s*, $x_{0.1,i} = x_{0.2,i} = 0.25$ cm, $c_1 = 0.24$ dyn/cm, $c_2 = 0.024$ dyn/cm, $K_c = 0.09$ dyn/cm. To integrate the equations, we used a Runge Kutta method of fourth order, with a time step d$t = 1.2\ 10^{-4}$ ms.

The suboscine syrinx shares with that used by oscine birds the existence of pneumatic valves modulating airflow. In the Kiskadee, the control of the sound frequency of song is not conveyed from neural instructions through syringeal muscles but directly transduced from pressure through biomechanical properties of the membrane folds. Previous work on the physical mechanisms involved in song production explored linear approximations for the restitution force as a function of labial displacement (Mindlin and Laje 2005). In the system studied here, this approximation generates sounds with very weak FM, whereas the nonlinear relationship allows the generation of sounds with similar dependence of frequency on absolute air sac pressure levels and similar FM characteristics as observed in the natural song syllables. Elastic restitution forces (F_{rest}) are odd functions of mass displacements (x). A first-order approximation consists of stating $F_{\text{rest}} = k\ x$. The next term in the expansion is then a cubic one, leading to $F_{\text{rest}} = k_1\ x + k_3\ x^3$. This is the rationale behind the approximation used in this work. Because the pressure between the membrane folds induces both, oscillations and an average displacement of the midpoint position of the membrane folds, this nonlinear elastic restitution force is enough to transduce pressure into frequency.

It is unclear whether or not this same or another relationship exists between air sac pressure and fundamental frequency in the denervated syrinx of other songbird species (Suthers 2001). In zebra finches, for which air sac pressure and acoustic data have been published after a bilateral tracheosyringeal nerve transection, fundamental frequency appears to follow air sac pressure, but frequency modulations are very small (Daley and Goller 2004), suggesting a linear relationship between pressure and syringeal restitution (see Fig. 5*A*). If this observation indicates a general difference between suboscine and oscine syringeal biomechanics, exploration of the underlying syringeal parameters will be of interest.

Song development has not been studied extensively in suboscines, but song may be innate in the whole group. In three tyrannid species, there is evidence that song develops innately (Kroodsma 1984; Kroodsma and Konsihi 1991). The lack of geographic variation in the song of the Kisakadee, despite its wide distribution from South to North America, suggests that it may develop innately as well. Eastern phoebes (*S. phoebe*) and Eastern kingbirds (*T. tyrannus*) also lack the forebrain neural circuitry (Kroodsma and Konishi 1991; Nottebohm 1980), which has been associated with song motor control and song development in vocal learners (e.g., Jarvis et al. 2000). This difference poses the question whether the degree to which vocal control plays a role in song production is linked to the presence or absence of telencephalic neural control. Although the comparison between tyrannids and songbirds suggests such a link, more comparative data on different bird orders with and without vocal learning are required to answer this question. Because there does not appear to be direct frequency control in the parrot syrinx, similar to that found in the songbirds, the presence of forebrain vocal motor control does not allow a strong conclusion about direct syringeal control mechanisms. This viewpoint is consistent with the situation in mammals, where sound frequency is typically under direct muscular control regardless of whether vocal learning occurs or not (e.g., Jürgens et al. 2004; Kent 1997; Suthers and Fattu 1973).

Much of the study of the behavior that enhances the survival and reproduction of an animal is focused on its neural control. The generation of a behavior, however, involves strong interactions among the nervous system, the morphology, and the environment. The biomechanics of a peripheral system imposes constraints on the neural control and also provide opportunities for the emergence of specific features in behavior (Chiel and Beer 1997). Here we show that an important feature, control of fundamental frequency, of a complex behavior (song) can be interpreted as the transduction of a respiratory motor pattern into frequency through the biomechanical properties of the syrinx. This relationship constitutes an interesting, previously undescribed mechanism for vocal control.

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