

Activity of three muscles associated with the uncinata processes of the giant Canada goose *Branta canadensis maximus*

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Accepted 10 January 2005

Summary

The activity of three muscles associated with the uncinata processes, (i) external intercostal, (ii) appendicocostalis and (iii) external oblique, were examined using patch and sew-through electrodes during sitting, standing and moderate speed treadmill running in the giant Canada goose. The external intercostal muscles demonstrated no respiratory activity, being active only during running, suggesting they play some role in trunk stabilisation. The appendicocostalis and external oblique muscles are respiratory muscles, being active during inspiration and expiration, respectively. The activity of the

appendicocostalis muscles increased during sitting, suggesting that the uncinata processes in birds play an important role in facilitating lateral flaring of the rib cage when sternal rocking is prevented. We suggest that the uncinata processes in birds facilitate movements of the ribs and sternum during breathing and therefore are integral to the breathing mechanics of birds.

Key words: bird respiration, lung ventilation, hypaxial muscle, intercostal muscles electromyography, locomotion, respiration, air-sac pressure, giant Canada goose, *Branta canadensis maximus*.

Introduction

Uncinata processes are ossified projections that extend posteriorly from the proximal surface of the vertebral ribs (Fig. 1) in all extant birds except the emu *Dromaius novaehollandiae* and the screamers (Anhimidae) (Bellairs and Jenkin, 1960). The function of the uncinata processes and associated muscle (Mm appendicocostales) is unknown. Based on a two-dimensional model reconstruction, Zimmer (1935) concluded that uncinata processes facilitate the movement of the ribs forward, which he interpreted as aiding inspiration. Alternatively, uncinata processes have been hypothesised to be an adaptation for flight (Welty and Baptista, 1988), a site for the attachment of respiratory and shoulder muscles (Hildebrand, 1982) or to strengthen the rib cage mechanically (Kardong, 1988). These hypotheses, however, remain untested.

The hypotheses that the uncينات are a prerequisite for flight, or components of an air-sac system, are refuted by the existence of the screamers (Anhimidae). Screamers are widespread in the wetlands of South America and consist of at least four species, all of which lack uncينات but are capable of powered flight, walking and swimming (Del Hoyo et al., 1992-1999). As for the importance of uncinata processes as sites of attachment for respiratory or locomotor muscles, these functions are not necessarily mutually exclusive, and definitive experiments have yet to be conducted to test hypotheses regarding breathing and locomotion in birds. There is,

however, evidence that the hypaxial muscles contribute to locomotion in birds (Boggs, 1997; Nassar et al., 2001).

There are few electromyographic (EMG) studies on the activity of the respiratory muscles in birds and, perhaps as the appendicocostalis muscle has traditionally been seen as an extension of the external intercostal musculature (George and Berger, 1966; Van den Berge and Zweers, 1993), electromyography has yet to be performed on this muscle (Fedde et al., 1964; Kadono et al., 1963). Furthermore, although dorsoventral movements of the sternum are known to be an integral part of the breathing mechanics in birds (Zimmer, 1935; Brainerd, 1999), EMG studies of respiratory muscles have been performed mainly on anaesthetised birds in a supine or restrained body position, using needle electrodes (Zimmer, 1935; Kadono, 1963; Fedde et al., 1964). There are no EMG studies examining the effect of restricting sternal movements on the activity of the respiratory muscle groups in birds, despite the fact that body positioning is known to alter respiratory movements due to the effect of gravity on the large muscle mass attached to the sternum (Zimmer, 1935). When placed in the supine position, tidal volume is reduced by up to half and the vertical displacement of the sternum is almost doubled with the higher respiratory frequency and larger end-expiratory carbon dioxide concentrations (Fedde, 1987). Furthermore, sternal movements must be restricted when the birds are resting on their sternum. The level of anaesthesia also has a marked effect

on muscle activity by depressing neuronal discharges (Fedde et al., 1964). Against the background of these methodological shortcomings, the external intercostal muscles of the 2nd, 3rd and 4th intercostal spaces are reported to be inspiratory and those in the 5th and 6th intercostal space are expiratory. The serratus dorsalis, scalenus, transverses thoracis, levatores costarum and costi-sternalis muscles are also reported to be inspiratory. All abdominal muscles, the external and internal oblique, rectus abdominus and the transverses abdominus, have an expiratory function (Kadono et al., 1963; Fedde et al., 1964; Zimmer, 1935).

Here, we report investigation of the function of three muscles associated with the uncinata processes: the external intercostal, appendicocostalis and external oblique, in non-anaesthetised and unrestrained giant Canada geese *Branta canadensis maximus*. Muscle activity was examined during normal quiet breathing in standing animals, and when sternal movement was restricted during spontaneous resting on the sternum. Muscle activity was also examined during treadmill running.

Materials and methods

Electromyograms were recorded in four giant Canada geese *Branta canadensis maximus* L. 1758 (body mass 3.56 ± 0.05 kg, mean \pm S.E.M.) during sitting, standing and treadmill running at moderate speed (0.87 m s^{-1}). Birds were obtained from a licensed breeder. Birds were fasted for 12 h prior to all surgery but water was available *ad libitum*. All surgery was performed using aseptic techniques. Heart rate and body temperature were monitored throughout surgery. Feathers were plucked, under anaesthesia, from the surgical sites the day before electrode implantation. The plucked region was treated with Betadine® (V.S.I., Modesto, CA, USA) antiseptic.

Following a 1-day recovery period after surgery, data were collected for 2–3 days. Post-operative analgesia (Flumeglumine, 0.5 mg kg^{-1} ; V.S.I., Modesto, CA, USA) and antibiotics (Baytril, 2.5 mg kg^{-1} ; Bayer, Shawnee, KS, USA) were given once a day for 4 days. Following completion of experiments, reversal surgeries were performed to remove electrodes and air sac catheters. After a recovery period of 7–10 days the geese were adopted as pets. All procedures conform to the guidelines of the University of Utah Institutional Animal Care and Use Committee.

Electrode and air sac catheter implantation

Birds were anaesthetised using Isoflurane (USP 99.9% Isoflurane ml^{-1} ; Bayer, Shawnee, KS, USA) inhalation anaesthetic (2–5%), intubated with paediatric endotracheal tubes and maintained on a ventilator (flow rate 3 l min^{-1} , tidal volume 100 ml) for the duration of all surgeries. We found it was necessary to ventilate the birds during surgery to avoid fatigue of the respiratory muscles (Ludders, 2001).

Electrodes were surgically implanted in the external intercostal, appendicocostalis and external oblique muscles (Fig. 1). Two incisions were made in the skin of the lateral

body above the sites for electrodes placement. The serratus muscle was cut and partially retracted to allow placement of the electrodes during surgery. After completion of the experiments the muscle was resealed. Sew-through electrodes were implanted in three sites in the external intercostal and appendicocostalis musculature (anterior: space between the 3rd and 4th ribs, middle: space between the 5th and 6th ribs and posterior: space between the 6th and 7th ribs) and patch electrodes were attached at three sites in the external oblique muscle directly below the intercostal spaces (Fig. 1). Electrode wires were tunnelled subcutaneously to the midline of the back and fixed to a Velcro® platform and were attached to a Velcro® collar secured around the bird. The collar was fashioned so as not to restrict sternal movements. Electromyographic signals were passed through separate connecting shielded cables (Cooner Wire Inc., Chatsworth, CA, USA), filtered above 1000 and below 100 Hz, amplified approximately 100 times with Grass P511 AC amplifiers and sampled at 4000 Hz on an Apple Macintosh Computer.

Electrodes were made from 0.3 mm diameter multi-stranded, Teflon-coated stainless steel wire (Cooner Wire Inc., part no: AS 631). For patch electrodes, two strands of wire were sewn through 5 mm square patches of Silastic sheeting (Dow Corning, Pittsburg, CA, USA). Exposed wire sections on the patch electrodes, about 1 mm long and 1 mm apart, were parallel to each other, and arranged at 90° to muscle fibre orientation. For sew-through electrodes, an overhand knot was tied in the two strands of wire and 1 mm of insulation was exposed on each, separated by 1–2 mm. Two 5 mm square buttons of Silastic sheeting were used to hold the electrode in place, electrodes were sewn directly into the muscle parallel to muscle fibres.

Inspiration and expiration were monitored using an air-sac catheter. The interclavicular air sac was cannulated using PE 200 Polyethylene tubing (Intramedic Clay Adams Brand, Parsipony, NJ, USA; 1.4 mm i.d., 1.9 mm o.d.) with side holes, held in place by cyanoacrylate glue and sutures. Silastic tubing was tunnelled subcutaneously, exiting and sutured in place on the back of the bird, next to the electrode platform. Miniaturised differential pressure transducers (Endevco 8507C-2, San Juan, CA, USA) were used to record pressure changes in the interclavicular air sac. Locomotion was monitored during treadmill running using a high-speed ($120 \text{ fields s}^{-1}$) video camera (Peak Performance Technologies Inc., Centennial, CO, USA). Locomotor movements were monitored using an accelerometer (Microtron 7290A-10, Mechelen, Belgium) attached to the Velcro® platform on the back of the birds. Video images were synchronised with locomotor events using a LED and synchronisation circuit.

Electromyography analysis

Electromyographs were analysed during sitting, standing and locomotion. To analyse the effect of sitting or standing on EMG activity, the integrated areas of rectified signals were calculated and averaged for 20 breaths. For the locomotion analysis, ensemble averages (Banzett et al., 1992) were

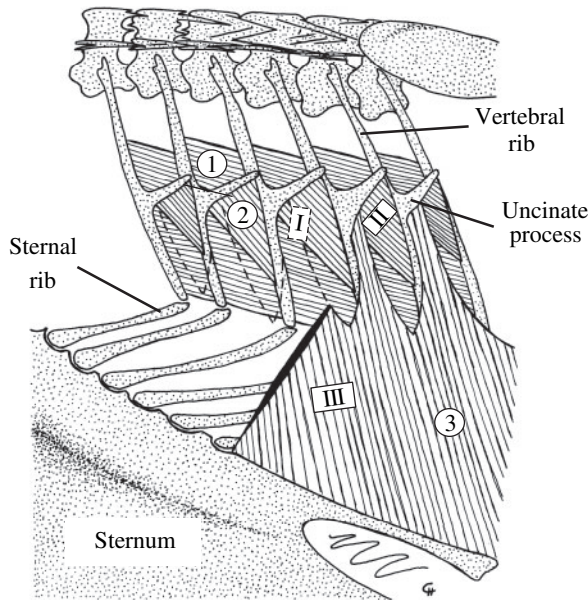


Fig. 1. Location of the external intercostal (1), appendicocostalis (2) and external oblique muscles (3) investigated in this study using both patch (I; external intercostals; lateral aspect of the muscle medial to the appendicocostalis) and sew-through electrodes (II; appendicocostalis), (III; external oblique). The anterior portion of the external oblique has been removed for clarity. One representative site (I, II, III) for electrode placements is shown for each muscle. Anterior is to the left, posterior to the right.

calculated relative to ventilation (20 breaths) and relative to stride (20 strides). A breath was defined as the time from the beginning of expiratory airflow to the end of inspiratory airflow (Fig. 2). A stride was defined as the time from peak contralateral limb support to the next peak contralateral limb support (Fig. 3). Both coupled (ventilation and breathing locked in phase) and uncoupled (ventilation and breathing out of phase) stride averages were analysed during locomotion. The distinction between coupled and uncoupled locomotion was based on visual examination of stride (accelerometer) and ventilation (air sac pressure) recordings. EMG activity was also examined during threat (hissing) responses when standing and running (Figs 2, 3).

To examine the relationship between EMG activity and ventilation and locomotion, ensemble averages (Banzett et al., 1992) were generated by dividing each rectified EMG trace into a series of 120 bins. The average activity in those bins was then analysed relative to stride and to breath. By generating ensemble EMG traces relative to stride and breath we are able to determine if the pattern of muscle activity corresponded with inspiration, expiration or footfall (Deban and Carrier, 2002). For trials in which breathing was not coupled to stride, data were 'whitened' by sampling equally from a range of locomotor and ventilation phase relationships for each ensemble average (Deban and Carrier, 2002). Whitening of the data ensured all phase relationships of drifting between breathing and stride were sampled equally (Deban and Carrier,

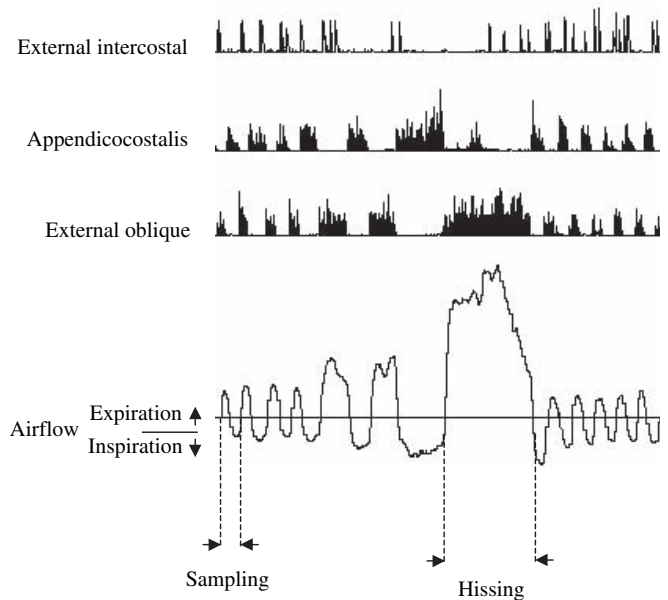


Fig. 2. Means of binned rectified EMG traces of the external intercostal, appendicocostalis and external oblique muscles during hissing when standing. The rectified EMG trace is sampled relative to breath (the onset of expiratory airflow to the end of inspiratory airflow). During hissing the external intercostal demonstrates no respiratory bursting, the appendicocostalis demonstrates inspiratory activity and increased bursting consistent with a larger inspiration and the external oblique demonstrates expiratory activity and increased bursting consistent with an increased expiration.

2002). Drifting of EMG activity relative to the locomotor or ventilatory cycles did not occur in coupled traces.

Interpretation of ensemble average traces

Analysis of muscle activity during periods in which ventilation and locomotion were not coupled provided an indication of a muscle's primary function. If a muscle is ventilatory one would expect EMG activity to correspond to either inspiratory or expiratory airflow and not to the locomotor cycle during uncoupled strides. If the muscle has a locomotor function, however, its activity will be correlated with the locomotor cycle during uncoupled strides. If the muscle has both a ventilatory and locomotor function some activity will be correlated with both cycles. Analysing a combination of breathing when the subjects are either sitting or standing and breathing during uncoupled locomotion, muscles can be assigned to the following groups (as described in Deban and Carrier, 2002): Group 1, locomotor function: phasic EMG activity correlated with uncoupled strides during running, and no activity correlated with breathing when standing or sitting. Group 2, primarily locomotor function: phasic EMG activity is correlated with breathing when the subject is not running, but is correlated with stride during periods of uncoupled running. This indicates that although the muscle contributes to ventilation when the subject is stationary, it has a locomotor function that overrides its ventilatory function. Group 3, dual

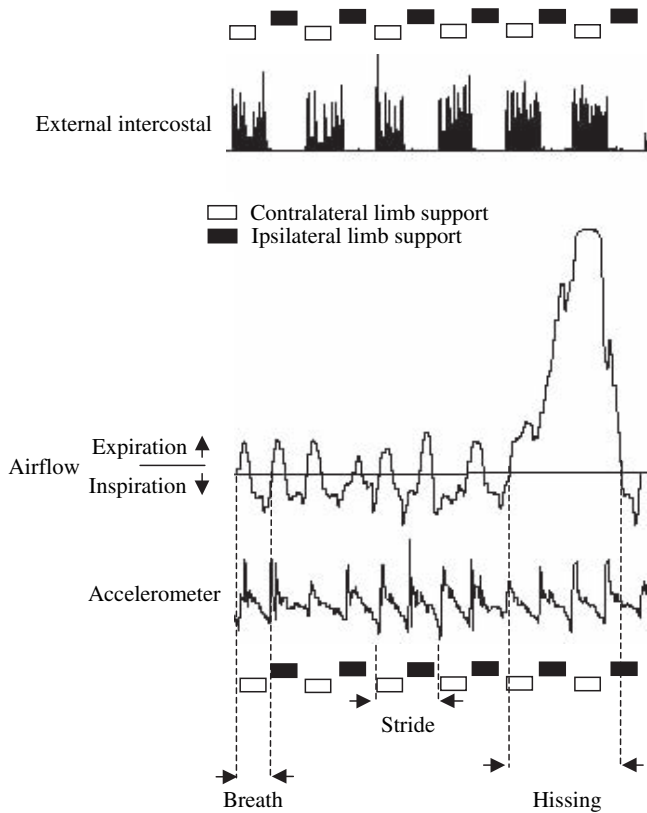


Fig. 3. Means of binned rectified EMG traces of the external intercostal muscle during hissing whilst running. The rectified EMG trace is sampled relative to breath (the onset of expiratory airflow to the end of inspiratory airflow) and relative to stride (peak contralateral limb support to next peak contralateral limb support). The external intercostal demonstrates no respiratory activity; rather bursting activity correlates with contralateral limb support.

ventilatory and locomotor function: phasic EMG activity is correlated with ventilation when the subject is stationary and is correlated, at some level, with both stride and ventilation during uncoupled running. This indicates that the muscle contributes to both ventilation and locomotion. Group 4, ventilatory function: phasic EMG activity correlated with breathing in stationary animals and correlated with breathing during periods of uncoupled running. This indicates the muscle is only active when it can contribute to breathing and has no locomotor effect.

To quantify the extent to which a particular muscle contributes to ventilation and/or locomotion during running we regressed the uncoupled ensemble averages against coupled ensemble averages (Deban and Carrier, 2002; Farley and Koshland, 2000). Calculating the coefficient of determination (r^2) illustrates how much the uncoupled averages (locomotion or ventilation) differ from the coupled averages. The value of r^2 when the uncoupled locomotor or ventilation averages are regressed against the coupled averages indicates the extent to which the muscle has a locomotor or ventilatory function, respectively.

Table 1. Average integrated sum of the inspiratory and expiratory activity of the external intercostal, appendicocostalis and external oblique muscles during sitting and standing

Goose	Muscle	Sitting	Standing	P-value
A	External intercostal	0.3±0.08	0.2±0.02	NS
	Appendicocostalis	11.6±1.2	6.9±0.8	<0.0001
	External oblique	4.2±0.3	5.2±0.4	0.02
B	External intercostal	4.9±0.9	4.9±0.6	NS
	Appendicocostalis	6.2±0.5	3.7±0.3	0.005
	External oblique	4.1±0.6	23.1±1.6	<0.0001
C	External intercostal	2.6±0.4	3.2±0.4	NS
	Appendicocostalis	6.4±0.5	2.9±0.3	<0.0001
	External oblique	0.1±0.03	1.2±0.1	<0.0001
D	External intercostal	0.02±0.001	0.03±0.001	NS
	Appendicocostalis	5.83±0.4	3.1±0.4	0.026
	External oblique	0.5±0.1	9.1±0.5	<0.0001

Statistical comparisons were made using two-sample *t*-tests of \log_{10} transformed data. NS, not significant.

Statistical analysis

Due to differences in electrode placement in different muscles and the location, depth and orientation of the electrode in a given muscle, statistical comparisons can only be made for electrodes for one day and not between electrodes or even on the same electrode on different days (Loeb and Gans, 1986). To compare the integrated area for sitting and standing (Table 1), and coupled and uncoupled locomotion (Table 2) two-sample *t*-tests were performed on \log_{10} transformed data, normalised to a percentage maximum of total EMG activity. All data are presented as means ± S.E.M.

Results

Each of the muscles examined demonstrated different patterns of muscle activity during sitting, standing and running on a treadmill. There were, however, no differences in the activity pattern of the muscles associated with the anterior, middle or posterior intercostal spaces.

External intercostal

The phasic activity of the external intercostal muscle was consistent with a locomotor but not a ventilatory function (Tables 1, 2; Figs 4, 5). In the ensemble averages from all geese the external intercostal muscle demonstrated no phasic activity that was correlated with ventilation during sitting (Tables 1, 2; Fig. 4A), standing (Tables 1, 2; Fig. 4B), or during the large inspirations and expirations associated with threat (hissing) displays when either running or standing (Figs 2, 3). In all geese during coupled and uncoupled (relative to stride) locomotion there was phasic activity that correlated with contralateral limb support (Table 2, Fig. 5A,C). No phasic

Table 2. Average integrated sum of the activity of the external intercostal muscle during inspiration, expiration and contralateral limb support

External intercostal muscle	Inspiration	Expiration	Contralateral limb support	P-value	
				Inspiration–contralateral	Expiration–contralateral
Goose A	0.6±0.3	0.4±0.2	2.73±0.4	<0.0001	<0.0001
Goose B	0.2±0.04	0.11±0.23	3.52±0.33	<0.0001	<0.0001
Goose C	0.43±0.06	0.15±0.07	4.18±0.03	<0.003	<0.0001
Goose D	0.04±0.001	0.05±0.02	3.03±0.3	<0.001	<0.002

Statistical comparisons were made using two-sample *t*-tests of log₁₀ transformed data.

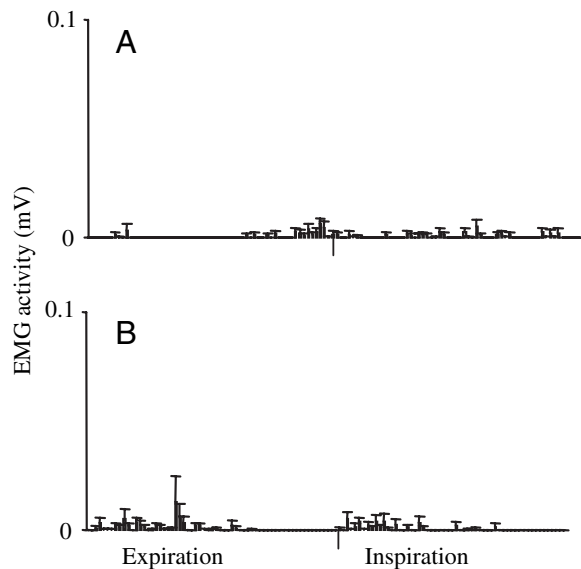


Fig. 4. Means of binned rectified EMG traces of the external intercostal muscle during (A) sitting and (B) standing. Data are presented from Goose A, averaged over 20 breaths and shown here for one breath cycle: the onset of expiratory airflow to the end of inspiratory airflow. The external intercostal demonstrates no respiratory bursting activity.

activity was seen during uncoupled locomotion when analysed relative to breath (Fig. 5B).

Appendicocostalis

The phasic activity of the appendicocostalis was consistent with an inspiratory function (Table 1, Figs 6, 7). Significantly greater phasic activity was seen when the geese were resting on the sternum (Table 1, Fig. 6A) as opposed to standing (Table 1, Fig. 6B). Before a hissing event (when standing) there was a corresponding increase in the bursting of the appendicocostalis muscle consistent with a larger inspiration (Fig. 2). The appendicocostalis muscle also exhibited some locomotor activity (Fig. 7). During respiration when locomotion was coupled there was phasic activity that correlated with the onset of inspiration (Fig. 7A). The biphasic activity during uncoupled locomotion (relative to breath) was consistently larger when associated with contralateral limb support (Fig. 7B). The EMG activity was cleaner during

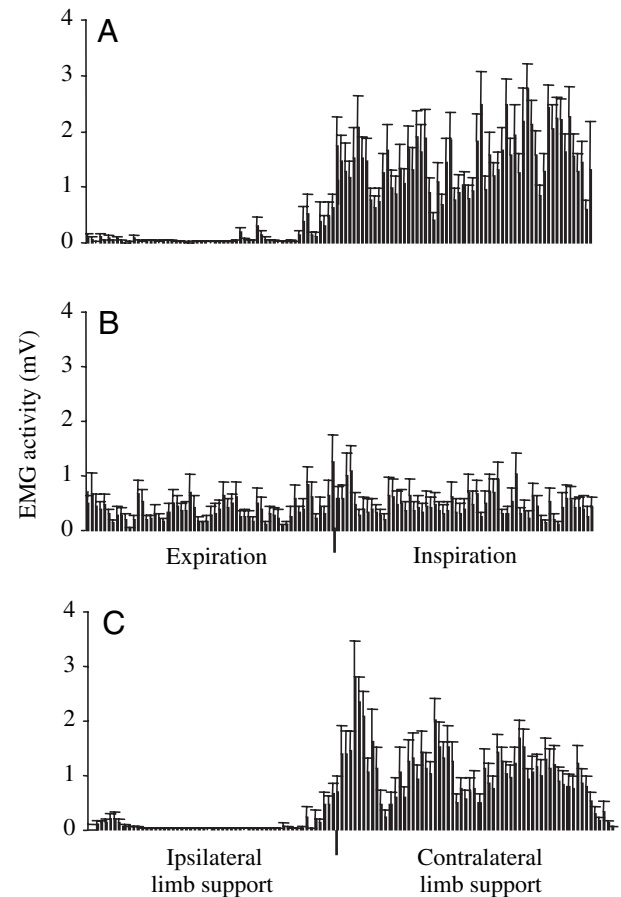


Fig. 5. Means of binned rectified EMG traces of the external intercostal muscle during (A) coupled walking: breathing and footfall in phase, (B) uncoupled running relative to breath: the onset of expiratory airflow to the end of inspiratory airflow and (C) uncoupled running relative to stride: one stride length. Data are presented from Goose A averaged over 20 breaths or 20 strides. The external intercostal is a locomotor muscle and its activity correlates with contralateral limb support.

uncoupled locomotion analysed relative to breath than relative to stride (Fig. 7C).

External oblique

The external oblique muscle demonstrated activity that was consistent with a role in expiration (Table 1, Figs 8, 9). There

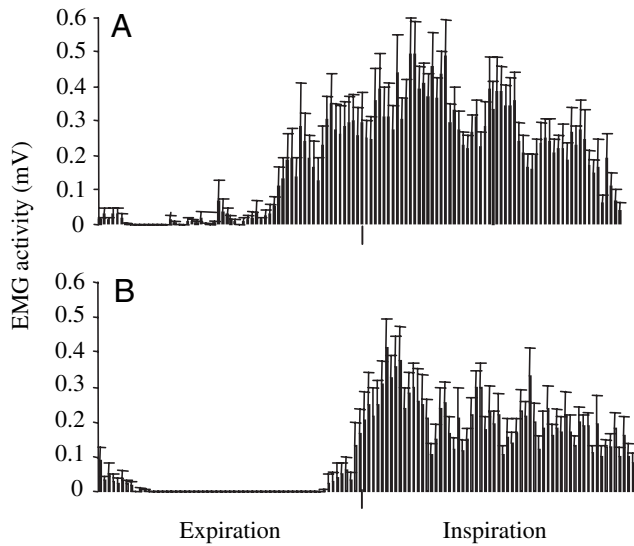


Fig. 6. Means of binned rectified EMG traces of the appendicocostalis muscle during (A) sitting and (B) standing. Data are presented from Goose A, averaged over 20 breaths and shown here from one breath cycle: the onset of expiratory airflow to the end of inspiratory airflow. The appendicocostalis is an inspiratory muscle and demonstrates increased activity during sitting.

was no bursting activity during sitting (Table 1, Fig. 8A). There was, however, phasic activity during expiration while standing (Table 1, Fig. 8B). During large expirations associated with hissing there was a corresponding increase in EMG activity consistent with a larger expiration (Figs 2, 3). When analysed for coupled and uncoupled breathing (relative to breath), there was phasic activity that was consistent with an expiratory function for this muscle (Fig. 9A,B). When uncoupled locomotion was analysed relative to stride no clear bursting activity was associated with either ipsilateral or contralateral limb support (Fig. 9C).

Discussion

The muscles investigated in this study can be grouped into the following functional categories (as outlined in Materials and methods). The external intercostal is a locomotor muscle (Group 1); the appendicocostalis muscle is primarily involved in inspiration but also supports locomotion (Group 3); the external oblique muscle is an expiratory muscle (Group 4).

The locomotor function of the external intercostal

The portion of the external intercostal muscle adjacent to the uncinat processes demonstrated no phasic activity associated with ventilation. This was true during quiet breathing when either sitting or standing, during the large expirations and inspirations associated with hissing, as well as during running. The low correlation between the uncoupled breath averages when analysed relative to breath or stride for all geese (Table 3) indicates that this portion of the external intercostal muscle has no significant ventilatory function during rest or

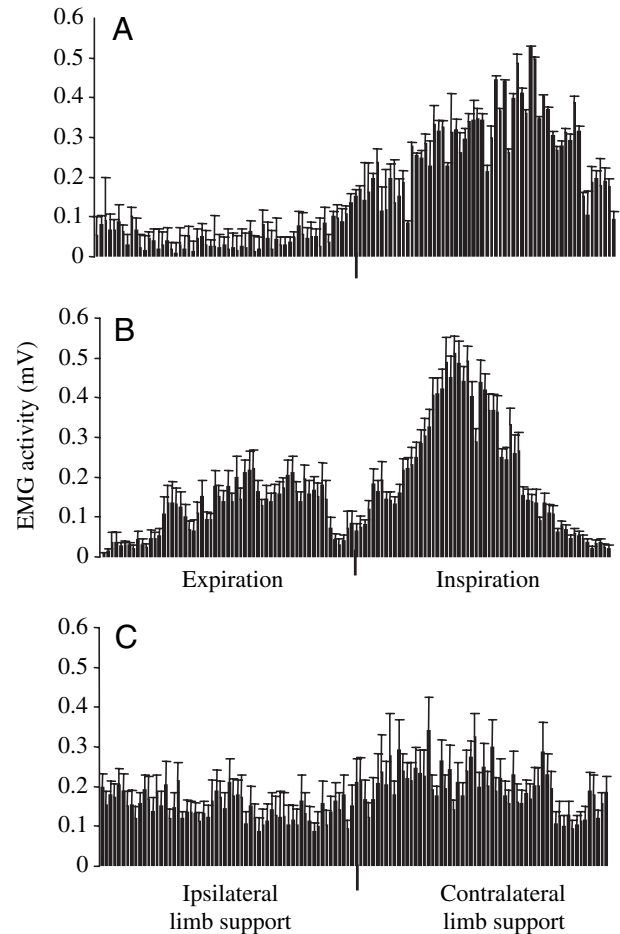


Fig. 7. Means of binned rectified EMG traces of the appendicocostalis muscle during (A) coupled walking: breathing and footfall in phase, (B) uncoupled running relative to breath: the onset of expiratory airflow to the end of inspiratory airflow and (C) uncoupled running relative to stride: one stride length. Data are presented from Goose A, averaged over 20 breaths or 20 strides. The appendicocostalis is an inspiratory muscle but does demonstrate some locomotor function which correlates with contralateral limb support.

running in the giant Canada goose. Activity of the external intercostal muscles was correlated, however, with contralateral limb support during running.

The idea that the external intercostal muscles are involved in locomotion is not new (De Troyer et al., 1985; Carrier, 1990, 1991, 1993, 1996). However, the locomotor role of the external intercostal muscles demonstrated in this study is not consistent with previous studies that have reported both inspiratory and expiratory activity (Kadono et al., 1963; Fedde et al., 1964). Methodological differences may account for these conflicting results. First, previous studies may have recorded from a different anatomical position in the external intercostal muscles. We monitored the lateral aspect of the muscle located medial to the appendicocostalis muscles (Fig. 1). Other portions of the external intercostal muscles may be involved in ventilation. Second, EMG in the current study was performed on non-anaesthetised and unrestrained birds using patch

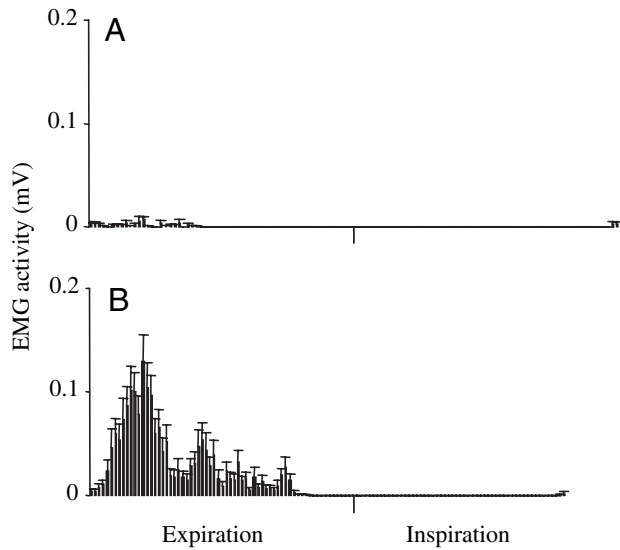


Fig. 8. Means of binned rectified EMG traces of the external oblique muscle during (A) sitting and (B) standing. Data are presented from Goose A and are averaged over 20 breaths and shown here from one breath cycle – the onset of expiratory airflow to the end of inspiratory airflow. The external oblique is an expiratory muscle and demonstrates no bursting activity during sitting.

electrodes. Kadono et al. (1963) performed EMG experiments on chickens that were fixed on their side to wooden boards, whereas Fedde (1964) conducted EMG experiments on anaesthetised birds in a supine position, restrained by the wings. Both experiments were performed with needle electrodes. Fedde et al. (1964) reported no change in EMG activity when birds were moved from the supine to upright body position. However, the exact placement of needle electrodes cannot be confirmed in living animals and failure rates of up to 50% can be expected due to needle movement (Loeb and Gans, 1986). Inherent difficulties in maintaining the exact placement of needle electrodes whilst moving the bird from the natural to supine position, coupled with the anaesthetising and restraining of the birds, may have masked any electrical changes in the muscles and could account for the lack of an observed difference in muscle activity (Fedde, 1987).

The locomotor role demonstrated in this study does not preclude the external intercostals from contributing to

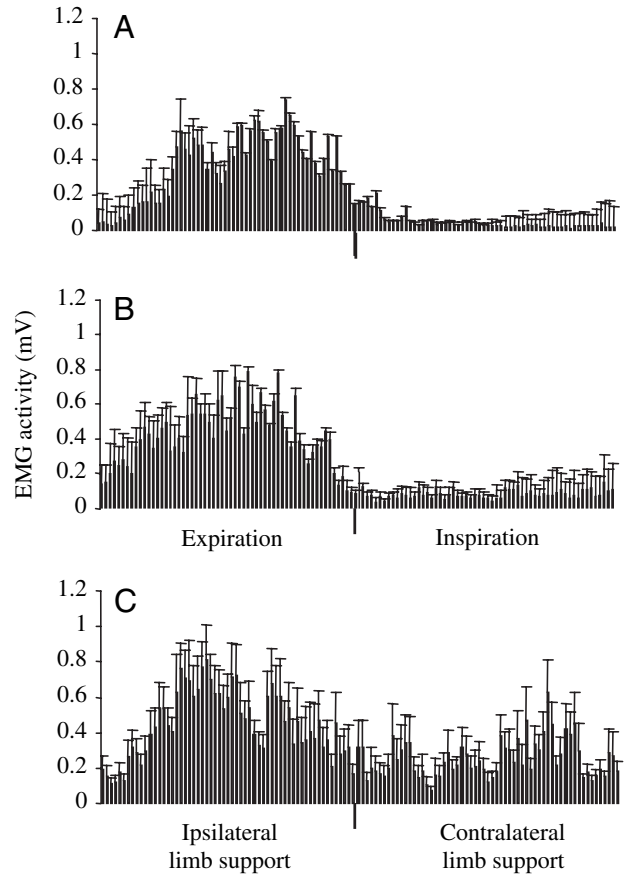


Fig. 9. Means of binned rectified EMG traces of the external oblique muscle during (A) coupled walking: breathing and footfall in phase, (B) uncoupled running relative to breath: the onset of expiratory airflow to the end of inspiratory airflow and (C) uncoupled running relative to stride: one stride length. Data are presented from Goose A, averaged over 20 breaths or 20 strides. The external oblique is an expiratory muscle and demonstrates no locomotor activity.

ventilation. In dogs the intercostal muscles have been found to be primarily locomotor muscles; however, they do participate in ventilation when dogs are running and can contribute to ventilation during running when the work of breathing is increased (Deban and Carrier, 2002). It is possible that during laboured breathing, in restrained (Kadono et al., 1963) or

Table 3. Coefficient of determination for coupled and uncoupled stride averages for the external intercostal, appendicocostalis and external oblique muscles analysed relative to stride and to breath

Muscle	Coefficient of determination (r^2)							
	Goose A		Goose B		Goose C		Goose D	
	Breath	Stride	Breath	Stride	Breath	Stride	Breath	Stride
External intercostal	0.05	0.73	0.04	0.59	0.006	0.47	0.06	0.60
Appendicocostalis	0.23	0.20	0.36	0.25	0.25	0.25	0.41	0.36
External oblique	0.85	0.004	0.71	0.01	0.81	0.03	0.70	0.003

Stride = one stride length; breath = the onset of expiratory airflow to the end of inspiratory airflow.

supine birds (Fedde et al., 1964), the external intercostals are recruited into assisting breathing.

Dual function of the appendicocostalis

The action of the appendicocostal muscles is independent of the external intercostal musculature, suggesting distinct motor control in these two muscle groups. The appendicocostalis muscles have a dual locomotor and ventilatory function, as they are active during the inspiratory phase of ventilation in standing and sitting *B. canadensis maximus*, and demonstrate biphasic activity during locomotion. The inspiratory activity of the appendicocostalis muscles demonstrated in this study confirms the hypothesis of Zimmer (1935).

During breathing while standing, contraction of the appendicocostalis muscles appears to move the vertebral ribs cranially in a fixed plane and, in conjunction with the other inspiratory muscles, rotates the sternum ventrally. Phasic activity of the appendicocostalis increased when movements of the sternum were restricted by sitting. When sternal movements were restricted, expansion of the thoracic cavity was achieved by lateral flaring of the rib cage. The corresponding increase in EMG activity of the appendicocostalis muscle during sitting suggests this muscle may play a key role in facilitating this lateral flaring. The activity of the appendicocostalis muscles during locomotion is consistent with a dual respiratory and locomotor function, as indicated by the equally low correlation between breath and stride averages (Table 3). The activity of the appendicocostalis muscle was consistently larger during contralateral limb support suggesting the muscle may play some role in stabilising the forces exerted on the trunk during running.

Function of the external oblique

Activity of the thoracic portion of the external oblique muscle indicates that it functions to produce expiration. The abdominal muscles of birds are known to contribute to expiration by moving the sternum dorsally (Kadono et al., 1963; Fedde et al., 1964). The importance of the external oblique in sternal movement is confirmed by the lack of phasic activity when sternal movements are restricted during sitting. The aponeuroses of the finger-like projections of the external oblique muscle insert onto the base of the uncinat processes in *B. canadensis maximus*. It appears that the uncinat processes may be acting as sites for the insertion of the projections of the external oblique muscle to move the sternum dorsally during expiration. Phasic activity of the external oblique muscle was cleaner during coupled and uncoupled breathing (analysed relative to breath) than uncoupled breathing analysed relative to stride. The low correlation between uncoupled stride averages (Table 3) indicates that the external oblique muscle is ventilatory and plays no role in stabilising the trunk during locomotion.

Conclusions

The results of this study suggest that the uncinat processes in birds are involved in movements of the ribs and sternum

during breathing. Contraction of the uncinat muscle, the appendicocostalis, during inspiration appears to assist in rotating the ribs cranially, which facilitates ventral rotation of the sternum. The uncinat processes may also act as a brace for the insertion of the finger-like projections of the external oblique muscle to move the sternum dorsally during expiration. While any putative stiffening function of the uncinat processes cannot be completely ruled out, the results obtained here confirm that the uncinat processes in birds are an integral component of breathing mechanics, involved in both inspiration and expiration. The activity of the appendicocostalis muscle increases when sternal movements are restricted, which suggests activity of these muscles may be particularly important during prolonged sitting such as during egg incubation. During flight the forces exerted onto the trunk may differ from those exerted during running, so the present experiments cannot be applied to locomotion in general. The serratus attaches onto the uncinat processes from the scapula and thus represents another vector for forces applied to the uncinat processes during flight. The absence of uncinat processes in the emu and screamers suggests that they may breathe more like other amniotes by swinging the ribs laterally. In light of the methodological differences between the present and previous studies, further EMG experiments of other putative respiratory or locomotor muscles in non-anaesthetised and unrestrained birds using patch or sew-through electrodes would be beneficial.

This work was supported by The National Science Foundation; IBN 9807534 and the Deutsche Forschungsgemeinschaft through the Graduiertenkolleg 'Evolution und Biodiversität in Raum und Zeit' Project No: 721/1. The authors would like to thank Timna Fischbein, Suzy Munns, Jessamyn Markley and Steve Deban for assistance with surgery. Carsten Heuer provided the artwork for Fig. 1. All experiments were performed under approval of the University of Utah Institutional Animal Care and Use Committee.

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