Anatomy and Histochemistry of Hindlimb Flight Posture in Birds. I. The Extended Hindlimb Posture of Shorebirds

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ABSTRACT

Birds utilize one of two hindlimb postures during flight: an extended posture (with the hip and knee joints flexed, while the ankle joint is extended caudally) or a flexed posture (with the hip, knee, and ankle joints flexed beneath the body). American Avocets (Recurvirostra americana) and Black-necked Stilts (Himantopus mexicanus) extend their legs caudally during flight and support them for extended periods. Slow tonic and slow twitch muscle fibers are typically found in muscles functioning in postural support due to the fatigue resistance of these fibers. We hypothesized that a set of small muscles composed of high percentages of slow fibers and thus dedicated to postural support would function in securing the legs in the extended posture during flight. This study examined the anatomy and histochemical profile of eleven hindlimb muscles to gain insight into their functional roles during flight. Contrary to our hypothesis, all muscles possessed both fast twitch and slow twitch or slow tonic fibers. We believe this finding is due to the versatility of dynamic and postural functions the leg muscles must facilitate, including standing, walking, running, swimming, and hindlimb support during flight. Whether birds use an extended or flexed hindlimb flight posture may be related to the aerodynamic effect of leg position or may reflect evolutionary history. J. Morphol. 269:967–979, 2008.

KEY WORDS: flight; posture; muscle fiber types

Birds use one of two hindlimb postures during flight. The legs are either held under the body with the hip, knee, and ankle (intertarsal) joints flexed, or are trailed beyond the body, with the hip and knee flexed, but with the ankle in extension. In general, shorebirds, raptors, ducks, geese, and parrots use the extended posture, while perchers, birds and woodpeckers use the flexed position (Barrett-Hamilton, 1903; Townsend, 1909; Shepard and Meyers, 2006). There has been little research regarding the reason one or the other posture is used, but we are in the process of evaluating several possible explanations (Shepard and Meyers, 2006; Shepard et al., 2008).

Our initial effort into this largely unstudied subject was to examine the hindlimb muscles of "extended" hindlimb flyers, such as American Avocets (Recurvirostra americana) and Black-necked Stilts (Himantopus mexicanus), for the presence of slow fibers associated with the extended hindlimb flight posture. These species were chosen because of their extremely long legs, lack of previous study, and local availability.

Slow twitch and slow tonic fibers have been well studied with regard to posture in birds, and a correlation between muscle function and fiber types has been documented (Rosser and George, 1986a; Rosser et al., 1994; see also Meyers and Stakebake, 2005 for review). Slow muscle fibers are considered efficient in the isometric contractions utilized during posture (Goldspink, 1980, 1981) and also during isometric contractions to maintain muscle length allowing tendons to store elastic energy (see Patak and Baldwin, 1993). Avian slow-twitch fibers react histochemically like those of mammals, but unlike these fibers in mammals, avian fibers are multiply innervated like slow tonic fibers (Baier and Gatesy, 2000).

Based on earlier work on the avian forelimb (Meyers, 1992), we expected to find a group of small, uniformly slow muscles functionally dedicated to posture. Our goals in this study were to 1) document the muscles believed to be involved in holding the hindlimb in the extended flight posture; 2) assay these muscles for the presence of slow, postural muscle fibers, and 3) discuss the evolution of hindlimb postures during flight. We believe we are the first (McFarland and Meyers, 2005) to take an interest in the muscles involved in the flight posture of the avian hindlimb, since previous studies have focused on walking (e.g., Jacobson and Hollyday, 1982; Johnston and Bekoff, 1996; Gatesy, 1999), running (Patak and Baldwin, 2000).
Materials and Methods

Six American Avocets (Recurvirostra americana) and six Black-necked Stilts (Himantopus mexicanus) were collected locally over the summer months of 2005 and 2006 for another project under possession of valid state and federal permits. We were given carcasses for tissue collection or dissection within 1 h of death. Muscles of the right leg were collected from each bird. The left leg and left half of the pelvis were removed intact and dissected fresh or fixed in formalin and stored in phenoxethanol for anatomical study. Skeletal material was obtained from the National Museum of Natural History (Smithsonian Institution).

After an evaluation of the available literature and initial manipulation of fresh specimens, we selected for study muscles believed to function in the extended flight posture. Most muscles were examined from the same three individuals (of each species); in some cases additional individuals were used when tissue quality was unknown. Muscles studied included Mm. iliotibialis cranialis, iliotrochantericus caudalis, iliotrochantericus cranialis, and iliotrochantericus medius, which act as hip flexors, Mm. iliofibularis, flexor cruris lateralis, and flexor cruris medialis, which function as knee flexors, and Mm. gastrocnemius and plantaris, which are ankle extensors. Mid-belly sections from each muscle, cut perpendicularly to fascicle orientation, were removed and mounted on a cork block, using 5% gum tragacanth, then flash frozen in isopentane cooled in liquid nitrogen to approximately −150 °C. Samples were stored at −70 °C until sections (10–12 μm thick) were cut on a cryostat at −20 °C and transferred to glass microscope slides.

To allow fiber type differentiation, serial sections were reacted for the presence of myofibrillar ATPase in either alkaline (pH 10.35–10.45) or acidic (pH 4.2) preincubations, following the procedure described in Meyers and Mathias (1997) and Kovacs and Meyers (2000). Reactions for glycolytic (α-glycerophosphate dehydrogenase, α-GPD) and oxidative enzymes (nicotinamide adenine dinucleotide diaphorase, NADH-D) followed the procedure described in Meyers and Mathias (1997) and Kovacs and Meyers (2000). Reactions for myofibrillar ATPase in either alkaline or acidic preincubations, following the procedure described in Meyers and Mathias (1997) and Kovacs and Meyers (2000) were carried out with the antibodies in a humidified chamber at room temperature for 2 h, or at 4 °C for 18 h. After rinsing in phosphate-buffered saline, samples were incubated in a goat anti-mouse antibody to recognize the primary antibody, and stained with a streptavidin peroxidase system (Zymed Labs, S. San Francisco, CA).

Slides were viewed under a Zeiss Axioskop 40/40 PL micro- scope and overlapping digital images of each muscle with a corresponding scale bar were obtained using either a Nikon Coolpix 995 or Olympus E-530 digital camera. Images were then uploaded in Adobe Photoshop and merged to provide a full cross-sectional photomontage of each muscle. Slow and fast muscle fibers were subsequently quantified using the ALD58 reaction, as this provided the best contrast for visualizing fiber borders (see Fig. 1). Fiber diameter measurements of one muscle from each limb segment were obtained by averaging the means of the narrowest and widest portion of each of 100 randomly selected slow and fast fibers per muscle. Diameters were compared using an independent sample paired t-test. Numerical data were stored and analyzed using Microsoft Excel.

Measurements of cross-sectional area were obtained by printing the cross section image of each muscle onto a single page of paper. This paper was then trimmed to conform to the photographed muscle boundaries. The mass of the photograph was then compared with the mass of paper representative of 1 mm², obtained by using the pre-photographed scale bar. Simpson’s Rule (Thomas and Finney, 1996) was used to verify our calculations.

Due to a difference in fiber sizes, a “weighted” cross-sectional area was also calculated for one muscle from each limb segment and represents the percentage of the muscle’s cross-sectional area made up of slow fibers. It was determined using the diameter measurements of slow fibers and total muscle cross-sectional areas.

Results

We describe the muscles of the hindlimb that we believe to function in hip and knee flexion and leg extension based on an analysis of the muscle morphology and the literature. Brief anatomical descriptions of the muscles are provided, along with our fiber type data. Nomenclature follows the Nomina Anatomica Avium (Baumel et al., 1993). Overall, the hindlimb muscles of both species are similar to those described for a wide number of birds (see Baumel et al., 1993). In contrast to other avian species (e.g., pigeons, Rosser and George, 1986b; Welsford et al., 1991) we found all of the muscles studied to possess some oxidative capability—from slightly to highly oxidative (see Fig. 1). Since there were no fast glycolytic (FG) fibers, we refrain from using the traditional system of nomenclature (SO, FOG, FG; see Kovacs and Meyers, 2000). Unless otherwise specified, data are for both avocets and stilts, which largely showed similar results.

Hip Flexion

M. iliotibialis cranialis (ITC) arises from the cranial aspect of the iliac crest, via a short tendon. It runs along the cranial border of the thigh, and passes medially to insert onto the anterior edge of the tibiotarsus, deep to M. gastrocnemius pars medialis. Its parallel-fascicled architecture suggests a functional role in large excursions. Slow fibers made up 12–19% of the muscle (N = 6; Tables 1 and 2) and were located along the medial half of the muscle, where they were interspersed with fast fibers. The lateral half was consistently uniformly fast (see Fig. 2).

M. iliotrochantericus caudalis (IlioCa) is a large muscle at the cranio-dorsal boundary of the hip joint. It has a wide origin from the iliac fossa, extending cranially and dorsally to the acetabulum. IlioCa inserts onto the femoral trochanter dorsally and cranially by a thick aponeurosis (see Fig. 2). There is an internal tendon that extends the length of the muscle and divides it into superficial and deep zones. In avocets and stilts, slow fibers range from 30 to 48% of the muscle (N = 6; Tables 1 and 2). Slow fibers make up all of the superficial muscle, lateral to the internal tendon.
The deep portion was mixed with both slow and fast fibers (see Fig. 2). The weighted cross-sectional area of slow fibers was determined to be larger than numerical percentages for all six individuals and ranged from 35 to 59% of the muscle area (Tables 1 and 2).

Mm. iliotrochantericus cranialis (IlioCr) and iliotrochantericus medius (ITM) are two small muscles that cross the cranio-ventral border of the hip joint; thus they should be considered protractors of the femur. IlioCr arises from the central border of IlioCa and the adjacent lip of the ventral ilium. Most of the muscle lies ventral and deep to IlioCa. The flat belly inserts onto the femur distal to the femoral trochanter (see Fig. 2). In two individuals there was no connection of IlioCr to IlioCa. ITM is a highly variable muscle; it was absent in half of our specimens. It lies deep to IlioCr and crosses the cranial aspect of the hip, extending from the ilium to the proximal femur (see Fig. 2). Slow fibers in IlioCr range from 8 to 16% of the muscle (N = 6; Tables 1 and 2) and are fairly evenly distributed

*Fig. 1. Recurvirostra americana. Serial sections of M. iliotibialis cranialis, with homologous fiber marked in each reaction (*). Top row: results of mATPase reaction with acid (pH 4.2) and alkaline preincubations (pH 10.35). Second row: results for reactions with ALD58 and MY32 to illustrate slow and fast fibers, respectively. Third row: glycolytic activity indicated by presence of α-GDP, and oxidative activity indicated by presence of NADH-D. Scale bar = 100 μm.*
with few to no slow fibers in the extreme cranial region (see Fig. 2). Slow fibers in ITM range from 17 to 65% of the muscle ($N = 6$; Tables 1 and 2) and are also evenly distributed (see Fig. 2).

**Knee Flexion**

Mm. iliofibularis (IF), flexor cruris lateralis (FCL), and flexor cruris medialis (FCM) are all flexors of the knee. IF takes origin from the dorsal iliac crest beneath M. iliotibialis lateralis. It is triangular in shape and tapers to a robust, round tendon, which passes through the ligamentous ansa IF (see Fig. 3). The ansa redirects the tendon’s course distally, where it inserts onto the proximal caudal fibula. In one individual, one-third of the tendon passing through the ansa also possessed fleshy fibers. The slow fibers of IF range from 15 to 25% ($N = 6$; Tables 1 and 2) and were mixed along the medial half of the muscle; few slow fibers were found along the lateral half (see Fig. 3). The weighted cross-sectional area of slow fibers was determined to be larger than numerical percentages for all six individuals and ranged from 16 to 29% of the muscle area (Tables 1 and 2).

FCL has a fleshy origin from the dorsal iliac crest, caudally to IF. It parallels and lies deep to IF, but is much narrower and is visible beneath the ventral border (see Fig. 3). FCL inserts onto a tendinous raphe, from which continues its accessory belly. The accessory belly attaches onto the caudal femur. The raphe continues caudally as a tendon that attaches onto M. gastrocnemius pars intermedia (see Fig. 3). The slow fibers of FCL range from 2 to 11% ($N = 6$; Tables 1 and 2) and are restricted along the cranio-lateral edge of the muscle (see Fig. 3).

FCM takes its origin from the ilium, caudal and ventral to the origin of FCL. It is approximately twice as wide as FCL, and lies beneath and ventral to that muscle. FCM inserts by a narrow flat tendon to the medial aspect of the proximal tibiotarsus (see Fig. 3). Slow muscle fibers range from 10 to 25% of the muscle ($N = 6$; Tables 1 and 2) and are restricted along the cranio-lateral edge of the muscle (see Fig. 3).

**Ankle Extension**

M. gastrocnemius is the principal extensor of the tarsometatarsus and has three parts in avocets and...
<table>
<thead>
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<th>Muscle</th>
<th>Bird</th>
<th>Slow</th>
<th>Fast</th>
<th>Mean # slow fibers ± SD</th>
<th>% slow fibers</th>
<th>Weighted CSA: % slow fibers ± SD</th>
<th>Mean # slow fibers ± SD%</th>
<th>Weighted CSA: mean % slow fibers ± SD%</th>
<th>Muscle CSA (mm²)</th>
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<td>6</td>
<td>9 ± 2</td>
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<td>10</td>
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*<sup>A</sup>*A third specimen demonstrated a similar distribution of slow fibers, but the fibers could not be discriminated for quantification.

*<sup>B</sup>*Value calculated using diameter measurements from specimen 5.
distal end of the tibiotarsus. Highly variable numbers of slow fibers made up 11–84% of the muscle of five individuals; a sixth was 100% slow (Fig. 4; Tables 1 and 2).

**Muscle Fiber Diameters**

One muscle from each limb segment was selected for measurement of muscle fiber diameters: from the hip flexors, IlioCa; from the knee flexors, IF; and from the ankle extensors, GI (see Table 3). A comparison of slow and fast fibers revealed that for all three muscles, slow fibers were significantly larger than fast fibers within a given muscle (IlioCa: $t = 3.99$, 5 d.f., $P < 0.05$; IF: $t = 2.43$, 5 d.f., $P < 0.05$; GI: $t = 3.83$, 5 d.f., $P < 0.05$; see Table 3).

**DISCUSSION**

**Muscles Involved in Extended Hindlimb Posture**

Our results revealed populations of slow fibers in muscles that flex the hip and knee, and extend...
the ankle in both American Avocets and Black-necked Stilts. We suggest that these slow fibers are utilized in maintaining the hindlimbs in a trailing, extended position during flight. Other postural activities, such as standing, would require slow fiber activity in hip and knee extensor muscles. Ankle extensors would need to function during both standing and during flight, and this may explain their large combined size and the total numbers of slow fibers in them.

**Hip flexion.** ITC is widely considered to be the principal protractor of the hip (Raikow, 1985; Verstappen et al., 1998; Smith et al., 2007). Suzuki et al. (1985) found the medial and lateral regions of chicken ITC to be 73 and 99% fast contracting, respectively. Torrella et al. (1996) described 22–26% slow fibers in the posterior part of the coot ITC, with 2% in the anterior half. In the Mallard, Torrella et al. (1998a) reported 17–19% slow fibers in the posterior part of the muscle, and ~2% in the anterior half. In gulls (Torella et al., 1998b), the posterior region had 6–16% slow fibers, almost three times that of the anterior half. Our finding that this muscle possesses 12–19% slow fibers fits with these studies.
Historically, IlioCa has been considered a femoral protractor. Raikow (1970) suggested that IlioCa functions as a rotator of the femur rather than a flexor. Cracraft (1971) suggested that this muscle might be a retractor of the femur, at least in pigeons and described several muscle sections as 50% slow tonic. He suggested that this muscle functions as a "shock-absorber" during landing. Gatesy and Dial's (1993) figure 6 shows low level electromyographic activity of IlioCa during flight. This low activity level may be slow muscle fibers active in hip flexion during flight. Since the muscle fascicle orientation changes through the muscle from cranial to caudal, it is likely that some of the slow fibers can function in flexion; the entire lateral aspect of the

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muscle was slow (see Fig. 2). Gatesy also found slow fibers along the external aspect of the muscle in this species (pers. comm.).

There has been some debate regarding the ability of IlioCr and ITM to protract the femur. Regarding ITM, Cracraft (1971) wrote that it “most assuredly does not protract the femur.” Raikow (1985) noted the small lever arm and size of both these muscles as making their role as protractors unlikely; Verstappen et al. (1998) went so far as to say they “cannot be considered femoral protractors.” Even if these muscles cannot move the femur, we believe that the 7–15% slow muscle fibers in these muscles may function to hold the femur flexed once other muscles (e.g., ITC) move the thigh into a protracted orientation.

**Knee flexion.** Very little is known about the fiber types of the avian knee flexors. Cracraft (1971) reported FCL as possessing “mixed” fiber types. In FCM, he noted that many of the fibers may be slow tonic, indicating a different functional role. In IF, Cracraft described fewer than 20% slow tonic fibers. Turner and Butler (1988) examined FCL in the Tufted Duck (*Aythya*) and found no slow fibers.

**Ankle extension.** Although the gastrocnemius muscle has been studied, its large size makes complete fiber analyses rare. Torrella and his colleagues determined that in the coot, the cranial half of GL possessed 11–19% slow fibers (with no slow fibers caudally) (Torrella et al., 1998a). Gulls and mallards showed the same pattern, with 17–34% slow in the cranial half of the gull GL (Torrella et al., 1998b) and 4–25% in the cranial half of the mallard GL (Torrella et al., 1996). Turner and Butler (1988) described the tibial part of the Tufted Duck GL as “much redder,” and was the only part to have slow fibers. According to Patak and Baldwin (1993), the emu had no slow fibers in any part of M. gastrocnemius. They assigned a postural function (and energy storage role) to the digital flexor muscles, which possessed ~30% slow fibers. The 18–45% slow fibers we found over all parts would seem sufficient to perform the multiple roles of this muscle in standing, walking, and maintaining the hindlimbs extended during flight.

Future *in vivo* stimulation and electromyographic studies are needed to determine potential muscle actions and usage during flight as well as in other behaviors.

**Muscle Compartmentalization**

It is well known that muscles can be functionally subdivided into regions or compartments having specific functions (English and Letbetter, 1982). For example, the avian pectoralis has two primary regions with discrete innervations, fascicle orientations and recruitment patterns (Dial

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**TABLE 3. Diameter and average diameter of fast and slow fibers (microns ± SD) by bird specimen number for three muscles of American Avocets and Black-necked Stilts.**

<table>
<thead>
<tr>
<th>Muscle</th>
<th>Avocet</th>
<th>Stilt</th>
<th>Average fiber diameter ± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>M. iliotrochantericus caudalis</td>
<td>45.03 ± 3.83</td>
<td>48.23 ± 5.18</td>
<td>50.55 ± 4.68</td>
</tr>
<tr>
<td>M. iliotrochantericus cranialis</td>
<td>37.67 ± 6.35</td>
<td>46.19 ± 7.18</td>
<td>56.98 ± 7.6</td>
</tr>
<tr>
<td>M. iliofibularis</td>
<td>38.09 ± 4.93</td>
<td>43.84 ± 5.36</td>
<td>51.06 ± 5.69</td>
</tr>
<tr>
<td>M. gastrocnemius pars intermedia</td>
<td>34.85 ± 4.15</td>
<td>42.04 ± 4.37</td>
<td>48.72 ± 4.55</td>
</tr>
<tr>
<td>M. gastrocnemius</td>
<td>32.02 ± 4.25</td>
<td>40.36 ± 4.38</td>
<td>46.82 ± 4.50</td>
</tr>
<tr>
<td>M. gastrocnemius</td>
<td>36.36 ± 4.20</td>
<td>49.67 ± 4.99</td>
<td>54.07 ± 4.53</td>
</tr>
</tbody>
</table>

*Indicates data from avocet 6.

*Indicates data from stilt 1.
et al., 1988), and has been shown to possess additional subregions with distinct innervations and EMG activity intensities (Boggs and Dial, 1993). With regard to different fiber types, there must also be a functional subdivision, as in the differing recruitment patterns of pigeon fast glycolytic (FG) and fast oxidative glycolytic (FOG) fibers (Sokoloff and Goslow, 1999), which also show a consistent pattern of fiber distribution (i.e., pigeon pectoralis with FG fibers at the periphery of fascicles: Dial et al., 1987). A similar subdivision might also be expected when a muscle's slow fibers are used for posture and the remaining fast fibers are inactive.

Most of the hindlimb muscles we examined showed a regionalization of slow fibers consistent across individuals. For example, FCM and IF showed a mixture of slow and fast fibers along the medial half of the muscle, but no slow fibers laterally in any of the six birds examined (see Fig. 5). IlioCa showed a consistent pattern in the six individuals; slow muscle fibers exclusively lateral to the internal tendon, and mixed fast and slow fibers medial to the tendon (see Fig. 5). ITC showed a mixed medial half, and no slow fibers along the lateral half in all individuals (see Fig. 5). Additionally, the lowest variability of fiber type ratios was observed in the largest muscles. For example, GM

Fig. 5. Cross section views of Mm. flexor cruris medialis (A–C), iliofibularis (D–F), iliotrochantericus caudalis (G–I), and iliobialis cranialis (J–L). A, H, K, and L are from *Himantopus*, remaining muscles are from *Recurvirostra*. Muscle cross sections illustrate anti-slow ALD 58 antibody reactions, where positively-reacting slow fibers appear red. Note similarity of muscle fiber distribution and the small degree of variation among individuals and species. Lateral is to the top of the page, cranial is to the right. All scale bars represent 1 mm.
and IlioCa, two of the largest muscles studied, ranged from 38 to 43% and 30–48% slow, respectively. In contrast, PI and ITM, two of the smallest muscles, had a much larger range of 11–100% and 17–65% slow, respectively.

We suggest that there is likely a functional reason for the patterns seen across individuals. Slow fibers must be able to be activated independently of the fast fibers, as they belong to their own motor units (Sokoloff and Goslow, 1999). Thus the superficial region of IlioCa may contract independently of the deeper region, and the slow fibers here may act in hip flexion during flight whereas the deep fast region may act in extension during walking.

**Muscle Fiber Sizes**

The traditional view of the various muscle fiber types is that the slow oxidative fibers (Type I, SO) are the smallest in diameter and the fast glycolytic (FG) fibers (Type IIb) are the largest (see Hildebrand and Goslow, 1998; Kernell, 1998). This provides an overall greater surface area for diffusion of oxygen into the muscle fibers. A number of studies have reported that this pattern is not typical of all muscles. SO fibers were reported to be larger than FOG (Type IIa) fibers in Mm. tensor fascia lata, gastrocnemius, and complexus in chickens (Wada et al., 1999) as well as those in the thigh muscles of rabbits (Rab et al., 2000). Likewise, Laidlaw et al. (1998) showed that in two of the seven hindlimb turtle muscles studied, SO fibers were larger than FOG fibers. It is worth noting that in all of these exceptions to the “rule,” anaerobic FG fibers (if present) were still the largest as both SO and FOG would gain a diffusion benefit from their smaller cross-sectional areas.

Our data of three measured hip, knee, and ankle muscles show that slow fibers in avocets and stilts are also significantly larger than the fast fibers (see Table 3). Since these bird muscles are largely oxidative (no FG fibers were found in any of the muscles examined), the increased diffusion distance may be a worthwhile tradeoff to increase cross-sectional area (CSA) and thus force production. Based on our weighted CSA calculations, these larger slow fibers do increase the percentage of slow fiber cross-sectional area. For example, in IlioCa from three avocets, slow fibers made up 51% of the muscle based on weighted cross-sectional area, although only 38% of the fibers were slow (Tables 1 and 2).

**Slow Fibers Deeper**

A number of studies have shown that slow muscle fibers are more likely to be distributed in deeper regions of muscles (e.g., Suzuki et al., 1985; Turner and Butler, 1988; Torrella et al., 1996, 1998a). Wang and Kernell (2001) discussed the functional reason for this type of fiber regionalization, including 1) heat conservation, 2) heat generation increasing muscle power, 3) biomechanical advantages, and 4) protection of fibers vulnerable to mechanical damage.

Our data for slow fibers in shorebirds do not support this observation. IlioCa has its greatest concentration of slow fibers within the superficial zone (see Fig. 2), a pattern displayed in all individuals of avocets and stilts. Other muscles (e.g., IF, FCL, FCM) had more slow fibers within the medial half than the lateral half (see Fig. 3), but these regions are not “deeper” in the sense that they are closer to bone.

As Wang and Kernell (2001) state, more research is needed to elucidate the possible functional advantages of deeply situated slow fibers. With IlioCa, we believe the lateral predominance of slow fibers specifically relates to their biomechanical advantage in rotating and holding the femur in a flexed position. Likewise, we believe that the location of slow fibers in other muscle groups (e.g., hip flexors) also relates to their biomechanical advantage, a factor that maintains the pattern across all individuals.

**Evolution of Hindlimb Posture**

Birds such as raptors, shorebirds, pheasants, and parrots utilize the extended leg posture during flight, where the ankle is in extension. Songbirds, hummingbirds, and woodpeckers adopt the flexed limb posture, with the ankle in flexion. We have developed four explanations, which, independently or summarily, may illustrate the reasons for the distribution of two different leg postures among the orders in class Aves.

Since the shape of the avian body has been shown to contribute aerodynamic lift (Csicsaky, 1977), we suspected that leg posture would affect the streamlined nature of the body in flight. Thus, large hindlimbs that may disrupt airflow would be trailed behind the body, while limbs small enough to be tucked below the body with minimal airflow disruption would be held in a flexed posture during flight. Supporting this idea, we have observed that pheasants and macaws use the flexed position during take off and short flights, and then reposition them to the extended position during sustained flights (Shepard and Meyers, 2006; Shepard et al., 2008).

Alternatively, in-flight hindlimb posture may be explained as a behavioral component of flight control. The use of feet extended behind the body as a rudder in birds with short tails (such as alcids), or the need to position the feet below the body for frequent takeoff and landing (as in perching birds), may dictate the position in which the legs must be placed during flight.
We also considered that limb posture during flight may be structurally constrained. Species utilizing the extended flight posture may lack the musculature (or joint mobility) to position their legs flexed beneath the body. However, having read numerous published accounts of birds that normally utilize the extended posture (Sandhill Cranes, Whooping Cranes, and Canada Geese) flying in cold weather with their ankles flexed (Barrett-Hamilton, 1903; Epp 1970; Nesbitt, 1978; Alonso, 1985), we determined that this explanation is unlikely. However, it may be that insufficient slow fiber populations in ankle flexors make long-term maintenance of this posture impossible.

Finally, we attempted to correlate the distribution of different flight postures among bird orders. Bird orders appear homogeneous for a specific hindlimb flight posture. For example, all members of the Falconiformes, Ciconiiformes and Anseriformes use the extended limb posture. All Passeriformes utilize the flexed position, even large birds such as crows and ravens. Thus hindlimb flight posture may have a phylogenetic component, and not related to body size nor otherwise adaptively driven. We are in the process of developing these ideas in more detail and testing them over a wide number of species.

CONCLUSIONS

Based on Meyers’ (1992) description of a set of uniformly slow muscles that act in maintaining the folded wing, we expected some muscles, notably small muscles, such as IlioCr and ITM, to be mostly or entirely slow in fiber makeup and postural in function. We were surprised to find no muscle consistently homogeneous for slow fibers. However, no muscle group was without slow fibers, indicating potential postural function at all joints. The distribution of these slow fibers was unusually consistent among individuals and across species. Furthermore, the larger cross-sectional area of slow fibers may account for a more robust postural component in the absence of greater fiber numbers.

We believe that the versatility of hindlimb function, such as standing, walking, and swimming, explains the lack of muscles dedicated to posture. This is in contrast to the wing, which is either in motion or at rest. Future work will determine muscle activity patterns during flight as well as fiber type distribution in the hindlimb muscles of “flexed hindlimb” flyers.

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