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Molt, feather growth rate and body condition of male and female Barn Swallows

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Abstract Molt and ecdysis are costly activities crucial to several animals. Most birds completely molt their plumage once per year. The diverse energy and aerodynamic costs of molt are expected to shape the evolution of molt schedules and rate of feather renewal. Differences in molt strategies are also predicted between the sexes because of sex-related variation in fitness payoffs of rapid and early feather renewal. Feathers show barring patterns that reflect daily growth pulses. Ptilochronological analysis of growth bar width (GBW) reflecting condition-dependent feather growth rate can provide important, though overlooked information on molt strategies, and on the effects of body condition on feather renewal and subsequent migration or breeding. In this study of migratory Barn Swallows (*Hirundo rustica*) wintering in Nigeria, for the first time we analyzed sex-related variation in molt schedules and feather growth rate using molecular sexing. We found no sex difference in molt schedules but faster feather growth in females. Hence, protandry in arrival of swallows to

European breeding grounds does not depend on sex-specific timing of molt or feather renewal rate. Individuals with advanced molt had larger GBW and muscle mass, suggesting that individuals in good condition complete molt earlier thanks to faster feather growth. Arrival date on the breeding areas may thus reliably signal male quality, if molt completion constrains migration. Finally, the frequency of low GBW values among wintering males was larger than during breeding in swallows that winter in Nigeria, suggesting selection against low GBW. This study provides the first unequivocal data on sex-related variation in molt schedules in a species with no discrete sexual dimorphism, and emphasizes the potential of ptilochronology in ecological–evolutionary studies, as it shows that feather growth rate covaries with timing of molt but may not mediate protandrous migration.

Keywords Barn Swallow · Body conditions · Feather growth rate · *Hirundo rustica* · Molt · Sex · Wintering

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Zusammenfassung

Mauser, Federwachstum und Kondition von Männchen und Weibchen der Rauchschwalbe

Mauser und Häutung sind zeitaufwändige Vorgänge, die für viele Tiere entscheidend sind. Die meisten Vögel mausern ihr Gefieder vollständig einmal im Jahr. Man erwartet, dass die vielfältigen Energie- und aerodynamischen Aufwendungen der Mauser die Evolution von Mausermustern und die Geschwindigkeit der Federerneuerung bestimmen. Unterschiede in Mauserstrategien werden auch zwischen den Geschlechtern auf Grund geschlechtsspezifischer Unterschiede in Fitnessvorteilen aus schneller und früher Federerneuerung vorhergesagt. Federn zeigen

Streifenmuster, die tägliche Wachstumspulse widerspiegeln. Ptilochronologische Analyse der Breite dieser Wachstumsstreifens, die die von der Kondition abhängige Federwachstumsrate widerspiegelt, kann wichtige, obwohl oft übersehene Informationen über Mauserstrategien und über die Wirkungen der Körperkondition auf Federerneuerung und folgenden Vogelzug oder Fortpflanzung liefern. In dieser Studie an in Nigeria überwinternden Rauchschwalben (*Hirundo rustica*) haben wir zum ersten Mal die geschlechtsgebundene Variation in Mausermustern und Federwachstumsrate analysiert, indem wir die Geschlechter molekular bestimmten. Wir fanden keinen geschlechtsgebundenen Unterschied im Mausermuster, aber schnelleres Federwachstum bei Weibchen. Folglich hängt die Protandrie in der Ankunft der Schwalben in europäischen Brutrevieren nicht von einem geschlechtsgebundenen Zeitplan für das Mausern oder die Federerneuerungsrate ab. Schwalben mit fortgeschrittener Mauser zeigten breitere Wachstumsstreifen und mehr Muskelmasse, was darauf hinweist, dass Vögel in guter Kondition ihre Mauser dank schnellerem Federwachstum früher abschlossen. Folglich könnte das Ankunftsdatum in den europäischen Brutgebieten auf zuverlässige Weise ein Zeichen von Männchenqualität sein, wenn die Vollendung der Mauser den Vogelzug einschränkt. Schließlich fanden wir, dass schmale Wachstumsstreifen bei überwinternden Männchen häufiger waren als bei Vögeln zur Brutzeit, die in Nigeria überwinterten, was auf eine Selektion gegen schmale Wachstumsraten hindeutet. Unsere Untersuchung liefert die ersten unzweifelhaften Daten über geschlechtsspezifische Variation im Mausermuster einer Art ohne diskreten Geschlechtsdimorphismus und betont das Potenzial der Ptilochronologie für evolutionsökologische Untersuchungen, indem sie zeigt, dass die Federwachstumsrate mit Mausermustern kovariert, ohne aber Protandrie zu bedingen.

Introduction

Photochemical and physical processes as well as biological agents cause feathers to undergo tear and wear at a fast pace relative to the lifespan of their bearer. Periodical plumage molt thus serves to retain thermal insulation and aerodynamic performance, with beneficial consequences for escape from predators and foraging. Molt can also be crucial to socio-sexual communication, as it may involve the expression or renewal of plumage signals (Andersson 1994; Fitzpatrick 1998). Because feathers contain ca. 95 % proteins (Murphy 1996) and plumage mass may build up to 20–30 % of total lean dry body mass, molt is highly demanding in terms of protein mobilization (Chilgren 1977; Murphy 1996) and entails a marked increase of metabolic rates (Lindström et al. 1993; Cyr et al. 2008).

Species differ considerably in the timing of molt, which in most cases is partitioned from other activities, such as breeding and migration, although overlap may occur because of time constraints (Ginn and Melville 1983; Jehl 1990; Jenni and Winkler 1994). The diverse time and energy, insulative, aerodynamic and predation costs of molt (Nilsson and Svensson 1996; Swaddle et al. 1999; Dawson et al. 2000; Hedenström 2003) may prevent simultaneous allocation of resources to other activities, and trade-offs with fitness traits (Siikamäki et al. 1994; Svensson and Nilsson 1997; Hemborg and Lundberg 1998) are thus commonly invoked as the evolutionary drivers of temporal segregation of molt from other major activities (e.g., Barta et al. 2008).

Variation in molt schedules also exists among conspecific populations (Ginn and Melville 1983; Buehler and Piersma 2008; Salewski et al. 2004), as well as among individuals according to age, timing of hatching or breeding, or physiological state (van den Brink et al. 2000; de la Hera et al. 2011; Newton 2011). In general, tightness of annual routines and thermoregulation or aerodynamic costs are expected to select for rapid molt. However, because speed of feather growth may have to be traded against feather quality (Dawson et al. 2000; Serra et al. 2007; Barta et al. 2008; de la Hera et al. 2009, 2010; Vágási et al. 2010), only individuals in prime condition should be able to afford rapid molt without hindering plumage quality.

Sex is also potentially important, though poorly investigated particularly in sexually monomorphic species, as a source of variation in molt schedules. Annual routines may diverge between the sexes because of unequal natural and sexual selection pressures on either sex (Andersson 1994; Newton 2011). In migratory species, males may be selected to arrive earlier than females to the breeding grounds (protandry), because of the potential benefits that early arrival accrues in scramble for access to territories and mates (Morbey and Ydenberg 2001; Møller et al. 2003; Spottiswoode and Saino 2010; Saino et al. 2010). If spring migration is directly constrained by completion of molt in the winter quarters (Rubolini et al. 2005), males may advance the start of molt and/or molt faster than females. Unfortunately, few published accounts exist of the timing of molt in either sex in general (Norman 1990), and information is virtually absent for species with no obvious phenotypic marker of sex at the time of molt.

A major limit to the performance of longitudinal analysis of molt is the difficulty of repeatedly sampling the same free-ranging individuals. However, feathers often show a transversal barring pattern, which varies in distinctness among species and plumage regions (Riddle 1907; Grubb 2006; Saino et al. 2012). Bars run (sub-)perpendicular to the rachis and are formed by consecutive pairs of dark/light bands, each a very few millimeters in width. Each 'growth

bar' (a pair of consecutive dark/light bands) is believed to correspond to a 1-day growth interval (Grubb 1995; de la Hera et al. 2011; Jovani et al. 2011; but see Kern and Cowie 2002). Thus, the analysis of growth bars can allow the estimation of the feather growth rate. Importantly, diverse factors may determine feather growth rate and thus growth bar width (GBW hereafter). Indeed, GBW covaries with habitat quality and urbanization (Carrascal et al. 1998; Perez-Tris et al. 2000; Vangestel et al. 2010), and with social environment (Carrascal et al. 1998), and is affected by diverse forms of stress and by nutritional condition (Grubb and Cimprich 1990; Grubb 1991; White et al. 1991; but see Murphy 1992). The analysis of GBW therefore qualifies as a potentially powerful, though underexploited, tool to study variation in molt speed and phenology, and of the impact that conditions during molt have on the entire process of feather renewal (de la Hera et al. 2011).

In the present study of Barn Swallows (*Hirundo rustica*) wintering in Nigeria, we first analyze the covariation of molt stage on wings, tail and crown feathers, and whether molt phenology differs between the sexes. Because Barn Swallows show protandrous arrival to Europe from spring migration (Møller 1994, 2007), we predicted advancement of molt in males relative to females. We then analyzed the association between molt stage and basic aspects of body condition, including subcutaneous fat depots and pectoral muscle mass, predicting that individuals in good condition would be in a more advanced stage of molt (Møller et al. 1995; van den Brink et al. 2000). In addition, molt of flight feathers entails increased wing loading, and birds can increase pectoral muscle mass to compensate for reduction in wing area (Holmgren et al. 1993; Lind and Jakobsson 2001), also leading to predict a positive relationship between the progress of molt (molt stage) and muscle mass. Under the likely assumption that individuals in prime condition do not postpone the start of molt, we predicted a positive correlation between feather growth rate and molt stage, because individuals in better condition should grow their feathers at faster pace. Along the same line of reasoning, we also predicted a positive relationship between GBW and body condition.

The present paper is companion to a study of molt phenology of primary wing feathers in relation to polymorphism at the *Clock* gene, which is involved in photoperiodic responses in vertebrates (Saino et al., submitted; see Caprioli et al. 2012).

Methods

Study organism

Barn Swallows are long-distance migratory, insectivorous passerines. European breeding populations winter in sub-Saharan Africa (Møller 1994; Cramp 1998). Barn Swallows arrive in Europe in March–May. Males arrive earlier than females but arrival dates extensively overlap between the sexes (Møller 1994; Cramp 1998). Breeding pairs may lay up to three clutches per season. Autumn migration mostly occurs in August–October. Barn Swallows molt wing and tail feathers once per year, mostly in the African winter quarters (Ginn and Melville 1983; Cramp 1998; van den Brink et al. 2000). Primary wing feathers are molted sequentially starting from the innermost one (P1), while the secondary wing feathers are molted sequentially starting from the outermost one (S1), and the tertials are molted separately. Tail feathers are not necessarily molted in a sequential order, but the innermost ones are normally molted earlier (Ginn and Melville 1983).

Sampling procedures

We mist-netted Barn Swallows at a nocturnal roost in Boje, eastern Nigeria (6°17'26″N, 8°55'51″E) on 12–16 February 2012. This traditional roost hosts hundreds of thousands of Barn Swallows originating from at least 12 western or central European countries (P. Micheloni, unpublished data). The roost maintains rather constant size until February, and then increases in size in March. The study was conducted in mid-February because we aimed at analyzing molt shortly before spring migration, in a period when 40–50 % of the birds, in normal years, have completed molt (P. Micheloni, personal observation). On the other hand, we also aimed at retaining variation in molt stages in the sample, and we therefore decided to collect a sample composed of three-quarters from birds that were still molting wing feathers and of one-quarter from birds that had completed their molt. Individuals within these categories were taken randomly. Because the aim of the study was to investigate the relationships among molt status, feather growth rates and body condition in either sex, rather than describing the phenology of molt, this protocol did not introduce any bias in the results.

Because we were interested in sex-related variation in the progress of molt shortly before migration, we intentionally sampled the birds in late winter. A drawback of this approach was that age of the birds could not be identified. This is because age of the individuals born in the previous spring can be identified based on plumage but only before the first complete molt has been completed. Thus, while it is likely that the majority of the birds that had completed molt were in their second (or later) winter, their age (1st winter or later) could not safely be assigned. In the analyses, no age effects are therefore considered and the potential effects of age are commented at length in the "Discussion".

Barn Swallows show moderate sexual size or color dimorphism at traits whose frequency distribution extensively overlaps between the sexes. Because individuals with intermediate phenotypic values may be difficult to sex unequivocally, particularly when tail feathers are missing because they have been shed and are not fully regrown, we relied on molecular sexing (Saino et al. 2008) using a small blood sample. Two standard indices of body condition were recorded: pectoral muscle mass (0–3 scale; Svensson 1992) and subcutaneous fat stores (0–5 scale; Kaiser 1993).

Molt status

We quantified molt at four plumage regions: primary and secondary wing feathers, tail feathers, and crown feathers. Each primary or secondary feather and tail feather of the right side was scored for molt using a standard 6-levels (0–5) scale (0: presence of the old feather; 5: fully grown new feather; 1–4: progressively increasing development of the growing feather) (Ginn and Melville 1983). Feathers on just one side were scored because molt was highly bilaterally symmetric. Molt stage on the crown was quantified as percentage (0–100 % in 10 %-increment discrete classes) of the crown area covered by newly grown feathers.

Ptilochronological measurements

We measured GBW on the 7th primary wing feather (P7; P1 = innermost primary) because growth bars can in most cases be identified with accuracy on this feather (Saino et al. 2012). In addition, P7 was the outermost primary that was at least partly grown in the vast majority of the individuals. To measure GBW, we first identified the proximal and distal limits of a feather segment, including 8 growth bars, starting from the first/second clearly visible bar at the distal end of the feather. The limits of the segment were marked on the rachis with a fine white fiber-tip pen. We then measured the length of the segment with a caliper (approximation of 0.01 mm, expressed in mm × 10). GBW on P7 was expressed (in mm × 10) as length of the segment/8 (=number of growth bars) (Grubb 2006; Saino et al. 2012). Hence, large GBW indicates rapid feather growth. In 24 individuals, we also measured GBW on P6 to test for consistency of GBW in different feathers. In a previous study, we have shown that repeatability of measures of GBW, which was assessed on tail feathers, is high (=0.86) (Saino et al. 2012). Owing to the difficulties of working at night in the forest, not all measurements could be taken from all individuals. In all analyses, however, we used the maximum sample size available. The size of the samples involved in the analyses is reported either precisely or as a range, when the results of several analyses were summarized.

Depending on the specific question at hand, estimates of GBW in, for example, either sex may need to be corrected for the length of the fully grown feather on which it is measured (e.g., de la Hera et al. 2009, 2011). In the present study, P7 as well as the outermost primaries were still growing in a large fraction of the birds we sampled, so that an index of wing length could have been obtained only for a non-random fraction of the birds. However, to test for a positive covariation between GBW and primary length, we used the data that we collected during the breeding season in Italy (Saino et al. 2012) in a sample of 41 females (23 yearlings, 18 older individuals) and 42 males (15, 27) and found weak, positive correlations ($0.191 < r < 0.380$; $0.051 < P < 0.457$) (see also "Discussion").

Statistical analyses

We mainly relied on non-parametric statistical tests because of right-truncation of the frequency distribution of molt scores and the ordinal categorical nature of several variables. PCA based on correlation matrix and run on either sex separately was used to summarize the information of the four molt score variables that were highly positively correlated (see "Results"). No axes rotation in PCA was applied. Scores of the individuals on the PC1 were used as an index of overall plumage molt status. In significance tests, the α value was set at $P \leq 0.05$.

Results

Sex differences in molt scores

Molt was scored at four plumage regions in 124 females and 136 males. The proportion of males and females in the subsamples of birds that had or, respectively, had not completed molt of the primary feathers did not differ (molt completed: 33 males, 32 females; still molting: 92 females, 103 males; contingency $\chi^2 = 0.02$, $P = 0.89$). Molt scores at individual plumage regions did not differ between males and females in the overall sample of birds (Table 1; Fig. 1), nor among the individuals that had not yet completed molt of the primaries (Mann–Whitney U test, $-1.61 < Z < -1.07$, $P > 0.10$ in all cases).

Within-sex, molt scores were strongly positively correlated among plumage regions (6 correlations among 4 body regions for each sex; $\tau > 0.70$, $P < 0.001$ in all cases) and the same held true when the analysis was restricted to individuals that were still molting their primaries ($\tau > 0.64$, $P < 0.001$ in all cases). In both sexes, the first principal component (PC1) reflecting molt stage (see "Statistical analyses") accounted for a very large proportion of the total variance in molt scores (females: 91.5 %,

Table 1 Median (25th and 75th percentile) molt scores at four plumage regions of male and female Barn Swallows (*Hirundo rustica*)

	Females	Males	Z	P
Primary feathers	39 (35, 45)	37 (34, 44)	1.02	0.307
Secondary feathers	25 (17, 30)	21.5 (16, 30)	1.31	0.191
Tail feathers	22 (16, 30)	18.5 (14, 30)	1.06	0.290
Crown	90 (42.5, 100)	80 (20, 100)	1.20	0.229

Sample sizes were 124 females and 136 males. The results of Mann–Whitney tests of sex differences are presented

the loadings of the four molt score variables on PC1 ranged between 0.924 and 0.975; males: 91.7 %, 0.919–0.973). As all variables had large, positive loadings on PC1, PC1 scores could be conveniently used as an increasing overall index of plumage molt stage (plumage molt index, PMI).

Molt scores and body condition

Among males, muscle and fat scores were not significantly correlated with PMI, although the correlation coefficient between muscle score and PMI was relatively large and positive (Table 2). Among females, there was also no significant relationship between fat and PMI, but the correlation between PMI and pectoral muscle score was significantly positive (Table 2). In both sexes, fat score was

significantly positively correlated with pectoral muscle score (Table 2). It should be noted that, irrespective of their associated significance level, the correlation coefficients were consistent in sign and of similar magnitude in either sex.

Feather growth rate, molt scores and body condition

Growth bar width was larger among females than males, though marginally non-significantly so [$t = 1.92$, $df = 193$, $P = 0.057$; mean (SE; sample size) for females: 40.72 (0.42; 94); males: 39.58 (0.42; 101)].

Information on molt score variables could also be efficiently summarized by PCA in the sample of birds for which information on GBW was available (variance explained by PC1 in females: 90.7 %, loadings range: 0.906–0.974; males: 89.1 %, 0.897–0.965). GBW was positively correlated with PMI in both sexes (Table 2; Fig. 2). In fact, a positive correlation between GBW and molt scores existed for all the plumage regions (males: $\tau > 0.16$, $P < 0.05$; females: $\tau > 0.20$, $P < 0.007$ in all cases). Moreover, in a subsample of individuals for which GBW of P6 was also measured, there was a significant positive correlation with GBW measured on P7 ($r = 0.34$, $P = 0.019$, $n = 24$). Overall, these results imply that individuals that were found to be in a later stage of molt also had faster growth of their 7th primary but likely also of

Fig. 1 Frequency distribution of total molt scores recorded at individual plumage regions (expressed as the sum of molt scores of individual feathers) in female ($n = 124$) or male ($n = 136$) Barn Swallows (*Hirundo rustica*)
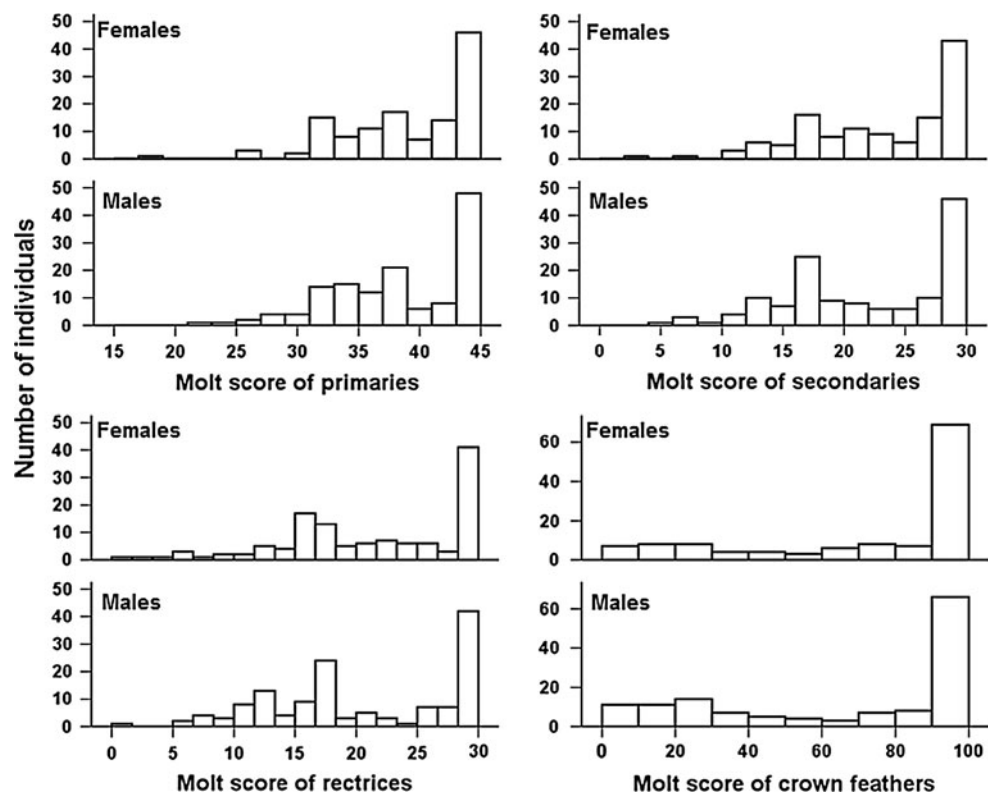
Table 2 Kendall's τ non-parametric correlation coefficients among plumage molt index (*PMI*) expressed as PC1 scores (see "Methods"), width of the feather growth bars (*GBW*) measured on the 7th primary wing feather and reflecting feather growth rate, and muscle and fat scores

	PMI	GBW	Muscle score	Fat score
PMI		0.21**	0.19*	0.05
GBW	0.17*		−0.01	−0.15
Muscle score	0.12	0.06		0.45**
Fat score	0.03	−0.17*	0.23**	

The analyses are run on females (above the diagonal) or males (below the diagonal) separately. Sample sizes ranged between 87 and 123 in the analyses of females and between 92 and 135 in the analyses of males

* $P < 0.05$, ** $P < 0.01$
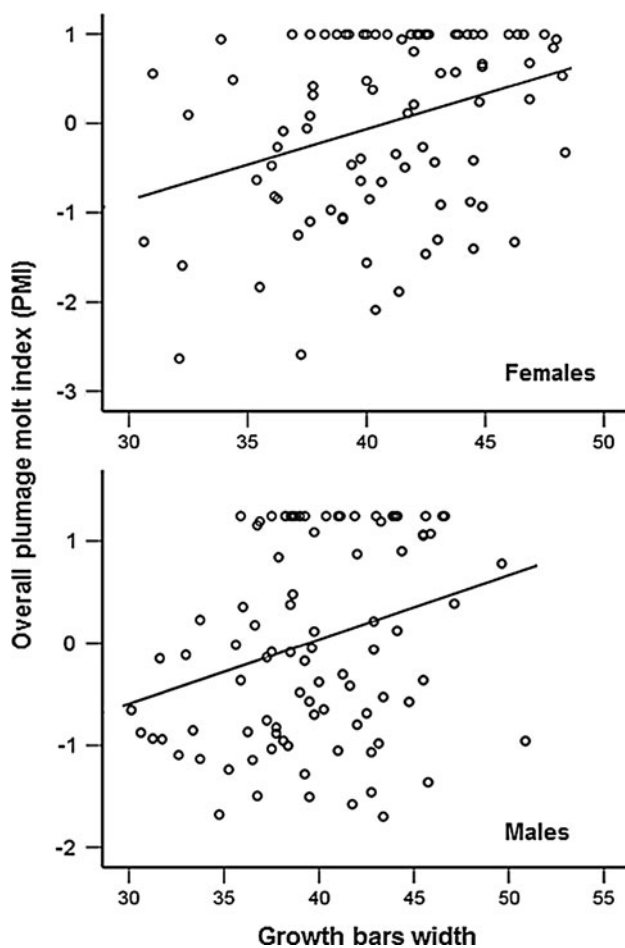
Fig. 2 Overall plumage molt index (PMI) in relation to growth bar width (in mm × 10) recorded on a wing primary feather (P7). PMI is expressed as score on the first principal component obtained from a principal component analysis (PCA) of molt scores at the four plumage regions (wing primaries or secondaries, tail feathers, crown). PCA was carried out on either sex separately. In both sexes, the relationship was positive and statistically significant (see "Results")

the other wing and tail feathers, as indicated by the consistency of molt stages across plumage regions and the correlation in GBW between different wing feathers and between wing and tail feathers (Saino et al. 2012).

Growth bar width was not significantly correlated with muscle score (Table 2), but was negatively correlated with fat store scores, though in females the relationship was marginally non-significant (Table 2; Fig. 3).
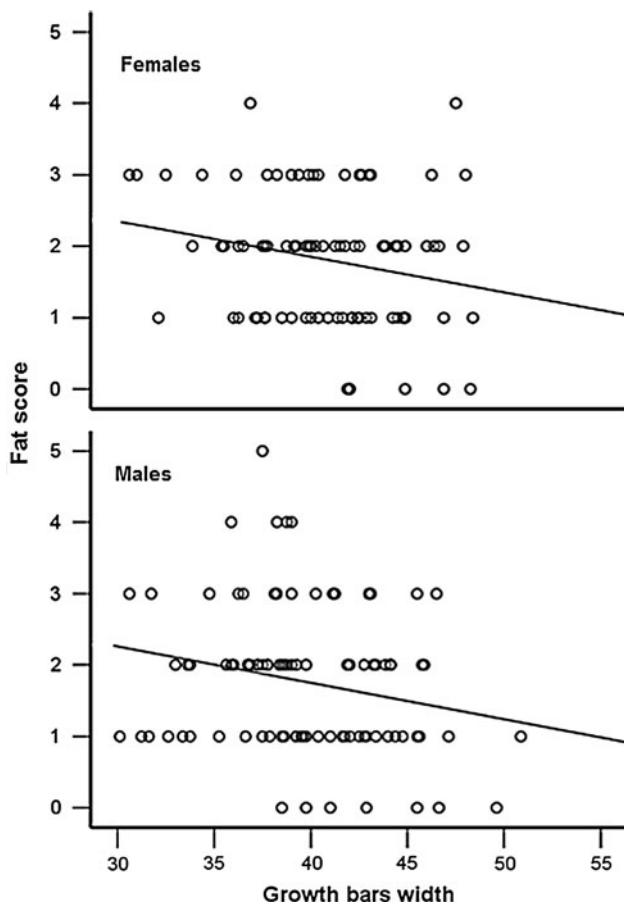
Discussion

In this study of Barn Swallows, we showed that the progress of molt shortly before spring migration does not differ between males and females. The growth rate of primary wing feathers of females was found to be slightly larger than that of males (see also Saino et al. 2012). Individuals whose wing feathers were grown faster completed molt earlier. Finally, molt stage positively covaried with pectoral muscle mass while feather growth rate negatively covaried with subcutaneous fat depots.

Molt stages of wing and tail feathers were positively correlated. If molt completion is a constraint to the start of migration dictated by the need to possess fully functional plumage, Barn Swallows should strive to complete molt at aerodynamically important plumage regions at the same time. General concordance of molt stages may partly reflect gross variation in the start of molt according to geographical origin, as swallows from several western and central European countries, with different timing of annual routines, have been found to winter in the study area (P. Micheloni, unpublished data). Age effects could also partly explain the positive correlation among plumage regions if young start molting later or renew their plumage at a different pace compared to older individuals (Ginn and Melville 1983; Møller et al. 1995, 2011). However, age is unlikely to be the sole mechanism behind the positive correlation among molt scores, as the correlation also persisted among the individuals that had not completed molt and likely were mostly young birds.

Shortly before spring migration, males and females did not differ in molt stage, suggesting no sex difference in the timing of molt completion. This conclusion is also supported by the observation that the frequency of males and females among the birds that had or, respectively, had not completed molt did not differ. Moreover, among birds that were still molting, males were unlikely to catch-up with females and eventually precede them in molt completion because of the larger GBW and shorter wing and tail feathers of females.

Migratory birds often show earlier arrival of males at the breeding grounds, likely as the evolutionary consequence of the fitness benefits that early arriving males accrue in

Fig. 3 Subcutaneous fat store scores, reflecting fat reserves and expressed on an arbitrary 0–5 scale of increasing fat depots, in relation to growth bars width (in mm × 10) recorded on a wing primary feather (P7). The negative relationships were significant in males and marginally non-significant in females

completion of molt does not constrain the start of migration or molt is in fact constraining the start of migration of both sexes and males start migration at the same time as females but migrate faster. In either case, molt seems not to be a determinant of protandry in this species.

An alternative, though much less parsimonious, interpretation is that males left the study area soon after molt completion and were continuously replaced by later molting males arriving from more southern wintering areas, whose influx maintained roost size constant during February. Because the roost included hundreds of thousands of individuals, its composition could not be monitored. However, there are several pieces of evidence that argue against such a sex-biased turnover effect. In the first place, the region where the study was carried out is at the northern limit of the wintering range of the species (Ambrosini et al. 2011). Second, data from geolocators indicate that individual swallows that winter in Nigeria and in neighboring countries tend to stay in the same, relatively small, wintering area at least until mid-February. Hence, in the period when the study was conducted, Barn Swallows seem not to show pre-migratory movements and are not found in regions far north of where the study area was located. Third, in February, the roost retains constant size, and this suggests that either no 'leakage' of individuals occurs or that there is a balance between emigrants that have completed molt and immigrants which should also have completed molt and should therefore not bias the sample. Fourth, the proportion of individuals of either sex among those that had or, respectively, had not completed molt did not differ, and, in addition, among the swallows that were still molting primaries, molt stage did not differ between the sexes, as would be otherwise predicted if either sex advanced molt relative to the other. Fifth, the sex ratio of individuals that we sampled was slightly male-biased and very similar to that recorded during breeding (our unpublished data; Møller 1994), and this suggests no earlier pre-migratory exodus of males compared to females.

Feather growth rate was positively related to molt stage. This is consistent with the expectation, because ptilochronological studies suggest that individuals in prime condition grow their feathers faster. This finding might be partly mediated by age effects. Young individuals during their first wintering in Africa may have lower feather growth rates because they are relatively poor foragers or possess poorer phenotypic quality compared to older individuals, which have passed more selection episodes. The positive relationship between GBW and molt score may thus result from older individuals both molting earlier and having faster feather growth. In a previous study of breeding Barn Swallows, older (≥2 years) females but not males showed larger GBW on a tail feather than yearlings (Saino et al. 2012). Like here, in that study we also measured GBW on

competition for mates and territories (Morbey and Ydenberg 2001; Spottiswoode and Saino 2010). However, the mechanisms behind such sex differences in migration phenology are poorly understood (Rubolini et al. 2004; Saino et al. 2010). Males may advance their arrival relative to females by an earlier start of migration, by migrating faster, by wintering closer to the breeding grounds, or a combination of these mechanisms (Spottiswoode and Saino 2010). If migration schedules are constrained by completion of molt, males could advance the start of migration by molting earlier or faster than females. In this study, the progress of molt in either sex did not differ and males did not show larger GBW than females. In fact, females had marginally non-significantly larger GBW than males, consistent with a previous study at the breeding grounds (Saino et al. 2012). Moreover, because females have shorter wing feathers than males, an analysis correcting for length of the fully grown feather (which was not possible in this study; see "Methods"), would emphasize any sex-related difference. The above findings imply that either

the 7th primary wing feather in a subsample of individuals to test for a relationship between GBW recorded on different plumage regions, which was found to be positive and significant. Hence, any age effect may indeed help in explaining the positive relationship observed between GBW and molt score of P7 among females but no age effect can be invoked for males, unless selection against males with low GBW (or any related trait) is the cause of the lack of age-related variation of GBW at the breeding grounds (Saino et al. 2012).

Interestingly, a comparison between the sex-specific frequency distributions of GBW during wintering and those recorded in spring among breeding individuals in Italy (Saino et al. 2012) shows a difference in the lower 'tail' of the distributions: low ($\leq$36) GBW values that were recorded in a non-negligible fraction of the wintering Barn Swallows were conversely not recorded at all in the breeding population (Fig. 4), yielding significant differences in the two distributions for males (Kolmogorv–Smirnov two-sample test; $P = 0.010$) but not for females ($P = 0.581$). This may suggest that survival selection

occurs during the late wintering or the spring migration period, which purges low-GBW young males that are in poor condition away from the population, or that young males with very low GBW breed only in their third calendar year. This would bias the results, however, only if such males do not happen to be captured at the breeding colonies, for which we have no evidence. These speculations must be taken with extreme caution, however. While ringing data show that part of the Barn Swallows breeding in Italy winter in Nigeria, they also show that Barn Swallows from several other European areas share the same wintering grounds (P. Micheloni, unpublished data). Variation in feather growth rate during wintering of swallows originating from different European regions might thus produce such a spurious result, although any such confounding effect would need to be sex-specific in order to apply. Moreover, the fact that the lower, but not the upper, 'tail' of the GBW distributions differ argues against the existence of such a geographical effect, unless the frequency distributions actually differ in skewness among populations.

Fig. 4 Frequency distribution of growth bar width on a wing primary feather (P7) of males and females during wintering in Nigeria (this study) or during the breeding season in Italy (females: $n = 41$; males: $n = 42$) (see Saino et al. 2012)
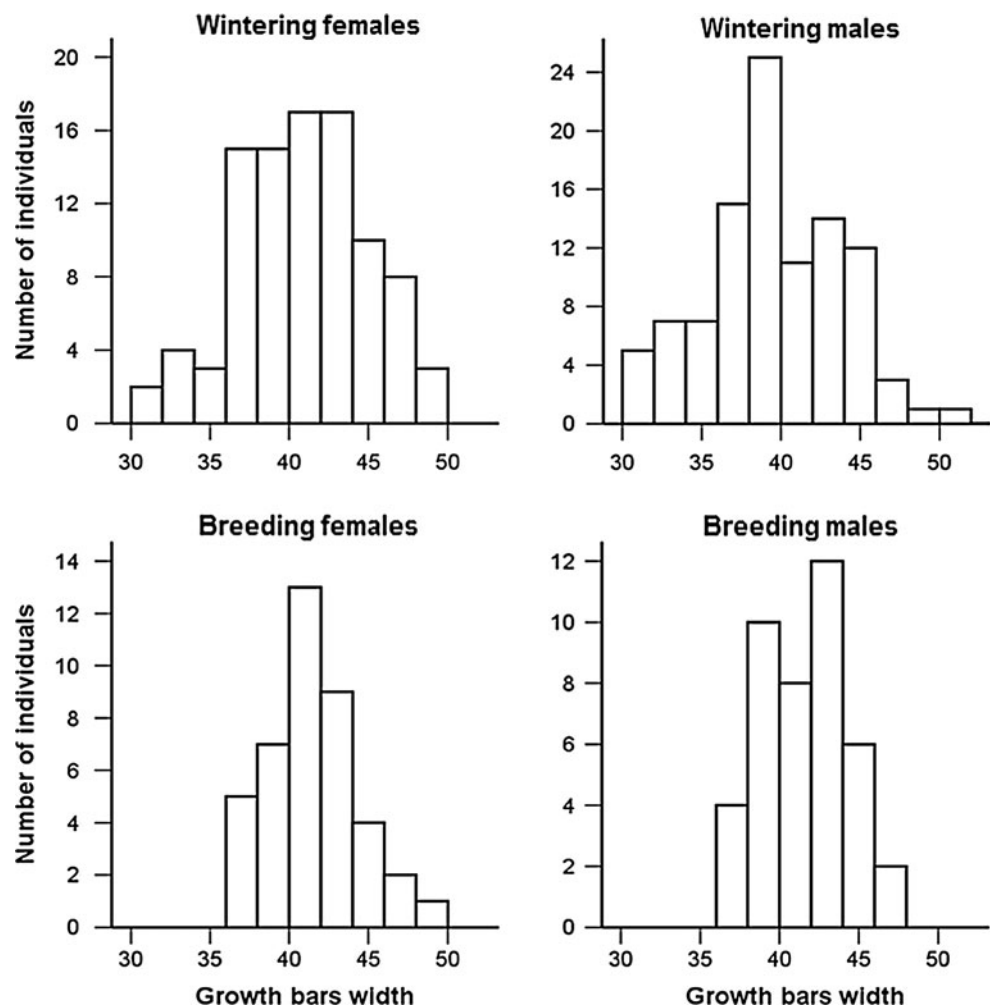
We found evidence for covariation between the progress of molt or GBW on the one hand and two indices of body condition on the other. Although individual relationships could be statistically non-significant, in both sexes pectoral muscle score was positively related to molt stage, whereas GBW was negatively related to fat stores. This finding is consistent with the hypothesis that molting Barn Swallows manage to compensate for the reduced wing area during molt by adaptively building up their pectoral muscles (Holmgren et al. 1993; Lind and Jakobsson 2001), or simply suggests that more exercise causes an effect on muscle mass with no marked consequences for flight performance. Alternatively, since 95 % of feather tissues are made up of proteins, the present findings suggest that individuals that are better at foraging and have access to better food sources can afford both advancing molt and retaining large pectoral muscle mass. Because the relationship between muscle mass and GBW was far from significant, it therefore appears that individuals that were in good nutritional conditions started molting earlier. On the other hand, GBW was inversely related to subcutaneous fat depots, suggesting that energetic demands of rapid feather renewal entail higher fat consumption. The lack of negative relationship between the progress of molt and fuel stores, that was documented in previous studies of post-breeding molt of migratory passerines (Merilä 1997; Lind et al. 2004), is possibly related to the fact that the swallows were in a non-migratory state. In fact, a physiological/energetic trade-off between fuel accumulation and molt may emerge only during active migration when birds show maximal fuel deposition rates (Perez-Tris et al. 2002).

The evidence that individuals with advanced molt stage had larger GBW and were also in better condition is relevant to subsequent sexual selection at the breeding grounds. Early migrating and arriving individuals may accrue benefits in terms of competition for mates or territories. However, arrival date may function as a reliable signal of quality, and choosy individuals may therefore benefit by mating with early arriving mates (Spottiswoode and Saino 2010). Albeit molt completion may not constrain relative migration and arrival schedules of either sex (see above), it may generate variance in migration/arrival schedules within each sex. Because individuals that advanced molt also had relatively large pectoral muscle mass and GBW, which is known to depend on nutritional condition and perhaps resistance to diverse forms of stress (see "Introduction"), only high quality individuals with large GBW may afford completing molt and thus migrating early. Hence, arrival date may reliably reveal physiological conditions during wintering via an effect of physiological condition on feather growth rates and timing of molt completion, and thus potentially mediate adaptive mate choice.

In conclusion, for the first time to the best of our knowledge, we studied sex-related variation in the progress of molt and in feather growth rate in a species where sex may not be unequivocally identified based on external morphology during winter. We found no evidence for sex differences in the timing of molt and evidence for slightly larger pace of feather growth in females. These results imply that timing of molt cannot be at the basis of protandry in Barn Swallows. Individuals in an advanced molt stage also had larger GBW, suggesting that nutritional conditions as reflected by feather growth rates affect the timing and/or speed of molt and thus potentially also the timing of the start of migration. These results are relevant to sexual selection as they are consistent with the assumption of sexual selection models of protandry that arrival date can serve as a reliable signal of individual condition before breeding. The frequency distribution of feather growth rates of males recorded during wintering was found to differ from that recorded in a sample of birds breeding in Italy, as individuals with low GBW were not recorded among breeding birds. Selection on feather growth rates may be responsible for a directional shift in frequency distribution because individuals with low GBW are expected to suffer higher mortality. In general, our study highlights the largely underexploited potential of ptilochronological approaches in the study of the evolution and expression of variation in molt strategies and circannual routines.

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