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Sexual dimorphism in the shape of the primary wing and tail feathers of the sky blue parakeet (budgerigar), *Melopsittacus undulatus* (Shaw, 1805) (Psittaciformes: Psittacidae)

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ABSTRACT

Difference between sexes was quantified in this study using outline basedgeometric morphometric analysis on the shape of the primary wing and tail feathers of the sky blue strain of parakeet bird *Melopsittacus undulatus* (Shaw, 1805). Results of Discriminant Function Analysis (DFA) and Principal Component Analysis (PCA) utilizing the coefficients derived from Elliptic Fourier Analysis (EFA) consistently showed that sexual dimorphism is present in the said species with differences primarily explained by the variations in the tip and outline of the flight feathers. Females generally have slightly rounded tip and partially curved (inward) feather outline while males have feathers with slightly pointed tip and more or less straight feather outline. However, no difference between sexes was observed in the shape of tail feathers. It is suggested that based from this study, a more complete understanding and proper identification on the biological basis of the observed wing shape differences are needed which could be elemental in the proper and effective conservation of this species.

INTRODUCTION

In birds, sexual dimorphism, the difference in morphology between male and female members of the same species (Klappenbach, 2010) can be manifested in size or plumage differences (Andersson, 2010). In most cases when size differences exist between the male and female of a species, it is the male that is the larger of the two sexes (Klappenbach, 2010). And in plumage dimorphism, males are typically more ornamented or brightly colored than females (Mcgraw *et al.*, 2002). In the case of the sky blue Parakeet birds, *Melopsittacus undulatus* (Shaw, 1805), sexual dimorphism is primarily identified by the color of their cere - fleshy part just above their beak which becomes more intense when the birds are sexually active. Male varieties have blue or purplish cere while females develop a white, tan, or brownish cere (Womach and Womach, 2010). However, new approach of determining sexual dimorphism in Parakeet birds is now available through the use of various statistical tools particularly outline-based geometric morphometric analysis (Richtsmeier *et al.*, 2002).

Geometric morphometrics is a collection of approaches for the multivariate statistical analysis of Cartesian coordinate data, usually (but not always) limited to landmark point locations. More generally, it is the class of morphometric methods that preserve complete information about the relative spatial arrangements of the data throughout an analysis. As such, these methods allow for the visualization of group

Keywords: sexual dimorphism, discriminant function analysis, elliptic Fourier analysis, principal components analysis.

and individual differences, sample variation, and other results in the space of the original specimens (Slice *et al.*, 2002).

To date, there have been no published articles studying presence or absence of sexual dimorphism in the sky blue Parakeet, *M. undulatus*, using outline-based geometric morphometric analysis. Thus, this study is conducted to determine presence of sexual dimorphism in the shape of the feathers in the left and right wing, as well as in the tail region of the Parakeet bird, *M. undulatus*, using various statistical/geometric morphometric tools from Thin-plate Spline (TPS) series (Bookstein, 1991) and Paleontological Statistical (PAST) software (Hammer *et al.*, 2001). Particularly, variations in the outline of the primary feathers from selected region between male and female Parakeet birds are being quantified in this study. Proper evaluation and identification of the biological and physiological meanings of the results would be helpful in assessing the flight mechanism, feeding habit, mating selection, and probably conservation of Parakeet birds.

MATERIALS AND METHODS

Collecting of bird samples was done by procuring a pair of Parakeet bird – one male and one female. Birds were visually sexed according to their cere (Fig. 1) – a fleshy bit above their beak, blue cere for males and white or brownish cere for females (Amaral, 2010). Bird samples were then taken to the laboratory and removal of the feathers followed. Only the primary feathers from both the left and right wings, as well as the feathers from the tail, were removed. The feather samples were fixed or arranged in such a way that the outlines are clearly defined, and after which, samples were scanned under 600-dpi resolution. Prior to outlining, a tps file was made for scanned feathers per region using the TPS software tpsUtil. Scanned feather images were then outlined using another type of TPS software called tpsDig. One hundred (100) curve points were used to outline the contour of the feathers. Outlined feathers were subjected to various analyses in the PAST software which include Elliptic Fourier Shape Analysis (EFA), Principal Component Analysis (PCA), and Discriminant Function Analysis (DFA). Scores (Eigen values) obtained from the analysis of the outline of the feathers using Elliptic Fourier Shape Analysis were used as the raw data for DFA and PCA. Sufficient numbers of 100 digitized points were used to generate x and y coordinates.



Fig.1: Photograph of the sky blue female (A) and male (B) Parakeet bird, *Melopsittacus undulatus* (Shaw, 1805) showing differences in the color of the cere (encircled part) designating whitish for female and blue for male (*Source: www.upatsix.comchatsbudgietopic74556.html*)

These coordinates were then arranged alternately in column. Coefficients or scores produced were copied to the main PAST spreadsheet for further analysis – DFA and PCA. In DFA, presence of sexual dimorphism was analyzed using the Discriminant scores. Percentage of correct classification equal or higher than the cut-off percentage score which is 75 was considered to be significant indicating that the two data sets being tested are sexually dimorphic. For PCA, Eigen values were analyzed producing principal component scores which account specific percentages of the total variance, and a Joliffe cut-off value which is the main basis for assessing each principal component scores as significant or not. Principal component scores greater than the Joliffe cut-off value are considered to be significant.

RESULTS

Presence of sexual dimorphism in the shape of the primary feathers of the sky blue Parakeet, *M. undulatus*, was determined using series of tests/analysis via PAST software. Coefficients obtained from the Elliptic Fourier Shape Analysis were subjected to DFA and PCA which warrant presence of sexual dimorphism in the Parakeet bird.

Discriminant Function Analysis was used not only to determine equality of the means of the two groups but also to reclassify specimens to previously defined groups. This analysis is a standard method for visually confirming or rejecting the hypothesis that two groups are morphologically distinct (Hammer et al., 2001). Two groups of multivariate, marked with different colors were used and plotted along x-axis using a histogram. Assessing figure (2), the two histograms (blue- and red-colored bins) are not overlapping each other. This implies that there is a complete separation between two data sets - male and female feathers, which warrants that sexual dimorphism is present in the Parakeet birds with respect to the primary feathers collected from the left wing region. To validate further, table (1) is presented below. The table shows the reclassification of the discriminant scores of the left wing feathers between sexes. Original count or number of feather samples was tabulated to compute the percentage of reclassification of group members. As seen in the table, all of the left wing feathers samples in the female group were correctly reclassified to the predicted female group membership. With regards to the male feather samples, same result was obtained wherein 100% of the original left wing feather was correctly reclassified to the predicted male group membership. Taken as a whole, the discriminant function analysis via the discriminant scores of the left wing feather samples between sexes, and as seen in the table, show that 100% of the original group cases are correctly classified.



Fig. 2: Frequency histograms of the female left wing feathers (red bins) and male left wing feathers (blue bins) showing presence of sexual dimorphism as shown by the complete separation of the blue and red bins.

blue I drakeet, M. unaututus (Shaw, 1865).							
COUNT	SEX	PREDICTED GROUP MEMBERSHIP					
		Female	Male	Total			
Original Count	Female	20	0	20			
	Male	0	20	20			
Percentage	Female	100	0	100			
	Male	0	100	100			

Table 1: Reclassification of the male and female feathers collected from the left wing region of the sky blue Parakeet, *M. undulatus* (Shaw, 1805).

100 % of original group cases correctly classified.

In fig. (3), it can be observed that the two colored bins overlap at some point implying no complete separation of the data sets. However, this result does not show that, sexual dimorphism is absent in the primary right wing feathers of the Parakeet birds, *M. undulatus*. We still have to check whether percentage of correctly classified group cases is higher than the 75 (the cut off value). To do so, table (2) is presented to show the reclassification of the right wing feathers in male and female Parakeet birds, *M. undulatus*. Assessing the female feather samples, out of twenty (20) total samples, nineteen (19) samples were correctly reclassified to the female group while the remaining one (1) feather sample was classified to the male group. However, if we have to assess the male group membership, all of the male feather samples (100 %) were correctly reclassification (that is by getting the ratio of the total number of feathers - both in male and female group cases, that are correctly classified and the total number feather samples), we get a value of 97.37 % which is higher than the cut off score/value (70%). Thus, this result indicates that, there is sexual dimorphism in Parakeet bird, *M. undulatus* with respect to the primary feathers in the right wing region.



Fig. 3: Frequency histograms of the female right wing feathers (red bins) and male right wing feathers (blue bins) showing presence of sexual dimorphism as shown by the separation of the blue and red bins.

Table 2:	Reclassification	of th	e male	and	female	feathers	collected	from	the	right	wing	region	of 1	the
	sky blue Parake	et, M	undul	atus	(Shaw,	1805).								

2	,					
COUNT	SEX	PREDICTED GROUP MEMBERSHIP				
		Female	Male	Total		
Original Count	Female	19	1	20		
	Male	0	18	18		
Percentage	Female	95	5	100		
	Male	0	100	100		

97.37 % of original group cases correctly classified.

Also, complete separation of the two data sets can be observed in figure (4). There is no overlap of the two colored bins which implies variation in the tail feathers between sexes of the Parakeet birds. The result is a good indication of presence of sexual dimorphism in the said species of bird with respect to the tail feathers. To validate the tentative result in the histogram presented in figure (4) as evident by the complete separation of the two groups of colored bins, table (5) is presented. The table shows the reclassification of the male and female tail feathers. For the female samples, all of the twelve (12) feather samples were correctly reclassified to the female predicted group membership. On the other hand, for the male group, out of eleven (11) feather samples, only one (1) feather is classified to the female group while the remaining ten (10) samples were correctly reclassified to the male predicted yields 95.6 % of correctly classified group cases which is higher than the cut-off score of 75 %. Thus, for the tail feathers between sexes of Parakeet bird, sexual dimorphism is present.





Table 3: Reclassification of the male and female feathers collected from the tail region of the sky blue Parakeet bird, *M. undulatus* (Shaw, 1805).

COUNT	SEX	PREDICTED GROUP MEMBERSHIP				
		Female	Male	Total		
Original Count	Female	12	0	12		
	Male	1	10	11		
Percentage	Female	100	0	100		
	Male	9	91	100		

95.65 % of original group cases correctly classified.

PCA was employed to effectively summarize information of the variations restrained in the coefficients which were obtained using the EFA. Thus, PCA is performed based on the variance-covariance of the coefficients (Torres *et al.*, 2008). Results for this analysis are presented in figure (5), wherein differences between sexes of *M. undulatus* in the mean shape of the primary tail and wing (left and right) feathers are shown as shape deformations. Consistent across three regions, a total of three significant principal components which account for the total feather shape variance were observed (Tables 4-6). Among these three significant principal

components, PC1 accounts the greatest percentage of all the variance both in male and female. Variations in these principal components primarily explain differences in the tip and outline of the feathers between sexes. For both the left and right wing feathers, females have slightly rounded tip and a more slightly curved inward outline, while males have slightly pointed and more or less straight feather outline respectively. With respect to the tail feathers, females tend to have greatly pointed tip while males have slightly pointed tip. However, in the outline of the tail feathers, both sexes generally have straight outline.



Fig.5: Differences between sexes in the mean shape of the primary feathers of the left wing (A, B), right wing (C, D) and the tail (E, F) of the sky blue Parakeet, *Melopsittacus undulatus* (Shaw, 1805).

Table 4. P	roportion	of variation	of the sha	pe of the	feathers of	of the le	eft wing.

	EIGE	N VALUES		PERCENTAGE	
PRINCIPAL COMPONENT (PC)	(must be greater than John	REMARKS	OF VARIANCE		
	Female	Male	1021/11/11/11/11	Female	Mala
	(Joliffe cut-off: 11303)	(Joliffe cut-off: 12473)			Male
PC1	2565700	3404100	significant	79.4%	95.5%
PC2	558587	79365	significant	17.3%	2.2%
PC3	81452	50180	significant	2.5%	1.4%

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1 auto 5.110	DOLUOII OI	variation	or the	snabe	or une	reactions	or the	112III	wing.
								<i>G</i> .	

EIGEN VALUES PRINCIPAL COMPONENT (must be greater than Joliffe cut-off value to be significant)			DEMADUS	PERCENTAGE OF VARIANCE		
(PC)	Female (Joliffe cut-off: 10167)	Male (Joliffe cut-off: 11904)	- REMARKS	Female	Male	
PC1	2406830	3147290	significant	82.9%	92.5%	
PC2	417046	186695	significant	14.4%	5.5%	
PC3	68725	45478	significant	2.4%	1.3%	

Table 6: Proportion of variation of the shape of the tail feathers.

PRINCIPAL COMPONENT (PC)	EIGEN (must be greater than J signi	REMARKS	PER V	CENTAGE OF 'ARIANCE	
	Female (Joliffe cut-off: 10167)	Male (Joliffe cut-off: 11904)	_	Female	Male
PC1	2946060	7484220	significant	89.0%	92.4%
PC2	267834	194725	significant	8.1%	2.5%
PC3	77685	146560	significant	2.3%	1.9%

DISCUSSION

Variations in the tip and the outline of the wing and tail feathers between sexes of *M. undulatus* could be attributed to several factors like mating system and parental care, flight mechanism, competition for food, and other factors. Differences between sexes have traditionally been explained by sexual selection, they may also result from natural selection favoring sex differences associated with other aspects of species ecology, such as effective resource partitioning (Darwin, 1871; Shine, 1989; Radford and du Plessis, 2003). Variations in the extent of sexual dimorphism among bird species are traditionally attributed to differences in social mating system and patterns of parental care (Owens and Hartley, 1998; Butcher and Rohwer, 1988; Andersson, 2010). Particularly, when freed from the burden of parental care, higher potential reproductive rates in males (Trivers, 1972) may lead to intense male-male completion for access to females and the elaboration of secondary sexual characters as well as size dimorphism (Trivers, 1972; Thomas *et al.*, 2006). Also sexual dimorphism is to some extent the result of intersexual competition for food.

One could expect to find the degree of dimorphism to vary within wide limits, even among closely related species, as somewhat different factors to be involved in every case (Ingolfsson, 1969). According to Tinbergen (1953), securing food for young will often require much flight, and hence sexual dimorphism may have a functional influence on flight mechanism in birds (Shaffer *et al.*, 2001; Freed *et al.*, 2009). Ecological stresses such as habitat destruction or eradication might have caused this variation since illegal logging is very rampant nowadays which consequently destroys a lot of bird breeding areas and habitats (Ingolfsson, 1969).

CONCLUSION

The results were consistent in showing significant variations in the shape/outline of the primary feathers collected from the selected bird regions – left and right wing, and the tail, thus consequently implying that sexual dimorphism does occur in the Parakeet, *M. undulatus*, bird species as manifested by the separation of two histograms (though some overlap at some point), the high percentages of principal component variances and reclassification, and the variations in the shape deformations of the feathers. Variations in the feather between sexes are explained by the differences in the tip and outline with females having slightly rounded tip and curved (inward) outline, while males having slightly pointed tip and straight feather outline. And these observed variations could be attributed to natural selection associated with the aspects of mating system, feeding habit, and flight mechanism of the bird.

A lot of other factors, such as evolutionary, genetic or ecological factors might also cause sexual dimorphism. To ascertain these factors, however, more specific and thorough studies are needed. Although the obtained results were only preliminary and exploratory, they have demonstrated that presence of sexual dimorphism in birds could be detected by geometric morphometric analysis. A complete understanding and proper identification of the biological basis of the observed presence of sexual dimorphism is elemental for the effective conservation of the species. Furthermore, the consistency of morphometric analysis in providing significant results indicates that it can become an important tool in taxonomic studies and in studying morphological variations of other biological forms.

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