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Comparative Morphology of the Avian Cerebellum: I. Degree of Foliation

Andrew N. Iwaniuk[a] Peter L. Hurd[a] Douglas R.W. Wylie[a, b]

[a]Department of Psychology, [b]Centre for Neuroscience, University of Alberta, Edmonton, Canada

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Abstract
Despite the conservative circuitry of the cerebellum, there is considerable variation in the shape of the cerebellum among vertebrates. One aspect of cerebellar morphology that is of particular interest is the degree of folding, or foliation, of the cerebellum and its functional significance. Here, we present the first comprehensive analysis of variation in cerebellar foliation in birds with the aim of determining the effects that allometry, phylogeny and development have on species differences in the degree of cerebellar foliation. Using both conventional and phylogenetically based statistics, we assess the effects of these variables on cerebellar foliation among 91 species of birds. Overall, our results indicate that allometry exerts the strongest effect and accounts for more than half of the interspecific variation in cerebellar foliation. In addition, we detected a significant phylogenetic effect. A comparison among orders revealed that several groups, corvids, parrots and seabirds, have significantly more foliated cerebella than other groups, after accounting for allometric effects. Lastly, developmental mode was weakly correlated with relative cerebellar foliation, but incubation period and fledging age were not. From our analyses, we conclude that allometric and phylogenetic effects exert the strongest effects and developmental mode a weak effect on avian cerebellar foliation. The phylogenetic distribution of highly foliated cerebella also suggests that cognitive and/or behavioral differences play a role in the evolution of the cerebellum.

Introduction

The neural circuitry of the cerebellum is remarkably conserved among vertebrates [Voogd and Glickstein, 1998], but this belies profound variation in the relative size and shape of the cerebellum [Larsell, 1967; Pearson and Pearson, 1976; Butler and Hodos, 1996]. One major difference in cerebellar morphology among the major clades of vertebrates is the degree of folding or foliation of the cerebellar cortex. At one end of the spectrum are frogs and non-avian reptiles that have a curved cerebellum, but no actual folds. At the opposite end of this spectrum are rays, mormyrids, birds and mammals that all possess cerebella with numerous folds. Lying in between these two extremes are species with a few simple folds, such as lungfish and most ray-finned and cartilaginous fish. The differences in the degree of foliation among these vertebrates are attributed to behavioral specializations, such as electroreception in mormyrids, and the evolution of increasingly complex motor behaviors in birds and mammals [Butler and Hodos, 1996]. Although this might explain large variations in cerebellar morphology, such as the dif-

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Andrew N. Iwaniuk
Department of Psychology
University of Alberta
Edmonton, Alberta, T6G 2E9 (Canada)
Tel. +1 780 492 7239, Fax +1 780 492 1768, E-Mail brainsize@yahoo.ca

ference between avian and non-avian reptiles, it remains unclear what behaviors or other factors are correlated with differences in the degree of foliation within vertebrate classes.

In birds, the cerebellum is complexly folded in all species, but the degree of foliation varies dramatically. For example, owls, chicken-like birds (i.e., Galliformes) and pigeons all possess the same number of folia [Senglaub, 1963; Larsell, 1967], despite the variation in body size, brain size and brain composition (i.e., volume of different brain regions) among the three groups. Similarly, corvids, raptors, seabirds, penguins and parrots all appear to have more complexly folded cerebella than other species [Senglaub, 1963; Larsell, 1967] despite marked differences in brain size and composition among these groups. Senglaub [1963] and others [Pearson and Pearson, 1976] suggested that variations in the degree of cerebellar foliation reflect body size in birds; larger birds have more and deeper folds in their cerebella. There are, however, several additional factors that might also influence the degree of foliation of the avian cerebellum. From an allometric perspective, it is possible that the degree of foliation is also correlated with the relative size of the brain and the cerebellum. In primates, the gyrification index, a measure of the degree of isocortical folding, is correlated with brain and isocortical volumes [Zilles et al., 1989; Striedter, 2004]. Therefore, it is reasonable to suggest that a similar correlation between cerebellar foliation and the relative size of the brain and the cerebellum is present in birds. Many authors have also emphasized the importance of hatchling developmental mode on the evolution of the brain in birds [Portmann, 1946; Bennett and Harvey, 1985; Iwaniuk and Nelson, 2003]. Specifically, the relative size of the brain [Portmann, 1946; Bennett and Harvey, 1985; Iwaniuk and Nelson, 2003], and some brain regions [Portmann, 1947; Bennett and Harvey, 1985], are correlated with developmental differences among species such that the longer it takes for a species to develop, the larger its brain or brain region. These same developmental 'constraints' might also affect the degree of foliation of the cerebellum.

Here we present a comprehensive analysis of the degree of foliation in birds and how it relates to allometry, phylogeny and development. Based on previous evidence [Senglaub, 1963], we predicted that higher degrees of foliation would be present in larger birds with relatively large cerebella. Due to the strong correlation between developmental differences and relative brain size in birds [Iwaniuk and Nelson, 2003], we also predicted that altricial species would have higher degrees of foliation than precocial species and that longer periods of embryonic and post-embryonic development will be positively correlated with the degree of foliation. Using both conventional and phylogenetically based statistics, we tested these hypotheses in a large comparative data set.

Materials and Methods

Specimens

The brains of several species were obtained from wildlife sanctuaries and veterinary clinics in Australia. Other researchers provided specimens and several were loaned to us from the Bishop Museum (Honolulu, Hawaii), Field Museum of Natural History (Chicago, Ill.) and the National Museum of Natural History (Washington, D.C.) (table 1). For all species, the brains were extracted from the skull, the meninges removed and the brain weighed to the nearest milligram with an electronic balance. All birds that we collected from wildlife sanctuaries, veterinary clinics and other researchers were immersion fixed in 10% buffered formalin or 4% paraformaldehyde. The museum specimens were also immersion fixed in 10% buffered formalin, but following adequate fixation, they were kept in 70% ethanol that was replaced on a regular basis. The museum specimens that were loaned to us were stored in 70% ethanol for between 2 and 68 years. To equilibrate the tissue, the museum specimens were placed in 4% paraformaldehyde in 0.1 M phosphate buffer (pH = 7.4) for several days prior to processing. Despite this marked difference in processing between the museum and other specimens, the measurements taken from the museum specimens did not appear different from related species obtained from other sources. In fact, there was no significant difference in relative or absolute CFIs (for both: Wilcoxon $Z = 0.44$, $n = 6$, $p = 0.66$). Similarly, excluding the museum specimens from our analyses did not qualitatively alter any of the results (i.e., p values were consistently significant or non-significant).

The brains were bisected in the sagittal plane and the cerebellum was detached by cutting through the cerebellar peduncle. The cerebella were then placed in 30% sucrose in 0.1 M phosphate buffer until they sank. The cerebella were subsequently embedded in gelatin, post-fixed in 30% sucrose paraformaldehyde and sectioned in the sagittal plane on a freezing stage microtome. We collected 40-μm-thick sections in 0.1 M phosphate-buffered saline and these were mounted onto gelatinized slides. After drying, the slides were stained with thionin, dehydrated through a graded ethanol series, cleared in Hemo-D and coverslipped with Permount. This procedure does not result in any significant tissue shrinkage (A.N. Iwaniuk, unpubl. data).

We supplemented our specimens by also including midsagittal sections shown in Larsell [1967], Senglaub [1963] and Matochik et al. [1991]. This enabled us to include some orders that we were not able to survey (e.g., Piciformes, ratites) as well as a larger number of species for orders that we had obtained only a limited number of species (e.g., Falconiformes, Anseriformes, Galliformes).

Measurements

To measure the degree of foliation of the cerebellum, we used the same approach that previous studies had used to examine gyrification of the mammalian isocortex [Hofman, 1985; Zilles et al., 1989]. First we measured the total length of the Purkinje cell layer along the rostro-caudal extent of the cerebellum across

Table 1. A list of the species surveyed and their respective sample sizes (n), body masses (g), brain volumes (mm^3), cerebellar (Cb) volumes (mm^3) and cerebellar foliation indices (CFI). The taxonomy broadly follows that of Monroe and Sibley [1997], with the exception that some parvorders and infraorders were differentiated at the order level

Order	Family	Genus	Species	n	Body mass	Brain volume	Cb volume	CFI	Source
Anseriformes	Anatidae	Mallard	*Anas platyrhynchos*	1	1,111	5,440	756.08	4.0788	This study, 1, 2
		Long-tailed duck	*Clangula hyemalis*	?	911	4,875	627.41	3.1148	2, 3
		White-winged scoter	*Melanitta fusca*	?	1,896	7,138	830.12	3.6081	2, 3
		Black scoter	*Melanitta nigra*	?	1,191	5,516	670.85	3.5387	2, 3
Apodiformes	Apodidae	Common swift	*Apus apus*	1	38	642	106.33	3.3383	This study, 1
		Glossy swiftlet	*Collocalia esculenta*	2	5	121	28.51	3.2431	This study
Apterygiformes	Apterygidae	Brown kiwi	*Apteryx australis*	?	2,120	11,300*	–	3.8957	This study, 1
Caprimulgiformes	Aegothelidae	Feline owlet-nightjar	*Aegotheles insignis*	1	71	1,540	242.40	3.6729	This study (BBM-NG 101365)
	Caprimulgidae	Spotted nightjar	*Eurostopodus argus*	1	121	1,013	135.52	2.9491	This study
		Parauque	*Nyctidromus albicollis*	1	53	910	200.56	3.2389	This study (USNM 504211)
	Nyctibiidae	Grey potoo	*Nyctibius griseus*	2	257	1,980	300.50	3.2389	This study (USNM 504185, 504184)
	Podargidae	Tawny frogmouth	*Podargus strigoides*	2	387	5,759	445.21	3.3850	This study
	Steatornithidae	Oilbird	*Steatornis caripensis*	1	414	3,900	586.29	3.1297	This study (USNM 431365)
Charadriiformes	Charadriidae	Northern lapwing	*Vanellus vanellus*	?	200	2,131	312.57	3.2934	1, 4
	Laridae	Common black-headed gull	*Larus ridibundus*	?	250	2,714	474.23	3.9148	1, 4
		Mew Gull	*Larus canus*	?	404	4,100	–	4.1751	2, 5
		Silver gull	*Larus novaehollandiae*	1	292	2,941	445.70	4.2401	This study
		Herring gull	*Larus argentatus*	?	1,000	4,312	663.80	4.4696	2, 4
		Arctic Tern	*Sterna paradisaea*	?	110	2,000	–	3.4888	2, 5
	Scolopacidae	Common sandpiper	*Actictis hypoleucos*	?	47	746	98.57	3.3815	2, 4
		Short-billed dowitcher	*Limnodromus griseus*	1	109	1,338	127.06	3.3926	This study
		Red-necked phalarope	*Phalaropus lobatus*	?	35.3	450	–	3.1285	2, 5
		Eurasian woodcock	*Scolopax rusticola*	?	290	2,503	313.20	3.8149	2, 4
Ciconiiformes	Ardeidae	Cattle egret	*Bubulcus ibis*	1	366	4,025	382.07	4.2061	This study
Columbiformes	Columbidae	Wood pigeon	*Columba palumbus*	?	450	2,315	337.71	3.6127	1, 4
		Peaceful dove	*Geopelia placida*	1	47	776.1	88.91	2.9451	This study
		Bush bronzewing	*Phaps elegans*	1	205	1,517.4	176.22	3.1237	This study
		Superb fruit-pigeon	*Ptilinopus superbus*	1	104	1,052	150.47	2.9729	This study
		African collared-dove	*Streptopelia roseogrisea*	?	155	1,100	–	3.2656	2, 5
Coraciiformes	Cerylidae	Belted kingfisher	*Ceryle alcyon*	1	148	1,606.27	184.21	3.5463	This study (USNM 430744)
	Dacelonidae	Laughing kookaburra	*Dacelo novaeguineae*	1	335	3,970	407.52	3.5214	This study
Falconiformes	Accipitridae	Brown goshawk	*Accipiter fasciatus*	1	403	4,631	634.91	4.2636	This study
		Wedge-tailed eagle	*Aquila audax*	1	3,350	15,997	1,850.45	4.7077	This study
		Common buzzard	*Buteo buteo*	?	900	8,452	1,169.15	4.3031	2, 4
		Bald eagle	*Haliaeetus leucocephalus*	?	4,419	18,040	–	4.3999	1, 5
		White-bellied sea eagle	*Haliaeetus leucogaster*	1	3,004	12,541	1,376.11	4.6655	This study
	Falconidae	Brown falcon	*Falco berigora*	1	562	6,032	631.60	3.8825	This study
		Common sparrowhawk	*Falco tinnunculus*	?	230	3,543	444.90	3.9325	2, 4
Galliformes	Phasianidae	Ruffed grouse	*Bonasa umbellus*	2	650	3,136	268.39	3.9399	This study
		Blue grouse	*Dendragapus obscurus*	?	1,010	3,070	–	4.0670	1, 5
		Turkey	*Meleagris gallopavo*	?	9,839	6,781	1,023.43	3.7991	2, 6
		Grey partridge	*Perdix perdix*	?	401	1,849	223.34	3.4847	2, 7
		Ring-necked pheasant	*Phasianus colchicus*	?	1,133	3,865	480.75	4.2058	2, 7
Gruiformes	Otidiidae	Australian bustard	*Ardeotis australis*	1	4,450	10,501	1,072.20	4.6750	This study
	Rallidae	American coot	*Fulica americana*	1	651	2,719	247.33	3.2863	This study

Table 1 (continued)

Order	Family	Genus	Species	n	Body mass	Brain volume	Cb volume	CFI	Source
Passeriformes	Bombycillidae	Bohemian waxwing	*Bombycilla garrulus*	?	55.5	1,102	140.15	3.2916	2, 4
	Corvidae	Common raven	*Corvus corax*	?	1,175	14,648	1,112.80	4.8274	2, 7
		Carrion crow	*Corvus corone*	?	537	9,382	753.06	4.6097	2, 4
		Little raven	*Corvus mellori*	1	675	9,834	797.37	5.0743	This study
		Jackdaw	*Corvus monedula*	?	200	4,593	382.03	4.3009	2, 4
		Eurasian jay	*Garrulus glandarius*	?	139	3,806	337.24	3.9679	2, 7
		Australian magpie	*Gymnorhina tibicen*	1	314	5,665	483.27	4.9232	This study
	Hirundidae	Barn swallow	*Hirundo rustica*	?	19	531	79.29	3.2841	2, 4
	Menuridae	Superb lyrebird	*Menura novaehollandiae*	1	644	10,163	801.58	4.2927	This study
	Muscicapidae	European robin	*Erithacus rubecula*	?	16.2	592	73.93	3.1841	2, 4
		European blackbird	*Turdus merula*	?	95	1,745	187.27	3.4260	2, 4
	Pardalotidae	Brown thornbill	*Acanthiza pusilla*	1	6	434	35.24	3.1428	This study
	Paridae	Great tit	*Parus major*	?	17.5	877	75.75	3.1619	2, 4
	Passeridae	Tree pipit	*Anthus trivialis*	?	18.4	600	–	3.0474	2, 5
		Gouldian finch	*Erythrura goulidae*	1	10	428	44.19	3.2431	This study
Pelecaniformes	Pelecanidae	Australian pelican	*Pelecanus conspicillatus*	1	5,850	24,880	1,357.75	4.8202	This study
Phoenicopteriformes	Phoenicopteridae	Greater flamingo	*Phoenicopterus ruber*	?	3,000	10,674	1,765.69	4.5568	2, 4
Piciformes	Picidae	Great spotted woodpecker	*Dendrocopos major*	?	80	2,609	270.12	3.3196	2, 4
		Eurasian green woodpecker	*Picus viridus*	?	200	4,232	404.35	3.9216	2, 4
Procellariiformes	Diomedeidae	Black-browed albatross	*Diomedea melanophris*	1	3,388	14,129	2,494.41	5.5338	This study
	Procellariidae	Northern fulmar	*Fulmarus glacialis*	3	544	6,330	–	4.2514	5, 8
		Short-tailed shearwater	*Puffinus tenuirostris*	1	490	4,758	911.80	4.3922	This study
Psittaciformes	Cacatuidae	Sulphur-crested cockatoo	*Cacatua galerita*	1	765	13,933	1,064.72	5.3408	This study
		Galah	*Cacatua roseicapilla*	1	355	7,456	521.91	4.8683	This study
		Long-billed corella	*Cacatua tenuirostris*	1	580	13,103	891.95	5.4016	This study
		Cockatiel	*Nymphicus hollandicus*	1	92	2,161	214.16	3.6187	This study
	Psittacidae	Masked lovebird	*Agapornis personata*	?	52.5	2,824	242.58	3.7498	1, 9
		Australian king parrot	*Alisterus scapularis*	1	160.4	4,902	412.54	4.3019	This study
		Green-winged macaw	*Ara chloroptera*	?	1,430	23,497	1,855.60	4.8904	1, 4
		Purple-crowned lorikeet	*Glossopsitta porphyrocephala*	1	37	1,855	164.46	3.8303	This study
		Budgerigar	*Melopsittacus undulatus*	1	43	1,487	166.36	3.9528	This study
		Crimson rosella	*Platycercus elegans*	1	129	3,628	295.00	4.2206	This study
Sphenisciformes	Spheniscidae	Little penguin	*Eudyptula minor*	1	715	7,584	1,282.29	4.9303	This study
Strigiformes	Strigidae	Saw-whet owl	*Aegolius acadicus*	1	86	2,857	214.64	3.5963	This study
		Short-eared owl	*Asio flammeus*	?	310	5,300	–	3.7698	2, 5
		Long-eared owl	*Asio otus*	?	250	5,321	421.23	3.8359	2, 4
		Great-horned owl	*Bubo virginianus*	?	1,416	14,730	–	3.5794	1, 5
		Southern boobook owl	*Ninox boobook*	1	231	6,339	492.00	3.5581	This study
	Tytonidae	Barn owl	*Tyto alba*	1?	290	5,857	444.12	3.8520	This study, 1, 4
Trochiliformes	Trochilidae	Green-fronted lancebill	*Doryfera ludoviciae*	1	6	139	27.42	3.0386	This study (FMNH 320498)
		Buff-tailed sicklebill	*Eutoxeres condamini*	2	9	257	41.53	2.9549	This study (FMNH 315304, 315300)
		Rufous-breasted hermit	*Glaucis hirsuta*	1	7	123	18.65	2.9638	This study (USNM 616825)
		Hummingbird	*Lampornis sp.*	?	6	200	–	3.0341	1, 5
		Green-backed firecrown	*Sephanoides sephanoides*	2	5	134	18.58	3.1133	This study (FMNH 316784, 316786)
Struthioniformes	Rheidae	Greater rhea	*Rhea americana*	?	25,000	19,228	2,973.89	4.5948	1, 10
	Struthionidae	Ostrich	*Struthio camelus*	?	90,000	39,631	5,844.31	5.3096	1, 2, 4

'?' indicate sample sizes that are unknown because the data was derived from the literature. Where data was obtained from the literature or a museum specimen, the sources of this information are provided.

BBM = Bernice Bishop Museum (Honolulu, Hawaii); FMNH = Field Museum of Natural History (Chicago, Ill.); USNM = National Museum of Natural History (Washington, D.C.). 1 = Senglaub [1963]; 2 = Larsell [1967]; 3 = Kalisinska [2005]; 4 = Portmann [1947]; 5 = Iwaniuk and Nelson [2003]; 6 = Ebinger and Röhrs [1995]; 7 = Rehkämper et al. [1991]; 8 = Matochik et al. [1991]; 9 = Iwaniuk et al. [2005]; 10 = Boire and Baron [1994].

* Note that only the brain volume measurement was provided by previously unpublished data collected by the authors whereas the CFI was measured from an illustration in Larsell [1967].

all sections (fig. 1). We then measured the length of the envelope, which is essentially the surface of the Purkinje cell layer without counting the depth of the folia (fig. 1). The total length of the Purkinje cell layer divided by the length of the envelope then provides an estimate of the degree of foliation of the cerebellum. We refer to this ratio as the cerebellum foliation index or 'CFI'. All measurements were made of digital photographs using the public domain NIH Image program v. 1.35 (http://rsb.info.nih.gov/nih-image/).

As mentioned previously, in addition to our own material, we also measured midsagittal sections from the literature [Senglaub, 1963; Larsell, 1967; Matochik et al., 1991]. To ensure that we could use midsagittal measures as an estimate of the CFI of the entire cerebellum, we tested for a significant relationship between the CFI of the entire cerebellum and the CFI of the midsagittal section of all of our specimens. The midsagittal CFI was significantly correlated with the volume CFI ($n = 31$; $p < 0.01$; $r = 0.93$). Given this strong correlation, we analyzed only the midsagittal CFI, which enabled us to almost double the number of species included in our analyses from 48 to 91 and include all of the species depicted in the literature.

Scaling and Developmental Variables

One of the primary aims of our study was to determine whether the degree of foliation was more strongly correlated with body size, brain size or cerebellar volume. For each of our specimens, we obtained body masses either from the actual specimen or, where this was not available, from the literature [see references in Iwaniuk and Nelson, 2003]. We determined brain volume by weighing each of our specimens to the nearest milligram and then dividing this weight by the density of fresh brain tissue [1.036 g/ml; Kretschmann and Wingert, 1969; Starck, 1989; Ebinger, 1995; Iwaniuk and Nelson, 2002]. Cerebellar (Cb) volumes were measured by multiplying the area of serial sections by the sampling interval and the section thickness (40 μm). The sampling interval varied from every second section to every eighth section, depending upon the size of the specimen. Varying the distance between sections did not, however, have an effect on the calculation of Cb volume. Volumes based on measurements taken every second section did not differ significantly from measurements taken every fourth, sixth or eighth section for species ranging in size from the cockatiel *(Nymphicus hollandicus)* to the Australian pelican *(Pelecanus conspicillatus)* (paired $t = -1.46$, d.f. = 16, $p = 0.18$). Furthermore, Cb volumes measured from larger sampling intervals (e.g., every eighth) were strongly correlated with Cb volumes measured from every second section ($r = 0.9992$; $p < 0.0001$). Cb and brain volumes for species represented in Senglaub [1963], Larsell [1967] and Matochik et al. [1991] were obtained from Portmann [1947], Rehkämper et al. [1991], Boire and Baron [1994], Iwaniuk and Nelson [2003], and Iwaniuk et al. [2005]. Most of these studies calculated Cb and brain volumes in a similar fashion to our study using fixed tissue. The exception is Portmann [1947], who appears to have measured fresh brains. Given that there was no overlap in data sets between Portmann [1947] and ourselves, we are unable to determine what effect using fresh versus fixed tissue masses would have on our analyses. This could result in additional variation in our analyses, but the consistency of our results (see below) suggests that such variation is unlikely to affect the significance of any of the tests performed.

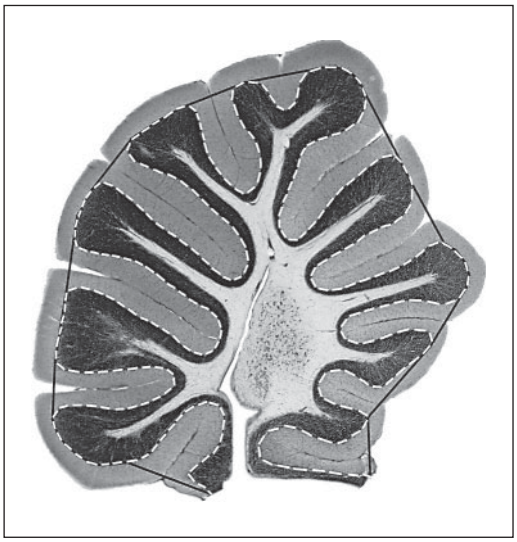

Fig. 1. A photo of a mid-sagittal section of the cerebellum of a Peaceful Dove *(Geopelia placida)*. The Purkinje cell layer, as indicated by the white line, is situated between the darkly-stained granule cell layer and the lightly-stained molecular cell layer of the cerebellar cortex. The envelope measurement of the Purkinje cell layer is indicated by the black line and follows the exterior surface of the Purkinje cell layer, thus discounting the deep folds and fissures.

In addition to body mass, brain volume and Cb volume, we also tested whether foliation reflected life history differences among species. As discussed previously, one possible correlate of foliation is the developmental mode at hatching, which is strongly correlated with relative brain volume [Portmann, 1947; Bennett and Harvey, 1985; Iwaniuk and Nelson, 2003]. To test whether developmental mode is also correlated with foliation, we categorized all 91 species according to the four categories used in Iwaniuk and Nelson [2003]: altricial, semi-altricial, semi-precocial and precocial. Because this categorization actually reflects continuous variation in development, we also tested for possible correlations between foliation and incubation period and fledging age. Data for both incubation period and fledging age were obtained from Iwaniuk and Nelson [2003] and references therein.

Allometric Effects

Prior to all analyses, the CFI and scaling variables were all log-transformed to normalize their distributions. We assessed allometric effects by calculating least-squares linear regressions of CFI against four scaling variables: body mass, brain volume, Cb volume and brain-Cb volume. This enabled us to determine which variable was most strongly correlated with CFI as well as measure the relative degree of foliation by calculating residuals from the regression lines. We also ran a multiple regression using body mass, Cb volume and brain-Cb volume as covariates of CFI and calculated residual CFI's from the multiple regression. Alternative regression models were compared by calculating parsimony using the Akaike information criterion (AIC) [Faraway, 2005]. The AIC evaluates

increased predictive value (the reduction in residual sums of squares, RSS) for a decrease in model simplicity as more parameters (k) are added according to the formula:

$$AIC = 2k/n + \ln(RSS/n)$$

where n is the number of samples. Smaller AIC values indicate a better model. All of these multiple regressions and AIC calculations were performed in the R statistical package [R Development Core Team, 2004]. Once we determined the best model using AIC, the residuals of the CFI were calculated for comparison among orders as well as for additional statistical analyses.

Developmental Effects

Correlations between CFI and developmental mode, incubation period and fledging age were assessed using two types of analysis. First, we ran multiple regression models with developmental mode, incubation period, fledging age and all three scaling variables as covariates of CFI. Multiple regressions were performed on each of the developmental variables independently in addition to including all of them in a single model. For all multiple regressions, developmental mode was considered an ordinal variable following that in Iwaniuk and Nelson [2003]. The ordinal ranking is as follows: 1 = precocial, 2 = semi-precocial, 3 = semi-altricial and 4 = altricial. When categorized in this fashion, the developmental modes represent a continuum of precocial to progressively more altricial hatchlings.

Second, we used ANOVAs of the CFI residuals and each of the developmental variables. In this analysis, mode was treated as a categorical variable whereas incubation period and fledging age were treated as continuous variables. This was performed on each set of residuals calculated from the regression analyses outlined above. Thus, we examined CFI relative to body mass, brain-Cb volume and Cb volume as well as relative to the multiple regression model of the three scaling variables.

Multivariate Model

In an attempt to determine how all of the scaling and developmental variables surveyed contribute to CFI across all species and for any interaction effects between the independent variables, we combined all of the data into a full multivariate model. Again, developmental mode was treated as an ordinal variable and the remaining traits, body mass, brain-Cb volume, Cb volume, incubation period and fledging age, were treated as continuous variables. As with our previous multiple regression models, we used AIC to determine the best model (see above).

Phylogenetic Effects

In addition to testing for allometric and developmental effects, we were also interested in determining whether phylogeny affected relative CFI. To test for a significant phylogenetic signal, we followed the procedure outlined in Blomberg et al. [2003]. This method examines the variance of independent contrasts (see below), which is a reflection of how well the phylogenetic tree fits the data. For example, if closely related species tend to share a similar relative CFI value throughout the tree, then the computed variance of the contrasts will be low. We tested whether the variance of the contrasts was significant or not by comparing the calculated variance with that obtained from a random permutation of the data across the tips of the phylogeny, irrespective of phylogenetic relationships. The distribution of the variances of the simulated data can then be used to determine if the observed variance falls outside of the 95% confidence interval. To perform this test, we calculated independent contrasts in PDTREE (see below) of absolute CFI values as well as the residuals from the regression analyses of the scaling variables. After diagnostic tests of the branch lengths, we recorded the observed variance as calculated in PDTREE. Using PDRANDOM, we then permuted the data randomly across the tips 1,000 times. This permuted data was then entered into PDERROR to calculate the variance of each permutation. Finally, we examined the distribution of the permuted variances in a histogram to determine the 95% confidence interval.

Although this method is useful for determining the presence of a phylogenetic effect, it does not actually yield any information regarding which clades have higher or lower CFIs than others. To determine whether there were significant differences among orders, we also used an ANOVA of CFI residuals with order-membership as a categorical variable. For the analyses of the residuals, all sets of residuals were compared among orders. Thus, we tested for significant differences in CFI among orders relative to body, brain, brain-Cb volume, Cb volume and our multiple regression model. Our order-level taxonomy broadly followed that of Monroe and Sibley [1997], with some changes made to reflect phylogenetic relationships and broad ecological/behavioral differences. These latter changes reflected parvorder/infraorder level taxonomy of Monroe and Sibley [1997].

To account for phylogenetic effects in the allometric and developmental comparisons, we calculated independent contrasts [Harvey and Pagel, 1991] of all continuous variables using PDTREE, a program within the PDAP software package [available from T. Garland upon request]. This method is used extensively in comparative biology, including neuroanatomical analyses [e.g., Timmermans et al., 2000; Hutcheon et al., 2002; Iwaniuk and Nelson, 2003; Iwaniuk et al., 2005; Sol et al., 2005]. All continuous variables were log-transformed prior to calculating the contrasts. A composite phylogeny was assembled using Sibley and Ahlquist [1990] for inter-ordinal relationships and resolution within each order provided by additional references [Christidis et al., 1991; Kimball et al., 1999; Barker et al., 2004; Altshuler et al., 2004]. Because we reconstructed this tree from a variety of sources, we used an arbitrary branch length model that set all branch lengths = 1. Diagnostic tests indicated that these branch lengths adequately standardized all of the data [Garland et al., 1992] and were therefore used in subsequent statistical analyses. We then repeated all of the analyses previously outlined. All least-squares linear and multiple regressions were forced through the origin [Garland et al., 1992].

Finally, for our one categorical variable, developmental mode, we used both an independent contrasts approach (see above) and a phylogeny-corrected ANOVA. Briefly, this latter method generates a phylogeny-corrected critical F distribution that can be used instead of a conventional critical F [Garland et al., 1993; Hutcheon et al., 2002; Pellis and Iwaniuk, 2002; Iwaniuk et al., 2005, 2006a]. We performed 1000 Monte Carlo simulations of the CFI residuals across the phylogeny using PDSIMUL. The simulations were constrained to biologically realistic values by setting the upper and lower limits just higher and lower than the extremes of the original data set. PDANOVA is then used to construct a phylogeny-corrected and empirically scaled F distribution and the critical F calculated.

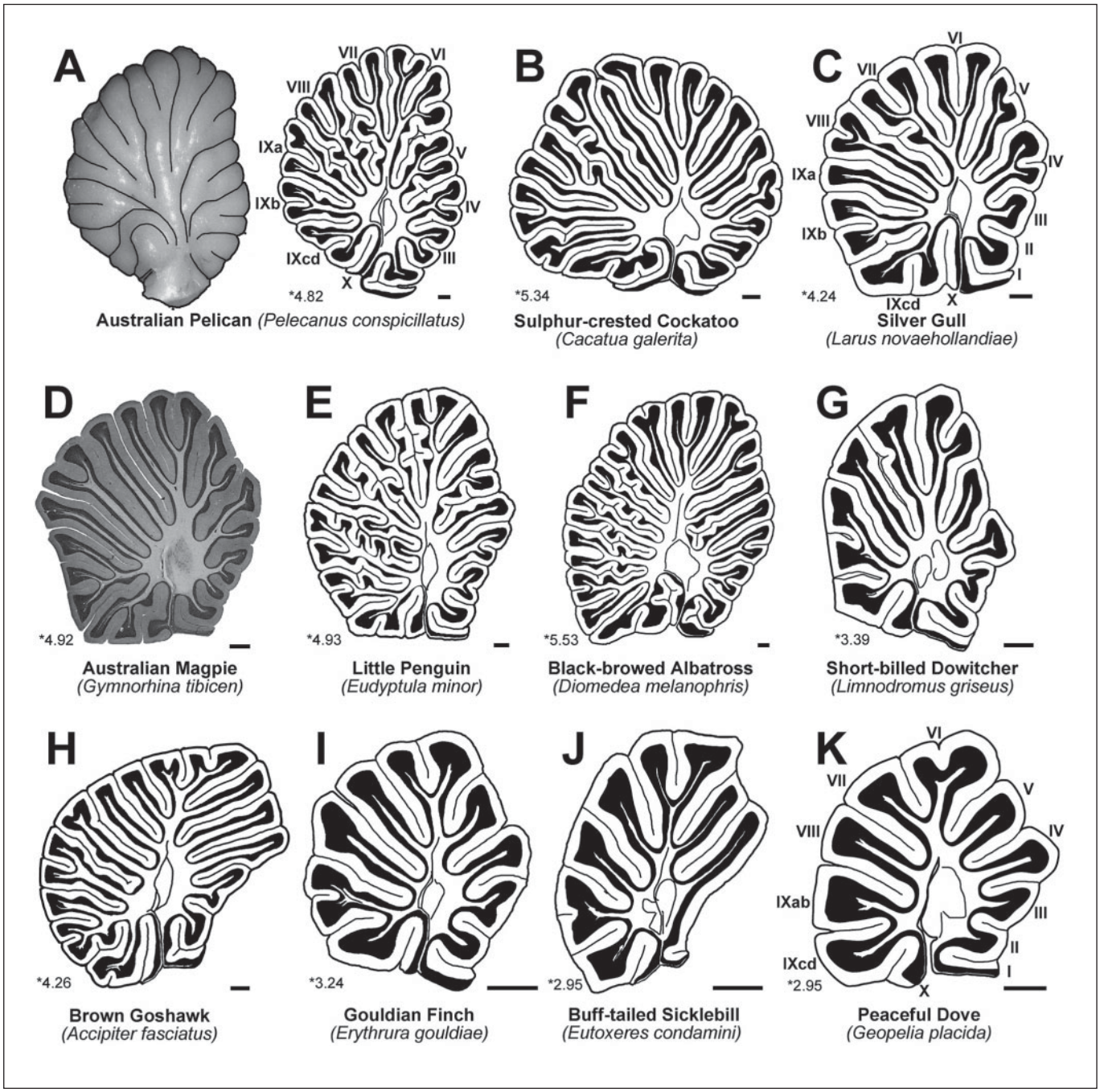

Fig. 2. A A lateral photo with the individual folia indicated in black and a midsagittal section through the cerebellum of an Australian Pelican *(Pelecanus conspicillatus)*. The remaining illustrations and photos of midsagittal cerebellum sections are: **B** Sulphur-crested Cockatoo *(Cacatua galerita)*; **C** Silver Gull *(Larus novaehollandiae)*; **D** Australian Magpie *(Gymnorhina tibicen)*; **E** Little Penguin (*Eudyptula minor*); **F** Black-browed albatross *(Diomedea melanophris)*; **G** Short-billed Dowitcher *(Limnodromus griseus)*; **H** Brown Goshawk *(Accipiter fasciatus)*; **I** Gouldian Finch *(Erythrura gouldiae)*; **J** Buff-tailed Sicklebill (*Eutoxeres condamini)*, FMNH 315304); and **K** Peaceful Dove *(Geopelia placida)*. For each species, the cerebellar foliation index (*) is indicated. Note that in **A**, **C** and **K**, the individual folia are labeled following Larsell's [1967] numerical nomenclature. Scale bars = 1 mm.

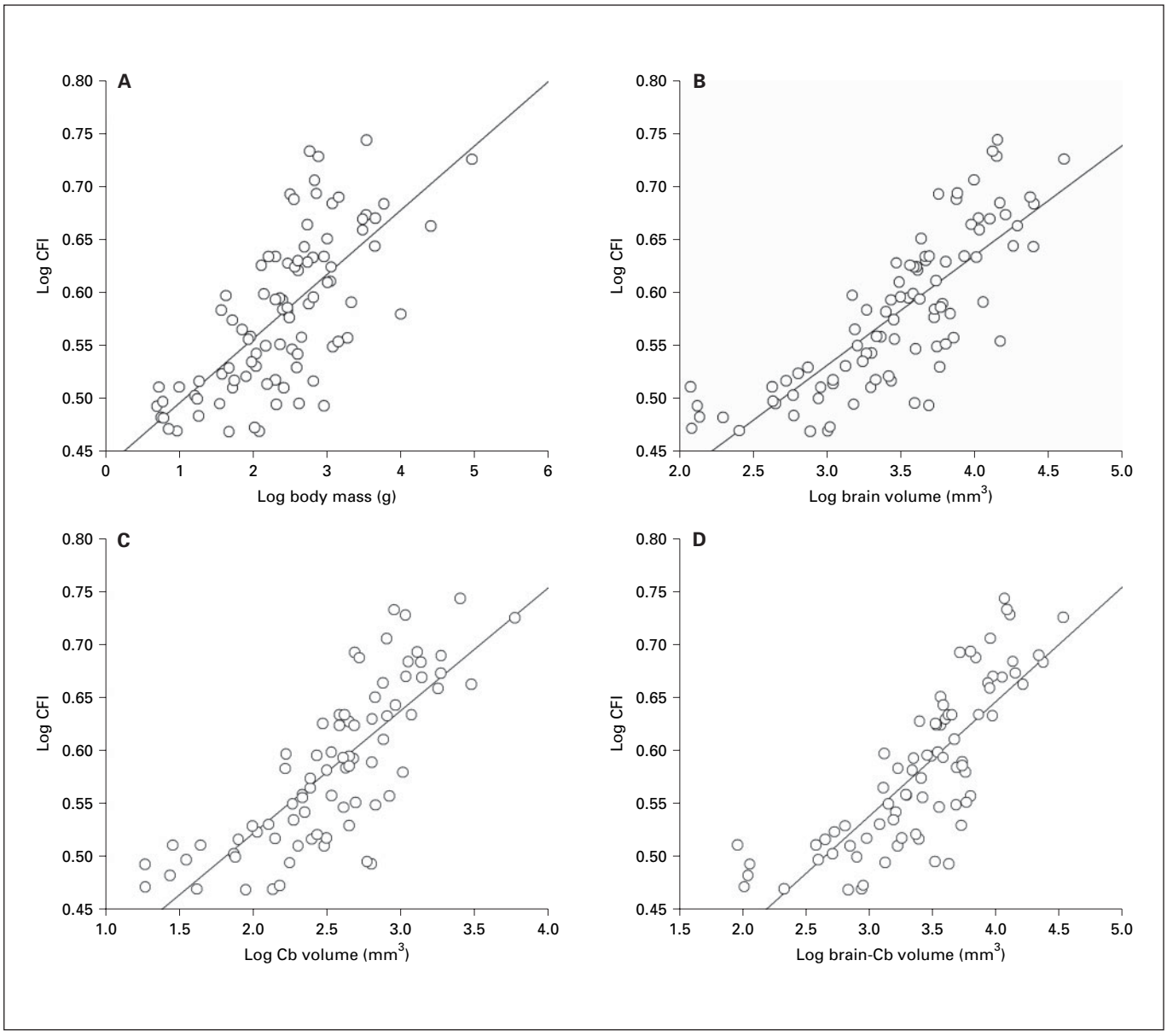

Fig. 3. Scatterplots of the log-transformed cerebellar foliation index (CFI) against each of the four scaling variables examined: **A** log-transformed body mass (g); **B** log-transformed brain volume (mm^3); **C** log-transformed cerebellar volume (mm^3), and **D** log-transformed brain minus cerebellar volume (mm^3). The least-squares linear regression line for each bivariate plot is indicated by a solid line.

Results

The degree of foliation varied considerably among the species surveyed. Figure 2 shows photos and line drawings of photos taken of midsagittal cerebellar sections of 11 species that encompass the variation observed. The generic foliation pattern, with the roman numerology adopted by Larsell [1967] is indicated in figures 2A, F and K. From anterior to posterior, the major folia are numbered I–X. Folium IX, the largest folium, is always divided into two major branches: IXab and IXcd. The swifts and hummingbirds, for example the hummingbird *Eutoxeres condamini* (fig. 2J), represent an exception as folia III of the anterior lobe is absent [see also Larsell, 1967; Iwaniuk

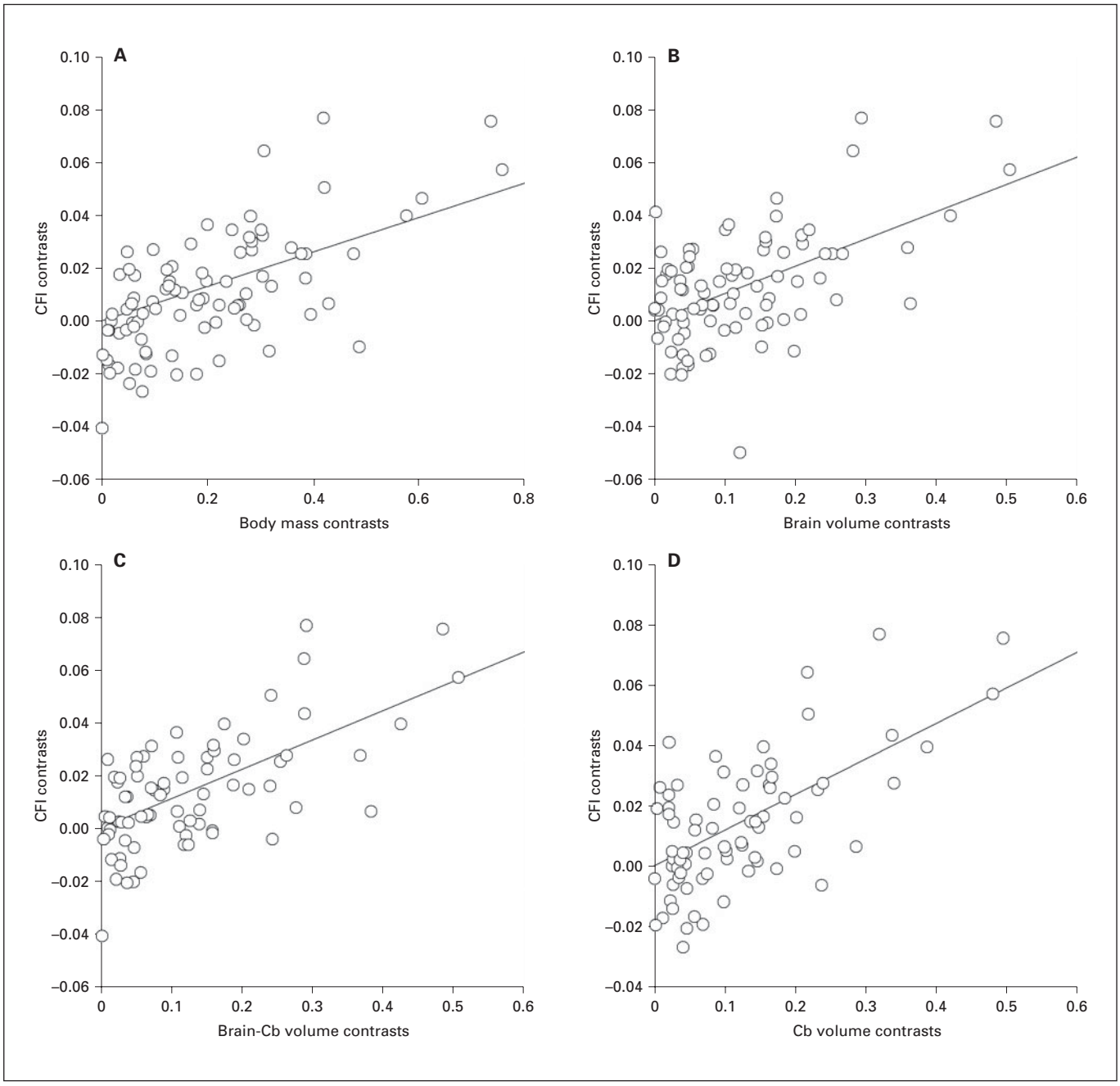

Fig. 4. Scatterplots of contrasts of log-transformed cerebellar foliation index (CFI) against: **A** contrasts of log-transformed body mass; **B** contrasts of log-transformed brain volume; **C** contrasts of log-transformed brain minus cerebellar volume and **D** contrasts of log-transformed cerebellar volume. The least-squares linear regression line for each bivariate plot is indicated by a solid line.

et al., 2006b]. For species with more complex foliation patterns, Larsell [1967] considered any extra folia as branches of these major folia. In most species, identifying the folia is relatively straightforward. For example, the Australian Pelican (*Pelecanus conspicillatus*; fig. 2A) and Silver Gull (*Larus novaehollandiae*; fig. 2C) where the cerebellum is highly foliated, the principal folia are easy to identify. For other species, such as the Black-browed Al-

Table 2. Variances of the actual data ('true variance') on the phylogenetic tree and the lower 95% interval of the variances calculated from randomizing the data across the phylogenetic tree ('randomized variance')

Measure	True variance	Randomized variance
Absolute CFI	3.062×10^{-3}	4.298×10^{-3}
CFI relative to body mass	5.868×10^{-4}	1.387×10^{-3}
CFI relative to brain volume	6.176×10^{-4}	1.002×10^{-3}
CFI relative to brain-cerebellar volume	5.565×10^{-4}	9.970×10^{-4}
CFI relative to cerebellar volume	5.355×10^{-4}	1.045×10^{-3}
CFI relative to multiple regression	5.352×10^{-4}	1.062×10^{-3}

batross (*Diomedea melanophris*; fig. 2F), the task is more difficult.

CFIs ranged from 2.9451 in the Peaceful Dove (*Geopelia placida*, fig. 2K) to 5.5338 in the Black-browed Albatross (fig. 2F, table 1). High CFIs were found in several taxa, most notably the seabirds (fig. 2A, E, F), gulls (fig. 2C), parrots (fig. 2B), corvids (fig. 2D) and raptors (fig. 2H). At the opposite end of the spectrum, low CFIs were found in shorebirds (fig. 2G), some songbirds (fig. 2I), hummingbirds (fig. 2J) and pigeons and doves (fig. 2K).

Allometric Effects

We found significant relationships between CFI and all four of the scaling variables examined: body mass ($F = 85.50$; d.f. = 1, 89; $p < 0.01$; fig. 3A), brain volume ($F = 166.16$; d.f. = 1, 89; $p < 0.01$; fig. 3B), Cb volume ($F = 132.19$; d.f. = 1, 77; $p < 0.01$; fig. 3C) and brain-Cb volume ($F = 146.14$; d.f. = 1, 77; $p < 0.01$; fig. 3D). The amount of variation explained by each of these allometric equations increased in the following order: body mass ($r^2 = 0.48$) < Cb volume ($r^2 = 0.63$) < brain volume ($r^2 = 0.65$) and brain-Cb volume ($r^2 = 0.65$). A multiple regression that included body mass, Cb volume, brain-Cb volume and all possible interactions as covariates of CFI yielded no significant three-way interaction effect ($F = 0.87$; d.f. = 1, 71; $p = 0.35$), but there was a significant two-way interaction effect between Cb volume and brain-Cb volume ($F = 79.49$; d.f. = 1, 76; $p < 0.01$) and a significant effect of body mass ($F = 17.00$; d.f. = 1, 76; $p < 0.01$). Thus, birds with large cerebella relative to the rest of the brain and a large body mass have high CFIs. In terms of the amount of variation explained, this multivariate model contributed a 9% improvement ($r^2 = 0.74$) to the two allometric models using only brain volume or brain-Cb volume (both $r^2 = 0.65$).

Our analyses of the independent contrasts yielded similar results, but with lower correlation coefficients. Log CFI contrasts were significantly correlated with body mass contrasts ($F = 80.02$; d.f. = 1, 87; $p < 0.01$; $r^2 = 0.38$; fig. 4A), brain volume contrasts ($F = 80.13$; d.f. = 1, 87; $p < 0.01$; $r^2 = 0.29$; fig. 4B), Cb volume contrasts ($F = 101.35$; d.f. = 1, 75; $p < 0.01$; $r^2 = 0.41$; fig. 4C) and brain-Cb volume contrasts ($F = 100.69$; d.f. = 1, 75; $p < 0.01$; $r^2 = 0.41$; fig. 4D). Thus, as with the analysis of species as independent data points, CFI was positively correlated with each of the scaling variables.

A multivariate analysis of these contrasts yielded no significant three-way interaction, two significant two-way interactions (body mass × brain-Cb: $F = 4.65$; d.f. = 1, 72; $p = 0.03$; body mass × Cb: $F = 10.09$; d.f. = 1, 72; $p < 0.01$) and a significant effect of brain-Cb volume ($F = 16.98$; d.f. = 1, 72; $p < 0.01$). Thus, the independent contrasts analysis partially supported the analysis of species as independent data points; birds with large cerebella relative to the rest of the brain and a large brain had high CFIs. Overall, this model explained more of the variation in CFI ($r^2 = 0.65$) than any of the variables on their own (see above).

Overall, these analyses indicated that allometry exerts a significant effect on the degree of cerebellar foliation. For all subsequent analyses, we therefore removed allometric effects by either using residuals from the four allometric relationships described above or residuals from the multiple regression models that were significant (also see above). Hereafter, we refer to all CFI residuals as 'relative CFI' so as to distinguish these allometry-free measures of foliation from the absolute CFI measured from the specimens.

Phylogenetic Effects

We detected a significant phylogenetic signal in both absolute and relative CFI values. In all instances (table 2), the observed variance was lower than 95% of the variance calculated from randomly distributing the data across the phylogenetic tree. Given that the observed variances were lower than that expected from our randomization test, we

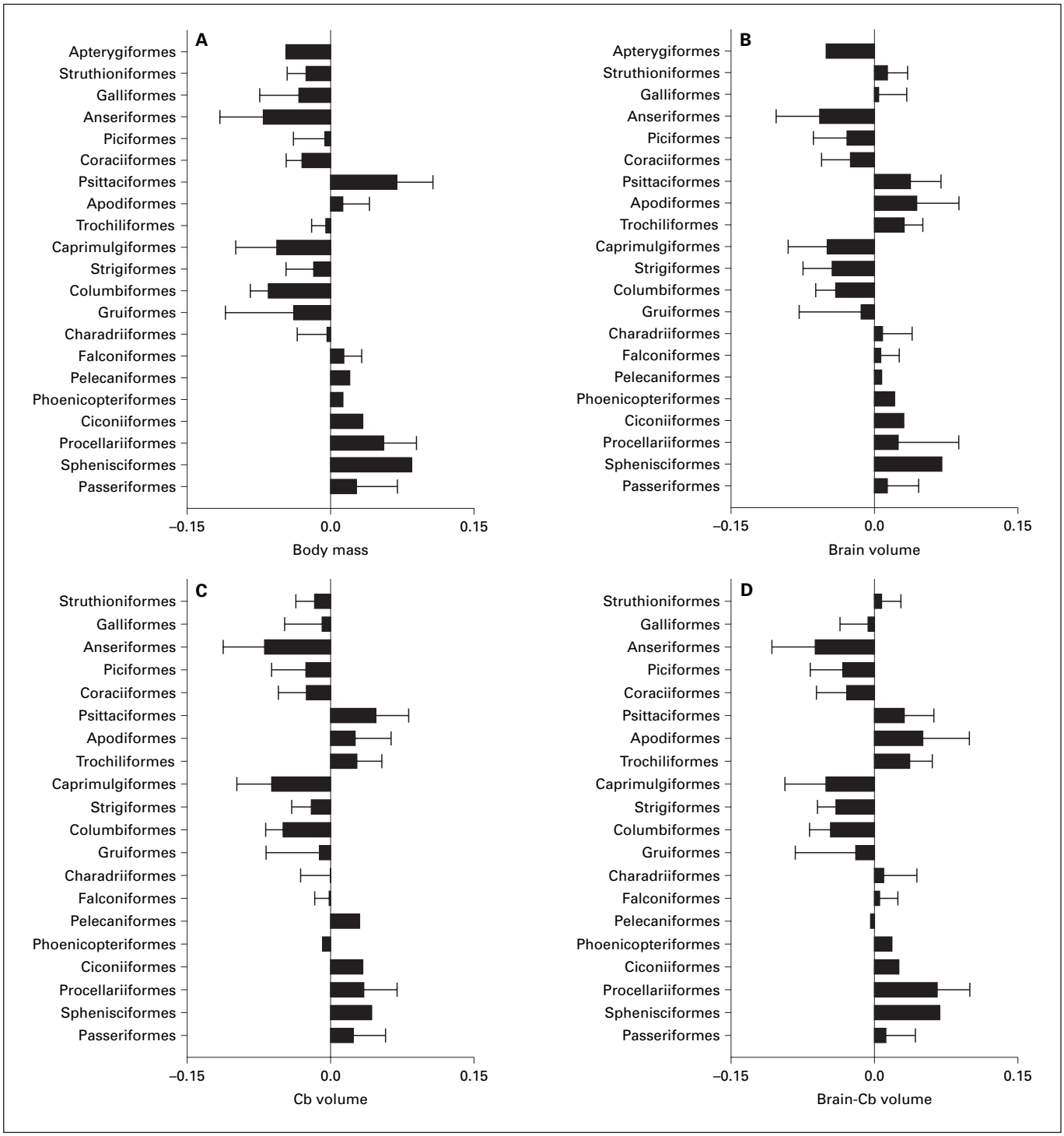

Fig. 5. Histograms of cerebellar foliation index (CFI residuals derived from allometric equations using each of the following scaling variables: (**A**) body mass; (**B**) brain volume; (**C**) cerebellar volume, and (**D**) brain minus cerebellar volume. Species are grouped according to order membership and are provided in the same sequence as the phylogenetic tree in Sibley and Ahlquist [1990]. The error bars indicate standard deviations for each order. Note that the Brown Kiwi *(Apteryx australis)*, the only representative of the Apterygiformes, is missing from **C** and **D** because we did not have any data on cerebellar volume for this species.

Table 3. The results of ANOVAs of relative cerebellar foliation index (CFI), with respect to four different allometric analyses, grouped according to order membership

CFI	F	d.f.	p	r^2
Body mass	5.92	20, 70	<0.0001	0.52
Brain volume	3.84	20, 70	<0.0001	0.39
Brain-cerebellar volume	4.22	19, 59	<0.0001	0.44
Cerebellar volume	4.74	19, 59	<0.0001	0.48
Multiple regression	4.65	19, 59	<0.0001	0.47

conclude that there is a significant phylogenetic signal in both absolute and relative CFI.

A comparison of mean relative CFI for each order revealed considerable variation among taxa (fig. 5). At one end of the spectrum, the parrots (Psittaciformes), seabirds (Procellariiformes) and penguins (Sphenisciformes) had relatively high CFIs. In contrast, relatively low CFIs were present in the waterfowl (Anseriformes), nightjars (Caprimulgiformes) and pigeons (Columbiformes). ANOVAs of relative CFI revealed significant differences among orders (table 3). Post-hoc Tukey-Kramer tests between all pairs of orders indicated that the waterfowl, pigeons and nightjars had significantly smaller relative CFIs than most other taxa whereas parrots had significantly higher relative CFIs than other taxa. Thus, our statistical analyses largely corroborated our observations of relative CFI variation based on the histograms (fig. 5).

It should be noted that there was substantial variation within some orders. For example, there is a large difference in relative CFI between the two gruiforms (table 1; fig. 5) and relatively high variation within the waterfowl and nightjars. Within the passerines, the sub-order Corvida had a significantly higher relative CFI than the sub-order Passerida (Wilcoxon $Z \geq -2.13$; $n = 14$; $p \leq 0.03$).

Developmental Effects

Relative CFI varied significantly among developmental modes, regardless of how the allometric effects were removed (table 4). As shown in figure 6, altricial species tended to have relatively higher CFIs and precocial species relatively lower CFIs, but there was a lot of variability within developmental modes. When compared to phylogeny-corrected critical Fs, however, this significant relationship disappeared. The phylogeny-corrected critical Fs were all much greater (all corrected Fs >12.70) than the calculated Fs (table 4). Thus, developmental mode did not appear to play a significant role in the degree of cerebellar foliation once phylogenetic effects were taken into account.

Incubation period was not correlated with relative CFI in any of our comparisons and fledging age was only correlated with CFI relative to body mass (table 4). Once we incorporated phylogenetic information, none of these relationships remained significant (table 5). From these results, we concluded that developmental differences were not significantly correlated with the degree of cerebellar foliation.

Multivariate Model

In our multivariate model, no significant five-, four- or three-way interactions were detected. Three significant two-way interactions were detected between body mass and brain-Cb volume ($p < 0.0001$); body mass and incubation period ($p < 0.0001$), and body mass and developmental mode ($p = 0.001$). In addition, we detected significant effects of Cb volume ($p = 0.007$), developmental mode ($p = 0.002$), incubation period ($p < 0.0001$) and fledging age ($p = 0.006$) on CFI. Overall, this model explained 80% of the interspecific variation in CFI ($r^2 = 0.80$) and was therefore a slight improvement over the allometric model described above ($r^2 = 0.74$). Given the number of significant variables detected, interpreting these results is problematic, but can best be summarized as follows: CFI increased with Cb volume, relatively large brains, relatively long incubation periods and short fledging ages and varied among the developmental modes.

Using independent contrasts, most of these effects became non-significant. The only significant effects detected were Cb volume ($p = 0.001$) and the interaction between brain-Cb volume and body mass ($p = 0.01$) and this model yielded a correlation coefficient (r^2) of 0.63. Although one might expect the same effects as our allometric analysis (see above), fewer contrasts were calcu-

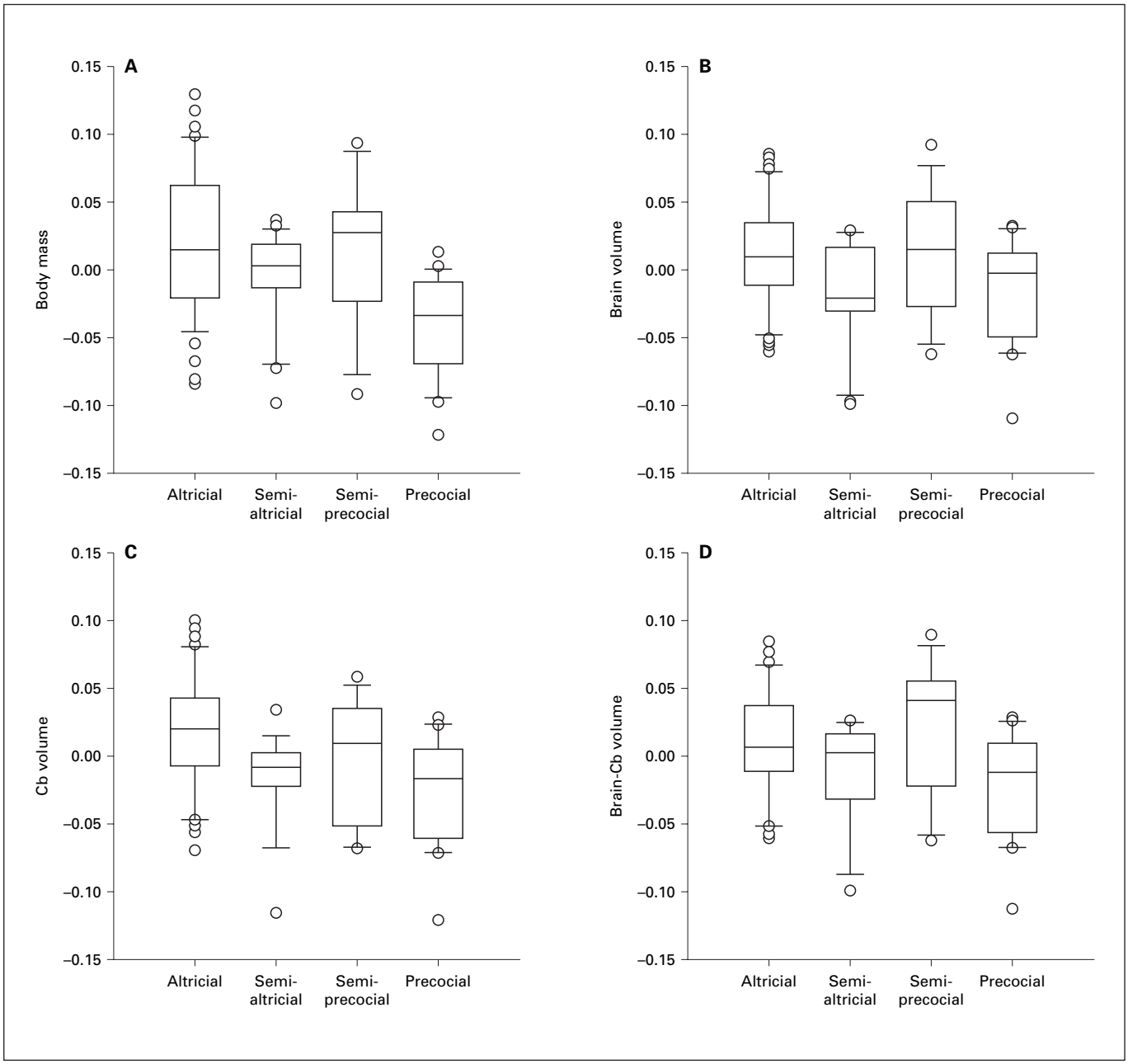

Fig. 6. Box-plots of cerebellar foliation index (CFI) residuals derived from allometric equations using each of the following scaling variables: **A** body mass; **B** brain volume; **C** cerebellar volume, and **D** brain minus cerebellar volume. Species are grouped according to developmental mode and are arranged in order of increasing precociality. The open circles represent individual species that fall outside of the 95% confidence interval for each developmental mode.

lated in this analysis because we lacked data on incubation period and fledging age for several species. Thus, no significant interaction effect between Cb volume and body mass was detected ($p = 0.66$). We therefore concluded from the multivariate model, that allometry exerted the strongest effect on CFI and this is specifically due to the strong relationship between CFI and the relative size of the brain and Cb volume.

Table 4. Results of ANOVAs of CFI relative to our scaling variables and each of the developmental variables surveyed

CFI	Variable	F	d.f.	p
Body mass	Developmental mode	6.44	3, 87	**0.0005**
	Incubation	1.93	1, 86	0.17
	Fledging	10.56	1, 83	**0.002**
Brain volume	Developmental mode	3.71	3, 87	**0.01**
	Incubation	0.24	1, 86	0.62
	Fledging	2.13	1, 83	0.15
Brain-Cb volume	Developmental mode	3.86	3, 75	**0.01**
	Incubation	1.20	1, 74	0.28
	Fledging	2.60	1, 71	0.11
Cb volume	Developmental mode	5.11	3, 75	**0.003**
	Incubation	0.00	1, 74	0.95
	Fledging	0.90	1, 71	0.35
Multiple regression	Developmental mode	4.88	3, 75	**0.004**
	Incubation	0.02	1, 74	0.90
	Fledging	0.67	1, 71	0.42

Significant p values are shown in bold.

Table 5. Results of ANOVAs of CFI contrasts relative to our four scaling variables and each of the contrasts of the developmental variables surveyed

CFI	Variable	F	d.f.	p
Body mass	Incubation	0.43	1, 84	0.52
	Fledging	1.30	1, 81	0.26
Brain volume	Incubation	0.05	1, 84	0.83
	Fledging	0.43	1, 81	0.51
Brain-Cb volume	Incubation	0.77	1, 72	0.38
	Fledging	0.09	1, 69	0.76
Cb volume	Incubation	0.40	1, 72	0.53
	Fledging	0.00	1, 69	0.94
Multiple regression	Incubation	0.33	1, 72	0.57
	Fledging	0.01	1, 69	0.94

Discussion

The degree of cerebellar foliation is significantly correlated with body, brain and cerebellum size. In addition, there is a strong phylogenetic effect such that some orders have significantly more folded cerebella than other orders. Of the developmental differences examined, only developmental mode at hatching varied significantly with relative CFI; neither incubation period nor fledging age were significantly correlated with relative CFI in a consistent fashion. Although the combination of these effects explained most of the variation in relative CFI, there remains the possibility that behavioral and/or cognitive differences also play a role in the evolution of a more or less foliated cerebellum.

Allometric Effects

Allometric effects account for most of the variation in CFI, which corroborates the findings of previous authors [Senglaub, 1963; Larsell, 1967; Pearson and Pearson, 1976]. That is, CFI increases along with increases in body, brain and cerebellar size. The strongest effect (as measured by the correlation coefficient) was exerted by brain volume. Thus, it is a large brain, and not necessarily a large body, that contributes most to interspecific variation in the CFI. The variation in scaling exponents and the

amount of variation explained by each of the scaling variables reinforces the importance of testing multiple scaling variables in allometric comparisons [Sherry et al., 1989; Deaner et al., 2000; Pellis and Iwaniuk, 2002; Day et al., 2005; Iwaniuk et al., 2005].

Interestingly, the correlation between Cb volume and CFI was weaker than that of brain volume. Thus, the degree of foliation is more affected by the size of the entire brain rather than the size of the cerebellum alone. It could be that an increased CFI is a means of overcoming the processing limitations of a relatively small Cb. By increasing the length of the Purkinje cell layer of a cerebellum with a restricted volume, it might be possible to functionally increase the processing power of the cerebellum. This could partially explain the relatively high CFIs in parrots and corvids. Both parrots and corvids have relatively larger brains and telencephala than other birds, but the cerebellum is relatively small [Portmann, 1947; Rehkämper et al., 1991; Iwaniuk et al., 2005; Iwaniuk and Hurd, 2005]. Increasing the Purkinje cell length, without altering Cb volume, might therefore be a means of increasing the processing power of the Cb in these species.

Developmental Effects

Unlike relative brain volume, which is correlated with a variety of developmental variables [Iwaniuk and Nelson, 2003], relative CFI was not strongly correlated with differences in development among species. In general, we detected no significant effects of incubation period or fledging age and detected only a weak effect of developmental mode. This weak effect specifically reflected differences between altricial and precocial species; altricial species tend to have relatively higher CFIs than precocial species. Although this could reflect differences in relative brain size between altricial and precocial species [see review in Iwaniuk and Nelson, 2003], the fact that all allometric effects were removed from the CFI suggests that this is not the case. Furthermore, in between these two extremes, there was substantial variation such that semi-altricial and semi-precocial species were equally likely to have a relatively high or low CFI. From this, we conclude that developmental differences do play a role in the evolution of CFIs, but that this role accounts for far less variation than allometry and phylogeny. Whether developmental effects have a more significant role in the evolution of other aspects of cerebellar morphology, such as cerebellar volume or sizes of individual folia, has yet to be tested.

Phylogenetic Effects

Using two different methods, we found a significant effect of phylogeny on absolute and relative CFI. With Blomberg et al.'s [2003] method, we found that related species tend to share a similar absolute and relative CFI and that the variation in absolute and relative CFI across the phylogeny is significantly less than that expected from chance alone. The degree of cerebellar foliation in birds can therefore be added to the growing list of morphological traits that have a significant phylogenetic signal, including brain size [Blomberg et al., 2003].

The extent to which relative CFI is reflected by the phylogeny is made clear by our statistical analysis among orders, which demonstrated that some groups have significantly higher relative CFIs than others. In particular, the seabirds, parrots and penguin all had relatively high CFIs whereas the pigeons, nightjars and waterfowl had relatively low CFIs. Why these taxa in particular have vastly different CFIs after removing body size effects is difficult to determine at this stage. One possibility is that biomechanical constraints on the morphology of the cerebellum prevent it from expanding in some species [Gould and Lewontin, 1979]. For example, the large adductor muscles of parrots, which attach to the braincase surrounding the cerebellum, could constrain the size and shape of the endocranial cavity containing the cerebellum such that the best means of increasing the processing capacity of the cerebellar cortex is to develop a more folded cortical sheet. As with isocortical folding in primates [Striedter, 2004], biomechanical constraints represent only a partial explanation for interspecific variation in relative CFI. Kingfishers have hypertrophied adductor mandibulae externi muscles compared to other birds, which appears to result in a medio-laterally flattened brain [Legge, 2004], but contrary to the biomechanical hypotheses, both of the kingfishers we examined have relatively low CFIs (fig. 5). Similarly, corvids do not have particularly large jaw muscles, but have relatively high CFIs. Thus, alternative theories to biomechanical constraints are required to explain the observed variation in relative CFI among avian orders.

A second possible explanation for the phylogenetic effect is that relative CFI reflects variation in behavior and/or ecology among orders. Phylogenetic history has a significant effect on the evolution of behavioral and life history traits in birds. For example, there are significant effects of phylogeny on body mass, brain size, metabolic rate, breeding habitat, morphometric measurements, age at maturity, diet, display behaviors, vocalizations and a variety of other traits [e.g., Irwin, 1996; Kennedy et al.,

1996; Böhning-Gaese and Oberrath, 1999; Johnson et al., 2000; Price and Lanyon, 2002; Bennett and Owens, 2002; Blomberg et al., 2003]. The phylogenetic effect could reflect correlations between any of these or other traits and relative CFI. Determining which of these traits are correlated with relative CFI is beyond the scope of the present study. We can, however, speculate that traits reflecting cognitive differences, such as tool use [Lefebvre et al., 2002] and feeding innovations [Lefebvre et al., 2004], are likely to be correlated with neuroanatomical features that improve processing capacity and a relatively high CFI represents just such a feature.

Functional Implications of Foliation

An increase in the amount of folding of a structure is a means of increasing surface area within a constrained volume [Striedter, 2004]. For laminar structures, such as cerebellar or cerebral cortex, increasing the surface area may be a better means of improving function than increasing volume [Sultan, 2002, 2005]. In the cerebral cortex, folding reduces the distance neurons need to travel for connections within the cortex, thereby enhancing its interconnectedness [Van Essen, 1997; Rilling and Insel, 1999; Changizi, 2001; Harrison et al., 2002; Klyachko and Stevens, 2003]. An increased demand for intracortical processing could therefore result in a more folded isocortex. This pattern appears in primate evolution whereby increases in isocortical gyrification correspond with cognitive abilities [Rilling and Insel, 1999].

The cerebellar cortex is, however, much different from the mammalian cerebral cortex. The cerebellar cortex is thinner than cerebral cortex and envelops a smaller volume of nuclei and ventricular space. In addition, because of the relative paucity of isocortical-like connections within the cerebellum, the position of the cerebellar folds cannot reflect patterns of intracortical connections in the same fashion as the cerebral cortex [Van Essen, 1997]. As a result, the argument that folding increases processing power by improving cortical connectivity is unlikely to apply to the cerebellum. This does not, however, mean that cortical folding does not improve processing capacity or efficiency in the cerebellum. An increase in the degree of foliation functionally translates into a greater number of Purkinje cells per unit volume of cerebellum. The Purkinje cells provide the sole route out of the cerebellar cortex [Voogd and Glickstein, 1998] and as such, they could represent a processing constraint or 'bottleneck' on information entering the cerebellar nuclei from the cortex. In terms of the degree of foliation, by increasing the CFI and thereby increasing the number of Purkinje cells, there could be a concomitant increase in the processing capacity of the cerebellar cortex that, in turn, results in an increase in behavioral complexity and/or cognitive ability. This is not dissimilar to other proposed brain-behavior relationships whereby an increase in the number of neurons translates to improved functional capacity [e.g., Ward et al., 2001].

Whether foliation truly reflects cognitive or behavioral abilities awaits further analysis. Recently, Sultan [2005] suggested that the expansion of the cerebellar cortex is correlated with tool use and other cognitive abilities in birds. In fact, a preliminary analysis [Iwaniuk and Wylie, 2005] indicates that there is a significant relationship between cognitive abilities and cerebellar foliation in birds. If true, this correlation would provide further support for experimental and other comparative studies suggesting that the cerebellum plays a critical role in cognitive processing [Paulin, 1993; Desmond and Fiez, 1998; Imamizu et al., 2003; Day et al., 2005; Rodriguez et al., 2005; Weaver, 2005]. Other functions of the cerebellum, such as motor learning and coordination [Ito, 1984; Paulin, 1993] and sensory integration [Bower, 1997] could also be improved by increased folding of the cerebellar cortex. For example, increases in motor skills, motor-based behaviors or sensory-based behaviors are also likely to occur with higher degrees of foliation. Increased processing demands arising from multiple behaviors may have therefore driven the evolution of a more foliated cerebellum in birds and possibly other vertebrates.

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