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RESEARCH ARTICLE

Brain Structure Variation in Great Apes, With Attention to the Mountain Gorilla (*Gorilla beringei beringei*)

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This report presents data regarding the brain structure of mountain gorillas (*Gorilla beringei beringei*) in comparison with other great apes. Magnetic resonance (MR) images of three mountain gorilla brains were obtained with a 3T scanner, and the volume of major neuroanatomical structures (neocortical gray matter, hippocampus, thalamus, striatum, and cerebellum) was measured. These data were included with our existing database that includes 23 chimpanzees, three western lowland gorillas, and six orang-utans. We defined a multidimensional space by calculating the principal components (PCs) from the correlation matrix of brain structure fractions in the well-represented sample of chimpanzees. We then plotted data from all of the taxa in this space to examine phyletic variation in neural organization. Most of the variance in mountain gorillas, as well as other great apes, was contained within the chimpanzee range along the first two PCs, which accounted for 61.73% of the total variance. Thus, the majority of interspecific variation in brain structure

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observed among these ape taxa was no greater than the within-species variation seen in chimpanzees. The loadings on PCs indicated that the brain structure of great apes differs among taxa mostly in the relative sizes of the striatum, cerebellum, and hippocampus. These findings suggest possible functional differences among taxa in terms of neural adaptations for ecological and locomotor capacities. Importantly, these results fill a critical gap in current knowledge regarding great ape neuroanatomical diversity. Am. J. Primatol. 63:149–164, 2004.

Key words: gorilla; chimpanzee; orang-utan; great ape; comparative neuroanatomy; magnetic resonance imaging

INTRODUCTION

The genus *Gorilla* occurs in two geographically discontinuous forest habitats, in the west and east of equatorial Africa. Across their range of distribution, gorilla populations display considerable diversity in their morphology [Groves, 2001; Leigh et al., 2003], ecology, and behavior [Doran & McNeilage, 2001]. Furthermore, some researchers believe that mtDNA variation among gorilla populations is sufficient to recognize separate eastern (*Gorilla beringei*) and western (*Gorilla gorilla*) species, based on reproductive isolation between these populations that may date back as far as 2 or 3 million years [Garner & Ryder, 1996; Ruvolo et al., 1994].

With the advent of magnetic resonance imaging (MRI), considerable data have been amassed regarding the macrostructural organization of the brain in great apes [e.g., Rilling & Insel, 1999; Semendeferi & Damasio, 2000]. Virtually nothing is known, however, about how neuroanatomical structure in great ape taxa varies with sex, age, or geographic origin. To date, brain structure volumes have been reported for several western lowland gorillas (*Gorilla gorilla gorilla*) [MacLeod et al., 2003; Rilling & Insel, 1999; Semendeferi & Damasio, 2000] and a single eastern lowland gorilla (*Gorilla beringei graueri*) individual [Stephan et al., 1981] (H.D. Frahm, personal communication). In this context, it is of interest to know whether neuroanatomical structure varies between the distinct gorilla populations of western and eastern Africa.

As part of an ongoing effort to document the behavior and biology of the gorillas living in the east African volcanic highlands of the Virunga mountains (*Gorilla beringei beringei*), we present neuroanatomical data from brains obtained postmortem at the Parc National des Volcans, Rwanda. As an initial step in examining neuroanatomical organization in the members of this critically endangered mountain gorilla population, we obtained high-resolution MRI scans and measured volumes of major brain structures. The purpose of this study was to compare the brain structure of mountain gorillas with that of other great apes, and to examine patterns of phyletic variation.

MATERIALS AND METHODS

Specimens and Preparation

A total of 35 postmortem great ape brains were used in the present study (Table I). Brain specimens of *G. g. gorilla*, *Pan troglodytes*, and *Pongo pygmaeus* were collected in the context of the Great Ape Aging Project and derived from

TABLE I. Structure Volumes From Formalin-Fixed Brains (Volumes in cm^3)

Species	Sex	Age	Whole Brain	Neo-cortex	Hippo-campus	Striatum	Thalamus	Cerebellum
Gorilla beringei beringei	F	~2.5	401.6	182.5	2.4	9.0	8.2	51.6
	M	5	460.6	214.9	3.4	8.2	8.3	52.3
	M	25–30	486.7	208.4	3.7	8.0	10.2	59.5
Gorilla gorilla gorilla	F	50	490.2	224.0	3.5	6.3	9.9	64.5
	M	25	459.4	225.6	3.4	8.6	11.4	76.6
	M	42	564.3	228.8	4.3	7.3	11.0	78.0
Pongo pygmaeus	F	17	311.2	149.8	2.0	5.5	6.3	46.4
	F	23	394.1	170.1	3.1	9.6	9.7	50.2
	F	33	298.6	138.6	2.2	5.6	6.8	36.2
	M	11	374.1	170.8	2.6	9.0	8.5	46.2
	M	24	344.7	144.2	2.5	7.5	7.5	46.4
	M	37	370.3	174.3	3.0	6.8	7.2	43.8
Pan troglodytes	F	13	299.0	142.8	2.7	6.8	6.7	42.0
	F	15	262.0	108.0	2.0	5.8	7.2	43.8
	F	18	343.9	154.7	2.5	6.3	7.6	44.4
	F	19	229.2	142.1	2.5	4.6	5.9	35.6
	F	25	354.3	166.6	3.1	7.9	6.8	44.2
	F	27	314.3	154.0	3.1	6.4	7.4	45.2
	F	33	373.5	165.6	3.1	7.9	7.7	50.2
	F	35	348.1	184.8	2.9	6.2	9.0	50.6
	F	36	355.9	166.6	2.3	6.6	6.5	48.0
	F	37	297.7	129.5	2.5	5.5	6.4	40.0
	F	38	370.7	174.3	3.3	6.7	7.7	42.8
	F	40	345.3	155.4	3.6	7.5	6.8	43.6
	F	41	327.8	152.6	2.9	6.8	9.1	47.8
	F	42	324.2	149.1	3.4	5.8	7.4	48.8
	F	44	332.9	163.1	3.4	6.4	7.0	50.8
	F	45	312.9	168.7	2.8	5.2	6.7	41.0
	F	50	344.6	159.6	3.1	6.5	6.8	45.2
	M	10	353.5	162.4	3.0	7.7	7.4	47.8
	M	17	384.0	200.9	3.1	7.5	7.9	59.8
	M	19	364.6	172.9	3.2	6.5	8.4	60.6
	M	20	414.3	180.6	2.7	9.6	11.0	58.6
	M	39	345.4	149.8	3.0	6.4	7.7	50.4
	M	40	341.2	155.4	3.2	7.2	8.0	41.4
	M	41	377.2	167.3	3.5	7.6	7.5	46.8

captive animals housed in zoological and research facilities. Mountain gorilla brains were collected postmortem by staff of the Mountain Gorilla Veterinary Project and the Dian Fossey Gorilla Fund International from wild animals located at the Parc National des Volcans, Rwanda. A total of eight mountain gorilla brains were obtained; however, only three specimens were preserved entirely free of morphologic distortion (Fig. 1).Volumetric data from these three *G. b. beringei* (a ~25–30-year-old male, a 5-year-old male, and a ~2.5-year-old female) specimens were included with our database of three *G. g. gorilla*, 23 *Pan troglodytes troglodytes*, and six *Pongo pygmaeus pygmaeus*. Only brains from

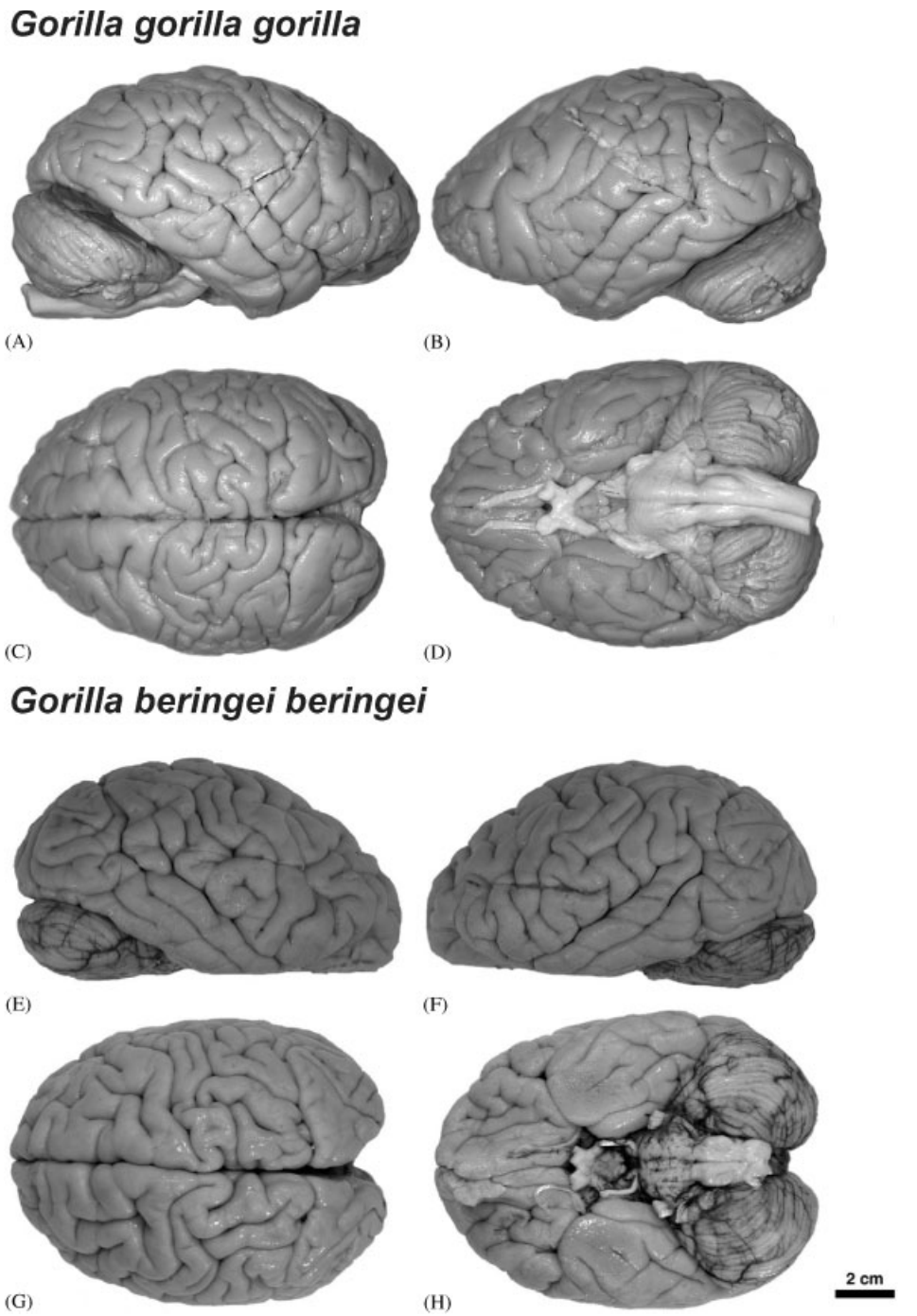

Fig. 1. External morphology of western lowland (*G. g. gorilla*) and mountain gorilla (*G. b. beringei*) brains. An adult male western lowland gorilla brain is shown in right lateral (**A**), left lateral (**B**), dorsal (**C**), and ventral (**D**) views. An adult male mountain gorilla brain is shown in right lateral (**E**), left lateral (**F**), dorsal (**G**), and ventral (**H**) views.

individuals >10 years of age were included, with the exception of the 2.5-year-old female and 5-year-old male mountain gorillas. Adult brain size is attained by 7 years of age in chimpanzees [Herndon et al., 1999] (the only great ape for which there are significant data) several years prior to sexual maturation [Harvey et al., 1987]. Since gorillas and orang-utans reach sexual maturity approximately 3 years earlier than chimpanzees [Harvey et al., 1987], the brain of the 5-year-old male mountain gorilla is probably very close to adult size. The brain of the 2.5-year-old female mountain gorilla should be considered that of a juvenile. Nonetheless, given the rarity of mountain gorilla brains in comparative neuroanatomic studies, we deemed it valuable to retain this individual in our analysis. Furthermore, the statistical results we report were not significantly affected by the inclusion of this specimen.

All animals used in this study had died of non-neurological causes, and all brains were screened for pathology upon radiological assessment. Whole mountain gorilla brains were removed 24–36 hours after death and immediately fixed by immersion in 4% paraformaldehyde. The brains of the other great apes were removed within 12 hr after death and immersion-fixed in 4% paraformaldehyde. All of the brains remained in fixative for a minimum of 2 weeks prior to the MRI acquisitions. Whenever possible, the brains were transferred from fixative to 0.1 M phosphate-buffered saline (PBS) + 0.1% sodium azide after 2 weeks to prevent excessive tissue shrinkage. We accounted for differences among specimens in shrinkage due to fixation by analyzing only relative measures of brain structure volumes as a fraction of whole-brain volume (described below). We did not correct the data for within-brain differential shrinkage of regions or tissue types (gray matter vs. white matter). Studies of formalin-fixed and embedded brains have shown that differential shrinkage of regions and tissue types is minimal. Kretschmann et al. [1982] found a greater amount of shrinkage (mean = 9%) of gray matter compared to white matter in brains that had been embedded in paraffin. Bush and Allman [2004] reported a 0.5–1.6% difference in shrinkage between regions of the neocortex in celloidin-embedded specimens. The error in our data due to differential shrinkage is likely to be even less than that in the previous reports because the brains were not subjected to further processing for embedding. In preparation for scanning, the brain specimens were submerged in a plastic container filled with 0.1 M PBS, vacuumed to remove air bubbles, and packed tightly with gauze to reduce movement artifacts.

MRI Acquisitions

MR images of mountain gorilla brains were acquired on a Siemens 3T Allegra (Siemens Medical System, Erlangen, Germany) running Syngo 2002B software. Coronal T1-weighted MR images were acquired through the entire brain with repetition time (TR) = 2500 ms and echo time (TE) = 4.4 ms with an echo-train of 1. Slices were obtained as 0.7-mm-thick contiguous sections with a matrix size of 256×256 and a field of view (FOV) of 18.0 cm $\times$ 18.0 cm, resulting in a final voxel size of $0.7 \times 0.7 \times 0.7$ mm. MRI scans of other great ape specimens were acquired on a commercial 1.5T GE high-gradient MRI scanner equipped with 8.3 software (GE Medical Systems, Milwaukee, WI). Coronal T1-weighted MR images were acquired through the entire brain with TR = 666.7 ms and TE = 14.5 ms with an echo-train of 2. Slices were obtained as 1.5-mm-thick contiguous sections with a matrix size of 256×256 and an FOV of 16.0 cm $\times$ 16.0 cm, resulting in a final voxel size of $0.625 \times 0.625 \times 1.5$ mm. Due to logistical constraints, different scanners and protocols were used for the mountain gorilla specimens compared to

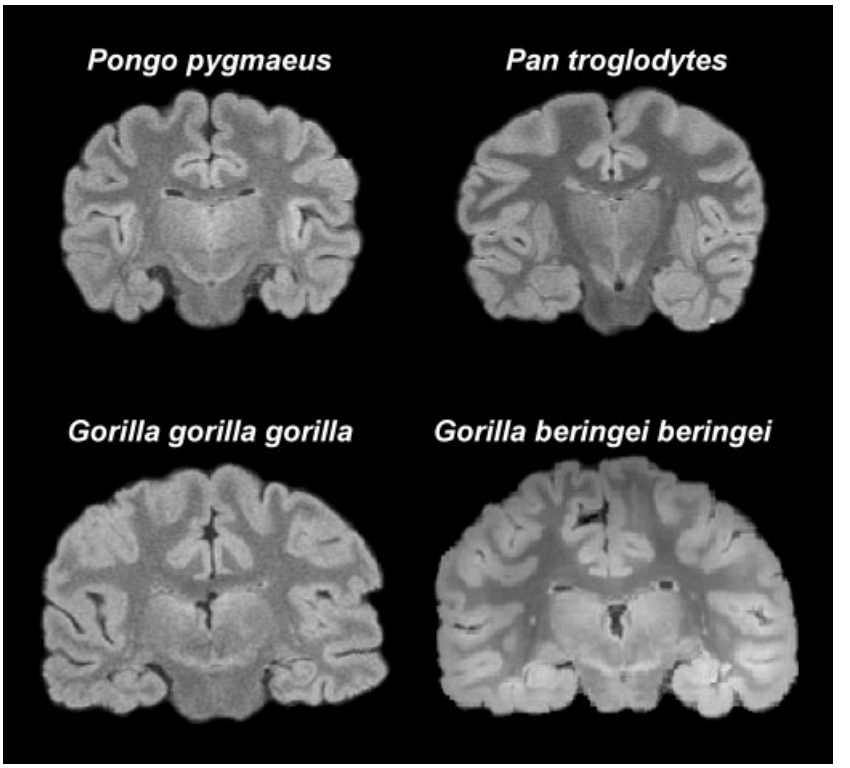

Fig. 2. Coronal T1-weighted MRI sections through great ape brains at the level of the substantia nigra.

other great apes. However, in-plane effective resolution was similar between the samples (0.7×0.7 mm vs. 0.625×0.625 mm), and there was sufficient contrast in all of the images to clearly identify the boundaries of the anatomical structures measured (Fig. 2). All brains were scanned in standard anatomical orientation with the transaxial plane parallel to the anterior commissure-posterior commissure (AC-PC) line and perpendicular to the interhemispheric fissure [Talairach & Tournoux, 1988]. We obtained initial localizing MR images in three orthogonal planes to visualize specimen orientation. We then planned the true coronal scans by orienting the imaging plane perpendicular to the AC-PC line calculated from the localizing images. Data were transferred electronically to eFilm software (version 1.5.3; eFilm Medical, Toronto, Canada) for offline processing. Computer files were numerically coded prior to measurement to prevent observer bias.

ROI Segmentation

All acquired slices were converted into the ANALYZE 3D volume file format, and measurements were performed with the use of MRIcro software version 1.27 [Rorden & Brett, 2000] on a PC workstation. With the MRIcro software package, one can interactively define and edit regions of interest (ROIs) in three orthogonal planes, and accurately segment structures based on visualization of

several anatomical views. We measured the ROI volumes by manually tracing the anatomical boundaries of structures in two-dimensional slices. We calculated the total ROI volume by multiplying the sum of the contour areas in each slice by the interval distance. The caudate nucleus, putamen, thalamus, and hippocampus were measured in all slices in which they appeared. The cerebellum was measured in every other section, and the neocortex was measured in every sixth slice. For ROIs that were not measured in every slice, the slices were sampled in a systematic random fashion. This approach resulted in measurements of at least 12 equidistant slices for each ROI (Fig. 3). Each structure was measured by a single observer who was blind to the identity of the specimen. A subset of five randomly-selected individuals was used to determine measurement reliability for each ROI. Across this set of 30 measurements, intra-rater measurement error was calculated as the percent difference between repeated measurements. Inter-rater measurement error was calculated as the percent difference between measurements by two separate observers. The mean intra-rater measurement error was 4.29%$\pm$3.38% (mean$\pm$standard deviation (SD)), and the inter-rater

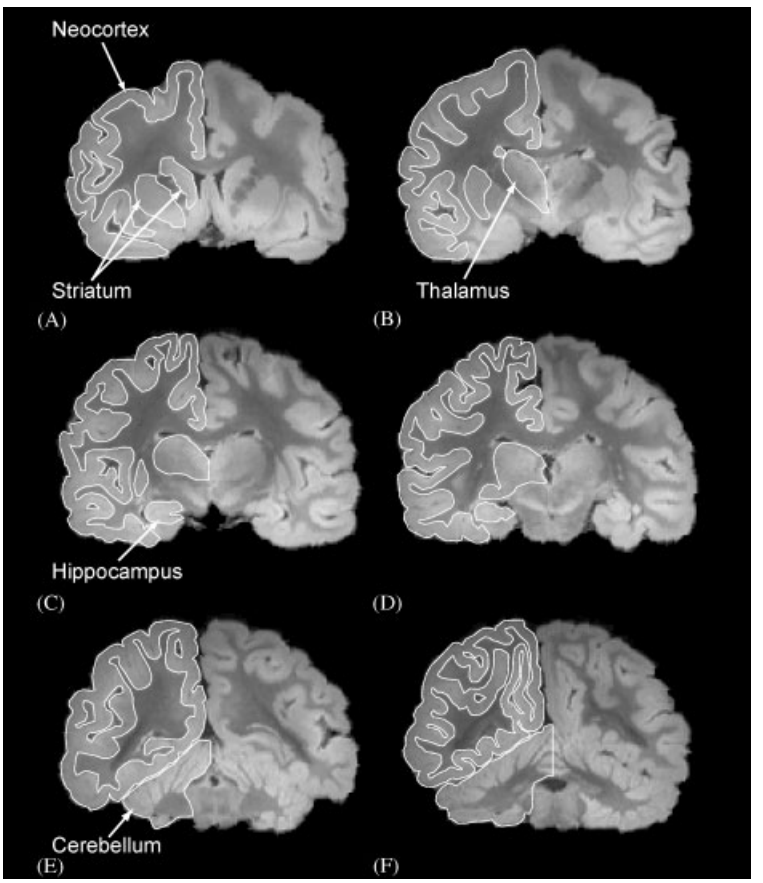

Fig. 3. Coronal T1-weighted MRI sections through the brain of a mountain gorilla. Sections are arranged rostral to caudal (**A–F**). Boundaries of ROI segmentation are shown on the left hemisphere.

measurement error was 8.61% ± 6.34%. The level of measurement error was not correlated with structure size.

The ROIs were segmented according to protocols described in previous volumetric MRI studies of human and nonhuman primate brains [Honeycutt et al., 1998; Lemieux et al., 2000; Matochik et al., 2000; Rilling & Insel, 1999; van Der Werf et al., 2001], so they are described here only in brief. The volume of the striatum was calculated as the sum of the caudate nucleus and the putamen. The caudate nucleus was bounded medially by the lateral ventricle, and laterally by the anterior limb of the internal capsule. To exclude the nucleus accumbens from the caudate measurements, the inferior boundary of the caudate was consistently set as the transaxial level immediately superior to the appearance of the AC. The tail of the caudate nucleus was not included. The putamen was bounded by the external capsule laterally, and the internal capsule and globus pallidus medially. Segmentation of the cerebellum included cerebellar gray matter, white matter, deep nuclei, and peduncles. The main body of the thalamus was bordered medially by the third ventricle, dorsally by the lateral ventricles, and laterally by the internal capsule. The medial and lateral geniculate nuclei were included in outlines of the thalamus. The images had adequate contrast and resolution for us to clearly identify the zona incerta, subthalamic nucleus, substantia nigra, and red nucleus. Therefore, we were able to define the ventral boundary of the thalamus to exclude hypothalamic and midbrain regions. The hippocampal formation, including the dentate gyrus, hippocampus proper, and subiculum, was measured as a single structure. The mesial boundary of the hippocampus was defined where the subiculum transitions into the parahippocampal gyrus. This boundary was delimited as a line running from the underlying white matter to the crown of the parahippocampal gyrus. Tissue located mesial to the uncal notch was excluded from the hippocampal measurement. Finally, neocortical (isocortical) gray matter measurements included the entire cortical mantle, excluding cortex located mesial to the rhinal sulcus. According to these criteria, our definition of neocortical gray matter also includes the proisocortex (cingulate gyrus, rostral insula, and temporal pole), but does not include the periallocortex (entorhinal cortex) or allocortex (olfactory cortex and hippocampus).

Since the sample sizes for the gorillas and orang-utans were small, we did not analyze interhemispheric asymmetry. We added the volumes of bilateral structures to obtain a total ROI volume. For a few cases in which only one brain hemisphere was available (one *G. g. gorilla* and two *P. troglodytes*), we doubled the measurement results to obtain the total ROI volume.

Whole-Brain Volume Measurement

Whole-brain volume was measured with the use of ImageJ software version 1.26t (http://rsb.info.nih.gov/ij/). In each slice, we identified voxels representing gray matter, white matter, and ventricular space, and separated them from the surrounding gauze using a combination of semiautomated thresholding and observer judgment to define the brain's outer margin. The entirety of the cerebral hemispheres, cerebellum, midbrain, and brainstem were included in the measurement of brain volume. The volume corresponding to these voxels was calculated for each slice and summed for the whole-brain volume. Because of the high level of contrast between brain tissue and the surrounding medium, this method yielded reliable estimates of brain volume, with an intra-rater

measurement error of $1.03\% \pm 0.57\%$, and an inter-rater measurement error of $2.92\% \pm 5.12\%$.

Statistical Methods

To compare brain composition among the taxa, we calculated the fraction of total brain volume comprised by each brain structure. Previous studies of relative brain structure volumes normalized in this manner have demonstrated correlations with behavioral traits across mammals and birds [Burish et al., 2004; Clark et al., 2001]. We chose this method because our goal in this study was to assess phyletic variation in the overall organization of brain structures using principal components analysis (PCA) as a data reduction and visualization method. For this purpose, the composite of brain structure fractions represents a trade-off between the relative sizes of the structures that comprise the total brain volume. For the current study, this method was favored over adjusting structure sizes for allometric scaling effects by using residuals from the best-fit line relative to brain volume. The residuals method of normalization has several drawbacks (for discussion see Burish et al. [2004]) that are especially problematic for the current data. In particular, an empirical estimation of the best-fit line is dependent upon the line-fitting method used, as well as the taxonomic level and composition of the reference group [Harvey & Krebs, 1990; Holloway & Post, 1982]. This can lead to idiosyncratic artifacts based on sampling that are difficult to interpret in a strictly biological sense. Since the current data set consists of only a few closely related taxa, with uneven samples of individuals within each taxon, it is unclear how a best-fit line to these data would afford an unbiased correction of allometric scaling. In contrast, when one normalizes structure sizes to overall brain size by taking a simple fraction, their values are not affected by the ambiguities associated with estimating interspecific scaling relationships. In addition, a further advantage of using ratio values is that they serve as an internal correction for variation in shrinkage due to fixation differences across specimens. Despite these advantages, however, ratio measures have been criticized for being prone to autocorrelation, with the largest structure in the brain (e.g., neocortex) disproportionately determining the other ratio values. In our data set, however, the neocortex fraction was not correlated with the total brain volume ($r = -0.239$, $P = 0.167$).

RESULTS

As previously observed, the overall shape of the brain in both western lowland and mountain gorillas is dorsoventrally flattened, with a less globular frontal lobe than that in other great apes [Connolly, 1950]. The external sulcal patterns of mountain gorillas ($n = 8$) also closely resemble those of western lowland gorillas (Fig. 1). In specific, both taxa tend to have a continuous inferior precentral sulcus that runs parallel to the central sulcus. Chimpanzees, on the other hand, exhibit a greater degree of variation in the pattern of sulci in the region of the inferior frontal gyrus, with more frequent bifurcations and discontinuities of the inferior precentral sulcus [Sherwood et al., 2003]. Other aspects of mountain gorilla sulcal anatomy closely resemble patterns observed in other great ape species. The superior parietal lobule is highly fissurated, and the lunate sulcus can be seen on the dorsolateral surface of the occipital lobe in the typical anterior location of most anthropoid species. Unfortunately, postmortem

gross anatomic distortion prohibited examination of petalia patterns in the majority of the specimens.

The results of volumetric measurements based on MRI of postmortem formalin-fixed specimens are provided in Table I. Species mean brain volumes in the present study fall within the range of species means found in the published literature based on fresh postmortem tissue, in vivo MRI, and cranial capacity [Herndon et al., 1999; MacLeod et al., 2003; Rilling & Insel, 1999; Stephan et al., 1981; Tobias, 1971; Zilles & Rehkämper, 1988]. It should be emphasized, however, that the volumetric data reported in Table I are subject to shrinkage artifacts from fixation effects in individual cases. Therefore, our analyses were performed on brain structure volume fractions, which take overall brain volume shrinkage into account. Figure 4 shows the distribution of brain structure fractions in each taxon. Across all specimens, the sum total of regional brain

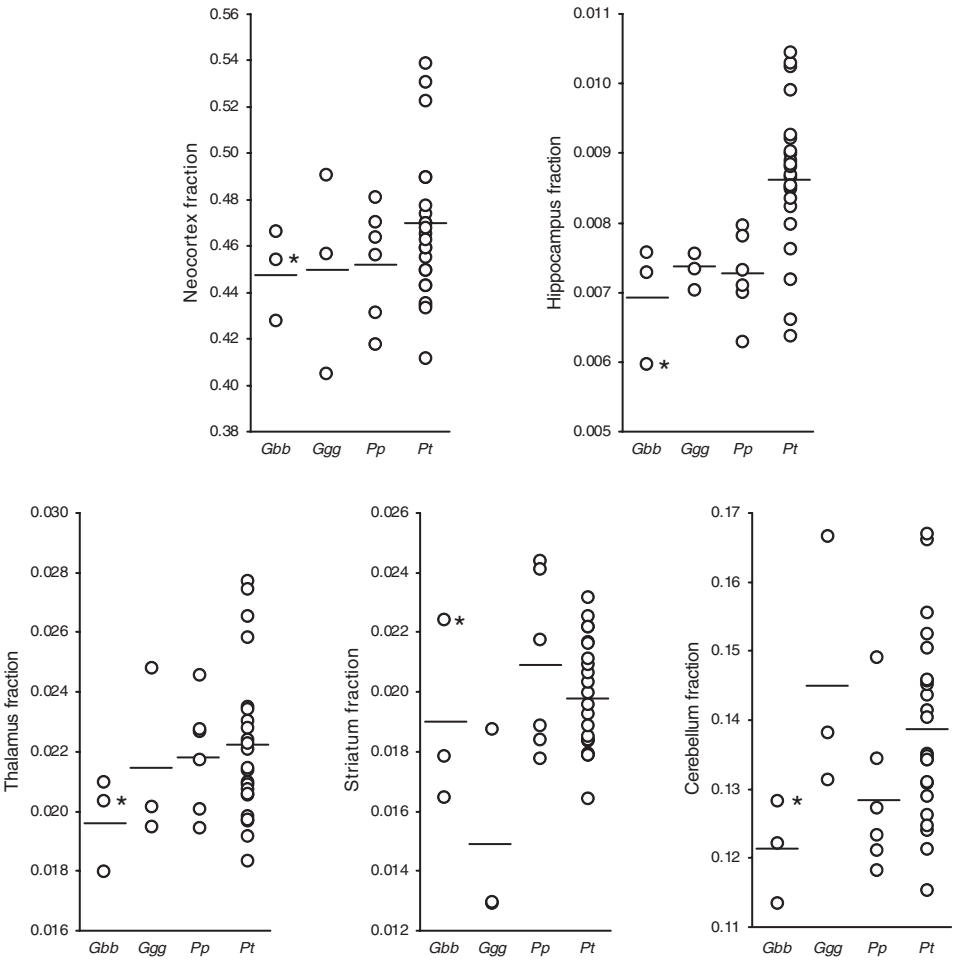

Fig. 4. Distribution of brain structure fractions. Means are indicated by horizontal lines. *Gbb, Gorilla beringei beringei; Ggg, Gorilla gorilla gorilla; Pp, Pongo pygmaeus; Pt, Pan troglodytes.* The 2.5-year-old female mountain gorilla data are indicated by an asterisk.

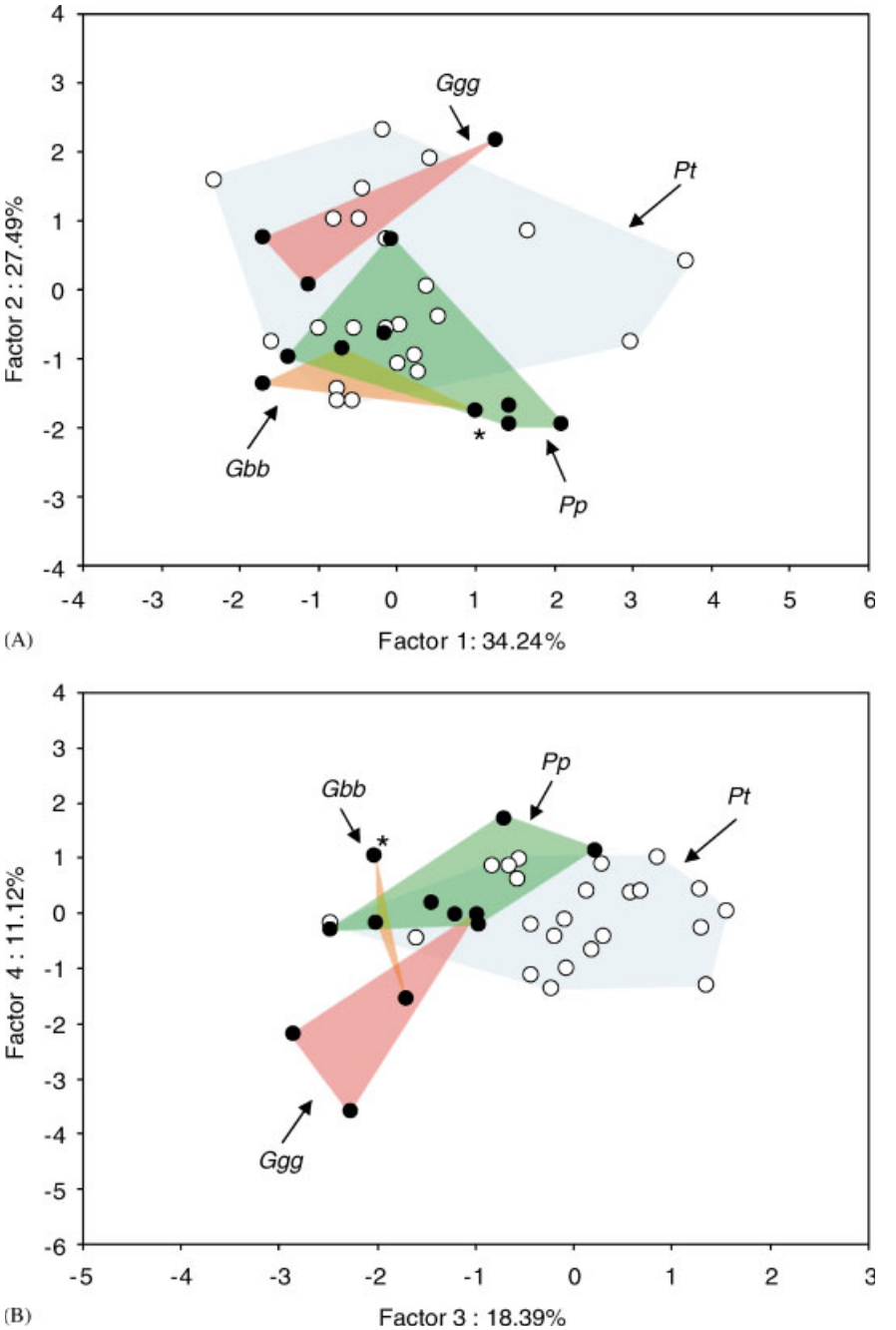

Fig. 5. PCA plots of brain structure fractions (shown in Fig. 3). The PC axes were calculated from only *P. troglodytes* data. All great ape data are plotted in the vector space defined by chimpanzees. *Gbb, Gorilla beringei beringei; Ggg, Gorilla gorilla gorilla; Pp, Pongo pygmaeus; Pt, Pan troglodytes.* The 2.5-year-old female mountain gorilla data are indicated by an asterisk.

TABLE II. Principal Components Analysis (PCA) Loadings: Based on the *P. troglodytes* sample

Variable	Factor 1	Factor 2	Factor 3
Neocortex fraction	−0.551	0.633	−0.051
Hippocampus fraction	−0.449	0.071	0.880
Striatum fraction	0.607	−0.525	0.332
Thalamus fraction	0.750	0.439	0.138
Cerebellum fraction	0.525	0.707	0.118
Percent of variance	34.24%	27.49%	18.39%

structure volumes constituted 66% $\pm$ 5% of brain volume. Of note, for every brain structure, the range of values in the chimpanzee sample encompassed much of the range of variation found in all other species. A one-way analysis of variance (ANOVA) of genus-level differences in brain structure fractions revealed that chimpanzees have a significantly larger hippocampus fraction than the other apes ($F_{2,32} = 9.79$, $P < 0.001$; Bonferroni post hoc: $P < 0.01$), and gorillas have a smaller striatum fraction ($F_{2,32} = 4.69$, $P = 0.016$; Bonferroni post hoc: $P < 0.05$). No other comparisons were significant.

We defined a multidimensional space by calculating the PCs from the correlation matrix of brain structure fractions in the sample of 23 chimpanzees. Only the chimpanzee sample was used to compute the PCs. Because of its relatively large sample size, it is assumed that the chimpanzee sample best captures the variance structure among brain parts. Brains from young as well as elderly adults of both sexes were included in the chimpanzee sample. Therefore, our calculation of variation in the chimpanzee data includes potential sex- and age-related differences in brain composition. To determine whether there were systematic age or sex differences in our data, we replotted points in the multivariate space and labeled them according to age class and sex (data not shown). Individuals were not differentiated on the PC functions according to these denominations. Data from all great ape taxa were then plotted onto the vector space defined by chimpanzees, and minimum convex polygons were constructed to illustrate morphometric distances (Fig. 5).

The first three PCs accounted for 80.12% of the total variance in the chimpanzee data, and were used for further data interpretation. Table II displays the factor loadings of these PCs, as well as the percentage of the variance in chimpanzees accounted for by each PC axis.

PC 1 shows marked overlap among gorilla and orang-utan individuals, and contains all the variance of these great ape species within the subspace defined by chimpanzees. This factor loads fairly equally on all variables. It is positively correlated with subcortical structures (i.e., the thalamus, striatum, and cerebellum), and negatively correlated with cortical structures (i.e., the neocortex and hippocampus), indicating the reciprocal nature of the largest dimension of variation in great ape brain composition.

PC 2 has positive loadings on the cerebellum, thalamus, and neocortex, and loads negatively on the striatum. Like PC 1, this important axis also captures most of the variance in other great apes within the range of chimpanzees, although a few orang-utans fall outside this region in the negative direction. Of note, *G. g. gorilla* and *G. b. beringei* occupy opposite poles along PC 2, with no overlap between them. Orang-utan data points on this axis are located

across the range defined by both gorilla taxa, with the bulk clustering with *G. b. beringei*.

PC 3, which loads predominantly on the hippocampus, separates a large cloud of chimpanzee data from the data of other great apes. On this PC, the *G. b. beringei* points are contained within the range of *G. g. gorilla.* As already indicated, chimpanzees have a larger hippocampus fraction than other great apes.

DISCUSSION

This study reports a comparison of brain structure composition in mountain gorillas and other great ape species based on volumetric MRI and PCA. We used a well-represented chimpanzee sample to calculate the PC axes, and plotted the other great ape data within this multidimensional space. Overall, this analysis shows marked commonalities across great ape taxa in terms of brain structure. Nonetheless, subtle anatomic differences may distinguish the brains of mountain gorillas and other taxa.

Levels of Variation

We found that most of the variance in mountain gorillas, as well as western lowland gorillas and orang-utans, is contained within the chimpanzee range along the first two PC axes, which together account for 61.73% of the variance. This suggests that there is a common *Bauplan* to great ape brain macrostructural organization, and most of the observed interspecific variation does not extend beyond the level of normal interindividual within-species variation, as seen in chimpanzees. This result is also interesting considering that our sample represents the added variance of including both wild and captive animals. Unfortunately, the possible effects of impoverished captive environments cannot be disentangled from phylogenic differences because the only wild individuals in the sample were of the same taxon, *G. b. beringei*. However, it is worth noting that the data points from these wild mountain gorillas were found within the range of variation defined by the captive chimpanzee and orang-utan polygons in multivariate space.

An important feature of the present study was the inclusion of a fairly large sample of chimpanzee brains ($n = 23$). To date, these data represent the largest sample of volumetric brain proportions available for any great ape species. Therefore, our findings call attention to potential sampling errors that may confound other studies based on limited sample sizes. For example, the data on hominoid brain structure volumes reported by Stephan and colleagues [1970, 1981] were derived from a single individual representing each of the following species: *Hylobates lar*, *Gorilla gorilla* (*Gorilla beringei graueri*), *Pan troglodytes*, and *Homo sapiens*. Nevertheless, this data set has been employed by many authors to draw conclusions about the coevolution of neuroanatomical structure and a panoply of life history and socioecological adaptations in great apes and humans [e.g., Dunbar, 1992; Joffe & Dunbar, 1997; Reader & Laland, 2002; Sawaguchi, 1992]. In these cases, however, the effects of sampling error may have profound consequences for data interpretation. As an illustration of this problem, consider what could happen if a single mountain gorilla were selected at random from our small sample. Depending on which individual was selected, its brain proportions may either be contained entirely within the chimpanzee range of variation along all PC axes, or, alternatively, it may fall outside the chimpanzee range for several PC factors. In sum, the present findings on

intraspecific variation in chimpanzees indicate that researchers should exercise considerable caution in making conclusions about species-typical attributes when only small intraspecific samples are available for obtaining data on brain structure.

Potential Variation Among Mountain Gorillas, Western Lowland Gorillas, and Other Great Apes

Based on the small samples currently available, our preliminary findings suggest that there are some differences in brain structure among great ape species. Mountain gorillas and western lowland gorillas were separated along PC 2, an axis that loads positively on the cerebellum and neocortex, and negatively on the striatum. Here, *G. b. beringei* individuals are differentiated from *G. g. gorilla* individuals in that the former have a relatively smaller cerebellum in combination with a relatively larger striatum. Previous comparative volumetric studies have noted that, compared to other hominoids, *G. g. gorilla* have a proportionally large cerebellum relative to brain size [Rilling & Insel, 1998; Semendeferi & Damasio, 2000; Stephan et al., 1981]. Our data corroborate and extend these observations. Although we found that *G. g. gorilla* has a relatively large cerebellum compared to other great apes, we also found that the cerebellum of *G. b. beringei* is relatively small. Interestingly, chimpanzee and orang-utan data were plotted along PC 2 between the two gorilla taxa. Another striking pattern was the distribution of data along PC 3. This is a less important factor, but one that is associated most strongly with relative hippocampus size. Individuals of *P. troglodytes* were separated from other great apes on this PC axis, based on a large hippocampus relative to brain size.

Our data suggest that the brain structure of great apes differs mostly in the relative sizes of the striatum, cerebellum, and hippocampus. These findings are especially interesting considering the central role of these structures in ecological and locomotor functions. Volumetric studies in a range of animals, including polygynous male voles [e.g., Jacobs et al., 1990], a number of food-storing birds [e.g., Krebs, 1990; Shettleworth, 2003], and London taxi drivers [Maguire et al., 2000] have shown that the size of the hippocampus, a brain region essential to spatial learning and memory, is relatively enlarged in individuals who face ecological challenges that place extra demands on this cognitive capacity. Hence, the larger hippocampal size in chimpanzees may reflect a greater dependence on a frugivorous diet [Malenky et al., 1994] and the accompanying increased demands on spatial memory; however, this is speculative. The size of the lateral cerebellum, the largest component of the cerebellum in hominoids, has been linked to locomotor adaptations for the planning of sequential movements, such as those employed in arboreal quadrumanuous climbing, and suspensory postures when moving in trees [MacLeod et al., 2003]. The more terrestrial lifestyle of mountain gorillas compared to western lowland gorillas [Doran & McNeilage, 2001] would appear to be consistent with our finding of a relatively smaller cerebellum in this taxon. However, it is unclear why orang-utans cluster with mountain gorillas on PC 2, as they are highly skilled at quadrumanuous clambering in tree crowns [Fleagle, 1999; Povinelli & Cant, 1995], a feat that would seem to require the type of precise timing of motor sequences supported by cerebellar circuits.

Interpretations of macrostructural variation in these taxa must remain provisional until further specimens can be analyzed and sample sizes are increased. Furthermore, histological studies of cyto- and chemoarchitectural

staining patterns are needed to investigate possible microstructural specializations of the mountain gorilla brain. In this respect, the preliminary data reported here underscore the need to study these unique animals. Considering the critically endangered conservation status of mountain gorillas, documentation of their biological distinctiveness is essential for their management and protection. In the present study, we took the opportunity afforded by a collaborative research effort including field veterinarians, hospital radiologists, anthropologists, and neuroscientists to provide these first data on the brain organization of mountain gorillas. This collaborative model should be applied to further our knowledge concerning the biology of other endangered species.

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