Diffusion tensor imaging of the carnivora brain: A pilot study

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Abstract

Fiber tractography is relatively novel method for reconstructing the course and location of white matter tracts in the mammalian brain. While this approach has been applied extensively in human studies, very few studies have looked at the application of this approach to the study of non-traditional animal models. Here we look to explore the use of fiber tractography to reconstruct the major commissural, association and projection fiber pathways in a range of closely related Carnivora. The null hypothesis is that fiber pathways in closely related species of carnivora have remained largely unchanged and are indicative of the constraints (architectural, developmental or phylogenetic) that unite species within a given Order. Alternatively, deviations from this known bauplan are interpreted as species specific variation, which is directly related to the unique ecological context of each species (i.e., hunting strategies, social organization and or cognition).

Introduction & Background

Since their evolutionary divergence in the early to late Eocene, canids and felids have developed separate morphological and behavioral adaptations, particularly focused on the honing of hunting and prey capture abilities (Van Valkenberg, 1985; Nowak, 1991). A flurry of recent work on the brain and nervous system of wild canids and felids have built an important framework for comparison and have revealed both striking similarities (e.g., Oddes et al., 2023; Nguyen et al., 2020) in form and function as well as unique species-specific attributes which underlie the behavioral ecology observed within this clade (e.g., Grewal et al., 2020; Chengetenai et al., 2020; Jacobs et al., 2018).To date, no comparative diffusion tensor imaging studies have been undertaken on the carnivora brain. Consequently, there is a gap in our knowledge about how fiber differences across canids and felids are related to variation in brain size or behavioral ecology.

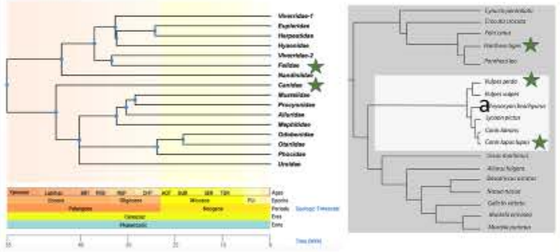

Figure 1: Phylogenetic trees showing the evolutionary relatedness of the three species used in the current pilot study. Drawn using TimeTree.org.

Figure 2: a- An Image of a Siberian tiger (*Panthera tigris*); b- adult fennec fox (*Vulpes fennecus*); c- domestic dog (*Canis lupus lupus domesticus*)

Materials & Methods

Magnetic resonance (MR) imaging was performed on a postmortem whole brains of 3 adult (male) Carnivora specimens obtained through collaborative links with Blank Park Zoo, St Louis Zoo, Iowa State University . MR imaging was performed in the Department of Radiology, Icahn School of Medicine at Mount Sinai, NY. In brief, post mortem fixation was performed within 4 hours of death 94% paraformaldehyde for 10 days). Before scanning, the brain was bathed in Fomblin solution and packed in gauze in a container for MR image acquisition on 7 T Bruker Biospec MR System. A 3D FLASH sequence was performed: (TR=36ms, TE=23ms, Flip angle=15 degrees, FOV=8cm, matrix size 384*384*384). Structural scan data was converted to Analyze format for post processing and image capture. Image intensity was optimized for visualization Of the 384 images in the image stack, thirty of the images, (roughly every 13th image) was saved to the work station for identification and labelling.

Segmentation & Diffusion Tensor Imaging: For white matter imaging we used a Diffusion Tensor Imaging (DTI) sequence with 515 b-directions/values: TR=25,000ms, TE=32.6ms, maximum bvalues=0-8000s/mm2, FOV=8cm, matrix size 128x128x80. The total acquisition time was 18.5h for the specimen. Fractional anisotropic (FA) maps and vector files were generated and tensor files were imported into DTI-Query for tractography of select fiber tracts.
3D brain volumes were also reconstructed using ITKSNAP. The T2 weighted images were loaded and outlined using semi-automatic segmentation via thresholding. Resulting segmentations of the hemisphere were smoothed in Meshlab before screenshots were taken for visual documentation and comparison.

Results

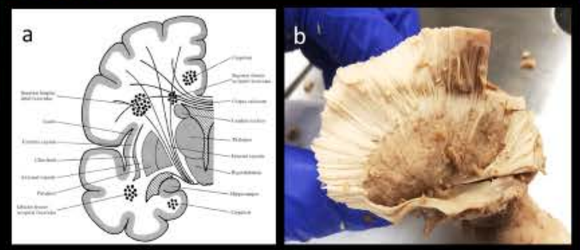

Figure 3: a- Schematic showing the spatial relationship of the corpus callosum to the internal capsule and basal nuclei; b-Sharp dissection followed by blunt dissection of the lateral cortical surface in the human to reveal the fibers of the internal capsule and adjacent putamen

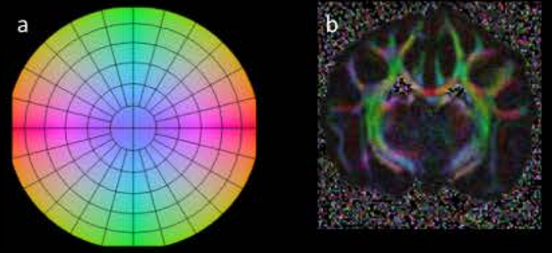

Figure 4: a- Image showing the color wheel used for interpreting and displaying the FA maps; b- also shown is representative coronal FA image through the dog brain at the level of the thalamus. Color code: RAS->RBG. Color schemes produced in MIPAV in accordance with Pajevis & Pierpaoli (1999).

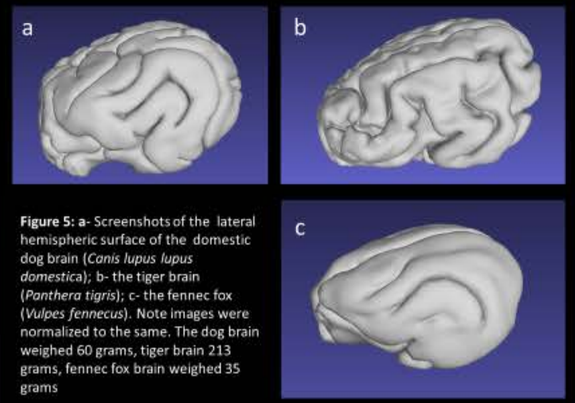

Figure 5: a- Screenshots of the lateral hemispheric surface of the domestic dog brain (*Canis lupus lupus domestica*); b- the tiger brain (*Panthera tigris*); c- the fennec fox (*Vulpes fennecus*). Note images were normalized to the same. The dog brain weighed 60 grams, tiger brain 213 grams, fennec fox brain weighed 35 grams

Results

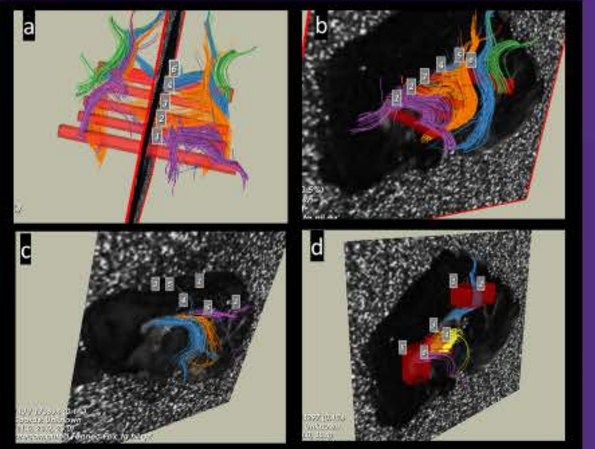

Figure 6: A series of screenshots taken in DTI QUERY after whole brain seeding and tractography of the corpus callosum (a-g), internal capsule and corona radiata (h & i) in the tiger, domestic dog and fennec fox .

For the corpus callosum, VOI's were placed equi distances apart within the major segments of the corpus callosum

Conclusion

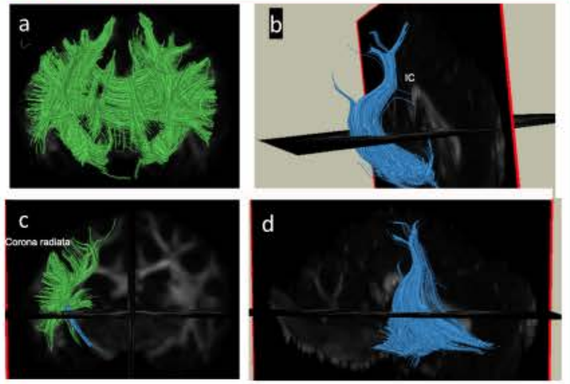

Figure 7: Tractography using whole brain seeding followed by dynamic querying to reveal the internal capsule and corona radiata of the canid

We noted that VOI's that were were placed equi distances apart within the major segments of the region of interest performed better at reconstructing the underlying fiber pathways. Such targeted "seeding" prevented the inclusion of projection fibers into other commissural pathways.

We did note some individual variation in our ability to fully resolve the underlying fiber pathways. We believe this variation might be due to fixation differences prior to scanning. In follow-up studies our goal is to resolve the major commissural, projection and association fibers shared amongst the Carnivora and investigate if there are any fiber density/ number differences as well as differences in fractional anisotropy.

Further explorative analysis of the major fiber pathways is necessary to demonstrate the consistency of the approach and help identify if there exists any species-specific fiber pathways which underlie the behavior of each species.

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Acknowledgments

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