WHAT WE DO

Present studies and future directions

Our laboratory is currently involved in two major areas of research. The first area includes our studies on the evolution of complex nervous systems, and the genetic and activity dependent mechanisms in development that contribute to phenotypic variability. The second area includes our studies of areas of the neocortex in human and non-human primates involved in manual dexterity, bilateral coordination of the hands, proprioception, and the generation of body centered coordinates for goal directed reaching.

I. The evolution of complex brains in mammals: Genetic and activity dependent mechanisms that contribute to the phenotype.

The driving question of these studies is: How are large brains with multiple, functional parts generated, and what is the evolutionary (inherited, genetic) component of these changes?
We know that mammalians brains differ in size, and that mammals with large brains generally have more complex sensory, perceptual, and cognitive behaviors than mammals with small brains. Comparative studies on the organization and connections of the neocortex, including those done in our laboratory, indicate that size alone is not the source of this variation. Rather, it is the number of functional areas and their connections that are responsible for the remarkable differences in complex behaviors. This is particularly true for the neocortex. Fossil records together with comparative studies indicate that the first mammals had small brains with a very little neocortex, which had only a few areas. Thus, small brains with few parts were the forerunners of the large, complexly organized brains. The following group of studies are designed to determine the processes by which this occurs.
Because we cannot study evolution directly we have examined the products of the evolutionary process and make inferences about the process itself. In our previous comparative studies we used electrophysiological recording techniques and neuroanatomical tracing techniques in a variety of mammals to examine the functional organization of the neocortex. This allowed us to ascertain which features of organization and connection patterns are the same across mammals, due to common ancestry (homologous), and which features are derived. More important, these studies allowed us to examine the types of changes to the neocortex that occur in evolution, and the constraints imposed upon the evolving neocortex. From these experiments we generated a number of hypotheses regarding brain evolution. Recently we began to test our hypothesis by manipulating, in developing brains, what we believed was being manipulated genetically in evolution. The types of genetically mediated changes that normally occur, and which our comparative studies suggest account for much of the phenotypic variability observed in extant mammalian brains, include changes in the size of the cortical sheet, changes in the number of cortical fields, and changes in peripheral morphology (e.g. receptor number, distribution density and type).

Studies in Monodelphis domestica

In our first series of experiments we directly altered the size of the developing cortical sheet before the arrival of thalamic afferents to determine the extent to which the development of cortical areas, is dependent on mechanisms extrinsic to the neocortex, such as the presence of thalamic input (Huffman et al., 1999). These experiments resulted in a shifting of thalamocortical afferents on the reduced cortical sheet. Thus, our studies demonstrated that with a change in the size of the cortical sheet, cortical field boundaries can be shifted dramatically. This year, we are beginning a series of experiments in which we will increase the size of the cortical sheet by transplanting age matched embryonic tissue from GFP mice into Monodelphis domestica. This will be done very early in development, before any CNS connections have formed. We hope

to determine the extent to which an increase in the size of the cortical sheet alone might account for changes in cortical field number.

Because we appreciate that changes in the size of the cortical sheet alone are insufficient to explain the large differences in cortical territory assumed by different sensory systems (sensory domains) in mammals with variable peripheral morphology, we have begun a new set of experiments designed to examine the role of the periphery in the allotment of sensory domains in the neocortex. These experiments are done in two different animals, Monodelphis domestica and congenitally deaf mice. In Monodelphis domestica, we have dramatically altered the pattern of activity of the sensory epithelium in different sensory systems by removing all or most of a receptor surface (bilateral enucleation; bilateral vibrissae removal). These removals were performed well before any connections are formed from the eye or from the vibrissae to the thalamus. These experiments bias the impact of one sensory system over another, and allow us to determine the extent to which the neocortex is capable of being reorganized in the absence of any direct alterations to the central nervous system. We have already performed some of these manipulations and have recently published a paper on the effects of early loss of visual inputs on the cortex (Kahn and Krubitzer, 2002). In these animals we use electrophsyiological recording techniques and neuroanatomical methods to determine the extent to which sensory domains have shifted across the cortical sheet. Our results indicate that sensory domains and the size of cortical fields therein appear to be largely regulated by peripheral inputs and associated patterned activity. However, some aspects of cortical architecture and connectivity appear to be highly constrained by intrinsic (likely genetic) mechanisms.

To exacerbate the effect of changes in peripheral morphology and relative activity patterns across the sensory epithelium, in future experiments we plan to perform bilateral enucleations in conjunction with vibrissae transplants in the same animal, or bilateral vibrissae removal + eye transplants (which have been performed successfully in Australian marsupials). These types of manipulations should result in an extreme re-weighting of the activity patterns from different peripheral receptors, much like that observed in highly derived mammals such as blind mole rats. Our objective is to create the types of morphological changes that occur naturally in evolution. In this way, we can determine the limits to which sensory domains are capable of shifting in the absence of direct alteration to the central nervous system, and if the changes to the neocortex reflect the types of cortical organization observed in highly derived extant mammals.

Studies in congenitally deaf mice

A new series of experiments designed to examine the specific contribution of patterned activity to the organization, function and connectivity of the neocortex has recently begun in our laboratory using mutant mice, which are congenitally deaf (NKCC1-/-). In these animals, changes in the functional organization of the neocortex and connections of subcortical structures are being examined (Hunt et al., 2002; Litinas et al., 2002; Punj et al., 2002). We are also interested in how activity might alter gene expression, which in turn regulates synaptic morphology and connectivity. To address this, we have begun a collaboration with Dr. John Rubenstein at UCSF. Dr Rubenstein is internationally known for his work on the contribution of genes to the development of structural boundaries and connectivity in the CNS. In his laboratory in situ hybridization techniques are used to determine how loss of patterned input from the cochlea (occurring during development) affects the spatial and temporal pattern of gene expression in cortical and subcortical structures, and how this in turn alters the connectivity and function of auditory structures. We are excited by the potential these studies hold for understanding the extent to which the developing nervous system is capable of reorganization, and the specific anatomical and functional changes that occur in congenitally deaf individuals.

Studies of gene expression in highly derived mammals

We have recently begun a very large collaborative effort with three other laboratories to compare patterns of gene expression in the cortex of highly derived mammals with patterns described in the mouse. Because highly derived mammals, such as echo locating bats and naked mole rats have a remarkably different cortical organization than that of the mouse, and because this organization strongly reflects peripheral morphological and behavioral specializations, studies of this sort will help unravel the precise contribution of genes vs. activity in the emergence of cortical fields in development. They will also allow us to understand the genetic changes in cortical and subcortical structures, which contribute to the phenotypic variability observed in the brains of extant mammals. This work is done in collaboration with Dr. Ellen Covey, Department of Psychology, University of Washington, Seattle, WA, Dr. Ken Catania, at Vanderbilt University in Nashville, TN, and with Dr. Rubenstein at UCSF.

II. Studies of the primate somatosensory system.

One of the most unique behaviors exhibited by humans is the ability to manipulate the world around them with their hands. While most mammals actively explore the world by making fine tactile discriminations with specialized body parts that manipulate objects of interest, this ability is particularly well developed in primates. We know that discriminations occur with the glabrous hands, lips and tongue in primates. However, the portions of the central nervous system, particularly the neocortex, that participate in the generation of tactile perception, recognition, proprioception, and goal directed reaching have yet to be elucidated. Our work examines the contribution of the somatosensory system to the generation of these behaviors.

We appreciate that even for seemingly simple tasks such as object manipulation and discrimination, inputs from all parts of all digits, from both hands must be integrated. In addition, the somatosensory system must provide the motor cortex, posterior parietal cortex, and subcortical structures with relevant information regarding the position of the limbs in space, and the location of contact at different points across the hand. It must also provide the motor cortex with feedback regarding changes in position of the digits of the hand, and stimulation patterns across the hand. In this way, exact motor commands specifying how and where to move digits for further exploration can be initiated. To actually discriminate between a novel object and something recognized from a previous experience, information would need to reach regions of the brain involved in memory, such as entorhinal cortex, the amygdala, and the hippocampus. Finally, for characteristics such as perception and cognition, the somatosensory system must provide information to areas of the cortex involved in attention, future planning, and non-stimulus bound task performance. We know from previous work that at least nine different cortical areas process inputs from the skin, muscles and joints. In order to understand the complex interactions of cortical fields and thalamic nuclei necessary for sensory discrimination, sensorimotor integration, manual dexterity, bilateral coordination and goal directed reaching, it is critical to understand the details of the circuitry that subserve such functions.

There are two major goals of this work. The first is to establish a reliable monkey model for the organization, interconnections and neural processing in somatosensory cortex of the lateral sulcus and posterior parietal cortex. For these experiments, we are using macaque and titi monkeys. The second is to use this model as a framework to generate testable hypothesis regarding the number, organization, and functions of areas of the lateral sulcus and posterior parietal cortex in humans.

Studies of non-human primates

In our monkey experiments, we have begun to describe in detail the organization and interconnections of areas in posterior parietal cortex (PP; areas 5 and 7a) that are involved in integrating inputs from behaviorally relevant body parts, and in generating body centered coordinates necessary for active reaching into immediate extra-personal space (Disbrow et al., 2001; Padberg et al., 2002. Areas in the lateral sulcus (LS), such as S2, PV, VS and RL are involved in integrating inputs from both hands and are thought to be involved in tactile discrimination and tactile recognition. In these experiments (in PP and LS), we use multiunit and single unit electrophysiological techniques to determine the types of stimuli that best activate these neurons, and examine the specific patterns of interconnections that characterize cortical fields such as 3a, 3b, 1 and 2, and thalamic nuclei such as VP (e.g. Disbrow et al., 2002). In this way, we can determine the precise body part representations that project to different fields. Ultimately, strict topographic interactions between higher order fields and those that provide input to these fields will not be maintained, since integration across different body parts is critical for bimanual coordination, and behaviorally relevant tasks that involve the hand and mouth. Our work indicates that there are differential interconnections from behaviorally relevant body part representations from proprioceptive areas such as 3a, to areas of posterior parietal cortex (Huffman and Krubitzer, 2001a; 2001b).

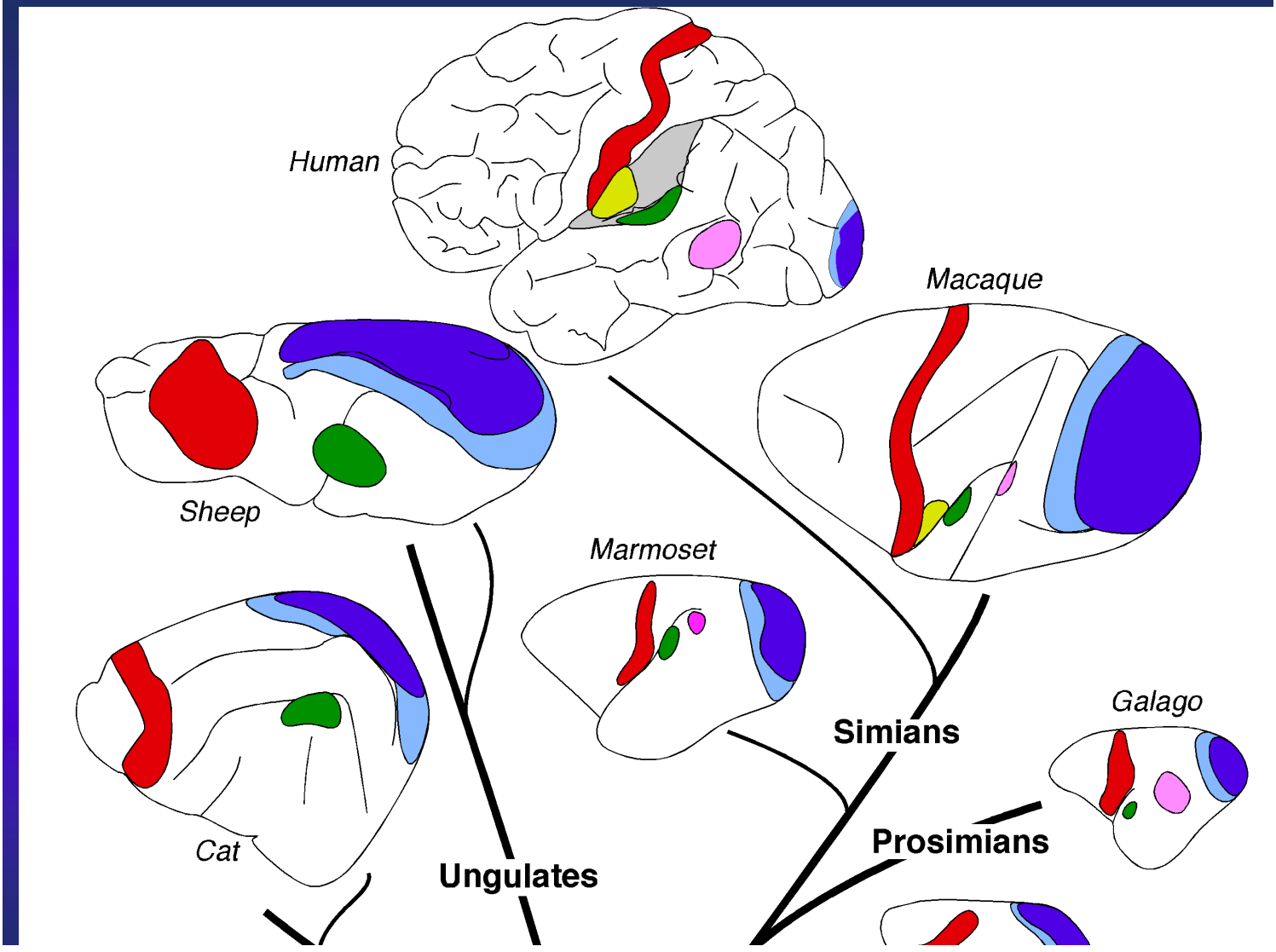
Recently we have begun a series of electrophysiological and anatomical studies in anterior and posterior parietal cortex in titi monkeys. Our results indicate that areas of cortex that were traditionally believed to be involved solely in somatosensory processing, also process visual inputs (Padberg et al,. 2002). These areas are hypothesized to be involved in visually guided reaching and grasping. To examine the role of some of these areas in specific aspects of this behavior, we have recently begun a collaboration with Drs. Bill Mason and Sally Mendoza from the Department of Psychology, UCD in which we will train animals on visually guided reaching tasks, lesion a particular area of interest, and examine the effects of our lesion on the reaching behavior. Ultimately, we would like to examine similar reaching and grasping behaviors in humans with lesions in similar cortical locations to those in titi monkeys.

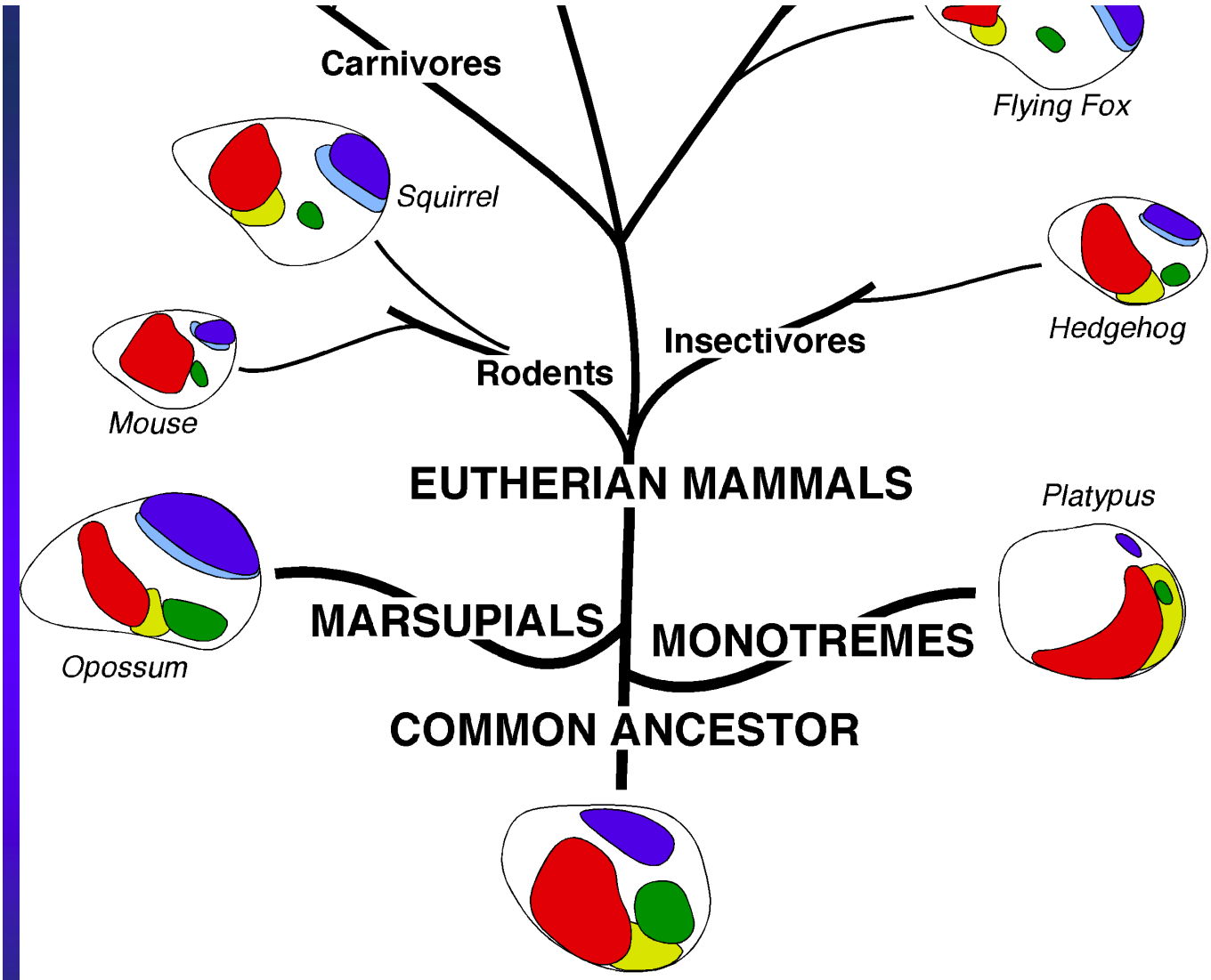
Studies of human primates

The use of non-invasive imaging techniques has revolutionized the field of neuroscience and has allowed us to directly study the activity of the human brain. Despite the recent technological advances in human imaging, there are still a number of questions regarding cortical field organization and connections that can only be answered using invasive, electrophysiological recording techniques. For these reasons, our laboratory has established a parallel monkey/human model to examine the somatosensory areas of the cortex involved in the abilities described above. Our studies in humans, which are done in collaboration with Dr. Timothy Roberts (UCSF), are naturally guided by studies in monkeys, which provide details on where such information is processed, and how cortical areas involved in these abilities are organized and interconnected.

Using fMRI, we have begun to explore regions of both the posterior parietal cortex and lateral sulcus to determine if they are organized in a similar fashion to those in monkeys (Disbrow et al., 2001; Hinkley et al., 2002). We used simple stimuli to examine the topographic organization of fields (using our results from monkeys to help interpret results in humans). This approach has proven very successful in our initial studies in human lateral sulcus in which the location and internal organization of S2 and PV have been described. The location, general types of stimuli that drive neurons, and the expected topographic organization of areas to be studied in humans will be garnered from our studies in monkeys. In our future experiments in humans, we will characterize the specific types of stimuli that produce maximum activation in a particular cortical field, using the controlled stimuli used in our monkey experiments. Again, these experiments will be driven by the results of single unit experiments in monkeys.

The major strength of our studies lies in linking our monkey data with human data. In short, we can directly test in humans a number of hypotheses regarding cortical field organization and function generated from our monkey experiments. In monkeys, we can extend our current understanding of posterior parietal cortex and lateral sulcus by defining the anatomical substrate for the complex neural responses identified in these areas. While either group of studies alone would be worthwhile, the combination of techniques, and the parallel design of experiments in both groups of primates make this an extremely compelling approach likely to yield important insights into higher order processing in the human neocortex.




Carnivores
Flying Fox
Squirrel
Hedgehog
Insectivores
Rodents
Mouse
EUTHERIAN MAMMALS
Platypus
MARSUPIALS
MONOTREMES
Opossum
COMMON ANCESTOR