**What constitutes the prefrontal cortex?**

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During evolution, the prefrontal region grew in size relative to the rest of the cortex. It reached its largest extent in the human brain, where it constitutes 30% of the total cortical area. This growth was accompanied by phylogenetic differentiation of the cortical areas. It has been argued that the human brain holds prefrontal regions that are both qualitatively and functionally unique. Present-day neuroscientists studying the prefrontal cortex increasingly use mice. An important goal is to reveal how the prefrontal cortex enables complex behavior. However, the prefrontal cortex still lacks a conclusive definition. The structure and function of this brain area across species remain unresolved. This state of affairs is often overlooked, warranting renewed focus on what the prefrontal cortex is and does.

Neuroscience as a unified academic field emerged in the 1960s (3). The progress in our understanding of the nervous system has been tremendous, aided by the inclusion of techniques from disciplines outside neuroscience and by bridging of studies of the brain to the rest of biological science. Modern neuroscience is not only a thriving scientific field but has also risen to the forefront of scientific and public interest. A central aim of modern neuroscience is to describe how the nervous system enables and controls behavior. Systems neuroscience combines different levels of analysis for interrogation of how neurons and circuits underlie neural computations serving behavior. Recently developed molecular and genetic tools enable scientists to record, observe, disrupt, and provide endogenous activity patterns in selected neuronal populations while examining the consequences for behavior and physiology. Because such studies require research animals amenable to genetic intervention, *Mus musculus* (the house mouse) has become extensively used as a model animal in neuroscience. Increased restrictions on studies in nonhuman primates will likely result in a growing focus on rodent studies in most parts of the world. It is thus increasingly important that we contemplate the translation of findings between rodents (here, limited to *Mus musculus* and *Rattus norvegicus*) and primates. Central to this is a consensus around terminology and the definition of concepts. However, many aspects of the structure and function(s) of the prefrontal cortex have been peculiarly difficult to reconcile.

**The prefrontal cortex gets in the ring**

The term “prefrontal” is suggested to have been first used in 1884, in a publication by Ferrier and Yeo (2, 3). However, already before the institution of the term, lesion experiments in this part of the brain were conducted, for example, in dogs by Hitzig (4). The first topographical description of regions we consider prefrontal is ascribed to Brodmann (5). Brodmann, on the basis of cytoarchitectural criteria, used the terms “frontal” and “precentral” to denote two main regions of the primate frontal lobe. He found the presence of a distinct granular layer IV to be a prominent characteristic of the frontal region in primates. In his interspecies comparative studies, Brodmann found the frontal (i.e., granular) region to be poorly developed or absent in nonprimates, which led him to conclude that the granular frontal region essentially is unique to primates (5, 6)—an influential view later blamed for delaying studies of the frontal areas in nonprimates (3). The granular frontal region described by Brodmann has over time interchangeably been referred to as granular frontal, frontal granular, or prefrontal (6). Defining the prefrontal cortex as the granular frontal region described by Brodmann inherently means that only primates have a brain area that could be called prefrontal (6). The perspective that primates possess a large part of the frontal cortex lacking homology in other species has found new arguments and persists today.

**The current, likewise old, definition**

Surprisingly, the current definition (insofar as there is one) of the prefrontal cortex is not common knowledge to neuroscientists. A contributing factor might be that the deducible definition has been built primarily on anatomical criteria, with no demonstrably direct correlation to a functional definition. For 70 years, the prefrontal cortex has in essence been defined as the part of the cerebral cortex that receives projections from the mediadorsal nucleus of the thalamus (MD). The core of this concept can be traced back to Rose and Woolsey (7). Rose and Woolsey proposed to use the projection field of the MD to define the frontal region of the cortex, as the MD was found to target a restricted frontal field in primates as well as nonprimate mammals. In primates, this cortical field includes the granular frontal cortex. Important to Rose and Woolsey’s theorem was the assumption that all thalamic projections reaching the frontal fields originate in the MD. This hodological (i.e., fiber connection) definition, according to Rose and Woolsey, allowed the conclusion that all mammals possess a cortical field equivalent to the granular frontal cortex of primates. The MD projection definition of the prefrontal cortex thus originated in proposed anatomical homology between mammalian species (7). Homology in evolutionary biology refers to structures or characteristics in different species that share resemblance due to a common evolutionary origin (6, 8). Rose and Woolsey did not study the prefrontal cortex in rats (nor in mice), and it would take until 1969 before the MD projection to the prefrontal cortex in rats was described (9, 10). This was followed by an era of intense research on the prefrontal cortex of rats, and the similarities to the prefrontal cortex in primates were of specific interest. In 1981, the MD projection to the mouse cortex was revealed, and on the basis of this projection, the topography of the mouse prefrontal cortex was described as resembling that in rats (9). However, although the MD was found to target a large part of the mouse frontal cortex, other thalamic nuclei were also shown to target most of this projection field.

The Rose and Woolsey definition of the prefrontal cortex has survived, despite substantial later redrawing of prefrontal-MD connectivity. It is now established that the MD projects to cortical fields outside of the prefrontal cortex (6) in cats, rats, mice, and rhesus monkeys (3, 11, 12). Other thalamic nuclei than the MD project to the prefrontal cortex. The Allen Mouse Brain Connectivity Atlas (13) (http://connectivity.brain-map.org/), which provides high-resolution quantitative connectivity mapping of a mammalian species...
in unprecedented detail, confirms these findings. Further, the organization of the MD and the connectivity pattern of efferents from the different MD regions differ across mammalian species (14). In summary, the projection from the MD cannot single-handedly define the prefrontal cortex across species.

Other indications of homology have been added to define the prefrontal cortex (12). The aspect of homology adds complexity to the comparison of cortical areas between different species. Structural or functional similarities are not automatically evidence for homology; similarities can be the result of convergent evolution. Likewise, homology does not necessarily say anything about how identical structures or functions are at the present time. In the discussions surrounding the prefrontal cortex, the focus on homology and usage of the term varies. At times the evolutionary development of the prefrontal cortex and homology are central, at other times not. Moreover, the term homology is used to declare a common evolutionary origin, but also to declare structural or functional similarities between now-living species (8). Nonetheless, at present, criteria to consider in the establishment of cortical homologies are the embryological development, functional properties, pattern, and density of specific connections, and the presence and distribution of neuroactive substances and receptors (12). Some researchers consider cytoarchitectural comparisons relevant only for closely related species (12). Plausible homology in cortex-based gangliothalamicortical circuits and connectivity with the amygdala have been suggested to define the prefrontal cortex (22, 14). As evident from the above, no single criterion has been identified that can adequately define the prefrontal cortex across species.

Parcellation of the prefrontal cortex

The anatomy of the prefrontal cortex in different species has been extensively covered (15). Different terminologies are used, depending on the species of focus and on the criteria (e.g., cytoarchitecture, connectivity and/or function) used to subdivide the structure (6, 12, 14, 16). Brodmann’s cortical scheme and cytoarchitectonic numbers are still widely used for primates, and efforts have been made to define the human and macaque monkey prefrontal cortex using uniform cytoarchitectonic and topographical criteria (17). The Brodmann areas (BAs) traditionally defined as prefrontal in humans are BA8 to 14 and BA44 to 47 (15, 18) (Fig. 1, A and B, and movie S1). These areas correspond roughly to the granular part of the prefrontal cortex. However, the granular prefrontal regions display systematic changes in the laminar structure and are therefore often divided into different cortical types: granular (homotypic or elanulate), with a conspicuous layer IV, and dysgranular, displaying a subtle layer IV (15, 19) (Fig. 2, A and B, and movie S2). An intermediate thin, lightly granular cortical type is regularly also distinguished. Several of the prefrontal BAs display more than one cortical type, and a few also hold agranular parts (i.e., they lack layer IV). The agranular anterior cingulate cortex (ACC; BA24, 25, and 32, in

Agranular Dysgranular Granular Thin, lightly granular

Fig. 2. Cortical types in the human and mouse prefrontal cortex. (A and B) Tilted frontal-side view (left) of the human brain. Schematic illustration of the four cortical types in the prefrontal cortex, including ACC. (C) Tilted frontal-side view (left) of the mouse brain depicting the agranular prefrontal cortex. Dashed black line indicates sagittal midline. Illustrations and movies were constructed using custom software written in MATLAB. Human architectonics were taken from (19) and the Brainnetome Atlas (50), the latter accessed through the Scalable Brain Atlas (51). Mouse nomenclature and architectonic data sets were taken from the Allen Brain Atlas (27) as accessed through the API. See also movies S2 and S4. Some accounts also BA33; Fig. 1, A and B, Fig. 2, A and B, and movie S1) is a midline, collar-formed structure surrounding the frontal part of the corpus colossus. The ACC receives projections from the MD, and some therefore consider this structure to be part of the prefrontal cortex (6). Many neuro-imaging studies focus on functional localization and division of the human prefrontal cortex. The subdivisions and their extent vary, but dorsolateral, dorsomedial, ventromedial, and orbital prefrontal cortex are common functional divisions (14) (Fig. 3, A and B, and movie S3). Of these, the dorsolateral part (BA46, 9, 9/46) has received special interest; in primates, this cortical field receives projections from the MD, whereas the MD in rodents does not target dorsolateral frontal areas. The granular dorsolateral prefrontal cortex is therefore considered a primate specialization (19). The MD targets medial and orbital cortices in rats, as in primates. Mice and rats possess fewer areas in the frontal lobe than primates, and all areas in the prefrontal cortex of mice and rats are agranular (Fig. 2C and movie S4). At times the agranular cytoarchitecture is used as

a definition of the rodent prefrontal cortex (20, 21).

Efforts have been undertaken to parcellate the mouse prefrontal cortex (20, 22, 25) (Fig. 4, A to C, and movie S5). In a recent study comparing different parcellations, the prefrontal areas were found to differ in location and terminology. Different terminologies have been used for the same structure, and areas with similar terminology display different extent and location in different atlases (27). The boundaries of the orbital areas were found to differ the most. Van de Well (20, 21) identified infralimbic and prelimbic areas in what Paxinos and Franklin (26) referred to as the medial orbital cortex. Van de Well also placed parts of the prelimbic cortex in what Paxinos and Franklin denoted as the cingulate cortex, area 2. The two parcellations also disagree regarding the boundary between the prelimbic and infralimbic cortex. Whereas the stereotactic Paxinos and Franklin atlas is widely used for planning and post hoc analysis of anatomical coordinates in experiments, the Allen Mouse Brain Atlas is used for structural annotation and comparison of gene expression patterns to neuroanatomical structures (27). The included anatomic reference atlas (atlas.brain-map.org) holds 738 annotated structures based on scanned Nissl-stained sections but does not display stereotactic coordinates. Allen mainly adopted the nomenclature from the Swanson rat brain atlas (28), which differs from Paxinos and Franklin regarding the nomenclature of the prefrontal cortex. The mediodorsal part of the rodent prefrontal cortex has been particularly riddled with varying terminology. This region is known as secondary motor cortex (M2), medial agranular cortex (Agm), secondary frontal area (PrF2), and yet other names, all denoting a varying cortical extent. Allen used the term secondary motor area (MOs) (4) (Fig. 4, A to C, and movie S5).

In summary, (i) as of 2017 there is not yet a consensus surrounding the anatomy (or nomenclature) of the prefrontal cortex in Mus musculus; (ii) because of this, miscommunication between researchers is a valid risk; and (iii) stereotactic coordinates are still indispensable for communication and comparison of experimental findings. The lack of a conclusive definition (14) hampers direct comparisons of the prefrontal cortex between species. Even with a definition, the vast variation in anatomy, connectivity, and possibly also function across species constitutes a major challenge to the establishment of what can be considered equivalent prefrontal regions between different species (29).

Is it meaningful to study the functions of the prefrontal cortex in the mouse or rat?

It is a tradition in cortical neuroscience to focus on a few model organisms and to search for general principles of neural mechanisms of behavior. Opponents point out that this approach inherently must assume a high degree of evolutionarily conserved characteristics and a somewhat shared cortical organization for studies on one species to have importance for the understanding of structural and functional principles
of a larger group of species (30). Conserved characteristics imply that brain evolution primarily has been a process of expansion and differentiation (6). The contrasting view holds that the brain—and perhaps the prefrontal cortex in particular—has become diversified during the course of evolution, resulting in unique modifications of structures and functions in different groups of animals. Even new, species-specific structures could have evolved; the dorsolateral prefrontal cortex in primates could be an example of this.

The matter of specialization has been central to the debate regarding whether rats have a prefrontal cortex (6, 12, 19). This question has a direct bearing on the suitability of rats as model animals for human brain functions and disorders. By contrast, little debate has surrounded the relevance of the mouse prefrontal cortex as a model for human cognition and behavior. Today, mice (Mus musculus) and rats (Rattus norvegicus) are often lumped together, and the general importance of rodent studies is being discussed (represented by two of >2000 rodent species; Macaca mulatta is one of ~200 primate species). Advocates of the view of brain evolution as a process of specialization often stress that if we are to better understand what the prefrontal cortex is and does, research should not be concentrated on only a few species. Further, the focus should be on identification of organization and functionality shared across species (6, 19, 31).

Arguments against the usefulness of rodents are often based on an apparent lack of homology. The modern human and the mouse separated from a common ancestor ~96 million years ago (humans from the Old World monkeys, such as macaques, 23 million years ago, and rats from mice 33 million years ago) (32). In the course of evolution, the cortical structure has undergone phylogenetic development. As pointed out earlier, the prefrontal cortex in primates is virtually entirely homotypical (i.e., it displays a six-layered internal structure (although with variation in laminar structure)), whereas all frontal areas in rodents are agranular. Agranular regions are considered phylogenetically more primitive than the granular regions of the prefrontal cortex (15). The spatial arrangement of frontal areas also differs between rodents and primates; the shared agranular areas lie adjacent to the allocortex (cortex with three or four layers). The granular prefrontal cortex instead lies next to agranular areas, not the allocortex, which renders this structure distinct from, and different from, the agranular cortex. Further, the dorsolateral prefrontal cortex in primates is suggested to send connections to the dorsal striatum; agranular areas lack such connections. These differences are seen as support for the notion that new cortical areas and connectivity have emerged in the course of mammalian evolution. In this view, rodents are not useful models for the functions and dysfunctions of the dorsolateral prefrontal cortex of primates (6). However, researchers in favor of this position often emphasize that rodents are valuable for the understanding of frontal areas emerging before evolutionary separation (6, 19, 33).

Arguments for the usefulness of mice and rats (apart from the declaration of shared anatomical organization) commonly concentrate on functional homology (or analogy). This inevitably relates to the fundamental questions of what the prefrontal cortex does (19) and why this structure has evolved (14). Although the six-layered dorsolateral regions might be a unique evolutionary feature of the prefrontal cortex of primates, rodents could hold the functions of these regions in their medial or orbital frontal regions (34). Prefrontal operations could be fundamental and shared between species, while the neural substrates responsible could be analogous (or homologous) and the behavioral output different (34). In essence, functions could be class-common (35). Class-common prefrontal functions find support in numerous lesion studies, as demonstrated by shared deficits. The temporal organization of behavior is suggested to be a class-common function of the mammalian prefrontal cortex (see below).

The dorsolateral prefrontal cortex in primates has long been considered essential to working memory, the active but temporary retention of task-relevant information. The information held is subjected to processing, which enables the prospective act to be performed successfully (36). Working memory thus has a future perspective (15). Delayed-response paradigms are most often used to probe working memory, and although what is encoded by the dorsolateral prefrontal cortex during the delay has come to be a matter of debate (37), there is agreement that in primates, activity in this area is central to working memory. Rats and mice can also perform delayed-response tasks, and working memory is suggested to be a class-common function (38). Further, the prefrontal cortex in rodents has been shown to participate in functions such as rule representation, response control, impulse control, attention, and strategy shifting, of which several in primates are ascribed to the dorsolateral prefrontal cortex (29, 34, 36). These demonstrations question the unique functions of the primates dorsolateral prefrontal cortex and the nature of specialization within subregions of the prefrontal cortex in general. Relevant to the latter is the proposal that the prefrontal cortex not only holds functional subregions, but also has a specific higher-order function that is more than the sum of its parts (16). This overarching function could be processing of temporal complexity and representation of events extended in time, possibly involving large and different areas of the prefrontal cortex (15, 16, 39). This integrative, nonlocalized processing is typical of higher cognitive functions (15). Studies of the prefrontal cortex as a whole, regardless of species, would be valuable for extension of these theories.

It is the behavior, not the cortex, that is selected by evolution (14). Although the same behavioral repertoire is not expected among species, mammals largely need to solve the same problems in their everyday life. General behavioral capacities can be seen across mammalian species, and these have been referred to as class-common behaviors (14). Although there is considerable variation in the elements of class-common behaviors, their selection could have been a contributing factor to the evolutionary development of the prefrontal cortex (14). The existence of class-common functions and behaviors is regarded as support for the view that elemental neural processes involving the dorsolateral prefrontal cortex can fruitfully be investigated in rodents. However, clearly, rodent studies do not investigate the primate prefrontal cortex as such.

Functions of the prefrontal cortex and the meaning of cognition

The functions of the prefrontal cortex have been abundantly theorized. This brain area is implicated in emotional, social, motivational, perceptive, and other processes. Research in both humans and animals has indicated a central role of the brain’s frontal regions in cognition. Attention, working memory, and decision-making are common cognitive functions attributed to the prefrontal cortex. In the literature, these functions are often organized under (or intermingled with, or made equivalent to) concepts such as executive function, cognitive control, top-down control, preparatory set, and inhibitory control. In essence, cognition has become an umbrella term encompassing a diverse and varying set of brain processes and functions. Studies probing cognition in model animals are increasing in number. A PubMed search on “cognition” currently
renders more than 220,000 entries, perhaps supporting the notion that cognition has become so broad that it can be anything that we want it to be (40, 41). How did cognition become so broad?

During the “golden age of cerebral localization,” 1870 to 1890 (42), experimental physiologists strived to assign physical and mental capacities to specific regions of the cerebral cortex. Although individually conflicting, collectively, the early studies of the prefrontal region indicated this part of the brain as central to mental processes. In the amalgam discipline of neuropsychology, which investigates how the brain relates to the mind and behavior, the prefrontal region was therefore particularly relevant to explore. Behaviorism, the philosophy of the science of behavior analysis (43), evolved to dominate scientific psychology for a long time. Skinner stated that mentalist terms (e.g., cognitive phrases such as consciousness and mind) should be avoided, and that the role of brain physiology is “to account for the mediation of behavior rather than of mind” (44). Behaviorism brought along the extensive use of animals in experimental psychology. New terms were introduced, such as conditioning, extinction, discrimination, reinforcement, and punishment; such terminology is still extensively used in rodent behavioral neuroscience.

Cognitive science, an interdisciplinary cooperation among disciplines such as philosophy, psychology, computer science, linguistics, anthropology, and neuroscience, viewed cognition as a form of information processing and the conceiving of thought as a computation (45). Information theory, artificial intelligence, and the first digital computers were therefore pivotal to the field’s information-processing approach to psychology. However, a central intention of cognitive science was to integrate “mentalistic” concepts with behavioral data and bring “the mind back into experimental psychology” (46).

Behaviorism and cognitive science exemplify movements that have heavily contributed to the present-day concepts, approaches, and terminology in neuroscience. Internal brain processes are nowadays universally acknowledged, and their neural mechanism is the focus of much research on the prefrontal cortex in model animals. Mentalist terms have been transferred to physiological neuroscience to assign psychological linkage to neural functions, the term cognition being a prime example. Although the term cognition certainly has aided discussions and synergies across fields for a long time, the use of artificial conceptual terms can misguide research, as they have no discrete reality in the brain, and there might not be brain mechanisms for them (40, 47). The close association between cognition and the prefrontal cortex warrants attention to how concepts and terminology are used, so as not to confuse discussions and comparisons between studies.

Most theories on the functions of the prefrontal cortex have been derived from studies in humans and nonhuman primates, awaiting translation in rodents. Technological advancements, however, make mice especially suitable for testing of hypotheses and probing of the neural computations of the prefrontal cortex. I will give a necessarily sketchy account of the suggested overarching function of this brain area, related to the question of why there is a prefrontal cortex in mammalian species. Temporal organization of behavior is a reoccurring proposition of what the prefrontal cortex is essential for. Goal-directed behavior requires action on the world for achievement of a goal. The behavior is organized in the time domain, which requires forward-looking (49). Previous experiences are used to foresee situations and to predict how different actions can change the outcome. Novelty and the future are central factors in the organization of behavior, and the necessity of the prefrontal cortex increases if new goal-directed actions are to be executed, and especially if the actions extend in time (15, 39). The fundamental function of the prefrontal cortex could thus be to represent and produce new forms of goal-directed actions—actions that can be, for example, mental and internal, emotional or motor-related (15). As part of this, executive functions are adaptively used depending on the goals, means, time scales, and motives relevant to the behavior. A cortex-wide infrastructure where knowledge and memory are represented is necessary for proper executive operations and prefrontal functioning. The representations in prefrontal cortex have been described as top-down or bias signals (49). The biasing signals select and shape processing in downstream brain areas in accordance with the task’s demands and the goal. As a result, information flows in pathways that can properly piece together the inputs, internal state, and output needed to pursue the goal (49). Note that much of the theory regarding the temporal organization of behavior builds on theories and findings involving the dorsolateral prefrontal cortex in primates.

Concluding remarks

Modern neuroscience has a bright future; progress in the understanding of the brain and its functions is happening at an unprecedented pace. Tool development is key to breakthroughs in neuroscience, and we have recently seen several revolutionary tools, including engineered transgenic animals and viral vectors, tools for large-scale electrophysiological recordings, and technology for the visualization and manipulation of neural activity in behaving animals. Mus musculus, amenable to genetic targeting of neuronal populations and circuits, has been central to the success of these technologies and has rapidly become extensively used in neuroscience. Nonetheless, much knowledge regarding the structure and function of the prefrontal cortex is still based on studies in rats, humans, and nonhuman primates. As studies of the mouse prefrontal cortex proliferate, it is vital that we remember to contemplate how their findings can be transferable to other species and how they might contribute to the overall understanding of what the prefrontal cortex is and why it has evolved.

Capitalizing on the current progress requires the parallel development of terminology and concepts with true relevance to the functional organization of the brain, guided by neural and behavioral data. Precise definitions and usage of descriptive terminology derived from brain processes shown to exist could aid comparisons across studies and species, and may clarify dissociations and interactions.

As for the understanding of why we have a prefrontal cortex, further studies in mice, rats, humans, and nonprimate humans, as well as other species, will be pivotal. But as stated more than four decades ago: “We can be confident that the prefrontal cortex did not evolve to enable animals to learn delayed response problems...” (31).


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SUPPLEMENTARY MATERIALS
www.sciencemag.org/content/358/6362/478/suppl/DC1
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