Published in final edited form as:

Brain Res Rev. 2007 October ; 55(2): 204–219.

The radial edifice of cortical architecture: From neuronal silhouettes to genetic engineering

Pasko Rakic

Abstract

The developmental principles that establish the columnar edifice of the cerebral cortex underlie its evolution and dictate its physiological operations and cognitive capacity. This article contrasts the initial discoveries made by Ramón y Cajal and his contemporaries, based on the ingenious interpretation of neuronal shapes and their relationships using the Golgi method, with new insights based on the application of the most advanced methods of molecular biology and genetics. We can now propose a realistic model of how the sequence of gene expression, cascade of multiple molecular pathways and cell-cell interactions establish the number of neurons, guide their migration and allocation into proper regions and determine their differentiation into specific phenotypes that establish specific synaptic connections. The findings obtained from different levels of analyses sustain the radial unit hypothesis as a useful framework for understanding the mechanisms of cortical development and its evolution as an organ of thought.

Keywords

cerebral cortex; neuronal migration; radial glial cells; cortical columns

Human civilization has advanced for centuries without understanding the organ that made it possible. It was only at the turn of the 19th/20th century that the application of the silver impregnation method (“la reazione nera”), developed by Camillo Golgi, allowed Santiago Ramón y Cajal to expose, until then, a hidden universe of unbelievable complexity and beauty formed by the black silhouettes of nerve cells and their intricate connections. These discoveries were rewarded by a Nobel Prize in 1906, for which we now celebrate its Centennial. Many consider this particular prize to be among the most significant because it acknowledged that understanding the biological basis of our mental abilities is a realistic possibility. Thus, it is paradoxical that a rich harvest made by Ramón y Cajal and his contemporaries was followed by about 70 years of relatively slow progress in this field. It was again the introduction of new methods in the second half of the 20th century that opened the door for new advances in understanding our organ of thought.

In this essay, I shall contrast the seminal discoveries revealed by the silver impregnation method with the concepts and insights into molecular and cellular mechanisms of cortical development obtained through modern technology. As this short overview could not possibly give justice to the exquisite drawings and documentation by the old masters, I urge readers to look for the originals in the libraries that are in the era of internet becoming museums. Instead, I will use the power of internet to provide two animated movies on the website to illustrate the dynamic nature of neuronal migration and cortical formation (Supplementary Movies #1: <http://>

rakiclab.med.yale.edu/MigratingCorticalNeuron.html and #2 <http://rakiclab.med.yale.edu/RadialMigration.html>).

The subject of this present essay is focused on the issue of what we have learned regarding the mechanisms of the development of the cerebral cortex, the organ that subserves complex cognitive tasks including human thought and creativity, how it emerges from the embryonic stem cells during individual development, and how it has expanded during evolution. It is not intended to be a comprehensive critical review of this burgeoning research field. Furthermore, since this is the major research theme in my laboratory, I will take advantage of the essay format to reflect on my personal experience in investigating this intellectually appealing subject. Due to the page limitations I can not possibly give credit to all of those who contributed to the understanding of this fascinating and complex subject.

Radial Edifice: Hardware for Cortical Operations

The cerebral cortex, in all species including human, is characterized by two distinct features: it is a sheet of neurons in organized (horizontal) layers intersected by (vertical or radial) columns. Although the laminar arrangement is more obvious, the radial deployment of neurons was already depicted in Ramón y Cajal's and Rafael Lorente de Nó's rendering of cortical circuits impregnated with the Golgi method (e.g., Ramón y Cajal, 1881, Ramón y Cajal 1891, Ramón y Cajal, Lorente de Nó, 1938). Subsequent research showed that the cortical columns consist of an array of iterative neuronal groups (also called modules) that extend radially (perpendicular to the pial surface) across cellular layers VI to II with layer I at the top (e.g., Mountcastle, 1997; Szentágothai, 1978; Goldman-Rakic, 1987). The neurons within a given column are stereotypically interconnected in the vertical dimension, share extrinsic connectivity and, hence, act as basic units subserving a set of common static and dynamic cortical operations (Eccles, 1984). This concept, inspired by the discoveries of functional columns subserving various attributes of somatic sensations (Mountcastle et al., 1957) and vision (Hubel and Wiesel, 1969), gained in significance by the finding that even in the so called "thinking" or association cortex, neurons with categorically specific responses reside in the same columns (Goldman-Rakic, 1988; Goldman-Rakic, 1995). The evidence for this concept came from a multiple electrode recording in the primate prefrontal cortex during a working memory performance displaying features analogous to the orientation preferences of the primary visual neurons (e.g., Funahashi et al., 1989; Wilson et al., 1993). Collectively, these data showed that neurons positioned adjacent to each other across the tangential axis of the cortex, which spans multiple columns, represents the memory of targets situated in adjacent locations in the visual field. Further, as predicted by the columnar hypothesis, neurons occupying the same column or location share the same tuning properties.

The evidence for the radial organization of neurons and neuronal processes in the association cortex not only provides an infrastructure for representing information symbolically, but also supports the idea that mental content is localizable and that combinatorial principles operate to arrive at a coherent structure of thought and language, just as they appear to do in vision and other sensory processes (Goldman-Rakic, 1987). Recent methodological advances in fMRI imaging are beginning to reveal changes in blood flow on the scale of millimeters; and, thus, will be able to reveal functional columns or modules in the living human brain and to test the validity of the columnar organization (e.g., McKinstry et al., 2002). We still do not have a full understanding of how and why columnar organization may be necessary for cortical operations from a functional point of view (e.g., Mountcastle, 1995). However, as reviewed below, we have already learned a great deal about cellular and molecular mechanisms that makes its neuronal organization possible.

Neuronal Migration and the Radial Unit Model of Cortical Development

Distant places of birthdays

If the assembling of cortical neurons into a crystalline-like array of layers and columns is not enough to provoke admiration, then the discovery that none of them are generated within the cortex itself should. In no other organ of the body is such a radical and precisely preprogrammed rearrangement of cell positions observed. Today, the fact that none of the myriad of cortical neurons are generated locally, and that they all acquire their proper areal and laminar positions through long-distance migration is common knowledge, so it is often forgotten how remarkable and unique in biology this phenomenon is. The initial concept was based on a simple histological observation in the human brain, that mitotic figures (which signify cell division) are situated along the lumen of the cerebral ventricles (at the time, called “germinal layer or matrix”) and are virtually absent in the developing cortex that is forming below the pial surface (His, 1874; 1904). Based on this finding, as well as on the observation of radially oriented bipolar cells in the middle of the cerebral wall, Swiss embryologist Wilhelm His concluded that after the last mitotic division near the ventricular surface, neurons probably migrate to the overlying cortex. With this revolutionary discovery and his other achievements, he would have been a serious candidate to co-share the Nobel Prize along with Cajal and Golgi, had he not died two years earlier, in 1904, the year when his monumental book on embryonic human brain development was published in Leipzig.

I became aware of the concept of directed neuronal migration during a histology course at the Medical School in Belgrade in the late 1960's. I noticed that in all the other organs of our body, cells are generated locally, or if they do migrate, like during the formation of gonads, their destination is not position-specific. Only in the brain specific cell classes are generated in one place and then subsequently migrate to the very precisely designated final position. This is particularly evident in the cerebral cortex where postmitotic cells migrate long distance and bypass each other before settling in the final position within specific layer within a given radial column. This extraordinary phenomenon has fascinated and inspired me to extent that I eventually interrupted the prospect of a career in neurosurgery to pursue the question of how and why this occurs. I was young and enthusiastic and thought that how the organ that mediates our humanity evolved is the most fundamental and important question that can be addressed. There are, of course, other great questions, such as origin of universe, but exploring how cerebral cortex develops actually could be accomplished and I was determined to participate in the search.

To begin to find some answers, as part of the work on my doctoral dissertation, I had incubated slices of fresh (postmortem) human fetal cerebral for 24 hours in a tissue culture medium containing ^3H -thymidine (Rakic, 1968). Today, the use of slice preparations is commonplace; but, to my knowledge, this was the first use of slices in developmental neurobiology. Furthermore, at the time, marking replication of DNA during cell division with ^3H -thymidine was also relatively new. Combining these two approaches, in forebrain slices from an 18 week old fetus, I found that dividing cells continue to synthesize DNA predominantly near the ventricular surface, even at this late stage of cortical neurogenesis when the cerebral wall is relatively thick and convolutions are becoming prominent (Rakic, 1968; Rakic and Sidman, 1968). More specifically, the cell nuclei become heavily radioactive, in the two transient proliferative layers (or zones) near the surface of the lateral cerebral ventricle: the narrow ventricular zone (VZ) at the surface, and the broader zone above, which I termed the SVZ or subventricular zone somewhat inappropriately, since the proliferative cells were situated above (supra) rather than below (sub) the VZ. I draw the schema of model of cortical development including transient embryonic layers using India ink, which gave me an opportunity to express my natural inclination and love for the visual art (as a medical student I was publishing cartoons for the Yugoslavian newspapers). I was also aware that Cajal used India ink to only to describe

images that he has observed on the microscope, but also to convey the dynamism and meaning of cellular events as they occur in time. This particular drawing had been presented by Richard Sidman to the Boulder Committee in Colorado, which published together with it with recommended suggested nomenclature (Boulder Committee, 1970). The important scientific point for the main subject of the present essay is that, apart from the occasional radioactive nucleus in the intermediate zone and marginal zone (prospective layer I), the cortical plate in the slices was devoid of radioactive labeling. I was impressed by this finding and decided to spend my professional life investigating why and how this occurs.

The study of supravital DNA synthesis in the embryonic human and mouse brain (Rakic and Sidman, 1968) was probably among the factors contributing to the offer I have received to join faculty of the Harvard Medical School faculty in 1969. After establishing my laboratory in Boston, I initiated a comprehensive ^3H -thymidine autoradiographic study of neurons *in vivo* in non-human primates to determine the time of origin and routes of their migration (e.g., Rakic, 1974; reviewed in Rakic, 2002). This method is particularly effective in the brain because neurons, unlike most other cells of the body, do not divide after the final division (birthday), leaving a trace of their trajectories and settling behavior. I selected the macaque monkey, because as an Old World Primate it was the species with the most similar cortical and visual system organization to that of human. Pregnancy in this species lasts six and half months (165 days as determined by the time of conception at the middle of the 28-day menstrual cycle). Since injected ^3H -thymidine is quickly eliminated from the bloodstream, the heavily labeled cells are only those that were in the last cell division and have not divided again (Nowakowski and Rakic, 1974). Over 120 animals, housed in the New England Regional Primate Research Center (Southborough, MA), were injected with ^3H -thymidine and sacrificed at various time periods, ranging from one day to six months and even several years. This research was supported by my first grant, on "Neurogenesis Processes in the Primate Brain" that is now in its 35th year. It was by necessity larger than the average grant, and the criticism that "for that money could be funded three average size grants" was deflected by my statement that "we still would not know what happens in primates".

The results revealed that corticogenesis in the macaque monkey begins almost simultaneously, between E38–E40, in all cortical areas examined (Rakic, 1974, Reviewed in Rakic, 2002). However, the time of cessation of neurogenesis was more variable and has ranged between E70 in the limbic cortex to E102 in the visual cortex. This project provided the normative data for the model in the Supplementary Movie #1: <http://rakiclab.med.yale.edu/RadialMigration.html>. The typical inside-out gradient of neurogenesis, known from initial studies in rodents (e.g., Angevine and Sidman, 1961) was even more pronounced in the primates; and, the cohorts of isochronously generated neurons eventually occupy a relatively narrow strata within the cortex (Rakic, 1974). The sole exception to the inside-out sequence of neurogenesis in primates was encountered in layer I, where, contrary to rodents, neurogenesis continues throughout the entire period of corticogenesis (Zecevic and Rakic, 2001; Rakic and Zecevic, 2003). Since post-mitotic neurons at later stages of development require more than two weeks to settle into their final positions, we estimated that about one month before birth (between E125 and 135), all cortical neurons in the macaque are settled in their final laminar and radial positions see the Supplementary Movie #1 in: <http://rakiclab.med.yale.edu/RadialMigration.html>. The biological significance for the inside-to-outside sequence of neurogenesis and settling of neurons in the cerebral cortex is not clear. One hypothesis is that it enables later generated neurons to interact and communicate with the neurons generated earlier when they pass by each other (Rakic, 1990). Teleological reasons are always hard to prove, but this one, actually, makes sense since it is generally agreed that the deeper lying neurons and their connections are phylogenetically older, while the more superficial neurons, which form the bulk of the corticocortical connections, are evolutionarily younger. Furthermore, the disturbance of this sequence by either genetic or environmental

factors leads to functional deficits (Reviewed in Caviness and Rakic, 1978; Rakic, 1988b; Ross and Walsh, 2001; Hatten and Mason, 1990; Hatten, 2002).

One unexpected and curious observation from this material was that, except for the granule cells in the cerebellum and hippocampus, we did not observe any labeled neurons in the animals exposed to ^3H -thymidine postnatally (Rakic, 2002). It should be emphasized that neuronal cells in the mature primate neocortex can be easily identified in autoradiograms and that non-neuronal cells such as astrocytes, oligodendrocytes and endothelial cells as well as the cells in other organs of the body have been labeled (Rakic, 1985). This observation prompted me to suggest that early completion of neurogenesis in the primate cerebral cortex, without addition or turnover throughout life, may be a critical step in the evolution that allows of mental prowess in *Homo sapiens*. The basic idea was that a permanent population of cortical neurons may be more valuable for survival of primates, than the replacement of neurons that involves introduction of “naïve” neurons that have not been exposed to previous experience and would cause a loss of acquired knowledge (Rakic, 1985). At the occasion of celebrating Cajal’s contribution, I would like to emphasize that he came to the conclusion that the neurons that subserve the most precious human mental functions are under normal conditions irreplaceable, based on a much cruder histological method. The observation that cortical neurogenesis in primates is indeed completed before the time of birth and that the number of cortical neurons remain stable during adulthood, has been conclusively proven in the human cerebral by the most advanced and sophisticated methodology of ^{14}C -carbon dating (Bhardwaj et al., 2006).

The early genetic determination of the species-specific number of neurons and radial units and the participating neurons in the adult cortex does not negate the existence of a considerable degree of plasticity during the critical periods of postnatal maturation. Studies in human and non-human primates show that both neurons and their axons are overproduced in the cerebral cortex during well-delineated stages of development. For example, there are about 40% more synapses in the macaque monkey visual cortex during development than in the adult (Bourgeois and Rakic, 1993; Rakic et al., 1986). Furthermore, the newborn monkey has almost four times the callosal axons than in the adult, which are lost at a rate of about 8 million per day or 50 per second during the first three weeks after birth; and, thereafter, 5 per second until the adult value is reached (LaMantia and Rakic, 1990). Other interhemispheric connections, including those between columns, display a similar phase of overproduction and loss of axons, dendrites and synapses (e.g., Lund, 2002; Lund et al., 1977). The functional significance of the loss of axons and synapses is not fully understood, but the prevailing hypothesis has been that activity dependent stabilization plays a critical role (Rakic, 1986; Shatz, 1996).

Mode of Migration: Role of Radial Glial Cells

The next question was how neurons find their way from the ventricular surface in the large and increasingly distant and convoluted human cortex? The application of combined histological, electron microscopic and anatomical methods that became available in the late 1960 and early 1970’s, on the embryonic monkey forebrain, provided the next step in understanding the possible mechanism of directed neuronal migration by showing that post mitotic neurons find their way to the appropriate radial position within the cortex by following the elongated shafts of glial cells that span the full thickness of the fetal cerebral wall (Rakic, 1972). The reconstruction of the relationship of the glial shaft, ultra-structurally clearly distinct from the migrating neurons, initially based on the serial EM reconstructions (Rakic et al., 1974) was animated (Supplementary Movie #2: <http://rakiclab.med.yale.edu/MigratingCorticalNeuron.html>). My studies in monkey renewed interests to these cells that were initially discovered in the human fetal brain by Golgi, Ramón y Cajal and their contemporaries using the silver impregnation method (Golgi, 1885; Ramón y Cajal, 1899; Magini, 1888; Retzius, 1893; Kölliker, 187–9). The classification of cells in the mammalian

embryonic cerebral wall by the use of the Golgi method alone was difficult even to the masters of this approach. Thus, for example, Cajal had changed his mind twice as what is the nature of some of these cells, until he came to the realization that the initial bipolar neuroepithelial cells (now often called neural stem cells) transform first into a fetal (radial) glial phenotype and eventually into an astrocyte. It was not until the introduction of electron microscopy and immunohistochemistry that their glial nature as a specialized form of astrocytes had been established based on ultra-structural and molecular characteristics (Rakic, 1972; Levitt and Rakic, 1980). Although there are substantial species-specific differences in the timing of the transformation of the neuroepithelial cells into more differentiated radial glial cells (RGSs), their classification into the glial phenotype has been accepted by the researchers in this field irrespective of the species (reviewed in Rakic, 2003; see also, articles in the Special Issues of Cerebral Cortex: Rubenstein and Rakic, 1999; Kriegstein and Parnavelas, 2003; 2006).

It is not surprising that the initial discoveries of the RGCs as well as their classification into glial phenotype and significance as scaffolding have been made in the primates, because, unlike in rodents, in which these cells become GFAP positive only after birth, in both monkey and human the large, elongated GFAP positive RGCs dominate the scenery of the fetal cerebral wall in mid gestation (Rakic, 1972; Sidman and Rakic, 1973; Sidman and Rakic, 1982; Gadisseux et al., 1985; Choi, 1986; Kadhim et al, 1988; deAzevedo, 2003; Zecevic, 2004; Rakic, 2003). At that stage RGCs in the human cerebrum are truly remarkable in their size reaching a length of up to 4,000 microns in the fetal frontal lobes. Furthermore, the RGCs in primates have distinct ultra-structural and biochemical properties from the much earlier stage than in rodents (Rakic, 1972; Levitt and Rakic, 1980). While some basal processes of the RGCs freely arborize in the intermediate zone and cortical plate or are attached to blood vessels, a subset of RGCs has their basal end feet firmly embedded in the outer cerebral surface that is called the “glia limitans” (Rakic, 1972; Schmechel and Rakic, 1979a). It is this subset of RGCs that in the fetal primate telencephalon does not divide for several months (Schmechel and Rakic, 1979b). We and the others so far could not detect equivalent population of RGCs in the mouse, which in the corresponding three months not only get born but already begin their reproductive cycle.

A substantial difference between RGCs in the rodent and primate cerebral cortex described above should not be very surprising, but I found that even sophisticated neuroscientists who easily accept that lateral geniculate nucleus of the thalamus, which has no laminae in rodents, has distinct layers in the primates, do not accept that a substantial difference occurs during evolution of the cerebral cortex where one should expect the largest advance. By looking only at the common properties we can miss the most important components - the differences - where the secrets of the human advent may lie. I speculated that maintenance of a stable, non-dividing population of RGCs that provides connections between the ventricular and pial surfaces in the large, rapidly expanding and increasingly convoluted primate cerebrum has evolved to allow immature bipolar neurons to migrate to the overlying cortical plate. Thus, in primates, RGCs may have evolved to mature early in order to provide a stable scaffolding for the formation of the convoluted cortex (Rakic, 1978; 2003). The generation of radial units in the developing cortex is provided in the animated form (Supplementary Movie #1: <http://rakiclab.med.yale.edu/RadialMigration.html>). The dynamic model illustrates how relative cell positions in the protomap of the proliferative VZ are preserved during their radial migration to the cortical plate even in the gyrencephalic primate cerebrum which shifts during formation of convolutions (For details see Rakic, 1988a; Rakic, 1988b; Rakic 1995).

At the first glance it may appear paradoxical that non-neuronal (epithelial) cells play such a pivotal role as the scaffolding for placing cortical nerve cells into their proper locations. However, it should be recognized that the vertebrate central nervous system is a derivative and

elaboration of the skin of the dorsal surface of the embryo that consists of the epithelial cells with organelles such as cilia that are characteristics of the tissues exposed to the outside environments. After neuronal tube closure the dorsal surface of the embryo becomes the inner lining of the ventricular cavity. During this period cells transform into pseudo-stratified neuroepithelium that form the neural tube and budding cerebral vesicles, retaining some of the features of skin cells including cilia which can be found in virtually every neuron that derives from the VZ (Breuniug J. and Rakic, P. unpublished observation). However, as outlined below they serve several important developmental functions before disappearing and/or transforming into different cell types.

Radial Glial Cells as Neuronal Stem Cells

Apart from its remarkable size and unusual shape, RGC has been recognized as a maverick among brain cells that has a pivotal role in development and evolution of the cerebral cortex. Although it has been known for some time that the primary or conventional RGC phenotype derives from the neuroepithelial form (now called neural stem cells) that can generate neurons (e.g., Cameron and Rakic, 1991), increasing numbers of *in vivo* and *in vitro* studies indicated that these can generate not only neuronal progenitors but also directly, post mitotic neurons (Chanas-Sacre et al., 2000; Hartfuss et al. 2001; Noctor et al., 2001; Tamamaki et al., 2001; Gaiano and Fishell, 2002; Heins et al., 2002; Fishell and Kriegstein, 2003; Malatesta et al., 2003; Tramontin et al., 2003; Gal et al., 2006; Rasin et al., 2007). More recent observations confirmed the existence of dedicated neuronal progenitors in the VZ/SVZ that are derived from the parent RGCs and do not inherit the basal fiber that terminates at the pial surface (Rakic, 2003; Noctor et al, 2004; Gal et al., 2006, reviewed in Martinez-Cerdeño et al., 2006). Use of the retroviral gene transfer method, which enables to trace cell lineages in the developing mammalian telencephalon, including primates, also shows a divergence of basic cell types (Luskin et al., 1988; Parnavelas et al., 1991; Kornack and Rakic, 1995).

Classification of cells using the silver impregnation method, which depends on subtle distinctions between cell size and shapes, has suggested, but could not prove or even expose, the full spectrum of cellular heterogeneity of the embryonic cerebral wall. More recently, the use of specific cellular markers, combined with computer-assisted serial EM cell reconstruction, *in utero* electroporation and time-lapse multiphoton imaging revealed that the embryonic cerebral wall contains multiple cell classes, including dividing, post-mitotic, radially and tangentially migrating and the stationary neuronal and glial cells that are organized into transient layers (Fig. 1 and Gal., et al., 2006).

Apart from the RGCs that span the entire neocortical cerebral wall there are precursors which retract their basal processes during mitotic division (Levitt and Rakic, 1980, Levitt et al., 1983; Gal et al., 2006). In primates, the relative number of GFAP+ and GFAP-precursors changes systematically over the course of cortical neurogenesis (Levitt et al., 1983). However, neural stem cells can be isolated from both the rodent and human cerebrum VZ/SVZ and their properties analyzed *in vitro* (e.g., Laywell et al., 2000; Carpenter et al., 2001; Kirschenbaum et al., 1994). They can be tagged by the retroviral gene transfer method, their phenotype and migratory pattern followed in slice preparations of mouse and human fetal tissue (Letinic et al., 2002; Gal et al., 2006). Furthermore, the neuron-restricted stem cells produce different classes of projections and local circuit neurons (Parnavelas et al., 1991; Tan et al., 1998). Eventually fetal RGCs transform into ependymal cells, astrocytes and/or glioblasts (Fig. 1) Thus, RGCs give rise to both neuron and astrocytic progenitors that each can produce several generations of dedicated progenitors before their terminal differentiation. The next challenge is to isolate and propagate various subclasses of neuronal stem cells to better understand cortical development, but also to devise a rational strategy for replacement therapy.

Deciphering Molecular Mechanism

While classical methods enabled observations and discoveries of the major principles that were not suited for deciphering molecular mechanisms, recent intense research on neuronal migration *in vivo*, using spontaneous and induced mutations, and *in vitro*, as well as dissociated cultures and slice preparations, has revealed that multiple mechanisms and a variety of molecules are involved in this event. The process of neuronal migration itself can be dissociated into several distinct steps: mechanisms that control the mode of cell division in the VZ; those which determine cell polarity and its detachment from the ventricular surface; the extension of the leading process and its selection of migratory pathway; the attachment migrating neuron to the surface of RG fibers; the control of nuclear translocation and rate of migration ending by derailment of the neuron from the RG and cessation of migration at the final destination. Each step possesses complex molecular machinery and only hints of what is actually involved. Progress being made in this field is provided below.

During and shortly after their exit from the asymmetric cell division in the VZ/SVZ, post mitotic daughter neurons become polarized with the leading process directed toward the cortical plate aligned along the RGC shafts (Rakic, 1972; Sidman and Rakic, 1973; reviewed in Fishell and Kriegstein, 2003; Rakic, 2003). The apical processes are interconnected via Cadherin-based adherens junctions (Fishell and Kriegstein, 2003; Gotz and Huttner, 2005; Rasin et al., 2007), so that the interkinetic (up and down), nuclear movement remains restricted to the VZ within the apical processes and their end-feet, as evident by classical *in vivo* and *in vitro* approaches (Misson et al., 1991; Temple, 2001) and confirmed with *in vivo* imaging (e.g., Noctor et al., 2004; Haydar et al., 2003; Gal et al., 2006). Neurons originated in the VZ/SVZ begin migration to the overlying cortex only after completing the last cell division and establishing their polarity. As a first step, Numb (an inhibitor of Notch) segregates at the basolateral side of dividing cells and becomes enriched during interphase along the apical-most end at the adherens junctions associated with Cadherins (Rasin et al., 2007). Thus, this molecular pathway appears to be important for the determination of symmetric/asymmetric divisions as well as the detachment of post-mitotic cells from the ventricular lining and the establishment of apical basal polarity of ventricular cells.

After detaching from the ventricular surface, bipolar migrating neurons extend their leading process towards the cortex. The initial EM-observation of a close neuronglial relationship in the fetal macaque cerebral wall (Rakic, 1972) has indicated the presence of a differential binding affinity and suggests the existence of a “gliophilic” mode of migration that may be mediated by heterotypic adhesion molecules present on apposing neuronal and glial cell surfaces (Rakic, 1991; Rakic et al., 1994). In contrast, the post-mitotic cells, which move tangentially along pre-existing axonal tracts (e.g., bipolar cells in Figs. 2 and 4), were considered “neurophilic” (Rakic, 1991).

In the past three decades, radial glial-guided migration has been observed in a variety of mammalian species, but it is particularly evident in primates where the pathways are long and curvilinear, RGCs differentiate both morphologically and biochemically within the first trimester and many do not divide during the second and third trimesters (Rakic, 1976; Schmechel and Rakic, 1979a, b; Kadhim et al., 1988; Zecevic, 2004). During the late period of corticoneurogenesis (or cortical neurogenesis) in the human brain, as many as 30 migrating GFAP negative neurons have been observed migrating along a single GFAP positive radial glial fascicle (Rakic, 2003). Multiple classes of putative recognition and adhesion molecules are involved in recognition, adhesion and cessation of neuronal cell migration (Rakic, 1981, Rakic et al., 1994; Hatten 2002; Hatten and Mason, 1990). These molecules are selectively and transiently expressed in the leading process of post-mitotic neurons at the surface adjacent to the radial glial fibers. Furthermore, they are involved in a cascade of multiple molecular interactions that eventually affect cytoskeletal remodeling which is essential for nuclear

translocation (see below). Glial membrane proteins between migrating neurons and adjacent radial glial fibers are involved in the attachment of neurons to the radial glial shafts (Cameron and Rakic, 1994; Anton et al., 1996).

Our recent study, using a series of genetic, molecular and cell biological studies, shows that proper radial and laminar positioning of cortical neurons during neuronal migration to the cerebral cortex critically depends on the activity of the Notch intracellular domain, which is directly stabilized by Reelin signaling (Hashimoto-Torii et al., 2007). Numerous additional molecular species have been associated with neuronal migration that indicate the complexity of this process (e.g., Schachner et al., 1985; Hatten and Mason, 1990; Fishell and Hatten, 1991; Cameron and Rakic, 1994; Rio et al., 1997; Anton et al., 1996; Anton et al., 1997; Anton et al., 1999; Wu et al., 1999; Wynshaw-Boris and Gambello, 2001; Gongidi et al., 2004; Xie et al., 2006) and their number is growing almost daily. Although most molecules probably have specific tasks and are engaged in an interactive cooperation between neurons and glia, it is premature to assess their relative contribution and relationship to each other; however, we are witnessing big strides in this area of research.

Nuclear and somal translocation

Neuronal migration to the cerebral cortex, similarly as in other laminated structures of the CNS, begins with extension of the leading process to a variable length that is followed by the displacement of the nucleus within the leading process, for which I used the term “nuclear and/or somal translocation” (Rakic, 1971; Rakic 1972; reviewed in Rakic, 1981; 1991). The “four dimensional” figure showing morphogenetic transformation of cerebellar granule cells as it develop the descending leading process within which nucleus and surrounding translocate to its final position has been, reproduced and/or quoted over 1000 times as a unique cellular event that explains formation of cerebellar cortical architecture. Although electron microscopic methods have not been suitable for studying the dynamic molecular mechanisms underlying movement on the nucleus, the ultra-structural findings indicated that a transformation of the cytoskeleton must play a role in initiation and cessation of nuclear movement (e.g., Rakic et al., 1996). The extension of the leading process and nuclear translocation are inseparable cellular events. Initially, while the cerebral wall is relatively thin, the tip of the leading process can reach the cortical plate and the nucleus needs to move only a short distance in both the small rodent cerebrum as well as in the human at comparably early embryonic stages (Sidman and Rakic, 1973; Nadarajah et al., 2003). However, when the total length of the migratory pathway increases and the leading processes of many migrating neurons do not span the entire width of the cerebral wall, the nucleus and surrounding cytoplasm are nevertheless being intermittently translocated to reach new distances (reviewed in Sidman and Rakic, 1982; Rakic, 1988a; Rakic, 1991). Time lapse movies of dissociated cells in cultures or in organotypic slices, where the conditions are similar to the environment *in vivo*, indicate that the leading process first extends slowly, and, after a pause, the nucleus begins to move at a faster rate, consistent with the rapid dissolution of cytoplasmic microtubules (e.g., Rakic et al., 1996; Feng et al., 2004; Schaar and McConnell, 2005; Bellion et al., 2005).

Neuronal migration can be considered as a highly specialized form of cell polarity. Analysis of the polarity of microtubule assemblies (Tubulin polymerization) within the leading and trailing processes reveals that the positive ends of the newly assembled microtubules situated in the leading process are facing the growing tip, while their disintegrating negative ends face the nucleus (Rakic et al., 1996). In the trailing process, by contrast, microtubule arrays are of mixed polarity. Thus, the extension of the leading process and translocation of the soma (nucleus and surrounding cytoplasm) within the membrane envelope may be orchestrated by a synchronized polymerization and disintegration of the microtubule that creates a rearrangement of the cytoskeletal scaffolding (Rivas and Hatten, 1995; Rakic et al., 1996; Feng

et al., 2004; Schaar and McConnell, 2005; Xie et al., 2006) that is regulated by Calcium fluxes through the activity of various receptor/channel complexes (e.g., Komuro and Rakic, 1993; Behar et al., 1999; Hirai et al., 1999; Haydar et al., 2000, Owen and Kriegstein, 2002). Synergistic action of other molecular pathways may be involved in cytoskeletal rearrangement, such as Doublecortin and MEKK 4, which is important for mobilization of another cytoskeletal protein, Filamin (Sarkisian et al., 2006).

After passing between previously generated neurons, already settled in the deeper strata of the CP, the leading process enters the MZ, but the movement of the nucleus abruptly stops at the CP/MZ interface. When migrating cells are prevented from detaching from the RGC's fibers at the CP/MZ interface, subsequently arriving neurons cannot bypass their predecessors and accumulate beneath the previously generated neurons, forming an outside-to-inside gradient of neurogenesis (Anton et al., 1996; Anton and Rakic, 2002; Gongidi et al., 2004). A similar event has been observed in the cortex of the neurological mutant Reeler mouse (Caviness and Rakic, 1978).

Radial Unit Hypothesis

The above reviewed observations are all consistent with the radial unit hypothesis of cortical development (Rakic, 1988a). According to this hypothesis, the two-dimensional positional information of the proliferative units in the ventricular zone is transformed into a three-dimensional cortical architecture: the X and Y axis of cells is provided by the site of cell origin in the VZ, whereas the Z-axis is provided from the time of their origin (Rakic, 1988). This hypothesis postulates that the radial edifice of the cerebral cortex forms by a migration of vertically oriented cohorts of neurons generated at the same site in the proliferative ventricular zone (VZ) of the cerebral vesicle (Rakic, 1978). As presented in animated form (see Supplementary movie #1: <http://rakiclab.med.yale.edu/RadialMigration.html>) each radial unit consists of neurons that share their birthplace and migrate radially to the cortex following transient scaffolding made of glial fascicles spanning the cerebral wall (Rakic, 1988). After arriving to the cortical plate, later generated neurons bypass earlier generated ones and settle in an inside-out gradient of neurogenesis, even in the large convoluted primate brain (Angevine and Sidman, 1961; Rakic, 1974). The radial unit hypothesis proposed three decades ago (Rakic, 1978, 1988) has acquired the status of a universal conceptual principle for understanding the complex cellular events underlying formation of the cerebral cortex (e.g., Mountcastle, 1997; Buxhoeveden and Casanova, 2002; Hatten, 2002; Chenn and Walsh, 2003; Tan and Breen, 1993; Kriegstein and Noctor, 2004; Zecevic, 2004).

One of the postulates of the radial unit hypothesis is that cells in a given radial column are clonally related. This was tested experimentally using the retroviral gene transfer method which allowed an analysis of cell lineages in the mammalian brain *in vivo* (Luskin et al., 1988). The use of the retroviral gene transfer method in the embryonic primate brain showed that even in the large and highly convoluted cerebrum, radial deployment of neuronal clones is remarkably preserved (Kornack and Rakic, 1995; Rakic, 1995a). Use of the same method in slice preparation of the human fetal cerebrum indicates that the same holds true for the human cortex (Letinic et al., 2002).

In addition to the radially deployed clones, there are a number of migrating cells that initially do not obey radial constraints but nevertheless contribute to the columnar edifice of the cerebral cortex. It has been shown that the interneurons, which originate from the ganglionic eminence (GE), migrate tangentially to the dorsal telencephalon before assuming their final place in the appropriate layers within the cortex. (e.g., deCarlos et al., 1996; Anderson et al., 1997, 1999; Tamamaki et al., 2001; Lavdas et al., 1999; Polleux et al., 2002; Tanaka et al., 2003; Wichterle et al., 2001; Ang, et al., 2002; Letinic et al., 2002). While in rodents, the majority of cortical interneurons originate in the GE (Rev. in Marin and Rubenstein, 2001) in the developing human

cerebrum, more than 65% of GABAergic cells originate from the SVZ (Letinic et al., 2002). It should be emphasized that even cells that initially migrate tangentially in the fetal cerebrum eventually take a radial route along glial shafts (Fig. 1 and O'Rourke et al., 1992; Ware and Walsh, 1999; Ang et al., 2002; Letinic et al., 2002). Thus, radial migration is the dominant mode of cell translocation, which is particularly evident in the human fetal cerebrum (Rakic, 2003). Despite these and many other species-specific differences, the cerebral cortex, in all species examined, including human, is characterized by two prototypical and common features: it is a sheet of isochronously generated neurons that forms horizontal layers intersected by radial (vertical) columns of neurons that share a common origin in the VZ (Rakic, 1988).

Protomap Hypothesis

Neither Cajal nor Golgi have paid much attention to the cytoarchitectonic subdivision of the cortex, though they were aware of it and did appreciate its significance. The most prominent students of the histological heterogeneity of the cortical mantle were German neurologists Constantin von Economo and Korbinian Brodmann whose major cytoarchitectonic atlas appeared in 2005 (Brodmann, 1905), one year before the announcement of the Nobel Prize for brain organization. This field was tainted with Joseph Gall's pseudo science, but the Brodmann's map has been included in virtually every book of Neurology and/or neuroscience, and is considered as seminal (Swanson, 2003).

Research on the development of cytoarchitectonic heterogeneity has been dormant until the introduction of new experimental methods. Apparent morphological uniformity of the neuroepithelium lining the cerebral ventricle gave birth to the concept, known popularly as "tabula rasa" which suggests that the ventricular zone produces equipotent cells destined for the cortical plate and that only selective input from the thalamus determines a species-specific pattern of cytoarchitectonic areas. It was an attractive idea that delegates the induction of areas to the periphery, and was well formulated and advocated by Otto Creutzfeldt three decades ago (Creutzfeldt, 1977). However, more recent experimental manipulations of cortical input indicated that cells of the embryonic cerebral vesicle may not all be created equal and have some intrinsic programs for the development of basic species-specific cortical regionalization as formulated in the protomap hypothesis (Rakic, 1988a). The initial evidence of the existence of heterogeneity of the cerebral wall before the arrival of input was initially based on the experimental reduction of size of thalamic input to the developing cortex (e.g. Rakic, 1988a; Rakic 1991). According to this concept known as the "protomap hypothesis", some region-specific cytoarchitectonic features can emerge before and at least initially, independently of the input. This is an important distinction as it opens the possibility that the species-specific changes in the cortex itself may be a direct target of evolution, rather than an indirect consequence of the changes in sensory systems at the periphery (Rakic, 1995). However, the word "proto" in the term "protomap" has been introduced to emphasize the malleable feature of this primordial map in which intrinsic cues, generated within cortical neurons, attract appropriate input that then cooperatively create a final area-specific, three-dimensional organization of each cytoarchitectonic field (Rakic, 1988a). Thus, although we found that input from the thalamus does not initiate regionalization of the cortex, we recognized that it is essential for its proper development and maturation (e.g. Rakic, 1988a, Rakic et al, 1991).

The evidence supporting intrinsic specification of cortical maps (or protomap) has over the years accumulated in numerous reports showing that a selective families of genes and morphoregulatory molecules are expressed in discrete gradients and cortical regions before or independently of the incoming input (Ferri and Levitt, 1993; Nakatsuji et al., 1991; Arimatsu et al., 1992; Simeone, et al., 1992; Gitton et al, 1999; Miyashita-Lin, et al, 1999; Bishop et al., 2000; Sestan et al., 2001; Grove and Fukuchi-Shimogori, 2003; O'Leary and Borngasser, 2006; Cholfin and Rubenstein, 2007; rev in Rubenstein and Rakic 1999). Furthermore, a

number of studies in chimeric and transgenic mice have provided direct evidence that the majority of post-mitotic, clonally-related neurons move and remain radially distributed within the intermediate zone and enter only restricted superjacent areas of the cortex (Nakatsuji et al., 1991; Tan and Breen, 1993; Parnavelas et al., 1991; Soriano et al., 1995; Tan et al., 1998; reviewed in Rakic, 1995a). As a consequence many cortical malformations in human such as polymicrogyria have a propensity to develop only in specific regions of the cerebral cortex (e.g. Berkovich et al., 2000; Piao et al., 2004; reviewed in Rakic, 2004). The protomap hypothesis has also gained support from evidence that abolition of thalamic input by genetic manipulation does not prevent cortical parcellation (Miyashita-Lin et al., 1999; Mallamaci, et al., 2000; Bishop, et al., 2000; Cholfin and Rubenstein, 2007). Finally, recent studies using genetic approaches have provided firm evidence that layer specificity of the cortical neurons that was indicated by early studies in Reeler mouse (Caviness and Rakic, 1978) and heterologous transplantations (McConnell, 1988) is determined already at the final cell division in the proliferative zones (Chen et al., 2005; Chen et al., 2005; Molyneux et al., 2005). These studies collectively exemplify the capacity of the modern methods to rearrange and create at will new cortical connections and maps using genetic tools that Ramón y Cajal and his contemporaries could not even predict. Now we can predict with considerable confidence that some additional genes and morphoregulatory molecules may be involved in cortical specification which then could be tested in rodents and possibly primate models of cortical dysgenesis.

Although, the clonally relationship of labeled cells using this approach was originally based on the law of probability, cell distribution strongly suggested that most progenitors originating in the same site at the ventricular surface remain radially deployed during migration and settle in a columnar fashion in the cortex (Luskin et al., 1988; Kornack and Rakic, 1995; Rakic, 1995a). Thus, radial migration directed by the radial glial scaffolding translates positional information (protomap) from the VZ to the overlying cortical plate (Rakic, 1988a).

Radial unit hypothesis of cortical evolution

Ramón y Cajal recognized that evolution provides nature's experiment and that comparative embryological and anatomical studies are very useful approaches. Cajal also recognized that the cerebral cortex is an organ that makes the most important distinction between human and any other organism. It is a paradox of our time that, seduced by the similarities, we often neglect the importance of distinctions. Indeed it should be expected that millions of years of evolution which selected enhanced mental capacity for survival would leave a significant mark on the organization of cerebral cortex. Although Cajal has often commented on the differences in cortical organization, particularly in terms of cell types (e.g. Ramón y Cajal, 1881, 1890, 1899), he did not think in terms of the genetic and cellular mechanisms of evolution, nor could he at that time.

Modern methods of molecular and developmental biology now provide the unprecedented opportunity to get a glimpse of what and how our cortex may have evolved. When one thinks about how to approach this question, there is an inescapable conclusion that analysis of the molecular and cellular events occurring at the early stages of embryonic development maybe the only way to learn what might have happened millions of years ago. This is not a resurrection of the discredited Haeckel's law that ontogeny "recapitulates phylogeny", based on morphological similarities, but a realization that genetic mutations that occurred in our ancestors have left imprints on our present phenotype and can be traced back. For example, the fact that the human, mouse or macaque telencephalic primordium is different in size even before the first cortical neuron has been generated, indicated to me that the mutation of the gene(s) that control the size of the neural stem cell pool must be the first step in the expansion of the human cortex. The particular aspect of cortical development that there is an expansion in cortical surface without expansion of cortical width, as predicted by Radial Unit Hypothesis.

The mechanisms underlying the expansion of the cerebral cortex is central to understanding both the potential and limits of our mental capacity including human-specific traits, such as language and abstract thinking. The human cerebral cortex is physically distinguished from other species not only by its larger size, but also by differential expansion and the addition of specialized cytoarchitectonic areas which are associated with an increase in the number of functional columns. Although the exact relationship between the functional columns in the adult cortex and embryonic columns has not been precisely determined, it is clear that the larger the cortex in a given species, the larger the number of participating columnar units (Bugbee and Goldman-Rakic, 1984; Rakic, 1988; Rakic, 1995b).

The evolutionary novelties in the cortex, as in other organs, are likely to be an outcome of relatively small genetic differences effecting the timing, sequence and level of gene expression that are introduced during the early embryonic stages. In this respect, it is important to recognize that the human forebrain is much bigger than in either the mouse or monkey, even at embryonic stages before the onset of neurogenesis of the local VZ (Sidman and Rakic, 1973; Sidman and Rakic 1982; Bystron et al., 2006). During the formation of the layer VI, its surface area enlarges another ten times. Thus, as predicted by the Radial Unit Hypothesis, the increase in cortical surface during evolution is associated with an initial increase in size of the VZ that generates mainly projection neurons. The idea of the Radial Unit Hypothesis of evolution has emerged from our concomitant, comparative analysis of ontogenetic development of the cerebral cortex in the mouse, monkey and human which shows a large and systematic increase in the number of radial columns during evolution without a comparable increase in the height of the columns (Rakic, 1995a).

According to the radial unit hypothesis, an increase in cell production at early stages (before the onset of neurogenesis) would result in an increase in the number of founder cells that give rise to radial columns, while an increase in production at later stages increases in number the neurons within each column. More specifically, a minor increase in the length of cell cycles or the number of cell divisions in the macaque monkey VZ before E40 can result in a large increase in the number of founder cells that form proliferative units (Rakic, 1988a). Since the initial proliferation in the VZ proceeds exponentially by the prevalence of symmetrical divisions, an additional round of mitotic cycles during this phase doubles the number of proliferative units and, consequently, the number of radial columns (Rakic, 1995b). According to this model, about three extra rounds of cell divisions can account for the 8 to 10-fold difference in size of the cortical surface between monkeys and human. In contrast, the 1000-fold difference between the size of the cerebral cortex in mouse and human can be achieved by less than 7 extra symmetrical divisions in the ventricular zone before the onset of corticogenesis (Rakic and Kornack, 2001). After the number of founder cells that will form individual radial units is set and corticogenesis has begun, many progenitor cells start to divide asymmetrically (Rakic, 1988a). Therefore, during this phase of development, an extra round of cell divisions would have a negligible effect on the thickness of the cortex (Rakic, 1995a; 2006). According to this model, one can predict that an approximate additional two weeks of corticogenesis in human compared to the duration in the macaque should enlarge the cortical thickness by only 10% to 20%, which is actually observed (Rakic, 1995b).

Can radial unit hypothesis of evolution be tested experimentally? I think that at least the cellular mechanisms of how it could have happened can be falsified. Based on our experience in studying the developing cortex in mouse, monkey and human, I proposed that a mutation in only one or a few genes can account for the dramatic increase in cortical surface, without a substantial increase in its thickness, provided that the gene(s) act early and that post-mitotic neurons follow the same rules and pattern of migration (Rakic, 1995b). Both excessive proliferation, as well as diminished programmed cell death (apoptosis) could affect the radial columns. An example of the later category is the development of the cerebral cortex in the

transgenic mouse deficient in caspase 3 and 9 protease, that are essential for the normal process of programmed cell death (apoptosis). In mice lacking both copies of these genes, the magnitude of apoptosis is reduced in the proliferative VZ at early stages, during production of the founder progenitor cells (Kuida et al., 1996, 1998). Thus, the affected mice have a larger number founder cells and RGCs and as a consequence larger cortical surface that begin to form convolutions (Haydar et al., 1999). A similar, though less dramatic effect on the programmed death of neuronal progenitors can be obtained by modulation of the ephrin-A/EphA signaling pathway (Depaepe et al., 2006; Rakic, 2006).

There are also examples in which an increase in cell proliferation, rather than decrease in cell death in the VZ/SVZ enlarges the surface of the cerebral cortex. Thus, the embryonic mouse brain over expressing β -catenin produces a larger number of neuronal progenitors and eventually more neurons by acting directly on the decision of progenitors to exit the cell cycle in the proliferative zones (Chenn and Walsh, 2005). The increase in the number founder cells lead to larger number of radial columns and the formation of elaborate cerebral convolutions in the mouse that normally has a smooth (lissencephalic) hemispheric surface (Chenn and Walsh, 2005). In contrast, over-expression of cyclin-dependent kinase (cdk) inhibitor p27, increases the production of neurons that form radial columns, thus increasing the thickness of the superficial layers of the cortex, without increasing its surface area (Tarui et al., 2005). Often the picture may be obscured because supernumerary progeny causes additional brain malformations. Nevertheless, genetic and cellular mechanisms by which the neocortex expands are realistic goals of modern developmental neurobiology and a number of candidate genes that control cell proliferation are expected to increase.

During evolution functionally and structurally new cortical areas are introduced and the existing areas do not expand in register with the overall surface expansion. Some of the recent experimental studies in mice can inspire ideas on how the expanded cortex may have acquired additional cytoarchitectonic areas during evolution. For example it is significant that ectopic barrel fields, created *de novo* by perturbations of the site and expression level of a family member of the fibroblast growth factor (Fgf8) have a reverse (mirror image) representation of the whiskers, as has occurred when an area becomes duplicated during evolution (Fukuchi-Shimogori and Grove, 2001). The recent finding that even subdivisions of the frontal lobes can be distinguished and their size regulated at by expression of *Fgf17* at early embryonic is a strong evidence of the role of genes in cortical regionalization (Cholfin and Rubenstein, 2007). The fact that it is now possible to enlarge and/or duplicate at will the sensory cortical representation of the periphery and create a new functional cytoarchitectonic area in the cerebral cortex presents an unprecedented opportunity to find out how these maps develop in individual as well as how they may been introduced during evolution (Rakic, 2001). I was amused that my statement how new cortical area can be created quickly in an individual embryo by manipulation of FGF8, that I made in my Commentary article in Science (Rakic, 2001) has been quoted by the creationists as a scientific evidence that human cortex can be created in a single day!

It is obvious that the increase in the number of neurons organized into a larger number of radial columns and areas is not sufficient to explain the evolutionary advances made in the cerebral cortex over millions of years of evolution that also involves elaboration of neuronal connections. However, since neurons have to be generated before they form connections, an increase in their number is an essential first step, without which the next steps cannot be achieved. The experiments in which the number of radial units is increased is only an example, which in the context of the radial unit hypothesis explains how a larger-than-normal number of founder cells can create a cortex with an increased surface area and the formation of convolutions. By a single gene mutation, a lissencephalic (smooth) mouse cortex can be transformed into a gyrencephalic (convoluted) cerebrum! These are the kind of experiments

that Cajal and Golgi could not do, or even perhaps could not imagine that could ever be accomplished. These experiments illustrate the remarkable power of molecular and developmental neurobiology: how by application of the genes identified initially in invertebrates can be applied to test the principles that were discovered in the human cerebral cortex. We can, for example, use a cell death gene, identified in a roundworm, to help us understand how changes in the regulation of cell death during cortical neurogenesis may have lead to cortical expansion during primate evolution. This type of research gives us confidence that human advent in universe, achieved through expansion and elaboration of cerebral cortex, can and will be eventually explained.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

References

- Angevine JB Jr, Sidman RL. Autoradiographic study of cell migration during histogenesis of cerebral cortex in the mouse. *Nature* 1961;192:766–768. [PubMed: 17533671]
- Anton ES, Cameron RS, Rakic P. Role of neuroglial junctional proteins in the maintenance and termination of neuronal migration across the embryonic cerebral wall. *J Neurosci* 1996;16:2283–2293. [PubMed: 8601808]
- Anton ES, Kreidberg J, Rakic P. Distinct functions of $\alpha 3$ and αv integrin receptors in neuronal migration and laminar organization of the cerebral cortex. *Neuron* 1999;22:227–289.
- Anton ES, Marchionni MA, Lee KF, Rakic P. Role of GGF/neuregulin signaling in interactions between migrating neurons and radial glia in the developing cerebral cortex. *Development* 1997;124:3501–3510. [PubMed: 9342043]
- Arimatsu Y, Miyamoto M, Nihonmatsu I, Hirata K, Urataini Y, Hatanka Y, Takiguchi-Hoyash K. Early regional specification for a molecular neuronal phenotype in the rat neocortex. *Proc Nat Acad Sci USA* 1992;89:8879–8883. [PubMed: 1409580]
- Behar TN, Scott CA, Greene CL, Wen X, Smith SV, Maric D, Liu QY, Colton CA, Baker JL. Glutamate acting at NMDA receptors stimulates embryonic cortical neuronal migration. *J Neurosci* 1999;19:4449–4461. [PubMed: 10341246]
- Bellion M, Courbot M, Jacob C, et al. Nucleokinesis in tangentially migrating neurons comprises two alternating phases: Forward migration of the golgi/centrosome associated with centrosome splitting and myosin contraction at the rear. *J Neurosci* 2005;25:173–181.
- Barkovich AJ, Hevner R, Guerrini R. Syndromes of bilateral symmetrical polymicrogyria. *Am J Neuropradiology* 2000;20:1814–1821.
- Bhardwaj RD, Curtis MA, Spalding KL, Buchholz BA, Fink D, Björk-Eriksson T, Nordborg C, Gage FH, Druid H, Eriksson PS, Frisén J. The age of human cerebral cortex neurons. *Proc Nat Acad Sci USA* 2006;103:12564–12568. [PubMed: 16901981]
- Bishop KM, Goudreau G, O’Leary DD. Regulation of area identity in the mammalian neocortex by *Emx2* and *Pax6*. *Science* 2000;288:344–349. [PubMed: 10764649]
- Boulder, Committee. Embryonic vertebrate central nervous system. Revised terminology *Anat Record* 1970;166:257–261.
- Bourgeois JP, Rakic P. Changing of synaptic density in the primary visual cortex of the rhesus monkey from fetal to adult stage. *J Neurosci* 1993;13:2801–2820. [PubMed: 8331373]
- Brodmann K. Beitrage zur histologischen localization der grosshirnrinde dritte mitteilung: Die rinderfelder niederen affen. *J Psychol Neurol* 1905;9:177–226.
- Bugbee NM, Goldman-Rakic PS. Columnar organization of corticocortical projections in squirrel and rhesus monkeys: similarity of column width in species differing in cortical volume. *J Comp Neurol* 1983;220:355–364. [PubMed: 6315786]
- Buxhoeveden DP, Casanova MF. The minicolumn hypothesis in neuroscience: A review. *Brain* 2002;125:935–951. [PubMed: 11960884]

- Bystron I, Rakic P, Molnar Z, Blakemore C. The first neurons of the human cerebral cortex. *Nature Neurosci* 2006;9:880–885. [PubMed: 16783367]
- Cameron RS, Rakic P. Glial cell lineage in the cerebral cortex: Review and synthesis. *Glia* 1991;4:124–137. [PubMed: 1827774]
- Cameron RS, Rakic P. Polypeptides that comprise the plasmalemmal microdomain between migrating neuronal and glial cells. *J Neurosci* 1994;14:3139–3155. [PubMed: 8182462]
- Carpenter MK, Inokuma MS, Denham J, Mujtaba T, Chiu CP, Rao MS. Enrichment of neurons and neural precursors from human embryonic stem cells. *Exp Neurol* 2001;172:383–397. [PubMed: 11716562]
- Caviness VS Jr, Rakic P. Mechanisms of cortical development: a view from mutations in mice. *Ann Rev Neurosci* 1978;1:297–326. [PubMed: 386903]
- Chanas-Sacre G, Rogister B, Moonen G, Leprince P. Radial glia phenotype: origin, regulation, and transdifferentiation. *J Neurosci Res* 2000;61:357–363. [PubMed: 10931521]
- Chen B, Schaevitz LR, McConnell SK. Fezl regulates the differentiation and axon targeting of layer 5 subcortical projection neurons in cerebral cortex. *Proc Natl Acad Sci USA* 2005;102:17184–17189. [PubMed: 16284245]
- Chen JG, Rasin MR, Kwan KY, Sestan N. Zfp312 is required for subcortical axonal projections and dendritic morphology of deep-layer pyramidal neurons of the cerebral cortex. *Proc Natl Acad Sci USA* 2005;102:17792–17797. [PubMed: 16314561]
- Chenn A, Walsh CA. Increased neuronal production, enlarged forebrains and cytoarchitectural distortions in beta-catenin overexpressing transgenic mice. *Cereb Cortex* 2003;13:599–606. [PubMed: 12764034]
- Choi BH. Glial fibrillary acid protein in radial glia of early human fetal cerebrum: A light and electron microscopic immunocytochemical study. *J Neuropath Exp Neurol* 1986;45:408–418. [PubMed: 3522808]
- Cholfin JA, Rubenstein JLR. Patterning of frontal cortex subdivisions by Fgf17. *Proc Natl Acad Sci USA*. 2007In press
- Creutzfeldt OD. Generality of the functional structure of the neocortex. *Naturwissenschaften* 1977;64:507–517. [PubMed: 337161]
- deAzevedo LC, Fallet C, Moura-Neto V, Dumas-Duport C, Hedin-Pereira C, Lent R. Cortical radial glial cells in human fetuses: Depth-correlated transformation into astrocytes. *J Neurobiology* 2003;55:288–298.
- DeCarlos JA, LopezMascaraque L, Valverd F. Dynamics of cell migration from the lateral ganglionic eminence in the rat. *J Neurosci* 1996;16:6146–6156. [PubMed: 8815897]
- Depaepe V, Suarez-Gonzalez N, Dufour A, Passante L, Gorski JA, Jones KR, Ledent C, Vanderhaeghen P. Ephrin signalling controls brain size by regulating apoptosis of neural progenitors. *Nature* 2005;435:1244–1250. [PubMed: 15902206]
- Eccles, JC. The cerebral neocortex: A theory of its operation. In: Jones, EG.; Peters, A., editors. *Cerebral Cortex*. 2. New York: Plenum Press; 1984. p. 1-36.
- Feng YY, Walsh CA. The many faces of filamin: A versatile molecular scaffold for cell motility and signaling. *Nature Cell Biol* 2004;6:1034–1038. [PubMed: 15516996]
- Ferri RT, Levitt P. Cerebral cortical progenitors are fated to produce region-specific neuronal populations. *Cerebral Cortex* 1993;3:187–198. [PubMed: 8324369]
- Fishell G, Hatten ME. Astrotactin provides a receptor system for CNS neuronal migration. *Development* 1991;113:755–765. [PubMed: 1821847]
- Fishell G, Kriegstein AR. Neurons from radial glia: the consequences of asymmetric inheritance. *Curr Opin Neurobiol* 2003;13:34–41. [PubMed: 12593980]
- Fukuchi-Shimogori T, Grove EA. Neocortex patterning by the secreted signaling molecule FGF8. *Science* 2001;294:1071–1074. [PubMed: 11567107]
- Funahashi S, Bruce CJ, Goldman-Rakic PS. Mnemonic coding of visual space in the monkey's dorsolateral prefrontal cortex. *J Neurophysiol* 1989;61:331–349. [PubMed: 2918358]
- Gadisseux JF, Evrard P. Glial-neuronal relationship in the developing central nervous system. *Dev Neurosci* 1985;7:12–32. [PubMed: 4029035]

- Gaiano N, Fishell G. The role of Notch in promoting glial and neural stem cell fates. *Annu Rev Neurosci* 2002;25:471–490. [PubMed: 12052917]
- Gal JS, Morozov YM, Ayoub AE, Chatterjee M, Rakic P, Haydar TF. Molecular and morphological heterogeneity of neural precursors in the mouse neocortical proliferative zones. *J Neurosci* 2006;26:1045–1056. [PubMed: 16421324]
- Gitton Y, Cohen-Tannoudji M, Wassef M. Role of thalamic axons in the expression of H-2Z1, a mouse somatosensory cortex Specific marker. *Cereb Cortex* 1999;9:611–620. [PubMed: 10498279]
- Goldman-Rakic, PS. Circuitry of primate prefrontal cortex and regulation of behavior by representational memory. In: Plum, F., editor. *Handbook of Physiology, The Nervous System, Higher Functions of the Brain Section I. V.* Am. Physiol. Soc; Bethesda, Md: 1987. p. 373–417.
- Goldman-Rakic PS. Topography of cognition: parallel distributed networks in primate association cortex. *Annu Rev Neurosci* 1988;11:137–156. [PubMed: 3284439]
- Goldman-Rakic PS. Cellular basis of working memory. *Neuron* 1995;14:477–485. [PubMed: 7695894]
- Golgi, C. Sulla fina anatomia degli organi centrali del sistema nervoso. Tipografia di Stefano Calderini e Figlio; Reggio Emilia: 1885.
- Gongidi V, Ring C, Rakic P, Anton ES. Sparc-like 1 is a radial glia-associated terminator of neuronal migration in cerebral cortex. *Neuron* 2004;41:57–69. [PubMed: 14715135]
- Gotz M, Huttner WB. The cell biology of neurogenesis. *Nat Rev Mol Cell Biol* 2005;6:777–788. [PubMed: 16314867]
- Grove EA, Fukuchi-Shimogori T. Generating the cerebral cortical area map. *Annu, Rev Neurosci* 2003;26:355–380. [PubMed: 14527269]
- Hartfuss E, Galli R, Heins N, Gotz M. Characterization of CNS precursor subtypes and radial glia. *Dev Biol* 2001;229:15–30. [PubMed: 11133151]
- Hashimoto-Torii K, Torii M, Sarkisian MR, Bartley J, Shen J, Radtke F, Gridley T, Honjo T, Sestan N, Rakic P. Stabilization of activated Notch by Reelin signaling is required for cortical neuronal migration. 2007Submitted
- Hatten ME. New directions in neuronal migration. *Science* 2002;297:1660–1663. [PubMed: 12215636]
- Hatten ME, Mason CA. Mechanism of glial-guided neuronal migration in vitro and in vivo. *Experientia* 1990;46:907–916. [PubMed: 2209800]
- Haydar TF, Ang ESBC Jr, Rakic P. Mitotic spindle rotation and mode of cell division in the developing telencephalon. *Proc Natl Acad Sci USA* 2003;100:2890–2895. [PubMed: 12589023]
- Haydar TF, Kuan CY, Flavell RA, Rakic P. The Role of cell death in regulating the size and shape of the mammalian forebrain. *Cereb Cortex* 1999;9:621–626. [PubMed: 10498280]
- Haydar TF, Wang F, Schwartz ML, Rakic P. Differential modulation of proliferation in the neocortical ventricular and subventricular zones. *J Neurosci* 2000;20:5764–5774. [PubMed: 10908617]
- Heins N, Malatesta P, Cecconi F, Nakafuku M, Tucker KL, Hack MA, Chapouton P, Barde YA, Gotz M. Glial cells generate neurons: the role of the transcription factor Pax6. *Nat Neurosci* 2002;5:308–315. [PubMed: 11896398]
- Hirai K, Yoshioka H, Kihara M, Hasegawa K, Sakamoto T, Sawada T, Fushiki S. Inhibition of neuronal migration by blocking NMDA receptors in the embryonic rat cerebral cortex: a tissue culture study. *Dev Brain Research* 1999;114:63–67.
- His, W. *Naturforscher*. Engelman: 1874. Unsere Körperform und das physiologische Problem ihrer Entstehung: Briefe an einen befreundeten.
- His W. *Die Entwicklung des menschlichen Gehirns während der ersten Monate*. Leipzig, Hirzel. 1904
- Hubel DH, Wiesel TN. Anatomical demonstration of columns in the monkey striate cortex. *Nature* 1969;221:747–750. [PubMed: 4974881]
- Kadhim HJ, Gadisseux JF, Evrard P. Topographical and cytological evolution of the glial phase during prenatal development of the human brain: Histochemical and electron microscopic study. *J Neuropath Exp Neurol* 1988;47:166–188. [PubMed: 3339373]
- Kirschenbaum B, Nedergaard M, Preuss A, et al. In-vitro neuronal production and differentiation by precursor cells derived from the adult human forebrain. *Cereb Cortex* 1994;4:576–589. [PubMed: 7703685]

- Kölliker, A. Grundriss der Entwicklungsgeschichte des menschen und der höheren Tiere. Engelmann: Leipzig; 1879.
- Komuro H, Rakic P. Modulation of neuronal migration by NMDA receptors. *Science* 1993;260:95–97. [PubMed: 8096653]
- Kornack DR, Rakic P. Radial and horizontal deployment of clonally related cells in the primate neocortex: Relationship to distinct mitotic lineages. *Neuron* 1995;15:311–321. [PubMed: 7646888]
- Kornack DR, Rakic P. Cell proliferation without neurogenesis in the adult primate neocortex. *Science* 2001;294:2127–2130. [PubMed: 11739948]
- Kostovic I, Rakic P. Developmental history of the transient subplate zone in the visual and somatosensory cortex of the macaque monkey and human brain. *J Comp Neurol* 1990;297:441–470. [PubMed: 2398142]
- Kriegstein AR, Parnavelas JG. Progress in corticogenesis. *Cereb Cortex*, Special issue 2006;1:i1–i165.
- Kriegstein A, John G, Parnavelas JG. Changing concepts of cortical development. *Cereb Cortex* 2003;13:541–699. [PubMed: 12764026]
- Kriegstein AR, Noctor SC. Patterns of neuronal migration in the embryonic cortex. *Trends Neurosci* 2004;27:392–399. [PubMed: 15219738]
- Kuida K, Haydar T, Kuan CY, Yong G, Taya C, Karasuyama A, Su SH, Rakic P, Flavell RA. Reduced apoptosis and cytochrome c-mediated caspase activation in mice lacking Caspase-9. *Cell* 1998;94:325–33. [PubMed: 9708735]
- Kuida K, Zheng TS, Na S, Kuan CY, Yang D, Karasuyama H, Rakic P, Flavell RA. Decreased apoptosis in the brain and premature lethality in CPP32-deficient mice. *Nature* 1996;384:368–372. [PubMed: 8934524]
- LaMantia AS, Rakic P. Axon overproduction and elimination in the corpus callosum of the developing rhesus monkey. *J Neurosci* 1990;10:2156–2175. [PubMed: 2376772]
- Laywell ED, Rakic P, Kukekov VG, Holland EC, Steindler D. A identification of a multipotent astrocytic stem cell in the immature and adult mouse brain. *Proc Nat Acad Sci USA* 2000;97:13883–13888. [PubMed: 11095732]
- Letinic K, Zoncu R, Rakic P. Origin of GABAergic neurons in the human neocortex. *Nature* 2002;417:645–649. [PubMed: 12050665]
- Levitt P, Cooper ML, Rakic P. Early divergence and changing proportions of neuronal and glial precursor cells in the primate cerebral ventricular zone. *Dev Biology* 1983;96:472–484.
- Levitt P, Rakic P. Immunoperoxidase localization of glial fibrillary acid protein in radial glial cells and astrocytes of the developing rhesus monkey brain. *Comp Neurol* 1980;193:815–840.
- Lorente de Nó, R. The cerebral cortex: Architecture, intracortical connections and motor projections. In: Fulton, JF., editor. *Physiology of the Nervous System*. London: Oxford University Press; 1938. p. 291–339.
- Lund JS. Specificity and non-specificity of synaptic connections within mammalian visual cortex. *J Neurocytology* 2002;31:203–209.
- Lund JS, Boothe RG, Lund RD. Development of neurons in the visual cortex (area 17) of the monkey (*Macaca nemestrina*): a Golgi study from fetal day 127 to postnatal maturity. *J Comp Neurol* 1977;176:149–188. [PubMed: 410850]
- Luskin MB, Pearlman AL, Sanes JR. Cell lineage in the cerebral cortex of the mouse studied *in vivo* and *in vitro* with a recombinant retrovirus. *Neuron* 1988;1:635–647. [PubMed: 3272182]
- Magini G. Sur la neuroglie et les cellules nerveuses cerebrales chez les foetus. *Arch Ital Biol* 1888;9:59–60.
- Malatesta P, Hack MA, Hartfuss E, Kettenmann H, Klinkert W, Kirchhoff F, Gotz M. Neuronal or glial progeny: Regional differences in radial glia fate. *Neuron* 2003;37:751–764. [PubMed: 12628166]
- Mallamaci A, Muzio L, Chan CH, Parnavelas J, Boncinelli E. Area identity shifts in the early cerebral cortex of *Emx2*^{-/-} mutant mice. *Nat Neurosci* 2000;3:679–686. [PubMed: 10862700]
- Marin O, Rubenstein JL. A long, remarkable journey: tangential migration in the telencephalon. *Nat Rev Neurosci* 2001;2:780–790. [PubMed: 11715055]
- Martinez-Cerdeño V, Noctor SC, Kriegstein AR. The role of intermediate progenitor cells in the evolutionary expansion of the cerebral cortex. *Cereb Cortex Suppl* 2006;1:i152–i61.

- McConnell SK. Fates of visual cortical neurons in the ferret after isochronic and heterochronic transplantation. *J Neurosci* 1988;8:945–974. [PubMed: 3346731]
- McKinstry RC, Mathur A, Miller JH, Ozcan A, Snyder AZ, Schefft GL, Almlil CR, Shiran SI, Conturo TE, Neil JJ. Radial organization of developing preterm human cerebral cortex revealed by non-invasive water diffusion anisotropy MRI. *Cereb Cortex* 2002;12:1237–1243. [PubMed: 12427675]
- Misson JP, Austin CP, Takahashi T, Cepko CL, Caviness VS. The alignment of migrating neural cells in relation to the murine neopallial radial glial fiber system. *Cereb Cortex* 1991;1:221–229. [PubMed: 1668365]
- Misson JP, Edwards MA, Yamamoto M, Caviness VS. Mitotic cycling of radial glial cells of the fetal murine cerebral wall: A combined autoradiographic and immunohistochemical study. *Dev Brain Res* 1988;38:183–190.
- Miyashita-Lin EM, Hevner R, Montzka Wasserman K, Martinez S, Rubenstein JLR. Early neocortical regionalization in the absence of thalamic innervation. *Science* 1999;285:906–909. [PubMed: 10436162]
- Molyneaux BJ, Arlotta P, Hirata T, Hibi M, Macklis JD. Fezl is required for the birth and specification of corticospinal motor neurons. *Neuron* 2005;47:817–831. [PubMed: 16157277]
- Mountcastle VB. The evolution of ideas concerning the function of the neocortex. *Cereb Cortex* 1995;5:289–295. [PubMed: 7580121]
- Mountcastle VB. The columnar organization of the neocortex. *Brain* 1997;120:701–722. [PubMed: 9153131]
- Mountcastle VB, Davies PW, Berman AL. Response properties of neurons of cats somatic sensory cortex to peripheral stimuli. *J Neurophysiology* 1957;20:374–407.
- Nadarajah B, Alifragis P, Wang ROL, Parnavelas JG. Neuronal migration in the developing cerebral cortex: observations based on real-time imaging. *Cereb Cortex* 2003;13:607–611. [PubMed: 12764035]
- Nakatsuji M, Kadokawa Y, Suemori H. Radial columnar patches in the chimeric cerebral cortex visualized by use of mouse embryonic stem cells expressing beta-galactosidase. *Dev Growth and Differ* 1991;33:571–578.
- Noctor SC, et al. Cortical neurons arise in symmetric and asymmetric division zones and migrate through specific phases. *Nat Neurosci* 2004;7:136–144. [PubMed: 14703572]
- Noctor SC, Flint AC, Weissman TA, Dammerman RS, Kriegstein AR. Neurons derived from radial glial cells establish radial units in neocortex. *Nature* 2001;409:714–720. [PubMed: 11217860]
- Nowakowski RS, Rakic P. Clearance rate of exogenous 3H-thymidine from the plasma of pregnant rhesus monkeys. *Cell Tissue Kinet* 1974;7:189–194. [PubMed: 4205927]
- O’Leary DDM, Borngasser D. Cortical ventricular zone progenitors and their progeny maintain spatial relationships and radial patterning during preplate development indicating an early protomap. *Cereb Cortex Suppl* 2006;1:i46–i56.
- O’Rourke NA, Dailey ME, Smith SJ, McConnell SK. Diverse migratory pathways in the developing cerebral cortex. *Science* 1992;258:299–302. [PubMed: 1411527]
- Parnavelas JG, Barfield JA, Franke E, Luskin MB. Separate progenitor cells give rise to pyramidal and nonpyramidal neurons in the rat telencephalon. *Cereb Cortex* 1991;1:463–491. [PubMed: 1822752]
- Piao X, Hil RS, Bodell A, Chang BS, Basel-Vanagaite L, Straussberg R, Dobyns WB, Qasrawi B, Winter RM, Innes AM, Voigt T, Ross ME, Michaud JL, Descarie JC, Barkovich AJ, Walsh CA. G protein-coupled receptor-dependent development of human frontal cortex. *Science* 2004;303:2033–2036. [PubMed: 15044805]
- Rakic P. Thesis for Sc D degree (in Serbo-Croatian). Belgrade: 1968. Studies on the proliferation, migration, and differentiation of neuroblasts during neurohistogenesis in mammals, particularly man; p. 191
- Rakic P. Neuron-glia relationship during granule cell migration in developing cerebellar cortex. A Golgi and electronmicroscopic study in *Macacus rhesus*. *J Comp Neurol* 1971;141:283–312. [PubMed: 4101340]
- Rakic P. Mode of cell migration to the superficial layers of fetal monkey neocortex. *J Comp Neurol* 1972;145:61–84.

- Rakic P. Neurons in the monkey visual cortex: Systematic relation between time of origin and eventual disposition. *Science* 1974;183:425–427. [PubMed: 4203022]
- Rakic P. Neuronal migration and contact guidance in primate telencephalon. *Postgraduate Medical Journal* 1978;54:25–40. [PubMed: 364453]
- Rakic P. Neuronal-glial interaction during brain development. *Trends in Neurosci* 1981;4:184–187.
- Rakic P. Limits of neurogenesis in primates. *Science* 1985;227:154–156. [PubMed: 17843070]
- Rakic P. Mechanism of ocular dominance segregation in the lateral geniculate nucleus: competitive elimination hypothesis. *Trends in Neuroscience* 1986;9:11–15.
- Rakic P. Specification of cerebral cortical areas. *Science* 1988a;241:170–176. [PubMed: 3291116]
- Rakic P. Defects of neuronal migration and pathogenesis of cortical malformations. *Prog Brain Res* 1988b;73:15–37. [PubMed: 3047794]
- Rakic P. Principles of neuronal cell migration. *Experientia* 1990;46:882–891. [PubMed: 2209797]
- Rakic P. A small step for the cell - a giant leap for mankind: a hypothesis of neocortical expansion during evolution. *Trends in Neurosci* 1995a;18:383–388.
- Rakic P. Radial versus tangential migration of neuronal clones in the developing cerebral cortex. *Proc Nat Acad Sci USA* 1995b;92:11323–11327. [PubMed: 8524778]
- Rakic P. Neurocreationalism: making new cortical maps. *Science* 2001;294:1011–1012. [PubMed: 11691974]
- Rakic P. Pre and post-developmental neurogenesis in primates. *Clinical Neurosci Res* 2002;2:29–39.
- Rakic P. Elusive radial glial cells: Historical and evolutionary perspective. *Glia* 2003;43:19–32. [PubMed: 12761862]
- Rakic P. Genetic control of cortical convolutions. *Science* 2004;303:1983–1984. [PubMed: 15044793]
- Rakic P. No more cortical neurons for you *Science*. *Neuroscience* 2006;313:928–929.
- Rakic P, Bourgeois JP, Eckenhoff ME, Zecevic N, Goldman-Rakic PS. Concurrent overproduction of synapses in diverse regions of the primate cerebral cortex. *Science* 1986;232:232–235. [PubMed: 3952506]
- Rakic P, Cameron RS, Komuro H. Recognition, adhesion, transmembrane signaling, and cell motility in guided neuronal migration. *Current Opin Neurobiol* 1994;4:63–69.
- Rakic P, Knyihar-Csillik E, Csillik B. Polarity of microtubule assembly during neuronal migration. *Proc Nat Acad Sci USA* 1996;93:9218–9222. [PubMed: 8799181]
- Rakic P, Sidman RL. Supravital DNA synthesis in the developing human and mouse brain. *J Neuropath Exp Neurol* 1968;27:246–276. [PubMed: 5646196]
- Rakic P, Stensaas LJ, Sayr EP. Computer-aided three-dimensional reconstruction and quantitative analysis of cells from serial electronmicroscopic montages of fetal monkey brain. *Nature* 1974;250:31–34. [PubMed: 4210169]
- Rakic S, Zecevic N. Emerging complexity of layer I in human cerebral cortex. *Cereb Cortex* 2003;13:1072–1083. [PubMed: 12967924]
- Ramón y Cajal S. Sur la structure de l'écorce cerebrale de quelques mammiferes. *La Cellule* 1881;7:125–176.
- Ramón y Cajal S. Sur les fibres nerveuses de la couche granuleuse du cervelet et sur l'évolution des elements cerebelleux. *Int Monatsch Anat Physiol* 1890;7:12–31.
- Ramón y Cajal, S. *Textura del sistema nervioso del hombre y vertebrados*. 2. Madrid, Moya: 1899.
- Rasin M-R, Gazula V-R, Breunig JJ, Kwan KY, Li H-S, Liu-Chen S, Jan LY, Jan YN, Rakic P, Sestan N. Numb and Numblake are required for Cadherin-mediated cell adhesion and polarity of forebrain neural stem cells. 2007In press
- Retzius G. Studien uber ependym und neuroglia. *Bio Untersuch*, Stockholm, NS 1893;5:9–26.
- Rio C, Rieff HI, Qi PM, Corfas G. Neuregulin and erbB receptors play a critical role in neuronal migration. *Neuron* 1997;19:39–50. [PubMed: 9247262]
- Rivas RJ, Hatten MB. Motility and cytoskeletal organization of migrating cerebellar granule neurons. *J Neurosci* 1995;15:981–989. [PubMed: 7869123]
- Ross ME, Walsh CA. Human brain malformations and their lessons for neuronal migration. *Ann Rev Neurosci* 2001;24:1041–1070. [PubMed: 11520927]

- Rubenstein JLR, Rakic P. Genetic control of cortical development. Special Issue Cereb Cortex 1999;9:521–523.
- Sarkisian MR, Bartley CM, Chi H, Nakamura F, Flavell RA, Rakic P. MEKK4 signaling regulates filamin expression and neuronal migration. *Neuron* 2006;52:789–801. [PubMed: 17145501]
- Schaar BT, McConnell SK. Cytoskeletal coordination during neuronal migration. *Proc Nat Acad Sci USA* 2005;102:13652–13657. [PubMed: 16174753]
- Schachner, M.; Faissner, A.; Fischer, G. Functional and structural aspects of the cell surface in mammalian nervous system development. In: Edelman, GM.; Gall, WE.; Thiery, JP., editors. *The Cell in Contact: Adhesions and Junctions as Morphogenetic Determinants*. John Wiley & Sons; New York: 1985.
- Schmechel DE, Rakic P. A Golgi study of radial glial cells in developing monkey telencephalon: Morphogenesis and transformation into astrocytes. *Anat Embryol* 1979a;156:115–152. [PubMed: 111580]
- Schmechel DE, Rakic P. Arrested proliferation of radial glial cells during midgestation in rhesus monkey. *Nature* 1979b;227:303–305.
- Sestan N, Rakic P, Donoghue MJ. Independent parcellation of the embryonic visual cortex and thalamus revealed by combinatorial *Eph/ephrine* gene expression. *Current Biology* 2001;11:39–43. [PubMed: 11166178]
- Shatz CJ. Impulse activity and the patterning of connections during cns development. *Neuron* 1996;5:745–756. [PubMed: 2148486]
- Sidman, RL.; Rakic, P. Development of the human central nervous system. In: Haymaker, W.; Adams, RD., editors. *Histology and Histopathology of the Nervous System*. C.C. Thomas; 1982. p. 3-145.
- Sidman RL, Rakic P. Neuronal migration with special reference to developing human brain: a review. *Brain Res* 1973;62:1–35. [PubMed: 4203033]
- Soriano E, Dumesnil N, Auladell C, Cohen-Tannoudji M, Sotelo C. Molecular heterogeneity of progenitors and radial migration in the developing cerebral cortex revealed by transgenic expression. *Proc Natl Acad Sci USA* 1995;92:11676–11680. [PubMed: 8524827]
- Swanson, LW. *Brain Architecture: Understanding the Basic Plan*. Oxford University Press; 2003.
- Szentágothai J. The Ferrier Lecture, 1977. The neuron network of the cerebral cortex: a functional interpretation. *Proc Roy Soc (London) B* 1978;201:219–248. [PubMed: 27801]
- Tamamaki N, Nakamura K, Okamoto K, Kaneko T. Radial glia is a progenitor of neocortical neurons in the developing cerebral cortex. *Neurosci Res* 2001;41:51–60. [PubMed: 11535293]
- Tan SS, Breen S. Radial mosaicism and tangential cell dispersion both contribute to mouse neocortical development. *Nature* 1993;362:638–640. [PubMed: 8464515]
- Tan SS, Kalloniatis M, Sturm K, Tam PPL, Reese BE, Faulkner-Jones B. Separate progenitors for radial and tangential cell dispersion during development of the cerebral neocortex. *Neuron* 1998;21:295–304. [PubMed: 9728911]
- Tarui T, Takahashi T, Nowakowski RS, Hayes NL, Bhide PG, Caviness VS. Overexpression of p27 (Kip1), probability of cell cycle exit, and laminar destination of neocortical neurons. *Cereb Cortex* 2005;15:1343–1355. [PubMed: 15647527]
- Temple S. The development of neural stem cells. *Nature* 2001;414:112–117. [PubMed: 11689956]
- Tramontin AD, Garcia-Verdugo J, Lim DA, Alvarez-Buylla A. Postnatal development of radial glia and the ventricular zone (VZ): A continuum of the neural stem cell compartment. *Cereb Cortex* 2003;13:580–587. [PubMed: 12764031]
- Wilson FAW, O'Scalaidhe SP, Goldman-Rakic PS. Dissociation of object and spatial processing domains in primate prefrontal cortex. *Science* 1993;260:1955–1958. [PubMed: 8316836]
- Wu W, Wong K, Chen JH, Jiang ZH, Dupuls S, Wu JY, Rao Y. Directional guidance of neuronal migration in the olfactory system by the protein Slit. *Nature* 1999;400:331–336. [PubMed: 10432110]
- Wynshaw-Boris A, Gambello MJ. LIS1 and dynein motor function in neuronal migration and development. *Genes Dev* 2001;15:639–651. [PubMed: 11274050]
- Xie Z, Samuels BA, Tsai LH. Cyclin-dependent kinase 5 permits efficient cytoskeletal remodeling--a hypothesis on neuronal migration. *Cereb Cortex Suppl* 2006;1:i64–i68.

- Zecevic N. Specific characteristic of radial glia in the human fetal telencephalon. *Glia* 2004;48:27–35. [PubMed: 15326612]
- Zecevic N, Rakic P. Development of layer I neurons in the primate cerebral cortex. *J Neurosci* 2001;21:5607–5619. [PubMed: 11466432]

Figure 1.

Schematic diagram of the composition, cellular events and the relationship in the developing cerebral wall, based on data obtained from species ranging from the mouse to human and non-primates. Initially homogeneous population of neural stem cells (green), which divide symmetrically transform into radial glial cells (RG) which divide asymmetrically and over time produce migrating neurons (MN, red) and dedicated neuronal progenitors with short processes (red mitotic divisions) that populate the ventricular (VZ) and subventricular (SVZ) zones and produce all projection neurons as well as the majority of interneurons in human and a small fraction in rodents. In addition a population of tangentially migrating neurons arrives to the dorsal telencephalon from the ganglionic eminence mostly via SVZ and marginal (MZ) zones to supply the majority of interneurons in rodents and about one third in human. Eventually RG undergo apoptosis (AP) or directly or indirectly generate ependymal cells, fibrillary astrocytes (FA), protoplasmic astrocytes (PA), glial progenitors (GP) or astrocytic stem cells that retain a neurogenic potential (NP).