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## The specification of neuronal identity in the mammalian cerebral cortex

S. K. McConnell

*Department of Biological Sciences, Stanford University, Stanford (California 94305, USA)*

*Summary.* The determination of neuronal fate in the developing cerebral cortex has been studied by tracking normal cell lineages in the cortex, and by testing the commitment of young cortical neurons to their normal fates. These studies together suggest that neuronal progenitors are multipotent during development and have the potential to produce neurons destined for many or all of the cortical layers. However, the laminar identity of an individual neuron appears to be specified through environmental interactions at the time of the cell's terminal mitotic division, prior to its migration into the cortical plate.
*Key words.* Neurogenesis; determination; central nervous system; lineage; migration.

During the development of the nervous system, the generation of neuronal diversity stands as a tremendous challenge, both to the proliferating cells that create this diversity, and to developmental neurobiologists who try to understand it. The last decade has seen several breakthroughs in our ability to explore these processes in the central nervous system of mammals. In several regions of the developing CNS, the processes by which a progenitor

cell produces specific neuronal phenotypes during development have been approached by exploring the lineal relationships of different cell types, and the intrinsic and extrinsic factors that influence their fates. Recent studies of the developing cerebral cortex suggest that the determination of neuronal identity in the cortex occurs early in a neuron's life history, around the time of its final mitotic division, and can be influenced by environmental factors that act on dividing progenitors or their newly-generated progeny.

Two features of the cerebral cortex make it a particularly attractive system for studying neuronal determination in the mammalian CNS. The first is that different neuronal phenotypes are segregated into different layers: within each of the six cortical layers, neurons tend to share similar morphologies, physiological properties, and patterns of long-distance axonal projections. In the visual cortex, for example, upper-layer neurons in layers 2 and 3 send axons to other cortical areas, whereas neurons in the deep layers 5 and 6 project to subcortical targets, such as the thalamus and superior colliculus [19, 41, 61]. Neurons within a layer are also generated at similar times during development [2, 33, 48]; thus, in higher mammals, the time of a cortical neuron's final mitotic division strongly predicts its eventual laminar position. This correlation between a neuron's birthday, its laminar fate, and its adult pattern of axonal projections has led to the hypothesis that the laminar identity of a young cortical neuron may be specified early on in its development, prior to its migration out into the cortical plate [8, 38, 50].

The second feature of the cortex that makes it an approachable model system is that neurons are also organized into columns, which are functional units of neurons that extend in a perpendicular stack through all the cortical layers. In the visual cortex, each column contains all the cells that are required to analyze a small region of the visual world [24]. Thus one can think of the cortex as being composed of a relatively small number of neuronal phenotypes which are segregated into different layers and then stacked one on top of another; each column is then reiterated many times over to constitute a whole cortical area. The laminar and columnar organization of the cortex make studies of its development attractive because cell birthday can be used to predict the adult phenotype of a neuron, and because the precursors that generate the neurons in one column of cortex should be essentially equivalent to precursors in neighboring columns. We and others have exploited these properties and used the cerebral cortex as a model system for exploring the processes that specify the phenotypes of young neurons in the developing CNS of mammals.

### *Approaches to exploring the determination of cell fate*

A cell may employ either (or both) of two basic mechanisms in the determination of its ultimate fate. The first is that a cell's fate may be specified by intrinsic factors (such as chromatin structure or cytoplasmic determinants) that are inherited from its ancestors. Thus the study of cell *lineage* can shed light on whether the progeny of progenitor cells develop in a reproducible and predictable manner, and whether their fates are restricted to a single or a narrow range of phenotypes. Lineage studies can provide important clues about the mechanisms that neurons use to achieve their normal fates. Highly indeterminate lineage patterns usually suggest that extrinsic cues provide the dominant force in shaping cell phenotypes; however, even a completely determinate lineage does not prove that cell-autonomous mechanisms direct development. In the nematode *Caenorhabditis elegans,* in which the pattern of cell lineage is identical from animal to animal, cell-cell interactions play crucial inductive roles in a surprisingly large number of cases (reviewed in Greenwald [21]). Thus in order to test whether intrinsic information or extrinsic instructions are essential to normal development, the *commitment* of a cell to achieving its normal fate can be tested by altering the cell's environment (through transplantation, the ablation of neighboring cells, mutation, or cell culture). These two strategies – mapping the fates of normal cells by tracing cell lineage, and testing the developmental potential of cells by manipulating their environment – have both been exploited in studies of the developing cerebral cortex.

### *Neurogenesis in the cerebral cortex*

Neurons in the cerebral cortex are generated in a region of proliferating cells called the ventricular zone, which lines the lateral ventricle of each telencephalic hemisphere [6]. The ventricular zone is initially a simple pseudostratified columnar epithelium. Precursors replicate their DNA in the outer half of the ventricular zone, but during prophase the nucleus moves inward to the ventricular surface, where the cell rounds up and completes mitosis [57]. This to-and-fro nuclear movement through the cell cycle is called interkinetic nuclear migration. Ventricular zone progenitors maintain an attachment to the ventricular surface at all stages of development; however, later in development, with the formation of the cortical plate and the widening of the intermediate zone, dividing cells lose their attachment to the pial surface of the brain [47, 49, 56]. A second region of mitotically active cells, the subventricular zone, forms late in neurogenesis just above the ventricular zone [6]. This zone lacks both the radial arrangement of precursors and the interkinetic nuclear migration characteristic of the ventricular zone proper. It is not yet known whether different classes of cells, such as neurons and glia, or different classes of neurons arise from these two mitotically active zones, but it has been suggested that the subventricular zone produces many of the upper-layer neurons in the cortex [1]. Several lines of evidence have suggested that the proliferating cells of the developing cortex are not homogeneous, even though they appear so by standard electron micro-

scopic criteria. Studies using antibodies as probes have revealed a molecular heterogeneity of ventricular cells and dynamic changes in their expression of different markers, particularly in the expression of intermediate filament proteins such as the RAT-401 antigen[17], neurofilaments[5,62], and glial fibrillary acidic protein[30]. The observation that subpopulations of mitotically active ventricular cells express immunoreactivity for neuron- or glia-specific protein markers has supported a notion that dates back to the turn of the century, that the ventricular zone contains separate precursors for neurons and glia[22]. A recent cell lineage study using retroviral vectors to directly mark the clonal progeny of a single precursor has supported the notion of separate neuronal and glial lineages, since clones of cortical cells labeled in vivo and in vitro tend to contain exclusively neurons or glia[31]. However, there is also evidence that at a minority of progenitors may give rise to both neurons and glia[46,63,68] (see Walsh and Cepko[69] for review). These results are not necessarily contradictory, since it is quite conceivable that at early points in cortical development a bipotential progenitor exists, but that later restrictions result in largely separate neuronal and glial lineages.

Once generated, newly-postmitotic neurons migrate out radially into the cortical plate, where they differentiate and form axonal projections. Studies in many species using ($^3$H)-thymidine as a marker of cell birthday have revealed that the cortical layers are generated in an inside-first, outside-last manner[2,33,48]. Layer 6 neurons were found among the first to be born and migrate out into the cortical plate; cells of cortical layers 5, 4, 3 and 2 are generated at progressively later times and migrate out past the oldest cells to the top of the cortical plate. The exceptions to the inside-out gradient of cortical neurogenesis are the earliest-generated neurons of the telencephalon. These cells become postmitotic early in embryonic life; they leave the ventricular zone and form a single cellular region underneath the pial surface of the brain. The early-generated cells are then split in two by the arrival of the neurons of the cortical plate[32,34]. The region above the plate is called the marginal zone during development; neurons in this region will constitute layer 1 in the adult animal. Neurons beneath the cortical plate, the subplate neurons[55], are an especially interesting population since these neurons will express neuropeptides[3,10,11,67], receive functional synaptic input[18], and generate both local and long-distance axons[3,10,34,40,66,67], but will then undergo massive cell death in postnatal life[12,32,65].

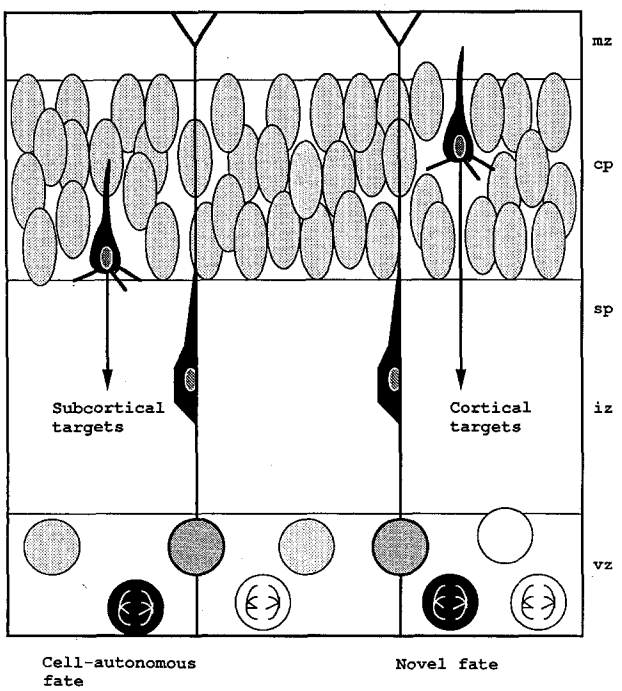
How are the cell types of the cortical layers generated during development? One possibility is that separate progenitors with restricted fates produce neurons in different layers. Contrary to this idea, however, lineage tracing studies using retroviral vectors have revealed that progenitor cells yield neuronal progeny that span several layers, suggesting that the precursors to these cells are multipotent[4,31,46,68]. Furthermore, no reproducible patterns of cell production have emerged from these studies, which strongly implies that lineage itself plays an indeterminate role in the specification of neuronal phenotypes. The obvious alternative mechanism is that neurons or their progenitor cells require instructive input from the environment that coordinates and controls the production of specific cell types during development.

### *Commitment to a laminar fate occurs at the time of a neuron's final division*

The hypothesis that undifferentiated cells employ cues in their environment to specify their identity can be tested directly. The classic test of whether a cell has become committed to its normal fate is to alter its environment by transplantation. The only requirement for performing such an experiment is that one has a way of predicting what the normal fate of a transplanted cell ought to be. In the developing cortex, the birthday of a neurons is a strong indicator of its normal laminar identity, since neurons within a cortical layer are generated at similar times during development. For example, in the developing ferret cortex, the vast majority of neurons generated on embryonic day (E) 31 or 32 are destined to sit in layers 5 and 6, and to form subcortical projections[37]. If environmental directives are normally employed by these neurons to achieve their normal fates, one would hypothesize that these cells might develop in quite a different manner if they were transplanted into an older brain, in which the neurons of layers 2 and 3 were being generated. Thus methods were designed to label presumptive deep-layer neurons with ($^3$H)-thymidine, remove the cells from the ventricular zone at various times after the labeling, then transplant them right back into the ventricular zone of an older host, in which layer 2/3 neurons comprise the current cohort of cells migrating out into the cortical plate[36,37]. The hope of these experiments was that the transplanted neurons would migrate out into the host brain side by side with host neurons, and would thus be subject to the same cues and influences that direct the development of upper-layer cells.

The two possible outcomes of such an experiment are illustrated in the figure. The first possibility is that the fate of the transplanted cells has already been specified prior to transplantation. If this were the case, one might anticipate that these cells would migrate out into the cortical plate and recognize layers 5 and 6 as their appropriate laminar positions, and that they might furthermore form subcortical axonal projections typical of deep-layer neurons. In contrast, if the transplanted cell had not been specified to its normal fate prior to transplantation, one would expect it to migrate out to the upper layers 2/3 along with surrounding host neurons, and to develop axonal projections to cortical targets as do normal upper-layer cells.

In control experiments designed to show that neurons are capable of normal migration and the formation of axonal




Possible outcomes of a heterochonic transplant in which embryonic ferret ventricular cells (black cells) are labeled with ($^3$H)-thymidine on E31 or 32, and then transplanted into the ventricular zone of a neonatal host. If the transplanted neurons are already committed to their laminar fates at the time of transplantation, they would be expected to migrate out along radial glia and into the cortical plate of the host, where they would stop in the deep cortical layers 5 and 6, and form subcortical axonal projections. This is the 'cell-autonomous' fate drawn on the left. If, however, the transplanted cells are multipotent at the time of transplantation, environmental cues in the host brain might induce them to adopt the upper-layer fate typical of host neurons (right). Abbreviations: mz, marginal zone; cp, cortical plate; sp, subplate; iz, intermediate zone; vz, ventricular zone.

projections following transplantation, presumptive layer 2/3 neurons were labeled and removed from the ventricular zone of neonatal ferrets, prior to the cells' migration away from their site of origin. These cells were then dissociated and transplanted into the ventricular zone of same-age host ferrets, in which newly generated neurons were also destined for layer 2/3. The transplanted cells migrated out normally into layer 2/3 in these 'isochronic' transplants, and there they formed axonal projections to association cortical areas, typical of normal layer 2/3 neurons[36,37].

The behavior of neurons in 'heterochronic' transplants, in contrast, differed markedly. In these experiments, ventricular cells were labeled with ($^3$H)-thymidine on E31 or E32, a time when layer 5 and 6 neurons were being generated, and were then dissociated and transplanted into older host brains. Many presumptive layer 5 and 6 neurons then migrated out into the host cortical plate, but here they assumed a bimodal distribution: 43% were found in layer 2/3, the laminar position of host neurons migrating at the same time; the remaining cells were found in the deeper layers, with the majority in layers 5 and 6, the positions characteristic of their birthday[37]. Thus, these cells made the decision to adopt either a position typical of their donor origin or one characteristic of newly-generated host cells; very few were found in layer 4, the laminar position typical of neurons generated between E32 and birth.

These results show that at least some of the transplanted neurons were committed to a deep-layer fate at the time of transplantation. Strikingly, not only did these cells recognize their proper laminar position in the deep layers, but at least some also developed axonal projections that are unique to deep layer neurons[37]. Several transplanted cells in layer 6 were shown by retrograde transport of horseradish peroxidase to extend axons to the lateral geniculate nucleus, a major long-distance target of layer 6 neurons in normal animals[19,37,41]. The question of the extent to which cell birthday specifies connectivity remains open to several interpretations. The transplant results are consistent with the possibility that deep-layer neurons are committed to forming a subcortical projection before they actually reach their final position. Alternatively, it may be that neurons initially commit only to migrate to a particular layer, and there their connections are determined by local interactions within that layer. To distinguish between these possibilities, one would have to deliberately transplant a committed deep-layer neuron into the wrong layer, and ask whether it would alter its projection patterns. It is clear, however, from other studies that at least some aspects of a neuron's projections remain plastic even during axogenesis[25,60]. Stanfield and O'Leary have in a series of experiments shown that layer 5 neurons in the rodent neocortex grow exuberantly to innervate multiple targets early in development[42,58,60]. A given neuron's decision to maintain a specific subset of its initial collaterals is determined later by extrinsic cues, which depend on the area of cortex in which the neuron sits[43,44,54,59].

The heterochronic transplants also imply that a subpopulation of the neurons that were transplanted had not yet been specified to attain a deep-layer fate. These cells, which assumed positions in layer 2/3, appeared quite similar to surrounding host neurons, at least on morphological grounds[37]. However, it will be extremely important to ascertain whether their axonal projections are also typical of normal layer 2/3 neurons – in other words, to ask whether these neurons have truly altered their identity or whether they maintain some aspects of the phenotype typical of their birthday. It could be that transplanted neurons sitting in the 'wrong' layer form axonal projections that are appropriate for their birthday, as do neurons in the *reeler* mouse. In *reeler*, an abnormality of neuronal migration results in an inversion and scrambling of the cortical layers[7–9,45]. Nevertheless, *reeler* neurons appear to develop physiological properties and axonal connections that are normal for their birthday[15,29]. Thus, it will be important to show directly whether the transplanted neurons in the upper layers are displaced deep-layer cells, or normal upper-layer neurons. Should the latter prove correct, this result would imply that the subpopulation of transplanted neu-

rons in layer 2/3 were still multipotent at the time of transplantation, and that they were directed by the older environment to adopt an identity different their normal fate. An alternative explanation would be that these cells represented a committed population of upper-layer neurons that are present during the genesis of layers 5 and 6, but that are normally eliminated from the population of postmitotic neurons by cell death. The problem of distinguishing between the selective survival of committed precursor cells, vs the induction of multipotent cells to adopt a new fate, is a problem intrinsic to transplantation studies employing whole populations of neurons.

The most puzzling question that arises from these results is, why don't all the transplanted neurons behave identically in the host cortex? It could be that the ventricular cells contains a heterogeneous mixture of committed neurons along with others that will not be specified until they reach the cortical plate – in other words, that different ventricular cells employ different mechanisms to achieve their final fates. Alternatively, it could be that the cells all employ the same mechanism, but that the population labeled with ($^3$H)-thymidine represents a mixture of cells, some of which have passed through a critical period for commitment, and others that have yet to reach it. The latter hypothesis seems likely – first, because ventricular cells do not cycle synchronously; thus, the population of ($^3$H)-thymidine-labeled cells represents cells at various stages of the cell cycle[57]. The pattern of ($^3$H)-thymidine labeling in the heterochronic transplants is also suggestive of the latter hypothesis. Because the transplanted cells were removed from their donor environment only four hours after they were labeled with ($^3$H)-thymidine, some cells may have had the opportunity to complete DNA replication and divide in their original environment, while the rest would have had to complete the cell cycle in the new, host environment. Indirect evidence indicates that the site of the cell's final division may determine its fate. Heavily-labeled neurons, which went through only one mitotic division after S-phase, could have divided in either the donor or the host environment; these neurons distributed themselves bimodally between the upper and lower layers. In contrast, all *lightly*-labeled cells (i.e., cells that went through at least one additional round of DNA replication and division after labeling) must have gone through their final cell division in the new, host environment, and these neurons migrated exclusively to the upper cortical layers[39]. This observation suggests that the site of a cell's last division (and environmental factors acting therein) determines its fate.

To test this hypothesis directly, we have begun to vary the time between the labeling of ventricular cells with ($^3$H)-thymidine and their removal for transplantation into the host brain. As a first step, we reasoned that if all cells are allowed to divide in their original embryonic environment, then all should display a commitment to their normal deep layer fate in the transplantation assay. When cortical neurons are removed from donor brains 24 hours after thymidine labeling (to allow for a full cell cycle in that environment) and then transplanted, essentially all the transplanted neurons that migrate into the cortex end up in deep layer positions characteristic of their origin (S. K. McConnell and C. E. Kaznowski, unpublished observations). This result provides direct support for the hypothesis that at some point during or just after the final cell cycle of a cortical neuron, its fate has been determined. We do not know whether a deep-layer fate is the 'default pathway' for ventricular cells, or whether environmental cues in the ventricular zone actively induce neurons to become layer 5 or 6 cells. It will be particularly interesting to identify the point in the cell cycle at which commitment occurs, as this may provide important clues about the factors, both intrinsic and extrinsic, that play a role in determining cell fate.

Another unexamined issue is whether the competence of ventricular cells to produce different neuronal phenotypes changes over time. Cell lineage studies of cortical neurons have produced indirect evidence suggesting that the primary mode of production of cortical neurons is a 'stem cell' mode. It is reasoned that ($^3$H)-thymidine birthdating studies of cortical neurons have shown that neurons in different layers are generated at different times during development[2, 33, 48], and lineage studies have revealed that clonally related neurons can span several layers[4, 31, 46, 68]. Thus a plausible hypothesis is that a single stem cell produces a postmitotic neuron, which migrates out into the cortical plate, and another stem cell, which will progressively produce neurons destined for more superficial layers. The finding that most clones in the cortex are relatively small in size[46] is consistent with this hypothesis; however, the occasional large clones generated over short periods of time are likely to employ at least some symmetric divisions as well (see Walsh and Cepko[69] for a full discussion of this issue).

Regardless of the specific mode of cell production, it appears that many individual progenitors will ultimately produce both deep- and upper-layer neurons. However, it is not yet clear whether progenitor cells retain the same degree of developmental potential over time. One possibility is that the production of deep-layer neurons early in development results in the loss or dilution of the capacity to generate this cell type later, when the main task of progenitors is to generate upper-layer neurons. This hypothesis could be tested by the heterochronic transplantation of neonatal ventricular cells into embryonic host brains. One would expect that a stem cell that retains its multipotency would have the capacity to generate deep-layer neurons in the presence of appropriate environmental cues.

### *A model of how specific neuronal phenotypes are generated during development*

Layered structures in the CNS may share common or similar mechanisms of neuronal determination. The het-

erochronic transplantation experiments, summarized above, have suggested (but not proven) that, prior to commitment to a specific laminar fate, early progenitor cells have the capacity to produce neurons as different as a layer 6 corticogeniculate neuron and an upper-layer neuron, the latter of which normally makes association cortical projections. These findings are consistent with the studies of cell lineage in the cerebral cortex, which show that individual progenitors have the potential to ultimately produce neurons in several layers. Thus, one might hypothesize that environmental (non-autonomous) factors act on ventricular cells at early times during development to direct the production of deep-layer neurons, and that these factors change over time as the ventricular zone's task turns to the generation of neurons destined for more superficial layers.

A similar hypothesis has emerged from work on neuronal determination in the retina, another layered structure in which specific neuronal phenotypes are generated sequentially over development. Lineage studies of retinal neurogenesis have shown quite definitively that retinal precursors are multipotential and may give rise to essentially all cell types found in the retina, including the Müller glia[23, 64, 70]. These results have pointed to the likelihood that environmental directives play a crucial role in the determination of retinal cell fates. Reh and coworkers have obtained evidence suggesting that the instructions to produce different cell types at different times may derive from neurons produced previously in development[51–53]. In one set of experiments performed in vivo, dopaminergic amacrine cells of the retina were selectively ablated with injections of the neurotoxin 6-hydroxy-dopamine[53]. In response to the ablation, retinal progenitor cells at the ciliary margin overproduce dopaminergic amacrine cells, suggesting that the presence of differentiated amacrine cells regulates the further production of this cell type. In a separate set of experiments, designed to test whether the pattern of generation of neurons can be altered in vitro, retinal precursor cells from embryonic retina were co-cultured with older retinal cells, and the phenotypes of the cells that differentiated from labeled progenitors were assayed using a battery of cell-specific antibodies[52]. Reh and Kljavin found that early progenitors cultured alone differentiate primarily into ganglion cells, the first neurons to be generated in the retina, and that none develop a rod phenotype[52]. However, when these cells are co-cultured with older retina, many early progenitors are induced to differentiate into rods, a cell type normally produced late in development[51]. Reh has thus proposed a 'feedback-control' model of neurogenesis in the retina: he hypothesizes that the differentiation of specific cell types during development provides a feedback onto the progenitor population, resulting in the suppression of production of that cell type, and/or the induction of the subsequently-produced phenotype[52]. This in vitro assay for cell specification shows high promise for use as a tool to identify environmental factors that act to determine cell fate.

*Mechanisms of migration and the formation of layers*

Although much progress has been made in understanding the cellular basis of migration in both the cerebral and cerebellar cortices[16, 47, 49], little is known about how neurons terminate their migration and form histotypic structures such as layers. The relationship between a neuron's birthday and its final laminar location in normal cortical development is consistent with lamination being a fairly passive process. Since deep-layer neurons are born first, it would seem that all they need do is migrate out to the end of the radial glia and step off at the top of the cortical plate. Likewise, later-generated neurons destined for more superficial locations could use the same simple mechanism to migrate past older cells into the upper layers. Thus, according to this hypothesis, the radial glia need simply act as a 'conveyor belt' to deliver each cohort of cortical neurons to the top of the cortical plate. However, this notion fails to account for the results of the transplantation experiments that were recounted above. The transplantation experiments suggest that individual cortical neurons are capable of actively recognizing their appropriate laminar position and terminating their migration early in the correct layer, before reaching the top of the cortical plate[37, 39]. Thus the decision to end migration may involve a specific recognition of positional cues in the developing cortex.

One possible mechanism by which neurons could recognize their correct laminar position involves a selective adhesion among neurons of the same layer. Experiments from several laboratories have provided relatively indirect evidence that supports the hypothesis of lamina-specific adhesion among cortical cells. First, when cortical neurons are dissociated and allowed to reaggregate in rotating cell culture, the cells tend to reform histotypically organized structures replete with cellular layers and cell-sparse zones of neuropil[13, 14]. Second, when presumptive deep-layer neurons are labeled with ($^3$H)-thymidine, dissociated, and placed in monolayer cell culture, they tend to group together in clumps, associating preferentially with one another compared to unlabeled cells[28]. Whether dissociated cells in culture regroup due to selective adhesion, and what the molecular nature of these interactions might be, remain open issues at this time.

In parallel to the observation that cortical neurons in vivo are capable of migrating to specific layers, the local axons of cortical neurons are capable of precisely targeting their growth into different cortical layers. Intracellular tracer injections into single neurons of the adult cortex have revealed that neurons in different layers maintain characteristic laminar patterns of local axonal ramification. Layer 2/3 neurons, for example, project within layer 2/3 and in layer 5 below[20, 35], whereas the

local axons of corticogeniculate neurons in layer 6 ascend into layer 4[20,26,35]. Katz and Wiesel[27] have studied the development of the projections of layer 2/3 neurons, and have found that axon outgrowth into layers 2/3 and 5 is remarkable precise from the start: upper-layer neurons appear to make few if any mistakes in recognizing specific layers as their appropriate axonal targets. The mechanism that axons use to accomplish this recognition is a mystery, as is the question of whether the same molecules are employed in recognition of layers by migrating neurons. Presumably there are molecules which, either singly or in combination, specify the identity of cortical layer; likewise, individual neurons must be endowed with a complement of recognition molecules that enable them to 'home' to different layers during migration and axogenesis.

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## Migratory patterns of clonally related cells in the developing central nervous system

G. E. Gray, S. M. Leber[a] and J. R. Sanes

*Department of Anatomy and Neurobiology, and [a] Division of Pediatric Neurology, Washington University School of Medicine, St. Louis (Missouri 63110, USA)*

*Summary.* Neurons and glioblasts that arise in the ventricular zone migrate to form discrete nuclei and laminae as the central nervous system develops. By stably labeling precursor cells in the ventricular zone, pathways taken by different cells within an individual clone can be described. We have used recombinant retroviruses to label precursor cells with a heritable marker, the *E. coli lacZ* gene; clones of lacZ-positive cells are later mapped histochemically. Here we review results from three regions of the chicken central nervous system – the optic tectum, spinal cord, and forebrain – and compare them with previous results from mammalian cortex and other regions of the vertebrate CNS. In particular, we consider the relationship between migratory patterns and functional organization, the existence of multiple cellular sources of migratory guidance, and the issue of whether a cell's choice of migratory pathway influences its ultimate phenotype.
*Key words.* Optic tectum; spinal cord; forebrain; radial glia; retrovirus; lineage.

### *Introduction*

Structurally diverse regions of the central nervous system arise from an apparently homogeneous neuroepithelium. One of the first steps in the histogenesis of each region is the migration of cells from the neuroepithelium to their final sites of differentiation. These migrations result in the delivery of appropriate numbers and types of cells to specific sites at particular times. Differences among regions in organization and function may arise in part from