The neocortex of primates, including humans, is thought to contain significantly higher numbers and more diverse forms of \(\gamma\)-aminobutyric acidergic (GABAergic) interneurons than that of rodents. The mouse cortex displays a number of other features that distinguish it from the cortex of primates and suggest a somewhat less complex pattern of organization. Nevertheless, dramatic findings on the origins and migratory patterns of newly specified GABAergic cortical interneurons in the embryonic mouse have led to a prevailing view that GABAergic cortical interneurons of all species are born in the ganglionic eminence and undergo the same long tangential migration toward the cortex that is seen in the mouse. Recent observations in fetal human and monkey brains, although clearly identifying GABAergic neurons that reach the neocortex via the tangential route, also demonstrate that substantial numbers of GABA neurons are generated in the lateral ventricular neuroepithelium and migrate into the cortex via the same radial route followed by glutamatergic neurons. In the course of evolution of the higher primate cortex, it is likely that new forms of cortical interneuron with origins in the ventricular neuroepithelium have been added to an older population derived from the ganglionic eminence.

**Keywords:** cerebral cortex, development, human, monkey, neuronal migration, radial route, tangential route

**Introduction**

The cerebral cortex of a mouse is not the same as that of a human, although the mouse cortex presents many features that are amenable to experimental interventions that can reveal a great deal about fundamental principles of cortical organization, function, and development. The mouse cortex is of infinitesimally small size in comparison with that of a human or monkey and possesses a relative paucity of well-differentiated cytoarchitectonic and functional fields in comparison with these primates; the cortex of a mouse is less than half the thickness of that of a human, and the ratio of supragranular and granular layers, where arguably all intracortical processing is carried out, to infragranular layers, from which all subcortical and many corticocortical connections arise, is decidedly less. Even if we accept that the number of neurons per arbitrary column through the thickness of the mouse cortex is about that same as that in a column of equivalent width in the primate cortex (Rockel et al. 1980), a view that is now open to question (Rakic 2008), this is but a reflection of the greater elaboration of the processes, dendritic and axonal, of primate cortical neurons, resulting in a vastly more extensive and probably a denser synaptic neuropil.

It is in the cortical neuropil that we find the axons and dendrites of the cortical interneurons, the majority of which are \(\gamma\)-aminobutyric acidergic (GABAergic) and inhibitory; they form about 25–30% of the cortical neuronal population in primates (Hendry et al. 1987) and perhaps 15–20% or less in the mouse (White and Keller 1999). It has never been satisfactorily determined if mice possess the same complement and varieties of cortical interneurons as humans and other primates. Many recent authors have, nevertheless, made the assumption that evolution has indeed bestowed an increased number and diversity of GABA neurons upon the primate cortex (DeFelipe 2002). Santiago Ramón y Cajal (hereafter, referred to as Cajal; 1911), much of whose work on the cortex was carried out on that of the human (DeFelipe and Jones 1988), was not in any doubt that at least one form of interneuron, a form that he called the bipenniform cell but whose name has come down to us via Leon Azoulay’s French translation as the double bouquet cell, was a population that was unique to the human cortex.

Subsequent to the era that had been dominated by Cajal, when most Golgi studies of the cortex were carried out on nonprimates, the double bouquet cell disappeared from view, only to reappear in the 1970s when renewed interest in the Golgi stain’s capacity to identify classes of cortical interneurons in primates, led to its rediscovery (Jones 1975). With time and the realization that the cortical GABA neurons could be further divided into classes characterized by differential coexpression of the calcium-binding proteins, parvalbumin, 28-kDa calbindin, and 29-kDa calretinin (Hendry et al. 1989; DeFelipe et al. 1999), the double bouquet cell became further highlighted as an archetypal calbindin cell (DeFelipe et al. 1990), and subsequent work has revealed it to be one of the most remarkable interneurons of the primate cortex.

All too readily, it has been assumed that an interneuron comparable to the double bouquet cell exists in the mouse cortex, but to my knowledge, no one has ever seen a full blown double bouquet cell in any cortical area of a rodent and only rarely have they been demonstrated in other nonprimates (Balesteros-Yáñez et al. 2005). Indeed, I am convinced that it is only in the primate cortex that the classical cortical interneurons—double bouquet cells, basket cells, neurogliiform cells, chandelier cells etc.—exhibit full expression of the axonal and dendritic characteristics that enabled Cajal and others to give them their names. In saying this, I am by no means suggesting that comparable elements of intrinsic circuitry are lacking in the mouse but if they are there, the parent cells seem to have taken on a less highly elaborated form. If Cajal was right, one particular form of interneuron may be lacking in the mouse. And if that is true, why not others?
With the foregoing words of caution, it is interesting to turn to the developmental history of the cortical GABAergic neurons for, in the experimental studies that have revealed this history, there is much that was unexpected and within these unexpected results there are the seeds of controversy. In the 1960s and 1970s, the use of tritiated thymidine labeling to track the birthdates and migratory patterns of neocortical neurons in rodents and monkeys gave us knowledge of the remarkable inside out sequence of cortical development in which young neurons generated in the neuroepithelium of the lateral ventricular wall migrated along radial guides to take up their positions in a temporally coordinated manner within the cellular layers of the cortex, the deepest layer being formed first with the neurons destined for subsequent layers migrating through each previous layer to take up a more superficial position (Angevine and Sidman 1961; Rakic 1974). In this manner, layer VI is formed first and layer II last.

For years, this scheme remained the fundamental one upon which knowledge of the development of the neocortex was based, and it required modification only when it became accepted that the earliest generated neurons actually formed a kind of protocortex or "preplate" (Marín-Padilla 1988) that was colonized by the waves of neurons invading from the ventricular neuroepithelium and then split by them, as they formed their 5 layers within it, into the marginal layer (layer I) and the subplate. Inherent in this Bauplan was the belief that interneurons, later shown to be a mixture of GABAergic and glutamatergic types, and pyramidal neurons, later shown to be glutamatergic, both had their origins in the ventricular neuroepithelium and migrated along the same radial lines into the cortex. Images of radially aligned GABAergic cells obtained from the cortex of fetal monkeys gave us no reason to believe that they could be arriving by any route other than a radial one (Huntley et al. 1988).

With the dramatic findings in embryonic rodents that neurons expressing GABA markers and generated not in the neuroepithelium of the ventricular wall but in the ganglionic eminence at its base find their way to the nascent cerebral cortex by a route tangential to the cortical plate and more or less following the cortical subplate, even reaching by this route the medial wall of the ventricle and the developing hippocampus, the longstanding gospel of neocortical development seemed to have been overturned (e.g., Anderson et al. 1999, 2002; Lavdas et al. 1999; Nery et al. 2002; Xu et al. 2003, 2004; Wonders and Anderson 2005). Indeed, the new revelations seem to have given us a new gospel in which cortical development is seen as a bipartite process in which the glutamatergic projection neurons, and presumably the glutamatergic spiny stellate interneurons as well, although they are rarely mentioned, are generated in the dorsolateral ventricular neuroepithelium and migrate into the cortical plate radially, whereas the GABAergic interneurons are born in the ganglionic eminence at the base of the hemisphere and make their way to the cortex over the extensive tangential pathway (e.g., DeDiego et al. 1994; Tamamaki et al. 1997; Anderson et al. 2001; Marín and Rubenstein 2001, 2003; Jiménex et al. 2002; Nadarajah and Parnavelas 2002; Polleux et al. 2002). Investigations that have tracked the origins of the glutamatergic projection neurons and the GABAergic interneurons in the mouse are in line with this dogma (Anderson et al. 2002). The molecular mechanisms whereby young GABAergic interneurons are guided over their long trajectory to the cortex are by no means so well established as those by which neurons born in the ventricular and subventricular zones of the lateral ventricular wall find their way into the cortical plate. In the cortical plate, the interplay between neuron and radial glial cell and the important role of calcium signaling are now well known (summarized in Rakic 2002). There are early indications that a series of receptors under the control of the Lhx6/lim homeobox gene are involved in guiding neurons that come to the cortex via the tangential route (Zhao et al. 2008). One can anticipate that this will be an area of intense research activity in the coming years.

The mouse studies have been so compelling that they have essentially led to rejection of the idea that even a subset of GABA neurons could be born from the lateral ventricular wall. Where evidence has been demanded for this radical new point of view, it has usually come from the perspective that cells expressing markers of a GABAergic phenotype observed in the subventricular zone of the lateral ventricular neuroepithelium have migrated there from the ganglionic eminence and are nonproliferative (Anderson et al. 2002).

There is still room for doubt in the rigidly dualist manifesto in which cortical neuronal development is currently presented. In the first place, in fetuses of transgenic mice null for transcription factors deterministic of a GABAergic phenotype in the ganglionic eminence, such as DLX1, DLX2, NKX2.1, and MASH1, the GABAergic interneuronal population of the neocortex, although reduced by as much as 75%, is not completely absent (Anderson et al. 1997, 1999; Casarosa et al. 1999; Horton et al. 1999; Sussel et al. 1999). Although in vitro studies suggest that for the mouse more than one type of interneuron, as marked by coexpressed peptides or calretinin, are born within different regions of the ganglionic eminence, the sources of other differently marked types still remain open (Xu et al. 2003). These observations continue to hold out the possibility that some cortical GABAergic interneurons, perhaps no more than a small subpopulation in the mouse, may indeed have its origins in the neuroepithelium of the lateral ventricular wall. Perhaps more importantly, however, investigations on GABAergic neurons and their precursors in the developing cerebral cortex of fetal human and monkey brains, while agreeing that many GABAergic interneurons have their origins in the ganglionic eminence, continue to point to substantial numbers of such neurons having their origins in the lateral ventricular neuroepithelium. At issue are the questions of whether investigators in the mouse have missed something or whether in the course of evolution of the primate cortex. Nature has recruited additional generative sources to populate the primate cortex with its full complement of interneurons.

In an earlier study of GABA cells in the human fetus, Rakic and Zecevic (2003) had observed radially aligned young neurons expressing transcription factors indicative of a future GABAergic phenotype in the ventricular and subventricular zones. In another, Letinic et al. (2002) while observing a substantial tangential migration of young GABA neurons from the ganglionic eminence into the lateral ventricular wall, akin to that seen in the mouse, also provided convincing evidence for the presence of substantial numbers of GABAergic neuronal progenitors in the ventricular and subventricular zones of the neuroepithelium beneath the developing neocortex. Against these observations, it was argued that the authors could not definitively determine whether the GABAergic cells had been generated there or whether they had arrived as part of the
tangential migration from the ganglionic eminence as precursor cells still capable of proliferation.

Two studies recently published in "Cerebral Cortex," one on monkeys and the other on the human, have provided convincing support for the view that in primates a far greater proportion of the cortical GABAergic interneurons have their origins in the lateral ventricular wall than in the mouse. These neurons migrate radially into the overlying cortical plate, apparently following the same rules as the glutamatergic neuronal population. In the forebrains of fetal cynomolgus (Macaca fascicularis) monkeys, from the 47th to 90th day of gestation, Petanjek et al. (2009) identified GABA neurons by immunocytochemistry or for expression of the mRNAs coding for GAD65 and GAD67, the isoenzymes of glutamic acid decarboxylase, and for the basic helix-loop-helix transcription factor, MASH1, a marker for precursors of GAD-expressing cells. They found that although MASH1-expressing cells were found predominantly in the ganglionic eminence at the early stages of development (E47–E55), where they coexpressed GAD65, a small but significant MASH1-expressing population could also be found in the ventricular and subventricular zones of the dorsal part of the lateral ventricular wall. At these stages, all GAD65-expressing neurons were either found in the ganglionic eminence or migrating from it as non-MASH1-expressing, postmitotic neurons through the intermediate zone and adjacent part of the subventricular zone of the cerebral hemisphere. They were en route to the developing neocortex where they would populate the marginal zone (layer I) and the subplate. Later at E64–E75, MASH1-expressing cells were found not only in the ganglionic eminence but also throughout the ventricular and subventricular zones of the whole lateral ventricular wall, and GAD65-expressing cells could be identified as 2 groups, one migrating from the ganglionic eminence and the other located within the ventricular neuroepithelium, by the coexpression of MASH1 in the neuroepithelial group and by its absence from the migrating group. Those in the ventricular neuroepithelium were characterized as proliferating GABA progenitor cells whose progeny migrated along radial lines into the cortical plate. Both groups—ganglionic eminence-derived and ventricular neuroepithelium-derived—appeared to provide GABA cells to the cortical plate in which layers II–VI are formed.

The clear evidence from the work of Petanjek et al. (2009) that a substantial number, perhaps a majority, of cortical GABAergic interneurons has its origins in the lateral ventricular neuroepithelium and that it enters the cortical plate following the time-honored radial route, by no means reduces the significance of the remarkable tangential radial pattern of migration of cortical GABAergic cells in the mouse. This tangential pattern is also observed in the monkey and was previously seen in the human forebrain (Leticin et al. 2002). Together, the observations in monkeys and humans lend strong support to the belief that the primate cortex, although built in part along lines manifest in the rodent, also depends for its construction upon additional mechanisms that are not present or only incompletely present in rodents. In the evolutionary recruitment of these other mechanisms, there may lie the key to the origins of the putatively greater numbers and greater diversity of inhibitory interneurons in the primate cortex.

Further insights into these ideas come from the second paper published recently in Cerebral Cortex. Fertuzinhos et al. (2009) studied cortical interneurons marked by immunostaining, histochemistry, or in situ hybridization histochemistry for various molecules specifically coexpressed in different populations of cortical GABA neurons in the brains of human fetuses or infants affected by holoprosencephaly with severe striatal hypoplasia. In these cases, the striatal hypoplasia reflects a failure of development or atrophy of the ganglionic eminence, and thus, cortical GABAergic neurons deriving from that site would be expected to be severely compromised. In these brains, cortical interneurons expressing nitric oxide synthase, neuropeptide Y, and somatostatin were either significantly reduced in numbers or absent altogether, whereas those expressing calretinin were present in normal numbers. Projection neurons of various types, as identified by SMI32 immunostaining, were also present in normal numbers. These findings strongly support the notion that the first group of interneuronal types would normally have had their origins within the ganglionic eminence and would have migrated to the neocortex via the tangential route, whereas the second class, along with the glutamatergic projection neurons, would have its origins in the neuroepithelium of the lateral ventricular wall and reach the cortex via the radial route. It is significant, a propos of the findings in the monkey of Petanjek et al. (2009), that the neurons of the first group identified by Fertuzinhos et al. are those normally found in their greatest numbers in the derivatives of the marginal zone and subplate of monkeys (Hendry et al. 1984), whereas the calretinin class is normally found within the principal cellular layers of the cortex, including in some of the layer II double bouquet cells (DeFelipe et al. 1999), thought by Cajal to be absent from the rodent cortex.

Missing from the Fertuzinhos et al. study is finite evidence for the origins of 2 other major classes of cortical GABAergic neuron, indeed the classes that are found in greatest numbers in the primate cortex, namely those coexpressing other calcium-binding proteins, parvalbumin or calbindin. These cells are found in far greater numbers in the primate cortex than those expressing the other calcium-binding protein, calretinin, although this is not often remarked upon by those whose studies have been confined to the mouse cortex. Fertuzinhos et al. showed some reductions in identifiable parvalbumin- and calbindin-containing neurons but, because these neurons do not become fully evident until after the time points that were investigated, the observations remain inconclusive. Parvalbumin is found in basket cells, chandelier cells, and perhaps other GABAergic cortical cell types as well, whereas calbindin is the major calcium-binding protein coexpressed with GABA in double bouquet cells (Hendry et al. 1989). If the parvalbumin and calbindin neurons were also found to have their origins in the lateral ventricular neuroepithelium, then we could say that by far the majority of the cortical inhibitory interneurons in primates have their origins in this site and that, unlike in the mouse, the ganglionic eminence contributes only a minor complement of interneurons to the primate cortex. If these statements are proven to be correct, then we may have even greater reason for assuming that the interneurons of the monkey and human cortex are more diverse than those of the mouse and reason for caution in too readily assuming that the mouse cortex represents a perfect model of that of the human or monkey.
Notes
Conflict of Interest: None declared.

Address correspondence to Dr Edward G. Jones, Center for Neuroscience, University of California Davis, 1544 Newton Court, Davis CA 95618, USA. Email: ejones@ucdavis.edu.

References