Self-organization and pattern formation in primate cortical networks

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Abstract. The primate neocortex is characterized by a highly expanded supragranular layer (SGL). The interareal connectivity of the neurons in the SGL largely determines the cortical hierarchy that constrains information flow through the cortex. Inter-areal connectivity is made by precise numbers of connections, raising the possibility that the physiology of a target area is dictated by the numbers of connections and hierarchical distance in each of the pathways that it receives. The developmental mechanisms ensuring the precision of these interareal networks is in part determined by (i) the numbers of SGL neurons generated by the OSVZ, a primate-specific germinal zone. Neuron generation rate in the OSVZ is determined by regulation of the G1 phase of the cell-cycle. This regulation is area-specific and is linked to thalamic projections to the OSVZ; (ii) Prolonged pre- and postnatal pruning of connections originating from the SGL when the infant monkey visually explores its environment. Remodelling serves to sharpen initial patterns of connections and establishes the adult hierarchy. These results suggest that primate cortical networks underlying high-level function undergo prolonged self-organization via regressive phenomena in the cortical plate (axon elimination) and progressive phenomena (directed growth of cortical axons).


The brain detects statistical regularities in the environment by sensing local and global correlations of neuronal activity. In this way invariant characteristics of the world can be inferred as illustrated by colour constancy and object segmentation. This notion of brain function tempers efforts to understand the brain uniquely by studying all of its components parts independently of the environment. It suggests that it is necessary to consider the statistical features of the environment that the brain is able to detect, and to directly link the structural/functional properties of the brain to its perceptual capacities (Shepard 2001). Likewise, to attempt to understand the development of the brain without taking into account the environmental factors that have moulded its phylogenetic history is to ignore the very factor that has driven its evolution. Corticogenesis cannot
be understood uniquely in terms of molecular pre-specification but must also take into account the environmental factors that modulate organization as cortical development unfolds.

The developing sensory apparatus produces environmental information from which the brain needs to extract behaviourally relevant patterns. By rewiring or re-weighting connections, it tunes itself to or learns about coherent (and presumably relevant) patterns in its input. This unsupervised classification procedure is used to generate self-organized maps. There is evidence that the neuronal mechanisms of ontogenetic self-organization actually persist into adulthood when they mediate adaptive changes in learning and memory.

The species as a whole is subject to environmental patterns that exert pressure through natural selection thus promoting the development of suitable circuits and processing modules that are tuned to the exigencies that led to survival of the current generation (Geisler & Diehl 2002). This proposed process carries the prediction that corticogenesis even at very early stages of development is influenced by extrinsic factors, echoing earlier stages of phylogeny. During the early 1970s there were considerable efforts to show that corticogenesis is significantly shaped by extrinsic factors related to the sensory periphery (Van der Loos 1977). This work was largely supported by the observation that visual experience plays an important role in the elaboration of the functional architecture of the primary visual cortex (LeVay et al 1980, Thompson et al 1983). This understanding of corticogenesis was later referred to as protocortex theory but has been largely superseded by protomap theory which postulates that corticogenesis is driven by intrinsic molecular mechanisms. While in recent years there has been overwhelming evidence in favour of a genetic specification of cortical areas, this evidence does not invalidate the numerous instances of so called afferent specification of the cortex and points to the need for a reappraisal of self-organization (O'Leary 1989, Killackey 1990, Sur & Rubenstein 2005).

The genesis of structures that arise through co-operative mechanisms leading to optimal equilibrium of the participating forces has been referred to as self-organization (Von de Malsburg & Singer 1988). Because self-organizing systems are initially in a relatively undifferentiated state and by definition respond over time to changing signals from the environment one might expect that they would be characterized by prolonged maturational processes. In the cortex the phenomena of self-organization has been traditionally linked to Hebbian plasticity by which competitive modification of synaptic strength underlies experience-dependent self-organization of the functional architecture of the visual cortex, so leading to postnatal plasticity in the orientation and ocular dominance domains (LeVay et al 1980, Thompson et al 1983). This form of developmental self-organization is intrinsic to the primary cortex and is largely confined to the middle layers of the cortex.
Compared to the other species, the primate cortex is characterised by an over-represented supragranular layer (SGL) compartment whose neurons are dedicated to forming local connections as well as the transfer of information between cortical areas (Fig. 1). In monkey cortex, the SGL is generated by a primate-specific germinal zone (Smart et al 2002), and shows an extended maturational period during which there is remodelling of its connectivity including during early life when the animal is visually exploring the environment (Kennedy et al 1989, Barone et al 1995, 1996). A further argument in favour of self-organization in the SGL is a dependency on activity for correct development. This has been shown to be the case for cortico-cortical pathways since immature cortical pathways are highly susceptible to manipulation of the ascending pathways (Dehay et al 1989). In the present review we shall examine the possibility that environmental factors might contribute to shaping the function of these cortical layers and that this might be a characteristic feature of primate cortical development.

Cortical hierarchy and the supragranular layers

Much of our understanding of cortical function comes from the work in the visual cortex where stimulus response function has been most extensively studied. Hubel and Wiesel’s pioneering work showed that the receptive field structure of neurons in the visual cortex is progressively elaborated exhibiting simple, then complex and finally hypercomplex features. This observation leads these authors to postulate that cortex processes afferent information through a feed-
forward (FF) hierarchy of progressive abstract detectors (Hubel & Weisel 1968). Anatomical studies showed that FF pathways originate from SGL and terminate in layer 4, while feedback (FB) pathways originate from infragranular layers and terminate outside of layer 4 (Kennedy & Bullier 1985). In the early 1990s David Van Essen’s group compiled an extensive data base of the FF and FB relations of the cortical areas. Pairwise comparison of these connections revealed the dorsal and ventral streams of the visual cortices as well as a strict hierarchical organization which extended to the prefrontal cortex (Felleman & Van Essen 1991) (Fig. 2a). Malcolm Young’s group performed a statistical analysis of the Van Essen database and confirmed the basic features of the Van Essen hierarchy, including the ventral and dorsal streams. However, while the Young et al organization appeared to be strictly hierarchical they found that it was highly indeterminate, in fact they found over 150,000 equally plausible solutions to the hierarchy (Fig. 2b).

Indeterminacy in the Van Essen model stems from the fact that there is no indication of hierarchical distance between nodes coupled to the fact that there are numerous parallel pathways in addition to the dorsal and ventral streams.
An anatomical solution to hierarchical distance is provided by the fact that long distance FF pathways arise uniquely from the SGL, and that as distance diminishes there is an increasingly important contribution to the projection form the infragranular layers (Fig. 3). Likewise long distance FB projections originate uniquely from infragranular layers and as distance is reduced there are increasing contributions from SGL (Kennedy & Bullier 1985, Barone et al 2000). Estimating the contributions of the SGL and infragranular layers to a given pathway involves quantitative estimations of numbers of neurons and defines the SLN% for a given pathway (SLN% = number of SGL neurons/numbers of SGL + infragranular layer neurons). Accurate SLN values makes it possible to construct a determinate model of the cortical hierarchy (Fig. 3). Further, we investigated whether the number of connections in a given pathway is constant across animals (Falchier et al 2002, Vezoli et al 2004). Previous attempts to estimate whether the strength of a pathway was a characteristic feature had failed (Scannell et al 2000), largely due to the fact that they were working on a database obtained across numerous

**FIG. 3.** Development of feedforward and feedback connections. Feedforward connections exhibit directed growth from the earliest stages of corticogenesis, selective elimination of axonal branches is minimal and serves to refine the already segregated adult pattern of connections. By contrast, feedback projections undergo a protracted remodelling phase during which selective elimination of axonal branches plays a major role in the establishment of the adult pattern of connections.
labs. While our data showed over dispersion we are able to model this over dispersion using a negative binomial distribution which confirmed that the cortical pathways projecting to a given target area have characteristic numbers of connections.

These findings show that the connectivity signature of a cortical area is defined by the individual strengths of 10–20 cortical areas that project to it, the hierarchical distance of each of these areas as reflected by its SLN and the numerical strength of the individual pathways. This would suggest that the physiological function of, or the range of information processing performed by the target area, is constrained by the particular profile of its inputs. Projections originating from SGL terminate in layer 4 and recurrent local circuits amplify the input signals before relaying them to the output neurons in the upper and lower layers (Douglas et al 1995). The output of the cortex is modulated by the infragranular layer projections to layer 1 (Cauiller 1995) and to a lesser extent to layers 5 and 6. In this way FF pathways construct the receptive field properties of the target and area while the FB pathways modulate the features of these receptive fields and the output of the target area. The numerical values and hierarchical features of the projections of a cortical area are determined during an extensive developmental period that stretches into postnatal life during which the contribution of the SGL undergoes extensive modulation (see below).

What is important here is the fact that the connectivity profile and the hierarchical organization depend on the number of neurons in the SGL and the convergence/divergence values of connectivity of these neurons. Both of these parameters are finely adjusted during development, during early corticogenesis by modulation of proliferation and at later stages by remodelling of the projections of SGL neurons.

**Developmental remodelling of supragranular layer axons and formation of cortical pathways**

Corticogenesis in the monkey begins at E40 with the generation of layer 1 and neuron production of the cortex is terminated by E100 (Rakic 1974). Layer 6 neurons are in place by E50 but they do not emit an axon toward distant cortical targets until E90 and when they arrive at their target area they contact preferentially the subplate, with a more modest invasion of the cortical plate. At this stage pathway length is considerably shorter given that the major phase of monkey brain expansion occurs between E120–165 (Batardiere et al 2002, Coogan & Van Essen 1996). SGL projections start to form at around E106 and reach peak levels at E120. The patchy FF projection of area V2 to V4 shows pronounced discontinuity from its onset, with only modest axon elimination at later stages which serves to sharpen the early formed pattern (Barone et al 1995), suggesting that FF pathways from
SGL are characterised by early target selection and directed growth (Barone et al 1996) (Fig. 4).

The directed growth exhibited by the FF pathways from the SGL contrasts with the growth properties of the feedback projections. Retrograde tracer injections in areas V1 and V4 in both primates and non-primates show that the characteristic SLN values in individual extrastriate areas are the consequence of a pre- and postnatal 45–90% reduction in the contribution from the SGL (Barone et al 1995, Kennedy et al 1989, Batardiere et al 1998, 2002).

In the adult cortex, cortico-cortical connections show high convergence and divergence values and therefore connect non visuotopic corresponding regions (Perkel et al 1986, Salin et al 1989, 1992). Quantification of convergence and divergence during cortical development shows that there is little modification in the topography of the pathways (Kennedy et al 1994). So it appears likely that the reduction of the projections from the SGL during development might serve to fine tune the SLN values. As discussed above the distribution of source neurons as measured by SLN values determine the cortical hierarchy. Since SLN values are changing during development it is possible that these changes reflect concomitant changes in the hierarchical organization of the cortex. However, when the SLN values in pre and postnatal cortex are analysed they show that the overall cortical hierarchy does not differ from that found in the adult, rather it would seem that the change in SLN values serves to sharpen the early-formed hierarchy (Batardiere et al 2002).

The remodelling of connections described above has been shown to result from changes in the convergence/divergence values of the SGL via pruning of axon collaterals neurons rather than neuron death (Barone et al 1995, 1998). The number

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**FIG. 4.** Laminar distribution of projection neurons determine hierarchy in macaque adult cortex. (A) Cartoon illustrating the distribution of labelled neurons in feedforward and feedback projections after injection of a retrograde tracer in the target area. Each area exhibits a specific SLN value, which determines its hierarchical distance from the target area. Distant feedforward projections have SLN values of 100 (e.g. Va). More proximal areas have lower SLN values. Distant feedback projections have SLN values of 0 (e.g. Vf). More proximal feedback areas have higher SLN values. SLN values of 40–60% correspond to lateral connections. Because of the cortical curvature, and the non-uniform distribution of labelled neurons in the projection, stable values of SLN require examining the distribution of labelled neurons at closely spaced intervals. Counts of neurons on successive sections show density profiles (B), the smoothness of which indicates appropriate sampling frequency. (C) Each area returns specific SLN values. (D) Hierarchical model of cortical areas connected to the target area according to the relationship between the SLN% and the distance rule (Barone et al 2000). (E) Total number of projections neurons is calculated for each area. This makes it possible to calculate the relative contribution of each area to the total afferent connectivity of the target area.
of connections made by the SGL is ultimately determined by the numbers of neurons in the different sublayers of the SGL and the specification of neuron number has been shown to be area-specific (Lukaszewicz et al 2005, 2006) and to be modulated by thalamic projections to the germinal zone of the primate (Dehay et al 2001).

**Generation of primate supragranular layer neurons**

In the primate visual cortex, more than 75% of cortical neurons destined for the upper layers originate from subventricular zone (SVZ) precursors (Lukaszewicz et al 2005). In the primate there is an important expansion of the SVZ that includes a thick outer component (OSVZ) which is not observed in the rodent (Smart et al 2002) (Fig. 5). The OSVZ exhibits a number of unique features. Contrarily to what is observed in the rodent, where the VZ is the major germinal compartment throughout corticogenesis, the primate VZ declines rapidly during the course of corticogenesis. This decline is associated with an early appearance of the SVZ followed by the OSVZ. This primate-specific organization, first described in the monkey has also been observed in the developing human cortex (Zecevic et al 2005).

Enlargement of the SVZ precursor pool in the primate might correspond to an evolutionary adaptive mechanism ensuring the increased neuronal output necessary to build a more highly developed neocortex with a pronounced cytological complexification of the SGL (Dehay et al 1993, Smart et al 2002, Lukaszewicz et al 2005). The rodent SVZ is only partially self-sustaining and instead has to receive a constant supply of precursors from the VZ (Reznikov et al 1997, Noctor et al 2004, Miyata et al 2004, Haubensak et al 2004, Wu et al 2005). In primates self-renewal (i.e. precursor division is leading to an increase in numbers of precursors) would appear to be much more pronounced in the OSVZ than in rodent SVZ (Smart et al 2002, Lukaszewicz et al 2005).

One major cell type of the cortical germinal compartments is the radial glial cells (RGC) (Rakic 1972). A major research breakthrough occurred when it was shown that the RGC are not only part of a glial scaffolding, but also constitute multipotent cortical progenitors (Malatesta et al 2000, Noctor et al 2001, 2002). There is further heterogeneity of RGC in the embryonic primate, where a fraction cease dividing and function as migration scaffolding for several months before reinitiating proliferation and generating astrocytes (Schmechel & Rakic 1979, Rakic 2003).

There is a major difference in the cellular composition of the primate OSVZ with respect to the rodent SVZ. Whereas in the rodent all RGC nuclei are restricted to the VZ, RGC somata are morphologically identified in the OSVZ of the primate (Lukaszewicz et al 2005, Levitt et al 1981). We have some preliminary evidence that substantial proportions of precursors of the primate OSVZ express Pax6,
FIG. 5. Comparison of human and mouse germinal zones at equivalent developmental stages. These drawings are transects through presumptive Area 17 in (A) monkey and (B) mouse dorsal cortex at comparable developmental stages. The depth of each layer is drawn to a common scale. In the primate, an early appearing outer fibre layer (OFL) forms a major landmark from E55 onwards. The ventricular zone (VZ) declines progressively after E65. The subventricular zone, by contrast, increases progressively in depth and by E72 is divided into an inner subventricular zone (ISVZ) and outer subventricular zone (OSVZ) by an intruding inner fibre layer (IFL). The increase in the OSVZ is particularly important between E65 and E72 and occurs as the VZ declines.

A number of observations link production of infragranular layers to VZ and SVZ to production of SGL. Although SVZ are derived from VZ precursors, there are clear differences in gene expression between the two precursor pools and these differences correlate with distinct neuronal progeny. For instance, Otx1 and Fez1 are expressed in VZ precursors, down regulated in SVZ and subsequently up-regulated in subsets of deep-layer neurons (Frantz et al 1994, Chen et al 2005a,b, Molyneaux et al 2005, Arlotta et al 2005). Furthermore, both Otx1 and Fez1 play a crucial role in specifying the axonal projections of subsets of lower layer neurons. Recent studies in mice show that several transcription factors (Cux2, Tbr2, Satb2 and Nex) (Britanova et al 2005, Zimmer et al 2004, Nieto et al 2004, Wu et al 2005) as well as the non-coding RNA Svet1 (Tarabykin et al 2001) are selectively expressed in both the SVZ and in upper layer neurons. This congruency of expression of genes first in SVZ progenitors and subsequently in supragranular neurons together with time-lapse microscopy observations suggest that the SVZ gives rise to upper layer neurons (Tarabykin et al 2001, Zimmer et al 2004, Noctor et al 2004) and that the SVZ is not uniquely a source of glial cells as was thought until recently (Bayer & Altman 1991).

Consistent with these findings, distinct molecular mechanisms have been identified for the specification of infra- and supragranular neuronal lineages. Studies from mutant mice show that Ngn1 and Ngn2 activity is required for the specification of a subset of infragranular neurons but not for the specification of SGL neurons. In contrast, \(Pax6\) and \(Tlx\), two genes required for the formation of the SVZ (Roy et al 2004, Nieto et al 2004, Zimmer et al 2004), are synergistically involved in the specification of SGL (Schuurmans et al 2004). It therefore be hypothesized that the selective expansion of the SGL compartment in the primate cortex results from modifications of the \(Pax6/Tlx\)-related specification without modification of the Ngn specification mechanisms (Schuurmans et al 2004).

### Control mechanisms of supragranular layer neuron production

The coordinated regulation of two cardinal cell cycle parameters of cortical precursors determines neuronal production via the regulation of the size of the precursor pool: the duration of the cell-cycle and the relative frequency of cell cycle reentry compared with cell-cycle exit. \textit{In vivo} and \textit{ex vivo} analysis of the cell cycle regulation of OSVZ precursors of the primate visual cortex has shed light on the molecular correlates of area-specific differences in proliferation that underlie area-specific differences in the thickness of supragranular layers.

Compared to area 18 OSVZ precursors, area 17 OSVZ precursors are characterized by both a shorter cell cycle duration—due to a reduction of the G1 phase—
and an increased relative frequency of cell cycle re-entry. These areal differences on OSVZ precursor cell cycle regulation are associated with significant differences in the level of expression of molecular regulators of the G1/S transition p27kpl and CyclinE. The *ex vivo* up and down modulation of their level of expression significantly affects cell cycle re-entry and the rate of cell cycle progression and stresses the role of the G1 phase regulation in corticogenesis (Lukaszewicz et al 2005, 2006). Mathematical modelling of the observed differences in both rates of cell cycle re-entry and in G1 phase duration show that the combined variation of these two parameters are sufficient to generate the enlarged SGL that distinguishes area 17 from area18. These results show that variations of G1 phase duration and the coordinated variation in mode of division (Lukaszewicz et al 2002, Gotz & Huttner 2005) contribute directly to regulate neuron number.

*In vitro* work on mouse cortical precursors (Dehay et al 2001) indicates that thalamic afferents control corticogenesis by modulating rates of proliferation. Embryonic thalamic axons release a mitogenic factor that increases the proliferative capacity of mouse cortical precursors during generation of SGL by decreasing the G1 duration and by promoting cell-cycle re-entry (Dehay et al 2001). In the monkey, the LGN axons that selectively project onto the OSVZ of area 17 could be responsible for the temporally and spatially restricted stimulation of proliferation that results in the transient upsurge of the size of SGL precursor pool in area 17 (Dehay et al 1993, Smart et al 2002, Lukaszewicz et al 2005). There is also in vivo suggestion in the primate that embryonic thalamic axons could impact on areal size and specification via an early influence on neuron production during cortical neurogenesis (Dehay et al 1989, 1996a,b, Rakic 1988). Because thalamic axons are precisely targeted on to distinct cortical areas (reviewed in Lopez-Bendito & Molnar 2003) they will be able to differentially affect rates of precursor proliferation and of neuron production across the germinal zones and therefore determine local areal cytoarchitectonic features.

**Concluding remarks**

Computation is a physical process implemented in a physical medium. The primate neocortex is the most sophisticated computational device known to humans and it has been argued that its mode of function and the local microcircuitry that subtend it might be identical across areas (Douglas et al 1989). Unlike conventional computers it does not rely on an external agency for its construction and programming. Instead, the entire circuitry is self-constructed by replication and interaction of the germinal cells and their derived neuronal types. Unlike the majority of tissues that emphasize local three-dimensional organization where cells contact their neighbours, the CNS is characterized by complex connectional topologies over very large spatial scales. The underlying need for this organization is due to
the fact that information processing is finally about selective communication between particular processors. Such functions can be represented as a graph-like topology composed of processing nodes (single or populations of neurons), and their connecting communication edges (axons).

One strategy for self-constructing these topologies is to assemble spatially organized cell types, constituting prototypical modules. This prototypical organization is then refined into more specific connections by both network and environmental informational data, and so provides the specific circuit functionality required for a particular information processing task. Thus, it is probable that the fundamental component of the neuronal organization arises relatively simply out of the genetic determination of cell populations, rules for migration, and cell growth. During the unfolding of this construction process, the neuronal networks in response to environmental factors begin to configure and tune themselves for effective behaviour of the organism.

An understanding of this self-construction and programming of neural computational networks would be a major contribution to the explanation of brain operation. One significant circuit regularity of the neocortex is the lattice-like patchy organization of the lateral connections between pyramidal neurons of the SGL that we refer to as Daisy architecture (Rockland & Lund 1983, Douglas & Martin 2004). With the exception of rodents (Van Hooser et al 2006), Daisy architecture is ubiquitous across areas and species and highly pronounced in primates (Douglas & Martin 2004). This lattice, plus the compexification of the SGL in carnivores and primates reaching their highest development in human, suggests that elaboration of certain common circuit principles could be associated with a huge increase in behavioural performance. These neuroanatomical clues, together with their strong implications for function, call for intensified research of the development of the SGL in species other than rodents and particularly in primates.

Here we have reviewed the evidence that thalamic afferents shape cortical cytoarchitecture via modulation of the cell cycle kinetics of the cortical precursors. There is also evidence that the pattern of activity in ascending pathways plays a key role in the elaboration of the Daisy architecture. In ferret the appropriate surgical manipulation of the fetus leads to a rewiring of the peripheral input to the thalamus such that retinal fibres innervate a deafferented auditory thalamic nuclei (Sur et al 1988). The rewired auditory thalamus acquires novel morphological features and visual responses. At the level of the rewired auditory cortex there are computational response features such as orientation selectivity and directionality. Interestingly, in the re-wired auditory cortex the Daisy architecture is expanded so that it no longer resembles auditory Daisy architecture but instead that Daisy architecture normally found in visual cortex (Gao & Pallas 1999, Sharma et al 2000). This reconfiguration of the cortex is likely the consequence of changes in
the pattern of activity given that following cross-modal reorganization the auditory thalamo-cortical projections are similar to those found in the normal ferret (Pallas et al 1990). These results show that the thalamic fibres initially set up the cortical cytoarchitecture and latter patterned activity, related to the statistical properties of the visual environment, then impact on the elaboration of the computational architecture of the cortex.

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DISCUSSION

Kriegstein: I am sure you have thought about what the substance or factor is that the corticothalamic fibres use to regulate progenitor cell division, and you have a good in vitro assay to sort this out. Has there been any progress on this?

Kennedy: We have used antibodies against FGF2, which strongly reduce the mitogenic effect in rodents. We think Sonic hedgehog might be involved. Beyond this we have no idea.

Hevner: There is some evidence that different types of progenitors may cycle at different rates, particularly neurogenic progenitors versus radial glia. In your measurements did you distinguish between VZ and SVZ, or were you pooling all the progenitors to measure the cell cycle length?

Kennedy: In the in vitro assay, explants of LGN are co-cultured with dissociated precursors from VZ and SVZ of area 17 and of area 18. We also have made measurements both in in vivo and ex vivo on organotypic slices in which we have been able to look at the most outer part of the OSVZ. We are comparing the part of the OSVZ that is up against thalamic fibres with the part that is not. We are finding here they cycle faster with more proliferative divisions The idea that cell cycle duration is homogeneous across precursors is simply very wrong. There is strong evidence from our lab and that of Huttner that proliferative divisions are fast and the differentiation divisions are much slower (Lukaszewicz et al 2002, Calegari et al 2005).

Rakic: I agree that cooperation between specific thalamic nuclei and cortical areas is essential. But in terms of specificity, it is important to recognize that the early set of geniculo-cortical axons in monkey project only to the part of the ventricular zone that will produce neurons for the area 17. This means that proliferative cells subjacent to this area have some kind of receptor or ligand that attract geniculate axons to the segment destined for area 17 but not area 18. In cat, for example, neurons of the lateral geniculate nucleus project to both areas 17 and 18. This is another proof of the early specification of these cells in the SVZ.

Kennedy: Absolutely. I showed unpublished data that dissociated cultures from area 18 that do not respond at all to the mitogenic factors released by the LGN axons.

Mallomaci: Are progenitors from other cortical areas responsive to influences coming from the LGN. In other words, if you put cells coming from, for example, the LGN and other non-visual areas in co-culture, do these progenitors respond in a similar way?

Kennedy: No.

Mallomaci: But, is there a gradient of responsivity along the rostrocaudal and mediolateral axes, or is the responsivity tightly restricted to areas?
Kennedy: That’s an important point. *In vivo* there is a gradient of high rates of mitogenesis in area 17 to lower values in area 18. We were initially disappointed, we had been hoping to find a boundary in the ventricular zone where there would be high rates of proliferation on one side and low rates on the other. It is not like that; there is a gradient. How we go from that gradient in the ventricular zone to the stepwise function in the cortex is a mystery. I think that the different rates of migration (fast in 17 slow in 18) coupled with differential tangential expansion must be involved. I agree with Arnold Kriegstein’s idea that the thickness of the subventricular zone and tangential expansion are interconnected. Tangential expansion differential rates of migration and proliferation all lead to determining the radial thickness and areal dimensions of the cortical areas. In passing, I’d like to add that the primate 17/18 border is a freak: it is a useful model, but there are no other borders like it.

Mallomaci: So, if you artificially combine the progenitors coming from area 18 with LGN neurons do you get a boost of proliferation?

Kennedy: We have done that, and our unpublished results are quite clear, the mitogenic effect of the LGN is specific to area 17 precursors.

Macklis: About four years ago Karl Deisseroth had a paper on mixed progenitors from forebrain that they claimed were glutamate-activated in the cell cycle increasing proliferation (Deisseroth et al 2004). This is contrary to what you were saying about the specificity. They were arguing that a proliferative zone that is activated might want to expand to receive more activity. It wasn’t clear from the data whether this was a survival effect on the progenitors or proliferation.

Kriegstein: The Deisseroth paper concerned adult hippocampus. They were looking at activity-dependent regulation of cell cycle events. Some time ago we looked at cortical neuroepithelial cells and saw that they had GABA and glutamate receptors (LoTurco et al 1995). We then looked at whether these were influencing cell cycle events. Activating either GABA or glutamate receptors had a net inhibitory effect on neurogenesis. Conversely, with receptor blockade there was an increase in the number of cells entering the cell cycle.

Macklis: I remember those data, and also thinking at the time when you published yours that the activation might be saying ‘let me stabilize and mature’, whereas here it says ‘let me divide more’. I wonder whether there could be differential effects.

Kriegstein: Pasko Rakic did a study in which he looked at the SVZ versus the VZ. He concluded that the GABA and glutamate effects were opposite in the two zones (Haydar et al 2000). The SVZ effect was the inhibitory one.

Rakic: That is correct. The study indicated that these cells are also having some kind of receptors that are differentially expressed in the progenitors in the VZ and SVZ, and made them to respond to GABA in a different way. Another interesting question that no one brings up is the length of cell cycle which is, at least four
times longer in primates than in rodents (Kornack & Rakic 1998), and why does such a paradoxical thing happen? Since there are much more cells in the primate cortex, you would predict that cells should divide faster, not slower. One possible reason for this could be that it gives them time to interact. In terms of lateral expansion and vertical expansion of the cortex during evolution, I think they are separate events that are regulated by different genes. For example, dolphin has a thin cortex with more radial units and fewer cells per unit. Ungulates have a thicker cortex with more cells per unit. They are separate cellular events and are probably induced and affected by different genes. The SVZ probably plays an important role in these events, but it isn’t sufficient to explain lateral tangential expansion. It is not only the superficial layers that need to expand, but also the deep layers (Rakic 1995).

Molnár: I think we can go even further back in looking at the role of thalamocortical interactions in cortical circuit formation. If we consider the whole idea of self-organization, then there are examples when just a modality-specific stimulus fed to the developing cortical circuit is enough to instruct aspects of development without even changing the thalamic relationship. This is the case for the patchy distribution of intracortical supragranular connections in the auditory cortex of the ferret. Normally, we don’t see these patchy supragranular intracortical connections in auditory cortex, yet when the retina is wired into the MGN as Mriganka Sur’s and Sarah Pallas’ group did then the auditory cortex starts to show the signs of patchy intracortical connectivity (Sur & Leamey 2001, Pallas 2001). With the very same thalamocortical relationship you have only changed the modality of the signals which are delivered to the forming circuitry and you will begin to see the patchy distribution of intracortical connectivity. It is the quality of the stimulus that could play on these wires. Even changing the thalamocortical relationship wouldn’t be so important for these kind of self-organizations.

Kennedy: I think we need to distinguish two things here. The first is the mitogenic effect of the thalamus on the cortex that we have described in the rodent and today in my presentation. The second is the intriguing effect that Sur showed in re-wired cortex. Sur and his colleagues showed that in ferret when retinal fibres are induced to innervate the auditory thalamus, the patchy connectivity of the supragranular layers in the auditory cortex takes on the characteristics of the patchy connectivity normally found in the visual cortex and not that in the auditory cortex because the auditory thalamic fibres have a configuration as in the normal animal these authors concluded that it is the firing pattern which is important.

Molnár: Sur and colleagues injected tracer into the ferret auditory cortex, which now processed visual input after rewiring after their optical recording experiments. The connections showed patchiness. In normal controls in the primary auditory cortex processing normal auditory signals there are no patches.

Kennedy: Yes there are.
O’Leary: You mentioned that thalamocortical afferents in the monkey grow into the cortical primordium before neurogenesis begins. What is the relationship between the waiting period in the SVZ compartment relative to the generation of layer 4 and thalamocortical input? Do thalamocortical axons wait until layer 4 neurons are generated?

Kennedy: This is what we wanted to look at, but it wasn’t that easy. It looks like nothing much is happening to the thalamocortical afferents, other than that they innervate the top part of the germinal zones. There isn’t a layer 4 until about E75–E80.

O’Leary: In principal, then, the distal ends of the thalamocortical afferents are in a position where they can be releasing a mitogenic factor.

Kennedy: Yes. Don’t forget that the subplate is huge, and some of the thalamic afferents go into the subplate.

References