Cellular and Molecular Mechanisms of Cortical Specification

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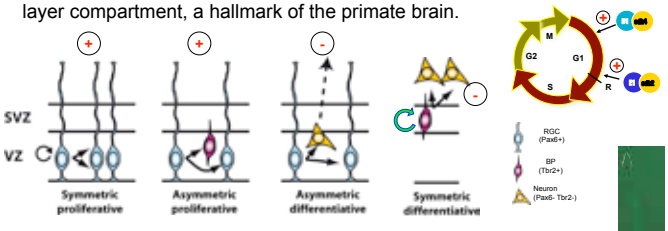
Introduction

The cerebral cortex is divided into anatomically distinct and functionally specialised areas. Understanding the mechanisms that control areal specification is a major challenge in developmental neurobiology. Our earlier work had pointed to an early specification of cortical areas identity, at the level of the germinal zones. A major aspect of our work supported by the ANR is the demonstration that cell-cycle control and more specifically G1 phase regulation regulate cytoarchitectonic features in cortical areas. Using in vivo and ex vivo approaches both in mouse and monkey, as well as theoretical modelling we have been able to uncover cell-cycle related mechanisms of corticogenesis that shed light on evolutionary mechanisms.

Main results

I) Role of the G1 phase duration in regulating the mode of division and identity of cortical precursors

We have experimentally modified the molecular control of TG1 in mouse via cortical precursors via in utero electroporation-mediated forced expression of CyclinE and cyclinD1 at E15. The resulting TG1 shortening influences differently Pax6 expressing apical precursors and basally dividing precursors (BPs): it promotes generation of BPs through Pax6 expressing precursors proliferative divisions and it increases cell-cycle re-entry of BPs at the expense of neurogenic divisions, leading to an increase of the BPs pool from the SVZ. We have also uncovered the role of a third germinal compartment, the dispersed mitotic compartment (DMC) made of sparsely crowded cycling precursors in the lower Intermediate Zone which significantly contributes to the generation of supragranular layer neurons. Together with the increase in the SVZ BP pool, the increase in size of the DMC, results in enlarged supragranular layers in the mature cortex. Modelling reveals that the long term consequences of TG1 variation on neuron production and laminar fate are mediated via the changes of mode of division (MOD). These experimental and modeling data converge to show that TG1 influences the MOD, which in turn is the major player in determining the size of the precursor pool and neuron production. This induced change in MOD appears to recapitulate the increase in the BP pool, a major characteristic of primate corticogenesis. This «primatisation» of the mouse cortex germinal zones translates in an enlarged supragranular layer compartment, a hallmark of the primate brain.



II) Characterisation of the primate-specific germinal zone (OSVZ) precursors phenotype and cell-cycle kinetics:

We have previously identified the Outer Sub Ventricular Zone (OSVZ), responsible for the generation of supragranular layer neurons in the primate. We have uncovered major differences between the rodent SVZ and the primate OSVZ, regarding the cellular composition and proliferative abilities.

Symmetric proliferative divisions (where the two daughters re-enter the cell-cycle) are responsible for the amplification of the OSVZ. This contrasts with the rodent where the SVZ progenitors undergo mostly differentiative divisions producing postmitotic neurons. Whereas in the rodent Pax 6 expression specifically characterises the VZ apical progenitors, we have shown that 90% of basal OSVZ mitotic precursors express Pax6 in agreement with the observed "radial-like morphology of OSVZ precursors (Lukaszewicz et al., 2005).

III) Role of embryonic thalamic axons in the early specification of cortical areas via modulation of the G1 phase duration.

One major extrinsic influence that is likely to be exerted on the developing cortex is that of the thalamic input. Our previous in vitro work in the mouse showed that embryonic thalamic axons modulate cell-cycle kinetics of cortical precursors. The ascending geniculo-cortical projections to area 17 of the monkey, unlike in non-primates, target the germinal zones, making it an ideal model to explore the influence of embryonic thalamic axons on rates of proliferation. Measurements of cell-cycle kinetics of E80 OSVZ cortical precursors in embryonic monkey visual cortex organotypic slice shows that the precursors located in the upper domain of the OSVZ, i.e. in close proximity with the bulk of ingrowing embryonic thalamocortical axons, exhibit faster cell-cycle rates-due to a reduced G1 phase duration- than their deeper counterparts. Co-culture of embryonic LGN (E80) explants over dissociated area 17 and area 18 OSVZ precursors, show a significant and selective increase in proliferative divisions of area 17 precursors (but not area 18 precursors) located at close proximity (<60 microns) of the growing axons, indicating that the mitogenic effect of embryonic geniculate axons is area-specific.

In vivo analysis in early anophtalmic monkeys where the thalamocortical projections to area 17 are depleted revealed decreased proliferation rates in the respecified cortex associated with a drastic modification of areal parcellation due to a reduction of the tangential extent of area 17.

E82 LGN Explant 6DIV

Conclusions

- The fine-tuning of a very basic biological mechanism, namely the G1 phase of the cell-cycle is the primary parameter that orchestrates, via the regulation of mode of division, the exquisitely ordered neuron production during corticogenesis.
- The increased proliferative abilities of basal progenitors in the primate can be considered to be one of the major evolutionary changes in corticogenesis
- The embryonic thalamic axons exert an unexpected major role in early areal specification via a modulation of G1 phase duration and mode of division.

Publications

Bonnefont J, Laforge T, Plastre O, Beck B, Sorce S, Dehay C, Krause KH. **Cell Death Differ**, 2010, 18(2):293-303.
Pilaz LJ, Patti D, Marcy M, Ollier E, Pfister S, Douglas RJ, Betizeau M, Gautier E, Cortay V, Doerflinger N, Kennedy H, Dehay C. **Proc Natl Acad Sci USA**, 2009, 106 :21924-9
Pinto L, Drechsel D, Schmid MT, Ninkovic J, Irmler M, Brill MS, Restani L, Gianfranceschi L, Cerri C, Weber SN, Tarabykin V, Baer K, Guillemot F, Beckers J, Zecevic N, Dehay C, Caleo M, Schorle H, Götz M. **Nat Neurosci**, 2009, 12(10):1229-37.
Dehay C, Kennedy H. **Neuron**, 2009, 62:455-457.
Fish JL, Dehay C, Kennedy H, Huttner WB. **J Cell Sci,** 2008 121(Pt 17):2783-93.
Dehay C, Kennedy H. **Nat Rev Neurosci**, 2007 8:438-50.

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