

LIFE IS A JOURNEY: A GENETIC LOOK AT NEOCORTICAL DEVELOPMENT

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Although the basic principles of neocortical development have been known for quite some time, it is only recently that our understanding of the molecular mechanisms that are involved has improved. Such understanding has been facilitated by genetic approaches that have identified key proteins involved in neocortical development, which have been placed into signalling pathways by molecular and cell-biological studies. The challenge of current research is to understand the manner in which these various signalling pathways are interconnected to gain a more comprehensive picture of the molecular intricacies that govern neocortical development.

NEOCORTEX

The most recently evolved part of the cerebral cortex. It is believed to orchestrate high-level motor, sensory and cognitive functions.

RADIAL DIRECTION

The movement of neurons from the inner to the outer brain surface. The inner brain surface lines the brain ventricles, and the outer brain surface lines the pia.

The mammalian NEOCORTEX contributes to the increasing functional complexity of the mammalian brain, partly because of its striking organization into distinct neuronal layers (FIG. 1). The development of the neocortex has been well studied, because it informs our understanding of cell-migration mechanisms and brain development in a more general sense, and because disrupted neocortical development results in several human diseases (TABLE 1). Although the basic principles of neocortical development have been known for some time, only recently have significant advances been made in our understanding of the molecular mechanisms that are involved. In large part, these molecular advances have been, and are, based on genetic studies. In this review, we discuss the findings of some key genetic studies that have led to advances in our understanding of the molecular signalling pathways that control neocortical development.

Layer formation in the neocortex

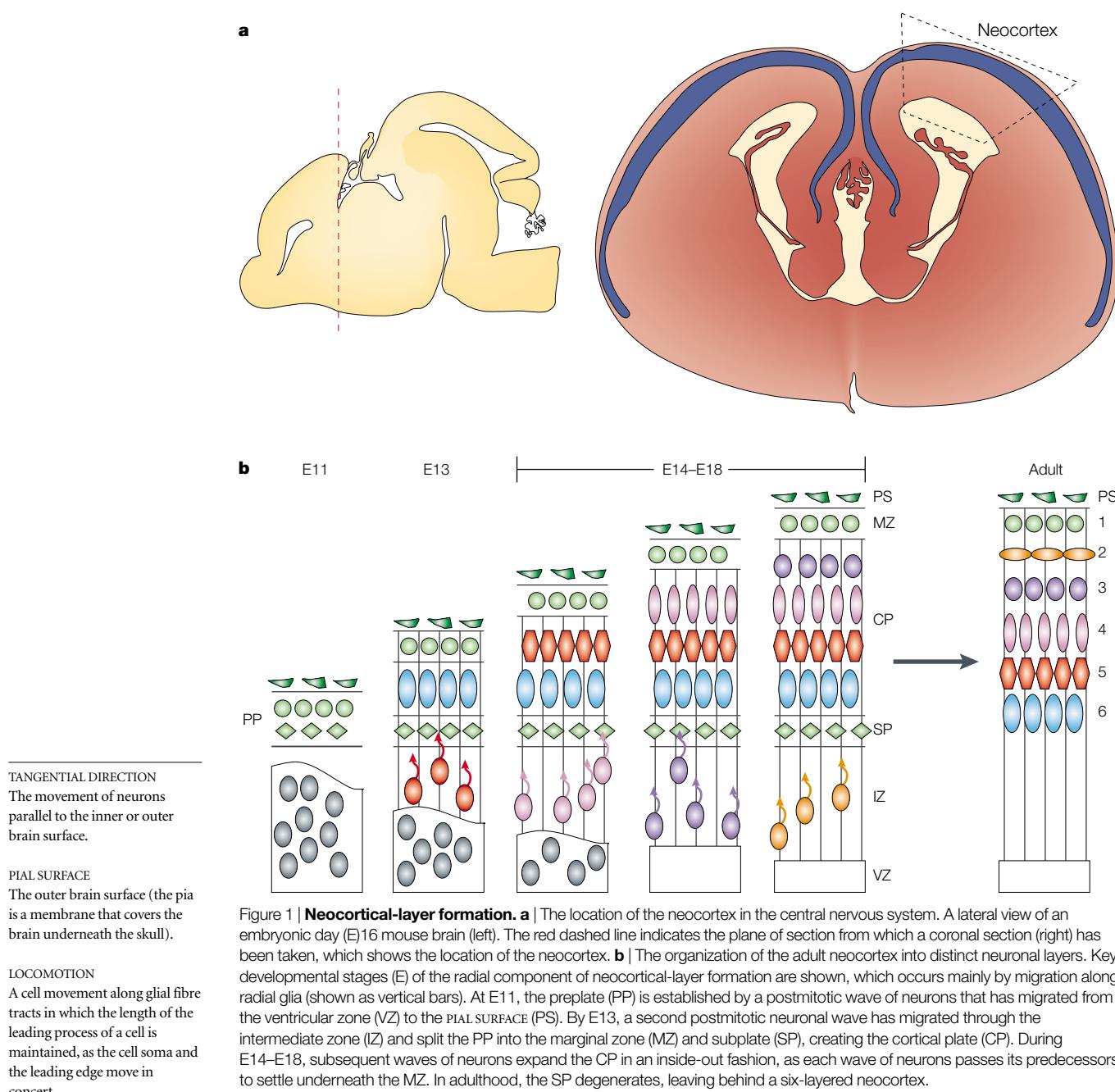
The neocortex consists of six layers of neurons that have distinct morphological and functional identities. The development of these neuronal layers involves the migration of neurons in RADIAL and TANGENTIAL DIRECTIONS to their final position. In this review, we discuss only radial migration because it accounts for the positioning of most neurons in the neocortex^{1–3}. Furthermore, most of the mutants with cortical layering defects have extensively disrupted

radial migration, whereas disrupted tangential migration is believed to be less common, or absent, in these mutants⁴.

Neuronal-layer formation by radial migration occurs in the mouse between days 11 and 18 of embryonic development (E11 and E18)^{1,5,6} (FIG. 1b). At E11, the neocortex consists of many proliferative precursor cells arranged in a layer known as the ventricular zone. A wave of postmitotic neurons then exits the ventricular zone, and these neurons move in a radial direction towards the pial surface of the brain, establishing a neuronal layer known as the preplate. At ~E13, a second wave of postmitotic neurons migrates radially away from the ventricular zone and splits the preplate into a superficial marginal zone and a deeper subplate. In this way, the second neuronal wave creates a layer between the marginal zone and the subplate, known as the cortical plate. Between E14 and E18, waves of postmitotic neurons continue to exit the ventricular zone in distinct phases, and migrate radially to traverse the subplate and form the sequential layers of the cortical plate. Notably, as revealed by studies that date cell birth, the layering of the cortical plate occurs ‘inside-out’, in which early-born neurons make up the deep layers of the cortical plate, whereas later-born neurons occupy the more superficial layers⁷. After the cortical plate has been fully established, the subplate degenerates and leaves behind a six-layered neocortex that persists throughout adulthood.

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Radial migration. Most radial migration occurs by LOCOMOTION (BOX 1), in which neurons migrate along the processes of RADIAL GLIA CELLS known as radial glial fibre tracts^{8–12}. These radial glial fibres provide an extensive radial lattice that guides radially migrating neurons (BOX 1). Indeed, neurons were shown to be attached to radial glial fibres in the developing nervous system and to slide along these fibres in cell-culture assays^{8,13,14}.

Glia-independent modes of radial migration also occur¹⁵, particularly by NUCLEAR TRANSLOCATION^{16,17} (BOX 1). Nuclear translocation has been observed in several structures of the developing central nervous system, including the cerebral cortex^{18–22}. Morphological studies have indicated that neurons that are translocating in

the cerebral cortex extend their leading process to the pial surface to reach their final destination^{21,22}, independent of radial glia²¹. A recent time-lapse study²³ of radially migrating neurons during early and late stages of neocortical development revealed that the modes of migration — nuclear translocation and locomotion — have different dynamics and occur at different times during neocortical development (BOX 1). These distinctive features have given rise to a refined idea of how radial migration is orchestrated to establish the cortical plate (BOX 1).

Having outlined the basic principles of how neuronal layers form in the neocortex, we now consider the underlying molecular mechanisms that control this

AGYRIA
The absence of gyri (folds in the surface of the brain).

PACHYGYRIA
A reduced number of broadened gyri.

HETEROPIA
A group of abnormally placed cells. In the context of neuronal migration defects, it refers to cells that are out of place in the cortex in either the grey or the white matter.

SUBCORTICAL BAND HETEROPIA
(SBH). A group of heterotopic neurons that form in the white matter.

Table 1 | Defects of neocortical-layer formation in humans

Gene affected	Disease	Gene locus	Mode of inheritance	References
<i>LIS1</i>	MDS and ILS	Chromosome 17p13.3	Sporadic	36–40
<i>DCX</i>	ILS and SBH	Chromosome X	X linked	40–44
<i>RELN</i>	LCH	Chromosome 7q22	Recessive	128
<i>FLNA</i>	PH	Chromosome Xq28	Dominant	120

DCX, doublecortin; *FLNA*, filamin A, α ; ILS, isolated lissencephaly sequence; LCH, lissencephaly with cerebellar hypoplasia; *LIS1*, lissencephaly 1; MDS, Miller–Dieker syndrome; PH, periventricular heterotopia; *RELN*, reelin; SBH, subcortical band heterotopia.

process. Genetics has had an essential role in discovering these mechanisms. Mutations that underlie severe cortical dysplasias in humans have provided clues as to the genes that are important for neuronal migration. This has provided an entry point into the pathways that control cortical development, which have been further investigated in mouse models. Nature and serendipitous gene targeting have also provided mouse mutants that harbour distinct defects in cortical-layer formation. By

identifying the genes and proteins that underlie the various layering defects, genetic approaches have been instrumental in establishing a framework around which cellular, molecular and biochemical studies of cortical development have flourished. In the following section, we briefly describe some key mutants of neocortical-layer formation, and outline the genetic and biochemical pathways that are disrupted in these mutants, and the role that these pathways have in cortical development (for more detail on the pathways, see REFS 24–30). We also propose a model of cortical development to tie together the known developmental mechanisms that are involved with our understanding of the pathways that are important for cortical-layer formation. Finally, we outline future studies that will begin to test the implications of this model.

Box 1 | Radial migration in the developing neocortex

Neurons in the neocortex undertake two main types of radial migration: nuclear translocation and locomotion.

• Nuclear translocation

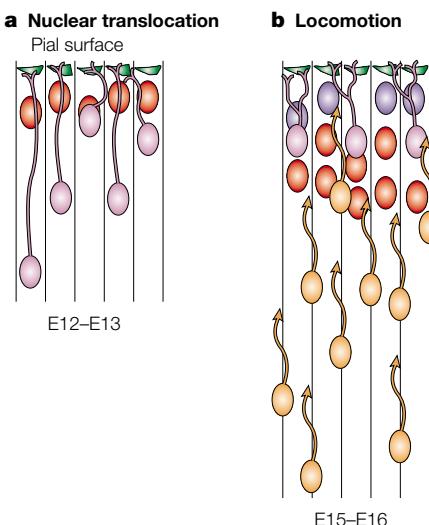
This type of radial migration occurs as a continuous, smooth movement, in which the cell nucleus moves towards the leading edge of the neuron, which results in the shortening of the leading process over time (pink cells in panel a). Nuclear translocation has also been called somal translocation²⁶, as the entire cell soma moves towards the leading edge, not just the cell nucleus. The leading edge is attached to the pial surface and is structured as a fork, with processes that branch off a common branch point. As nuclear translocation is independent of the glia, pial attachment suffices for a translocating neuron to migrate radially, and, if required, past its predecessors.

At embryonic day E12–E13 in mouse development (a), neurons use nuclear translocation to create and to split the preplate. Nuclear translocation occurs across the entire width of the neocortex, as the cerebral wall is thin and neurons are able to attach their leading processes to the pial surface while their cell soma is still in the ventricular zone.

• Locomotion

Locomoting cells (orange cells in panel b) maintain the length of their leading process, as the cell soma and the leading edge move together. Locomotion is interspersed by pauses, and is dependent on the glia. Radial glia might, therefore, be required in the absence of pial attachment.

At E15–E16, the preplate has already split and the cortical plate is expanding rapidly. Locomotion predominates during this time (orange cells), but nuclear translocation continues to occur in the upper half of the neocortex (pink cells in b). As the cerebral wall has widened greatly, neurons cannot translocate across the entire width of the neocortex. Instead, neurons first use radial glia to move by locomotion (shown as movement along vertical bars), and then switch to nuclear translocation when they have traversed the neocortex far enough to attach their leading processes to the pial surface.



Functional studies of *LIS1* and *DCX*

There are several types of human cortical dysplasia that are associated with defects in neuronal migration. Here, we focus on one type of migration defect, **lissencephaly** (meaning ‘smooth brain’), for two reasons. First, lissencephaly has been well studied and described in the medical literature. Second, two genes have been cloned that, when mutated, result in lissencephaly: lissencephaly 1 (*LIS1*; also known as Miller–Dieker syndrome chromosome region, *MDCR*) and doublecortin (*DCX*).

Lissencephaly. The key clinical features of lissencephaly are **AGYRIA** and **PACHYGYRIA**, abnormal cortical layering, enlarged ventricles and neuronal **HETEROPIAS**^{31–33}. Clinically, type I, or classical, lissencephaly presents itself in two main ways: **isolated lissencephaly sequence** (ILS) and **Miller–Dieker syndrome** (MDS). In ILS, patients show a variable degree of severe lissencephaly with no other major malformations. MDS patients, conversely, have lissencephaly that is more severe than in ILS, and also have characteristic craniofacial anomalies³⁴. Regardless of whether patients have ILS or MDS, they are severely retarded, suffer from epilepsy and usually die early in childhood³⁵.

Virtually 100% of MDS, and 40% of ILS, cases are caused by heterozygous mutations in the *LIS1* gene^{36–40}. A second gene, *DCX*, is responsible for X-linked ILS^{41–42}. Only males with *DCX* mutations develop X-linked ILS; females with *DCX* mutations are mosaics due to X inactivation and, instead, manifest **SUBCORTICAL BAND HETEROPIA**^{41–44}. However, as mutations and deletions of *LIS1* or *DCX* account for only 76% of cases of ILS^{40–41}, additional ILS-causing genes are still to be identified.

Mouse models of lissencephaly. To address the *in vivo* effects of dosage reduction and complete deficiency of *Lis1* in greater detail, a hypomorphic conditional knockout and two null knockout *Lis1* alleles have been produced by gene targeting in mice⁴⁵. Mice with a graded reduction in *Lis1* dosage show a *Lis1* dose-dependent disorganization of the cortical layers, the hippocampus and the olfactory bulb^{45–47} (M. J. Gambello *et al.*, unpublished data). *Lis1*-null mice show peri-implantation lethality, which indicates that *Lis1* is an essential gene⁴⁵. Mice that are heterozygous for a shorter form of *Lis1* (*sLis1*), which lacks the first exon, are viable but have a disrupted cortex with abnormalities in radial glia and thalamocortical innervation⁴⁸. *sLis1*-homozygous mice die around implantation and are indistinguishable from

Lis1-null mice, which indicates that the *sLis1* protein is functionally inactive⁴⁸.

The study of cortical-layering effects is complicated by the dosage dependence of *Lis1* function and by embryonic lethality in *Lis1*-null and *sLis1*-homozygous mice. The primary defect in *Lis1* or *sLis1* heterozygotes seems to be a cell-autonomous slowing of neuronal migration: a normal inside-out pattern of cortical layers forms, but more slowly and with less-distinct layers^{45,48} (FIG. 2a). However, as *Lis1* dosage is further reduced in compound heterozygotes that carry a null and a hypomorphic *Lis1* allele, cortical lamination is completely disrupted⁴⁵ (M. J. Gambello *et al.*, unpublished data), similar to lissencephaly in humans³⁶. The mutants also have defects in splitting of the preplate (M. J. Gambello

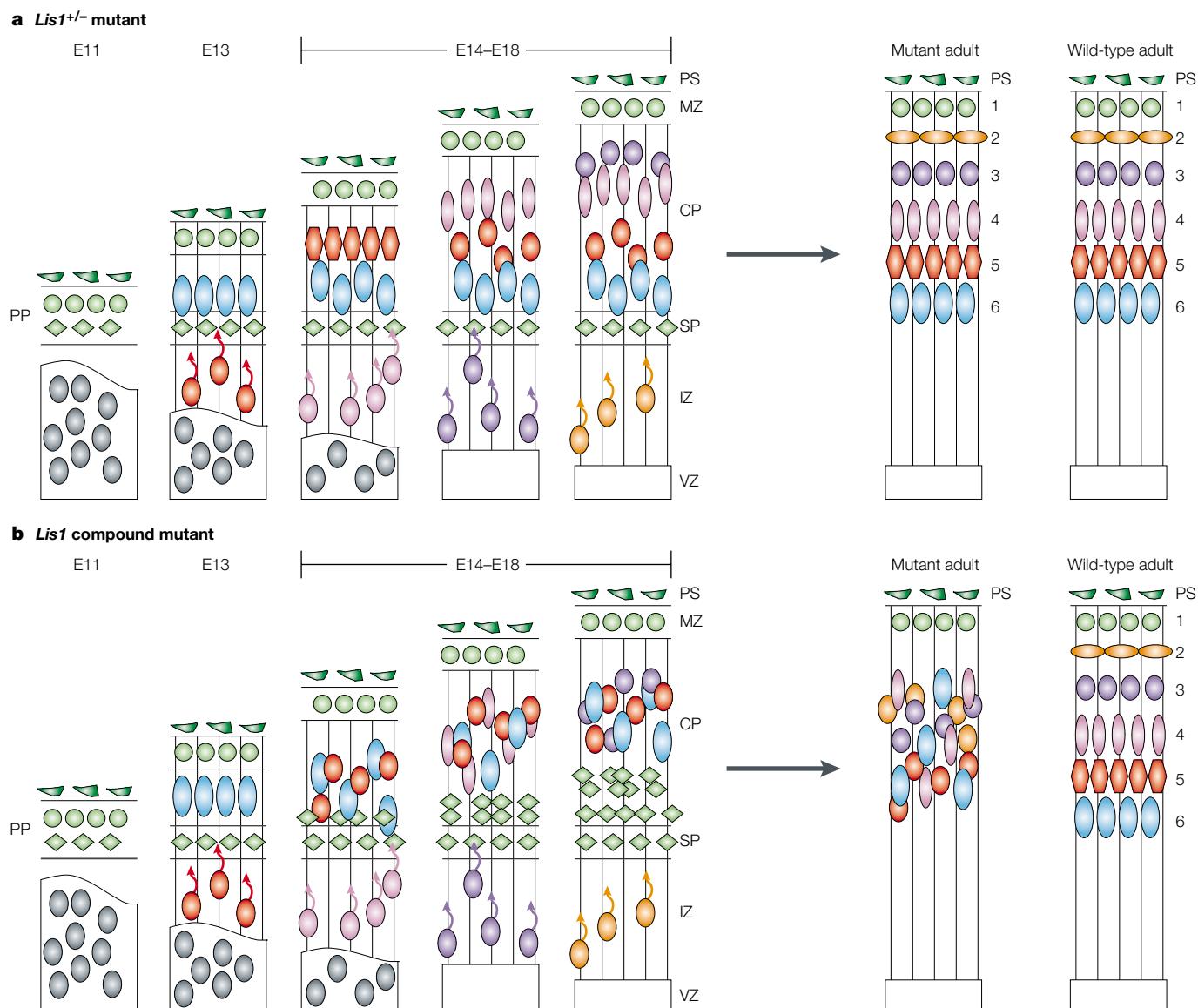


Figure 2 | Neocortical-layer formation in *Lis1* mouse mutants. **a** | Layer formation in *Lis1*^{+/-} mutant mice. The splitting of the preplate (PP) and the inside-out layering of the cortical plate (CP) are preserved in this mutant, but individual layers form more slowly. **b** | Layer formation in *Lis1* compound mutant mice. The splitting of the PP is perturbed in a dose-dependent fashion, which results in a subplate (SP) that is more diffuse and broader. Furthermore, the CP is in complete disarray, with no clear delineation of the individual cortical layers. E, embryonic day; IZ, intermediate zone; MZ, marginal zone; PS, pial surface; VZ, ventricular zone.

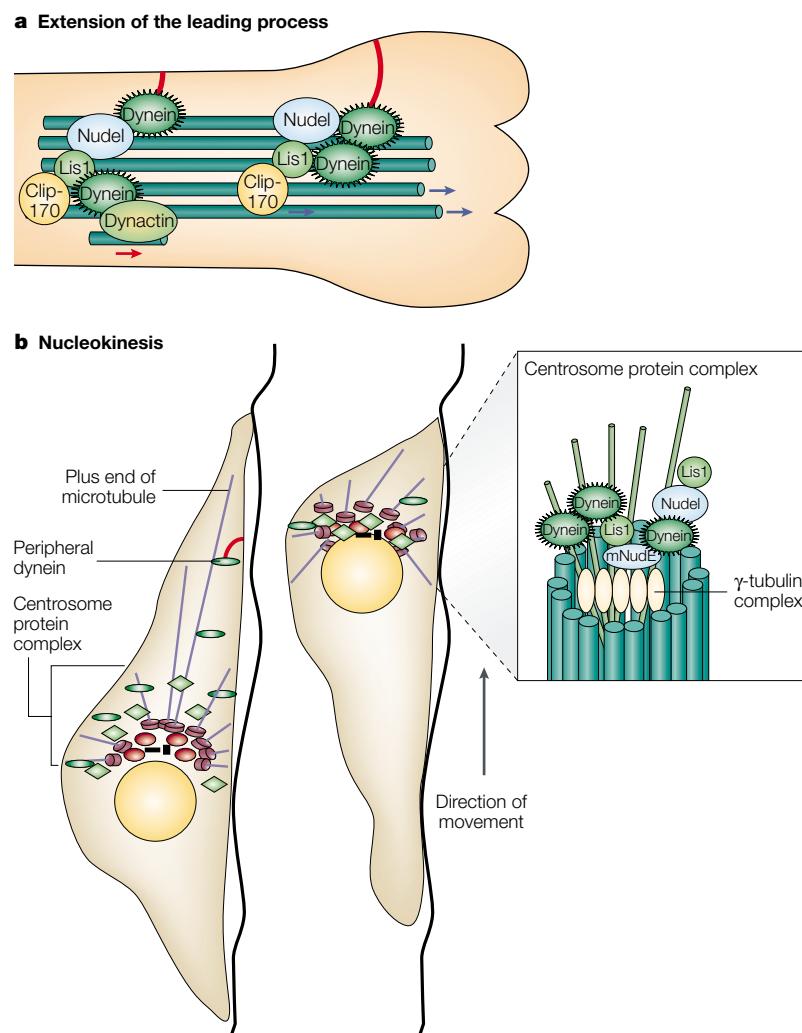


Figure 3 | Cell migration by leading-edge extension and reeling in of the cell soma. **a** | Leading-edge extension in mammalian neurons. The lissencephaly 1 (Lis1)-dynein interaction might modulate the extension of the leading process of a neuron by facilitating plus-end-directed transport of microtubule segments through dynein (red arrow) or by exerting plus-end-directed force on the existing microtubule network (blue arrow). In both cases, dynein is stationary (red anchor lines), as it is a minus-end-directed microtubule motor. This potential Lis1-dynein function might be modulated by Nudel (Nude-like). Clip-170 tethers the Lis1-dynein complex to microtubules. **b** | Reeling in of the cell soma. Peripherally anchored dynein might exert force on microtubules attached to the centrosome and so might pull the cell soma into the periphery. Aspects of microtubule dynamics at the centrosome are potentially regulated by the Lis1-dynein interaction (see insert), together with mNudE and Nudel. Panel **b** modified with permission from REF. 24.

WD40 REPEAT
A conserved finger-like structural motif that consists of repeats of tryptophan (W) and aspartic acid (D). In LIS1, there are seven WD40 repeats that are postulated to assume a 'propeller wheel' configuration similar to other WD40 repeat proteins.

et al., unpublished data)) (FIG. 2b) and, consequently, a diffuse, ill-defined and broad subplate forms. This might occur owing to a primary defect in the formation of the cortical plate and subplate, or to a secondary defect caused by a delay in neuronal migration and inefficient splitting of the preplate, or to a combination of both. Further experiments are needed to distinguish between these models.

LIS1 function. LIS1 is highly conserved across various mammalian species and encodes a protein that contains seven WD40 REPEATS^{46–50}. Two main functions have been described for mammalian LIS1 on the basis of both its activity as a non-catalytic subunit of platelet-

activating-factor acetylhydrolase isoform 1B (PAFAH1B), which is an inactivating enzyme for platelet-activating factor (PAF)⁵¹, and its association with microtubules. It has been argued previously⁵² that the PAFAH1B function of LIS1 might be a mammalian evolutionary adaptation. However, it is unclear how this function is related to PAFAH1B activity and what the implications are of LIS1 regulating PAFAH1B or PAF. It has been proposed that the hydrolysis of PAF might release LIS1 from PAFAH1B, and thereby provide a means of regulating free LIS1 concentrations²⁵.

The second function of LIS1 relates to its association with microtubules⁵³ and to its regulation of cytoplasmic DYNEIN, perhaps through the microtubule-interacting protein CLIP-170 (also known as restin, RSN), which bridges LIS1 to microtubules⁵⁴. Notably, the role of LIS1 in regulating dynein activity is highly conserved among eukaryotes^{55,56}.

The first link between LIS1 and cytoplasmic dynein was reported in studies of several nuclear distribution (*nud*) mutants that affect NUCLEOKINESIS in the filamentous fungus *Aspergillus nidulans*^{57–60} (reviewed in REFS 24,25). A direct genetic interaction between LIS1 and dynein was revealed by the suppression of phenotypic effects of mutations in *nudF*, the fungal homologue of *LIS1* (REF. 61), by mutations in *nudA*, which encodes the heavy chain of cytoplasmic dynein^{62–64}. Subsequently, NUDF was shown to interact physically with NUDA⁶⁵. The NUDF-dynein interaction is also influenced by other proteins, such as NUDE, which binds to NUDF and to the dynein light chain^{65,66}. Notably, NUDE is also a homologue of the mouse proteins Nudel (for Nude-like)^{67,68} and Nude (also known as mNudE)⁶⁹, as well as the rat protein Nude⁷⁰. All three proteins interact directly with LIS1, and the two mouse proteins have also been shown to interact with dynein subunits (see below).

In *Drosophila melanogaster*, loss-of-function mutants of *Lis1* (a homologue of LIS1; also known as *Dlis1*) and *Dhc64C* (which encodes the cytoplasmic dynein heavy chain) have similar phenotypes, which again implicates both proteins in nuclear positioning^{71–73}. In addition, the loss of function of *Lis1* or *Dhc64C* specifically in mushroom-body neurons results in defective neuroblast proliferation and dendritic arborization⁵⁶. Importantly, a gain-of-function allele of *Dhc64C* dominantly suppresses the *Lis1*-null homozygous phenotype⁷⁴, which provides further evidence of a genetic interaction between LIS1 and dynein.

In mammals, LIS1 interacts directly with the cytoplasmic dynein heavy chain (CDHC)⁶⁷. Moreover, LIS1 co-localizes with subunits of dynein and dynein regulatory complex, in the ventricular zone and in the cortical plate of the developing neocortex. This LIS1-dynein interaction is important for the organization of microtubules in the cell periphery, and LIS1 dosage regulates the dynein motor in this cellular process⁷⁵. CLIP-170 might have a role in LIS1-regulated microtubule organization at the cell periphery, as it can recruit LIS1 to the plus ends of microtubules⁵⁴. Another important function

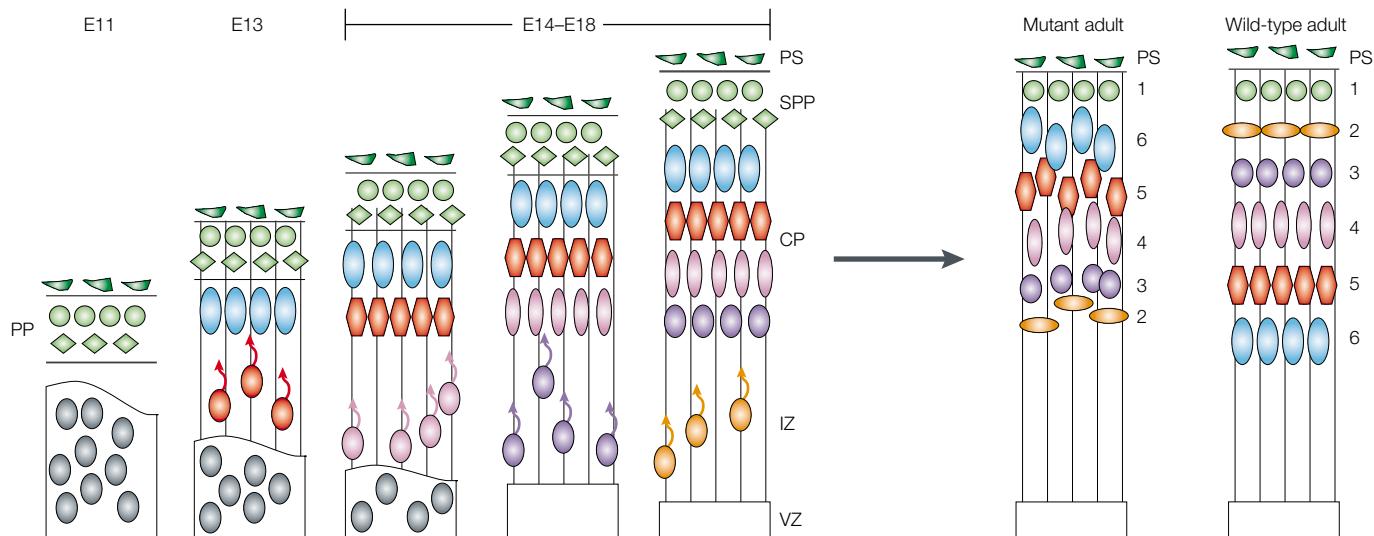
DYNEIN
A minus-end-directed microtubule motor protein that can move along microtubules and carry cell cargo along them towards the cell centre, where the minus ends of the microtubules are positioned. When it is tethered in the periphery to the plasma membrane or to the non-microtubule cytoskeleton, it can transport cell cargo towards the cell periphery.

of the LIS1–dynein interaction might be the regulation of mitotic cell division and chromosomal segregation⁷⁶. LIS1 localizes to mitotic spindle poles and kinetochores, and elevated or reduced levels of LIS1 in cultured cells disrupt mitotic progression, mitotic-spindle orientation and chromosome attachment. Such disruption is believed to depend also on cytoplasmic dynein⁷⁶.

How do the various cellular roles of the LIS1–dynein interaction contribute to the neuronal migration phenotype of lissencephaly mutants? As the timing of the last cell division affects the final

destination of a cortical neuron⁷⁷, it has been proposed that the altered distribution of neurons in lissencephaly might result from the effect of a disrupted LIS1–dynein interaction on cell division, particularly on cell-cycle-dependent interkinetic nuclear migration⁷⁸. This hypothesis is consistent with the effects of the *Lis1* mutation on neuroblast proliferation in *Drosophila*³⁶. Indeed, neurogenesis is altered in *Lis1* mutant mice, and interkinetic nuclear migration in the ventricular zone is disrupted in a dose-dependent manner (M. J. Gambello, unpublished data). Conversely, the LIS1–dynein interaction might independently regulate

a Mutants with preplate defects



b Mutants with post-preplate defects

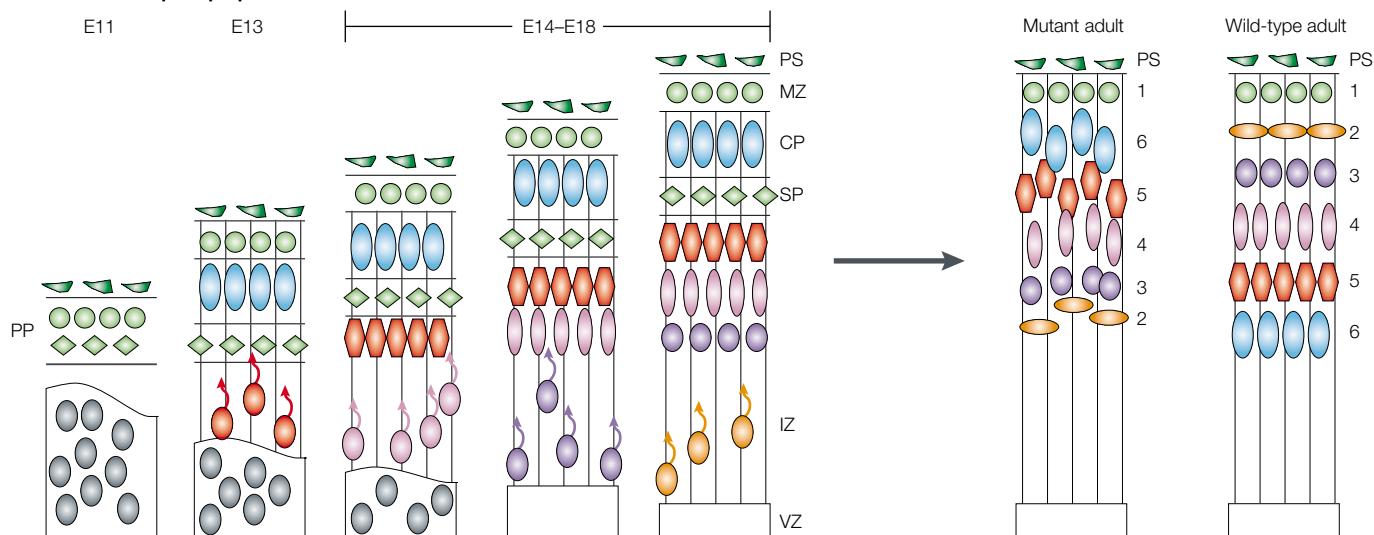


Figure 4 | Mouse mutants with preplate and post-preplate defects. **a** In mice with mutations in *Reln*, *Dab1*, and both *Vldlr* and *ApoER2*, the preplate (PP) does not split and forms a structure called the superplate (SPP). The cortical plate (CP) forms under the SPP and is inverted, which indicates that late-migrating neurons are unable to migrate past their predecessors. Early-migrating and late-migrating neurons are positioned in the superficial and deep layers of the CP, respectively, and layering is very disorganized. **b** In post-preplate mouse mutants — for example, in *p35*^{-/-} and *Cdk5*^{-/-} mutants — PP-splitting is fairly normal. The CP is organized in an inverted fashion, and individual layers are disorganized, albeit less than in the preplate mouse mutants. Notably, most of the CP initially settles beneath the SP in the upper intermediate zone (IZ). In *p35*^{-/-} mutants, CP neurons arrive at their final destination after birth (see text for details). The perinatal death of *Cdk5*^{-/-} mutants prevents the final position of CP neurons in these mice from being assessed. E, embryonic day; MZ, marginal zone; PS, pial surface; VZ, ventricular zone.

postmitotic neuronal migration *per se*. By affecting microtubule organization and dynamics, the LIS1–dynein interaction might regulate the extension of the leading processes of neurons (FIG. 3a), which is consistent with the dendritic arborization defects that are seen in *Lis1* mutant flies⁵⁶ and *Lis1* mutant mice⁴⁷. It is also possible that the LIS1–dynein interaction might facilitate the ‘reeling in’ of the cell soma into the leading process, which is analogous to the events that might occur during nucleokinesis in *A. nidulans*¹⁶ (FIG. 3b). LEADING EDGE and nuclear movements are possibly mediated by dynein motors that are anchored at the anterior tip of the leading process (FIG. 3). However, in the case of the reeling in of the cell soma, peripherally anchored dynein might exert a force on microtubules that are attached to the CENTROSOME and pull the cell soma in the direction of the dynein⁶⁷ (FIG. 3b). Interestingly, dynein has been proposed to regulate microtubule function at the centrosome, and this function might be influenced not only by LIS1, but also by the two mouse NUDE homologues Nudel and mNudE (FIG. 3b, insert). Lis1 and Nudel together bind to Cdhc, and this interaction presumably places these proteins at the centrosome^{67,68}. mNude also binds to Lis1, and to γ -tubulin and several other centrosomal proteins, including the dynein light chain⁶⁹. Hence, Nudel and mNudE might be crucial components of the Lis1–dynein–microtubule interaction at the centrosome and thereby might regulate centrosome-associated processes, including the reeling in of the cell soma in migrating neurons. Consistent with this interpretation, a physical interaction between Lis1 and mNudE is required for the neuronal function of Lis1, as point mutations that are associated with lissencephaly in humans disrupt the Lis1–mNudE interaction, and a dominant-negative mNudE construct disrupts cortical lamination in *Xenopus* embryos⁶⁹.

Doublecortin function. Little is known of how DCX is involved in neuronal migration. This is partly because DCX seems to be present only in mammals, and so has not been studied in fungi or *Drosophila*, and a mouse *Dcx* mutant has yet to be reported. However, both LIS1 and DCX are known to associate with microtubules in mammalian cells. LIS1 seems to increase microtubule polymerization and to decrease microtubule depolymerization rates *in vitro*⁵³, and DCX seems to bind to, and polymerize, microtubules both *in vitro* and in heterologous cells^{43,79,80}. It seems, then, that LIS1 and DCX work in concert to maintain microtubule polymerization, which indicates that they might act in similar pathways. This is supported by the recent finding that DCX and LIS1 interact with each other *in vitro* and in mouse embryonic brain extracts⁸¹, although it is unknown whether DCX has any effect on dynein motor function. DCX and LIS1 might function together to increase the pool of polymerized microtubules to promote neuronal migration, although how microtubule polymerization might facilitate successful neuronal migration remains to be elucidated. It is possible that DCX and LIS1 could function synergistically in the extension of the leading process.

NUCLEOKINESIS

A process in which nuclei migrate towards the tips of the developing hyphae of *Aspergillus nidulans* under conditions of nutrient deprivation. This migration of the nuclei is an example of nuclear translocation and is disrupted in nuclear distribution (*nud*) mutants.

LEADING EDGE

The thin margin of a lamellipodium that spans the area of the cell from the plasma membrane to about 1 μ m back into the lamellipodium. Lamellipodia are flattened, sheet-like projections from the surface of a cell, which are often associated with cell migration.

CENTROSOME

The main microtubule-organizing centre of animal cells.

Mouse mutants: cortical signalling insights

Apart from cortical defects that are associated with human disease, several spontaneous and gene-targeted mouse mutants also undergo defective cortical-layer formation. As we discuss below, these mutants, which can be categorized according to whether or not splitting of the preplate is affected, have considerably expanded our insight into neocortical development.

Defects that affect splitting of the preplate. Several well-known mouse mutants show preplate-splitting defects. These include the naturally occurring *reeler*, *scrambler* and *yotari* mouse mutants, as well as an engineered mutant that is a double homozygous null for the genes *Vldlr* (which encodes very-low-density lipoprotein receptor) and *Lrp8* (which encodes apolipoprotein E receptor 2 (ApoER2))^{82–86}. Notably, all four of these mouse mutants have the same defect in neocortical layering (FIG. 4a): the preplate does not split, and subplate cells remain adjacent to marginal-zone cells in a structure known as the superplate^{87,88}. Consequently, the cortical plate is established underneath the superplate and is itself severely affected, as layering is inverted and indistinct (FIG. 4a). It seems that neurons are not only unable to migrate past their predecessors, but also impaired in organizing themselves into distinct layers⁸⁶.

The finding that all four mouse mutants have the same cortical-layering defect indicated that the corresponding mutant proteins might be identical or might be components of the same signalling pathway. The *reeler* defect is caused by mutations in the reelin (*Reln*) gene⁸². *Scrambler* and *yotari* mice have two distinct mutations in the same gene, *Dab1*, which encodes the mouse homologue of the fly protein *Disabled*^{83–85}. Together with *Vldlr* and *ApoER2*, genetics has therefore provided not only four distinct proteins that are involved in neocortical-layer formation, but also four proteins that are part of the same signalling pathway.

This genetic analysis helped to hasten studies of the individual proteins. *Reln* is a secreted 388-kDa protein that associates with the extracellular matrix. *Vldlr* and *ApoER2* belong to the low-density lipoprotein receptor gene family and as such are multifunctional cell-surface receptors that mediate the endocytosis of extracellular ligands, including *ApoE*^{89,90}. *Dab1* was identified originally as a cytoplasmic adaptor protein that binds physically to family members of the non-receptor tyrosine kinase *Src*⁹¹. Conversely, its *Drosophila* homologue *Disabled*, interacts genetically with *Abl* (another non-receptor tyrosine kinase)⁹², as well as with *enabled* (the mouse homologue of which is *Enah*, also known as *MENA*)⁹³, and with other *Drosophila* proteins that include *fax* (failed axon connections) and *prospero*⁹². Interestingly, all of these interacting proteins are implicated in modulating migration or neurite outgrowth, probably through modification of the actin cytoskeleton⁹², which argues strongly for a role of *Dab1* in neuronal migration.

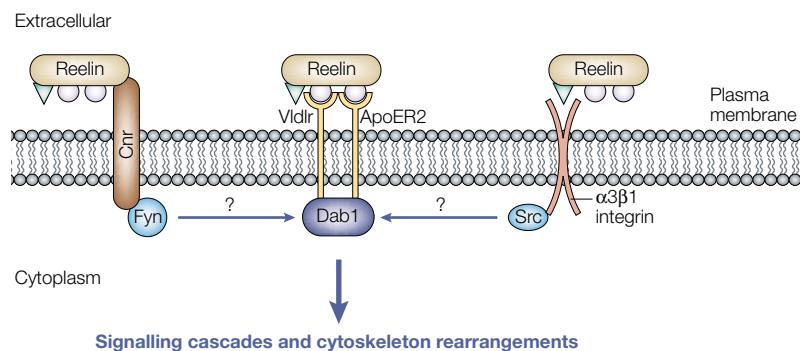


Figure 5 | Reelin signalling. Binding of reelin to very-low-density lipoprotein receptor (Vldlr) or ApoE receptor 2 (ApoER2) results in tyrosine phosphorylation of Dab1, the mouse homologue of Disabled, which interacts with the cytoplasmic tail of either receptor. Tyrosine phosphorylation of Dab1 is necessary for the activation of downstream signalling cascades that ultimately mediate the cellular effects of reelin. The identity of the kinases that tyrosine phosphorylate Dab1 has not been established, although non-receptor tyrosine kinases of the Src family are potential candidates. Consistent with this theory, Fyn and Src bind to cadherin-related neuronal receptors (Cnrs) and integrins, respectively, which are also receptors for Reelin.

Knowledge about the nature of the individual proteins has facilitated studies into how they relate to each other. Reelin is a ligand for both Vldlr and ApoER2 (REFS 94,95). Furthermore, Reelin is secreted by distinct cells of the marginal zone — the Cajal–Retzius cells — which results in the concentration of Reelin in the marginal zone or in its immediate vicinity. Dab1 has been discovered to bind to the cytoplasmic side of both Vldlr and ApoER2 (REFS 96,97). Vldlr, ApoER2 and Dab1 are expressed primarily in the neurons that migrate radially towards the pial surface. In summary, Reelin, the receptors and Dab1 constitute components of a common protein complex that relays an extracellular signal to the interior (FIG. 5). Moreover, in the light of data concerning its localization, Reelin might function as a local or as a graded signal.

Whereas Reelin protein levels are unchanged in *scrambler* mice, the level of Dab1 protein is elevated in *reeler* mice, as well as in *Vldlr/ApoER2* double-knockout mice^{96,98}. These protein relationships nicely support the molecular data that Vldlr, ApoER2 and Dab1 are all involved in transmitting a Reelin signal. Evidence of how this Reelin signal is transmitted came from the observation that Reelin induces the tyrosine phosphorylation of Dab1, and that Vldlr and ApoER2 are necessary for this effect^{95,99} (FIG. 5). However, the true significance of Dab1 tyrosine phosphorylation in active Reelin signalling could not be addressed by these molecular studies. Instead, a *Dab1* knock-in mouse was engineered, in which all of the tyrosine phosphorylation sites of Dab1 were mutated to alanine, making Dab1 completely inaccessible to tyrosine phosphorylation¹⁰⁰. The phenotype of this mouse is essentially identical to that of *reeler* or *scrambler* mice, which shows that the tyrosine phosphorylation of Dab1 is absolutely necessary for successful Reelin signalling. However, it is unclear which Src-family members — previously implicated in the tyrosine phosphorylation of Dab1 — are necessary for this effect *in vivo*¹⁰¹. Also, it is unknown how Dab1 tyrosine phosphorylation translates into the activation of signalling cascades and cytoskeleton rearrangements.

What can be learned from connecting the molecular data to the genetic data? Reelin signalling is crucial for normal splitting of the preplate, as a disruption of any component of the Reelin–receptor–Dab1 signalling complex results in a preplate-splitting defect. In fact, the significance of Reelin signalling in preplate splitting is emphasized by the recent finding that the preplate-splitting defect in *reeler* mice can be rescued partially by ectopically expressing *Reelin* in the ventricular zone¹⁰². Despite these findings, it is far from certain how Reelin signalling accomplishes this. As preplate splitting occurs principally by migrating neurons that carry out somal translocation, Reelin signalling might be instrumental for the proper execution of this glia-independent mode of migration. It is conceivable that, to split the preplate, the leading edge of a translocating neuron concentrates Vldlr or ApoER2 on its surface. When the leading edge attaches to the pial surface, either of the two receptors is able to bind to Reelin, as it is secreted by the Cajal–Retzius cells of the developing marginal zone. The Reelin–receptor interaction results in tyrosine phosphorylation of Dab1, which activates downstream signalling events that propel the cell soma towards the leading edge to split the preplate successfully.

This, of course, is not the whole story, as Vldlr and ApoER2 are not the only Reelin receptors. Reelin also binds to α3β1-integrin¹³ (FIG. 5), an adhesion molecule that is implicated in maintaining neuronal–glial interactions¹⁴. As α3β1-integrin is expressed in migrating cortical neurons, and Reelin can detach migrating neurons from glia in cortical-imprinting assays¹³, Reelin binding to α3β1-integrin might contribute to neuronal–glial disruption *in vivo*. Reelin also binds to members of the cadherin-related neuronal receptor (*Cnr*) family (FIG. 5), which are expressed in cortical-plate neurons that are adjacent to the marginal zone¹⁰³. Notably, Reelin-binding to Cnr depends on an RGD motif in Cnr. As this RGD motif is also a crucial feature of several integrin ligands¹⁰⁴, integrins and Reelin might compete for binding to Cnr. Integrins that are expressed on radial glia have also been implicated in promoting neuronal–glial interactions¹⁴. Reelin binding to Cnr might facilitate the detachment of migrating neurons from glia by sequestering Cnr away from glial integrins. So, the integrins and the Cnr family might have a role in common in modulating neuronal–glial adhesion. This role of Reelin might be relevant at later stages of neocortical development, when locomoting cells need to detach from radial glia to complete their journey by somal translocation (BOX 1). Both receptors also signal through Src-family kinases^{103,105,106}, which could contribute to Reelin-induced Dab1 tyrosine phosphorylation (FIG. 5). Future studies are required to sort out the complex interactions between Reelin, its various receptors, Dab1 and the signalling cascades that mediate its effects.

Defects that spare preplate splitting. Genetics has also provided mutants that have cortical-plate disruptions in the absence of preplate defects. Classical examples of such mutants include mice that are deficient in cyclin-dependent kinase 5 (*Cdk5*) or in *Cdk5r* (also known as p35), which is a regulatory activator of *Cdk5* (REFS 30,107–110). Mice that are null for either encoding gene

RGD MOTIF
A peptide motif that consists of the amino acids arginine (R), glycine (G) and aspartate (D), which is found in many ligands that bind to integrins.

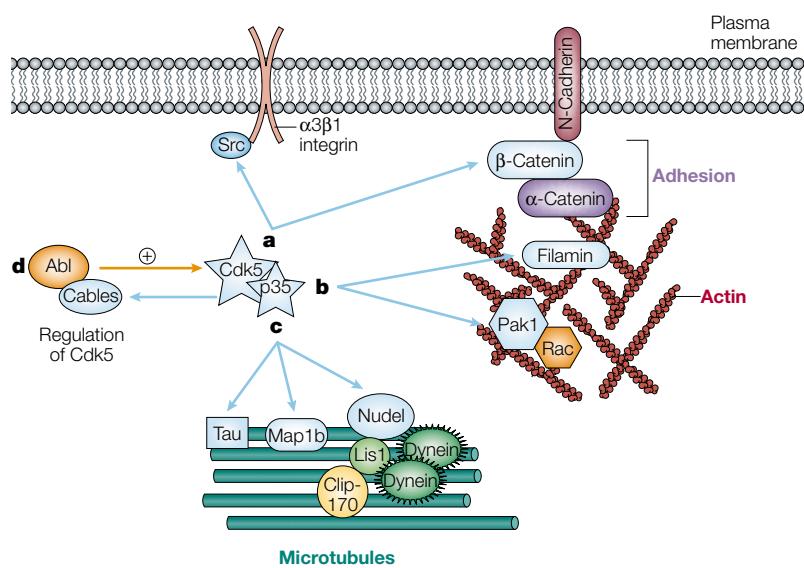


Figure 6 | p35–Cdk5 signalling. p35–Cdk5 and their substrates are depicted in light blue, as are the arrows that connect p35–Cdk5 to their substrates. **a** | p35–Cdk5 might modulate integrin-dependent cell–matrix adhesion by phosphorylating Src and might also affect N-cadherin-dependent cell–cell adhesion, as binding of p35–Cdk5 to β -catenin might disrupt the tethering of N-cadherin to the plasma membrane. **b** | p35–Cdk5 might also regulate actin dynamics by phosphorylating filamin, an actin-binding protein, or by inhibiting Pak1, on phosphorylating it, in a Rac-dependent manner. **c** | p35–Cdk5 might also influence microtubule dynamics by phosphorylating the microtubule-associated proteins Map1b or Tau, which both stabilize microtubules, or by phosphorylating Nudel, which interacts with the Lis1–dynein complex. **d** | Cdk5 activity itself might be modified by the non-receptor tyrosine kinase Abl, which can be targeted to Cdk5 by Cables and can activate Cdk5 by phosphorylating it (orange arrow). Furthermore, Cdk5 might fine tune its association with Abl and Cables by phosphorylating Cables. Cdk5, cyclin-dependent kinase 5; Lis1, lissencephaly 1.

have two main layering defects^{111–113}. First, layering of the cortical plate is inverted, which reflects the inability of later-born neurons to migrate past their predecessors (FIG. 4b). However, in contrast to mutants with defective preplate splitting, the inversion of the cortical plate is much more pronounced, as layering *per se* is reasonably well preserved. Second, although the early layers of the cortical plate are organized correctly between the marginal zone and the subplate, much of the cortical plate that develops later is situated aberrantly in the upper intermediate zone, underneath the subplate¹¹⁴ (FIG. 4b). It is only after the birth of p35 mouse mutants that this portion of the cortical plate becomes positioned between the marginal zone and the subplate, which reflects a significant delay in radial migration past the subplate (FIG. 4b).

How can the phenotypic consequences of the loss of p35 or Cdk5 be explained in molecular terms? Cdk5, like other Cdks, is only active as a kinase when complexed with an activator; however, unlike other Cdks, Cdk5 is not activated by a cyclin, but by the activator proteins p35 or Cdk5r2 (also known as p39)^{30,108–110}. In the neocortex, p35 is the dominant activator of Cdk5, as revealed by the fact that *p35*^{−/−} or *Cdk5*^{−/−} mice have very similar defects in neocortical layering^{111–113}, whereas *p39*^{−/−} mice are phenotypically normal¹¹⁵. Interestingly, as Cdk5 activity is restricted to postmitotic cells, the p35–Cdk5 complex might function in the migrating neuron itself to allow for correct neocortical layering.

N-CADHERINS

Adhesion molecules that bind to each other in a calcium-dependent manner, and thereby connect nerve cells to each other. N-cadherin is stabilized on the cell surface by β -catenin, which links N-cadherin to the actin cytoskeleton.

To explain how p35–Cdk5 might contribute to normal inside-out layering, biochemical and cell-biological approaches have identified a diverse group of p35–Cdk5 substrates. p35–Cdk5 might regulate cell–cell adhesion by binding to β -catenin¹¹⁶ (FIG. 6a). Overexpression of p35–Cdk5 reduces the interaction between β -catenin and N-CADHERIN, which results in a decrease of N-cadherin levels at the cell surface¹¹⁶. Furthermore, the observed increase of homotypic adhesion in *p35*^{−/−} neurons can be reversed partly by an N-cadherin-blocking peptide¹¹⁶. These results indicate that a *p35*^{−/−} neuron might not migrate past its predecessors, because it adheres to them in an N-cadherin-dependent way.

The p35–Cdk5 complex is also believed to orchestrate key neuronal–glial interactions. As mentioned before, the integrin family of adhesion molecules mediates some neuronal–glial interactions¹⁴. Notably, integrin signalling is mediated by the non-receptor tyrosine kinase Src, which is also a p35–Cdk5 substrate¹¹⁷, indicating that p35–Cdk5 might regulate neuronal–glial adhesion by affecting integrin signalling through the phosphorylation of Src (FIG. 6a).

The p35–Cdk5 complex might also regulate the motility of migrating neurons. Motility is based on cytoskeletal dynamics that determine migration speed and that coordinate cell–migration components, such as process extension and cell–soma propulsion. There is ample evidence that p35–Cdk5 modulates the dynamics of the actin cytoskeleton^{118–120} (FIG. 6b). p35–Cdk5 might also influence microtubule dynamics (FIG. 6c) by phosphorylating microtubule-associated proteins, which in turn might affect microtubule stability^{121,122}, or by phosphorylating Nudel to regulate the dynein motor complex^{67,68}. Taken together, the regulation of cytoskeleton dynamics by p35–Cdk5 might secure the proper radial movement of migrating neurons. In the absence of p35–Cdk5, radial movement might be erratic, which indicates defective neuronal extension or cell–soma propulsion¹²³, or slowed, which explains the delay of radial migration past the subplate.

Finally, as several p35–Cdk5 substrates are pertinent to cortical migration, the question arises as to how p35–Cdk5 activation is regulated spatially and temporally. A protein known as Cables connects the non-receptor tyrosine kinase Abl to Cdk5 (REF. 124) (FIG. 6d). Abl phosphorylates the Y15 residue of Cdk5, which promotes the formation of a p35–Cdk5 complex and subsequent activation of Cdk5. So, it is possible that the spatial and temporal activation of p35–Cdk5 is regulated, at least in part, by the spatial and temporal activation of Abl (FIG. 6d).

Connecting the signalling pathways

Different types of defect in neocortical layering arise from disruptions of distinct molecular signalling pathways. The common denominator of the mutants discussed above is that they all show disrupted radial migration, which raises the question as to whether the signalling pathways are interconnected. The answer is yes.

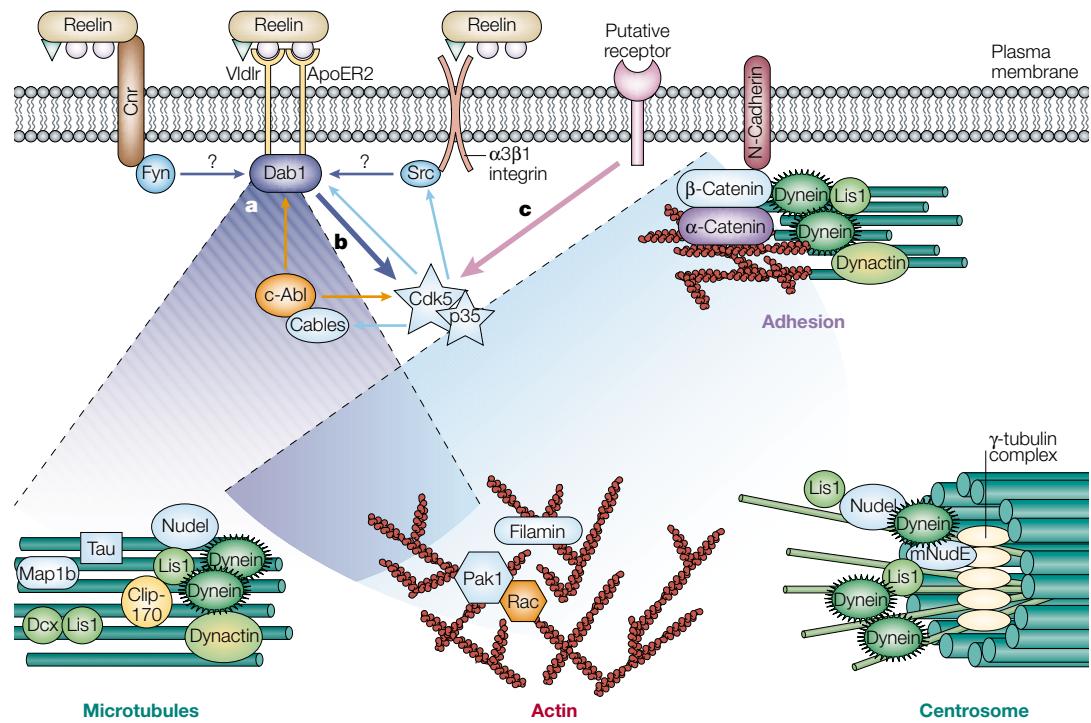


Figure 7 | Integration of signalling. **a** | Reelin (Reln) signalling is independent of p35–Cdk5. Reln signalling leads to tyrosine phosphorylation of Dab1, which, in turn, activates the signalling cascades that mediate the various cellular effects of Reln. Effects on cytoskeleton rearrangements are also shown (Dab1 shaded area), but effects on other cellular processes can equally be envisioned. **b** | Reln and p35–Cdk5 crosstalk. p35–Cdk5 might exert an influence on Reln signalling by indirectly or directly targeting the phosphorylation status of Dab1 (light blue arrows). Reln signalling, in turn, might regulate p35–Cdk5 activity through the tyrosine phosphorylated Dab1 (dark blue arrow), thereby affecting p35–Cdk5-dependent cellular processes that include adhesion, actin rearrangements, and microtubule dynamics in the periphery and at the centrosome (see p35–Cdk5 shaded area). **c** | p35–Cdk5 signalling is independent of Reln. The effects that p35–Cdk5 exerts on cellular processes (see p35–Cdk5 shaded area) are also mediated by cell-surface molecules other than Reln receptors. ApoER2, apolipoprotein E receptor; Cdk5, cyclin-dependent kinase 5; Lis1, lissencephaly 1; Vldlr, very-low-density lipoprotein receptor.

p35–Cdk5 and Reln signalling. The first evidence of the interconnectivity of signalling was again provided by genetics. Mice that are deficient for both *p35* and *Dab1* (or *p35* and *Reln*) have a neocortical-layering phenotype that differs from that of mice that are deficient for either gene alone¹²⁵. Some aspects of the neocortical phenotype are more severe in the compound mice than in the single-knockout mutant mice, which indicates that the p35–Cdk5 and Reln signalling pathways intersect to orchestrate cortical migration. Moreover, as some aspects of the neocortical phenotype are distinct, it is probable that p35–Cdk5 and Reln–Dab1 are also part of separate pathways. However, what cannot be deduced from these results is the hierarchical order in which, or the level at which, the signalling pathways relate to each other.

More evidence of the interconnectivity of these pathways can be obtained from a careful analysis of the *Cdk5*^{-/-} phenotype¹¹⁵. Although nearly identical to the *p35*^{-/-} neocortex, the *Cdk5*^{-/-} neocortex has an important difference in that preplate separation into marginal zone and subplate is not as distinct as it is in the *p35*^{-/-} neocortex¹¹⁵. This phenotypical difference raises the possibility that Cdk5 is involved, at least to some degree, in the completion of preplate splitting. As the Reln path-

way strongly affects splitting of the preplate, it is plausible that the proper completion of preplate splitting requires crosstalk between the Cdk5 and Reln signalling pathways. However, the issue of preplate splitting is complex, as chimeric studies with *Cdk5*^{-/-} mice have indicated that Cdk5 function might not be required for preplate splitting⁴. Further studies are required to clarify the degree of signalling crosstalk, such as the assessment of *Cdk5*^{-/-} neurons for marginal zone or subplate identity in chimeric mice.

Biochemical and molecular studies have also complemented genetic studies regarding the interconnectivity of these pathways. Tyrosine phosphorylation of Dab1 is crucial for active Reln signalling (as discussed above), and Src-family kinases have been implicated in this phosphorylation by the finding that inhibitors of Src-family kinases reduce tyrosine phosphorylation of Dab1 in cell culture¹⁰¹. Moreover, Src itself can tyrosine-phosphorylate Dab1 *in vitro*¹⁰⁰. As Src is also a p35–Cdk5 substrate *in vitro*¹¹⁷, Dab1 phosphorylation by Src might be regulated in a p35–Cdk5-dependent manner (FIG. 7). Alternatively, p35–Cdk5 might affect Dab1 tyrosine phosphorylation through Abl, which interacts with Dab1 and is connected to p35–Cdk5 through Cables (FIG. 7). More generally, it seems that

non-receptor tyrosine kinases could provide a bridge between the p35–Cdk5 and Reln signalling pathways. However, to give physiological significance to this potential molecular connection, it needs to be clarified which Src-family members are necessary for Dab1 tyrosine phosphorylation *in vivo*. Furthermore, it has not been established consistently whether the interaction of p35–Cdk5 with Abl or Src affects their tyrosine-kinase activity. Finally, if p35–Cdk5 regulates Src or Abl kinase activity, it needs to be shown that such regulation affects Dab1 tyrosine phosphorylation in the way that it has an impact on Reln signalling as a whole.

Is Dab1 itself a p35–Cdk5 substrate? Recent evidence indicates that p35–Cdk5 can phosphorylate Dab1 at serine and threonine residues *in vitro*^{94,125} (FIG. 7). However, *in vivo* data to support this are still lacking and, in contrast to tyrosine phosphorylation, the functional significance of Dab1 serine/threonine phosphorylation is unresolved. To determine whether direct Dab1 phosphorylation by p35–Cdk5 is required for proper Reln signalling, it will be necessary to assess the phenotype of a *Dab1* knock-in mouse, in which all Cdk5 phosphorylation sites have been made inaccessible to phosphorylation.

Finally, links between the p35–Cdk5 and Reln signalling pathways are supported by the observation that phosphorylation of Tau is increased in *reeler* mutant mice and in mice that lack both Vldlr and ApoER2 (REF. 95). Tau is a p35–Cdk5 substrate, and it seems that p35–Cdk5 activity might be influenced by Reln signalling. If true, this implies that Reln signalling impinges on p35–Cdk5 to orchestrate cytoskeletal changes (FIG. 7). However, as several kinases are able to phosphorylate Tau protein, they might mediate equally well Tau phosphorylation in these mice.

p35–Cdk5, Lis1 and cytoplasmic dynein. Nudel, one of the mouse homologues of fungal NUDE, might be another avenue of crosstalk between Cdk5 and Lis1 signalling. As mentioned previously, Nudel binds to Lis1 and Cdhc and thereby modulates the interaction of these two proteins. Nudel also contains five Cdk5 consensus phosphorylation sites and can be phosphorylated by p35–Cdk5 *in vitro* and in co-transfected cells^{67,68}. Therefore, p35–Cdk5-mediated phosphorylation of Nudel might modify its function to affect the interaction between Lis1 and dynein (FIGS 6c,7). Interestingly, as all five Cdk5 phosphorylation sites are located in the dynein interaction domain of Nudel⁶⁸, p35–Cdk5 phosphorylation of Nudel might affect the binding of Nudel to dynein or regulate dynein function through Nudel. In either case, altered Nudel–dynein interactions could affect the way in which Lis1 relates to dynein and the way in which the dynein complex functions on microtubules in the periphery and at the centrosome (FIGS 6c,7).

What about mNudE, the other homologue of fungal NUDE? As noted above, mNudE might also regulate microtubule-dependent processes at the centrosome, as it acts as a scaffold for several centrosomal proteins, including γ -tubulin and the dynein

subunit⁶⁹. As the Cdk5 phosphorylation sites in Nudel are conserved in mNudE, Cdk5 might also phosphorylate mNudE and affect its function to modulate the organization of centrosomal proteins (FIG. 7). However, as the interaction domains in mNudE have not yet been mapped, the precise type of mNudE interaction that is affected potentially by p35–Cdk5 phosphorylation awaits further studies.

The Cdk5 and Lis1 pathways might also converge on ADHERENS JUNCTIONS. Although the actin cytoskeleton is essential for the formation of adherens junctions, the maintenance of these junctions depends on microtubules¹²⁶. Recently, dynein was shown to interact physically with β -catenin at adherens junctions¹²⁷, which indicates that dynein might be tethering microtubules to these junctions. The link between dynein and β -catenin is noteworthy, as it allows for modulation by Lis1 and p35–Cdk5 (FIG. 7). Lis1 might affect microtubule tethering at adherens junctions by interacting directly with dynein. p35–Cdk5, by contrast, might modulate the same process through its interaction with β -catenin. As mentioned previously, p35–Cdk5 binding to β -catenin disrupts N-cadherin-dependent adhesion. It is possible that the effect of p35–Cdk5 in this context could be caused partly by a phosphorylation-dependent disruption between β -catenin and dynein.

RELN and LIS1. *RELN* mutations in humans cause **lissencephaly with cerebellar hypoplasia** (LCH), a disease that is characterized by autosomal-recessive lissencephaly and malformations of the cerebellum¹²⁸. These mutations disrupt the splicing of *RELN* mRNA, which results in low or undetectable amounts of RELN protein. Although LCH shows phenotypic differences from *reeler* mice and from type I lissencephaly in humans, interesting similarities between them also exist¹²⁸, which indicates that *RELN* mutations are possibly linked with other lissencephaly genes.

Concluding thoughts

Our understanding of neocortical-layer formation has come a long way — from discovering mutants of neocortical development to integrating the signalling pathways that control this process. Now that the connections between the three signalling pathways of Lis1, Reln and Cdk5 have become evident, we can speculate about a model that brings these pathways together (FIG. 7). Although simplified and incomplete, this model provides a way to understand the molecular mechanisms that underlie neuronal migration and positioning.

Putting the evidence in perspective, it is conceivable that the Reln–Vldlr/ApoER2–Dab1 axis orchestrates p35–Cdk5-independent aspects of neuronal migration (FIG. 7a). Additionally, Reln might crosstalk with p35–Cdk5 at the level of Dab1, either indirectly through Src-family kinases or directly through p35–Cdk5 itself (FIG. 7b). This interaction with Dab1, in turn, might result in the modulation of p35–Cdk5 activity and the subsequent regulation of p35–Cdk-dependent aspects of neuronal migration (FIG. 7b). Important molecular processes that underlie neuronal migration include

ADHERENS JUNCTIONS
Points of cell-cell contact. In adherens junctions, a member of the cadherin family usually binds to β -catenin, which in turn is linked to the cortical actin cytoskeleton through α -catenin.

N-cadherin-mediated neuronal–neuronal adhesion, integrin-mediated neuronal–glial adhesion, the regulation of actin dynamics and the modulation of microtubule dynamics through microtubule-associated proteins or the Lis1–Nudel/mNudE–dynein complex (FIG. 7). At the cellular level, the molecular processes might translate into the extension of the leading processes of migrating neurons, the ‘reeling in’ of the cell soma or the coordination of both. It is difficult, at present, to indicate the directionality of the crosstalk between Reln and Cdk5, but we favour the idea that the Reln and Cdk5 pathways are organized in a parallel, and not in a linear, fashion. In other words, some aspects of p35–Cdk5 signalling might depend on Reln signalling (FIG. 7b), whereas other aspects might be Reln independent (FIG. 7c). Consistent with this idea, mice that are deficient in the Reln and Cdk5 pathways have common and distinct defects in the neocortex.

The inability to split the preplate seems to be specific to Reln signalling. As somal translocation predominates at the time of preplate splitting, this mode of migration might be required for preplate splitting and might be mediated by cytoskeleton changes that depend on Reln signalling, but not on p35–Cdk5. Conversely, a disruption in the inside-out layering of the cortical plate seems to be common to mice that are deficient in either the Reln or the Cdk5 pathway. Perhaps, Reln signalling through p35–Cdk5 is necessary for cortical neurons to

migrate past their predecessors. Alternatively, Reln and Cdk5 signalling might be required independently, but at different times during the journey of cortical-plate neurons. At the molecular level, both the regulation of adhesion and migration dynamics might contribute to inside-out layering. Finally, *Lis1* mutant mice and *Cdk5^{-/-}* mice both show delayed neuronal migration. This indicates that migration speed might be a function of p35–Cdk5 signalling that requires Lis1-dependent modulation of microtubule dynamics.

Further advances in our understanding of neuronal migration will be best addressed by a combination of both genetic and developmental approaches. The *in vivo* studies of migration in various single and double knockout mouse mutants of each of the three pathways will shed light on their genetic interactions. Time-lapse imaging approaches in cortical slices can also be used to assess the various mutants for defects in somal translocation or locomotion, and are complemented by *in vitro* migration assays using cortical neurons from the single and double knockout mutant mice. Manipulating gene function by gene transfer into subsets of neurons will also help to order the effects of the pathway, and to determine what subset of functions are mediated by individual pathways. A combination of these approaches will be useful to determine the precise defects in migration in each of the mutants and how the pathways that regulate cortical layering are integrated.

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DATABASES

The following terms in this article are linked online to:

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 β -catenin | Abi | Cables | Cdk5 | Cdk5r | Cdk5r2 | CLIP-170 | Cnr family | cytoplasmic dynein heavy chain | Dab1 | DCX | Dcx | Dhc64C | Disabled | enabled | Enah | fax | Lis1 | Lis1 (fly) | Lis1 (mouse) | Lrp8 | Nude (mouse) | Nude (rat) | Nudel | PAFAH1B | prospero | Reln | Vldlr

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 isolated lissencephaly sequence | lissencephaly | lissencephaly with cerebellar hypoplasia | Miller-Dieker syndrome

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- The neocortex is the most recently evolved part of the cerebral cortex. Its development has been well studied for some time, partly because of its striking organization into six distinct neuronal layers and partly because its development is disrupted in several human diseases.
- Only recently have significant advances been made in our understanding of the molecular mechanisms that determine neocortical development, mainly based on genetic studies of human diseases that affect cortical development, such as lissencephaly, and of mouse mutants with cortical-layering defects. These studies have provided molecular biologists with several key proteins that are required for proper neuronal migration in the cortex.
- Genetic and cell-biological studies have identified how several key proteins that are required for the proper radial migration of neurons in the cortex, such as reelin, lissencephaly 1 and cyclin-dependent kinase 5, modulate signalling pathways that are involved in neuronal migration and adhesion. Recent work is beginning to identify how these pathways interact and modulate one another.
- Further advances in our understanding of neuronal migration and how the pathways that regulate cortical layering are integrated will be best addressed by a combination of both genetic and developmental approaches.

Amitabh Gupta received his M.D. from the University of Heidelberg Medical School, Germany, and then pursued a short postdoctoral fellowship with the late Edgar Haber at the Harvard School of Public Health. He went on to enter the Ph.D. programme in Biological and Biomedical Sciences at Harvard Medical School, where he is now in his fifth year, under the auspices of Li-Huei Tsai. His research interest focuses on the dynamic analysis of migration defects in mutants of cortical-layer formation.

Li-Huei Tsai earned her Ph.D. from the Southwestern Medical School. After a postdoctoral fellowship at the Cold Spring Harbor Laboratories and Massachusetts General Hospital Cancer Center, she became a faculty member at the Department of Pathology at Harvard Medical School, where she is now a professor and an assistant investigator of the Howard Hughes Medical Institute. Her laboratory studies the role of the Cdk5 kinase in the central nervous system, ranging from the involvement of this kinase in neurodevelopment and synaptic plasticity to neurodegeneration.

Anthony Wynshaw-Boris received his M.D. and Ph.D. from Case Western Reserve University, followed by a clinical genetics and postdoctoral fellowship at the Harvard Medical School. He was the head of the Mouse Models Unit in the National Human Genome Research Institute at the National Institutes of Health for five years, before moving to the Departments of Pediatrics and Medicine at the UCSD School of Medicine. His laboratory studies mouse models of human neurogenetic diseases and cancer, including mouse models of human neuronal migration defects.

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