Lineage-dependent circuit assembly in the neocortex

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Summary
The neocortex plays a key role in higher-order brain functions, such as perception, language and decision-making. Since the groundbreaking work of Ramón y Cajal over a century ago, defining the neural circuits underlying brain functions has been a field of intense study. Here, we review recent findings on the formation of neocortical circuits, which have taken advantage of improvements to mouse genetics and circuit-mapping tools. These findings are beginning to reveal how individual components of circuits are generated and assembled during development, and how early developmental processes, such as neurogenesis and neuronal migration, guide precise circuit assembly.

Key words: Lineage, Neuronal circuits, Neocortex

Introduction
The mammalian cerebral cortex is composed of the archicortex (hippocampal region), the paleocortex (olfactory cortex) and the neocortex, with the last being the evolutionarily youngest region. The neocortex is composed of two major classes of neurons: glutamatergic projection neurons (see Glossary, Box 1), which elicit excitation in postsynaptic neurons and generate circuit output; and GABA (γ-aminobutyric acid)-ergic interneurons (see Glossary, Box 1), which typically trigger inhibition in postsynaptic neurons and are essential for shaping circuit output. It is generally accepted that two defining structural and functional features of the neocortex are lamination and radial columns (Douglas and Martin, 2004). Together, these features provide the basic framework on which neocortical circuits are built. Interestingly, both of these features are tightly linked to early developmental events, including neurogenesis and neuronal migration. In this Review, we discuss recent findings on the generation, migration and organization of generating neurons and glia.

Lamination: a hallmark of the neocortex
The neocortex is a continuous six-layered structure. All components of neocortical circuits, including afferents, excitatory cells, inhibitory cells and efferents, are organized with respect to the laminae (Douglas and Martin, 2004). Cortical lamination is generated as a result of radial migration of newborn excitatory neurons during development (Hatten, 1999; Rakic, 1971; Rakic, 1972). Glutamatergic excitatory neurons are produced from progenitor cells (Fig. 1A) that reside in the proliferative zone of the

Box 1. Glossary
Cortical plate. A progressively thickening layer in the dorsal telencephalon that harbors newly born post-mitotic neurons and eventually develops into the future cortex.
Hebbian learning rule. ‘Cells that fire together, wire together’: a theory introduced by Donald O. Hebb (Hebb, 1949) for the mechanism of synaptic plasticity whereby repeated and persistent stimulation of the postsynaptic cell by a presynaptic cell increases synaptic efficacy.
Interneurons. Inhibitory neurons with short axons in the cortex that typically participate in only local circuits.
Marginal zone. A superficial layer that develops as the preplate is split during early corticogenesis; it eventually becomes layer 1 of the mature cortex.
Pia (or pia mater). Innermost layer of the meninges that surrounds the brain and spinal cord.
Preplate. Located between the pia and the ventricular zone, it contains the earliest born neurons and represents the beginning of corticogenesis prior to the emergence of the cortical plate.
Projection neurons. Excitatory neurons in the cortex that send long-range projections to different brain regions.
Radial glial cells. A major population of neural stem cells transiently existing in the developing brain that are crucial for generating neurons and glia.
Striatum. A subcortical structure derived from ventral regions of the developing telencephalon that receives input from the cortex.
Subplate. A transient zone comprising of some of the earliest generated neurons in the cortex; it is crucial for both structural and functional development of the cortex.
Subventricular zone. A region situated above the ventricular zone that harbors intermediate progenitor cells and migrating neurons.
Telencephalon. The anteriormost region of the developing CNS that gives rise to the mature cerebrum.

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Fig. 1. Generation and migration of neocortical excitatory and inhibitory neurons. (A, B) Excitatory and inhibitory neurons originate from different germinal zones of the embryonic telencephalon. (A) Cortical excitatory neurons are generated from progenitor cells (Pax6+, orange) residing in the ventricular zone (VZ) of the dorsal telencephalon. Newborn excitatory neurons undergo radial glial fiber-guided radial migration (orange arrows) and settle into the developing cortical plate (CP, light green). (B) Cortical inhibitory interneurons are predominantly generated from progenitor cells located in the proliferative zone of the ventral telencephalon, mainly within the medial ganglionic eminence (MGE; contains Nkx2.1+ cells, dark green) and the caudal ganglionic eminence (CGE). A small population of cortical inhibitory interneurons is produced from the preoptic area (PoA). Newborn inhibitory interneurons follow two tangentially oriented migratory streams to enter the cortex: a superficially migrating early cohort (pale-blue arrows) migrates through the marginal zone, and a deeply migrating second and more prominent cohort (dark-blue arrows) migrates through the lower intermediate zone and subventricular zone. Upon reaching the cortex, they switch to radial migration (pink double-headed arrows) and settle into their final laminar position in the CP. (C) Inside-out fashion of cortical layer (L) formation. In early developmental stages (E10-E11), the neural tube is composed of a single layer of neuroepithelial cells. A small fraction of these undergo asymmetric division to generate the first wave of postmitotic neurons that migrate out radially and form the preplate (PP). As development proceeds (E12-E13), newborn excitatory neurons split the PP into a superficial marginal zone (MZ) and a deeper subplate (SP). Successive waves of newly generated excitatory neurons migrate past the existing neurons to occupy a more superficial region in the CP (E13-E18), creating the mature six-layered cortex. Excitatory neurons in the mature cortex are heterogeneous. IZ/SVZ, intermediate zone/subventricular zone; WM, white matter.
and simultaneously generate daughter cells that are either postmitotic neurons or intermediate progenitor cells (IPCs). IPCs then undergo additional rounds of symmetric division in the subventricular zone (SVZ, see Glossary, Box 1) to produce neurons (Kowalczyk et al., 2009; Noctor et al., 2004). Newborn neurons undertake radial glial fiber-guided radial migration, splitting the existing preplate into a superficial marginal zone (MZ, see Glossary, Box 1) and a deeper subplate (SP, see Glossary, Box 1), and reside in the middle region, leading to the formation of the cortical plate (CP, see Glossary, Box 1) – the future cortex. Successive waves of newly generated neurons migrate past the existing early-born neurons and occupy more superficial layers in the CP, creating cortical layers (L) 2-6. Thus, cortical lamination occurs in an ‘inside-out’ fashion (Angevine and Sidman, 1961) (Fig. 1C).

It is becoming increasingly clear that the progenitor cells for excitatory neurons are not homogeneous, but rather diverse. In addition to RGCs and IPCs, two new types of neuronal progenitor cells were recently discovered in the developing neocortex: short neural precursors (SNPs) and outer subventricular zone (OSVZ) radial glial progenitors. SNPs maintain their ventricular end-feet, while their basal processes are of variable length (Gal et al., 2006) and, unlike RGCs, they generate neurons directly instead of going through an IPC stage (Stancik et al., 2010). OSVZ progenitors, by contrast, maintain the basal processes but lack the apical processes, and are capable of undergoing asymmetric division in the OSVZ. They were initially discovered in humans and later found in primates, ferrets, mice and other species (Fietz et al., 2010; Hansen et al., 2010; Kelava et al., 2012; Shitamukai et al., 2011; Wang et al., 2011). Notably, the abundance of the OSVZ progenitor population and their increased proliferative potential has been suggested to underlie the evolutionary expansion of the cortex from mouse to human (Lui et al., 2011). To add to the progenitor heterogeneity, a recent study indicated that a subset of RGCs expressing the homeobox protein cut-like 2 (Cux2) is pre-specified to give rise to upper-layer neurons (L2-4). The Cux2-expressing RGCs proliferate early in development when lower-layer neurons (L5-6) are generated, and only start to produce neurons at later stages (Franco et al., 2012). This is at odds with the prevailing model of neocortical neurogenesis through progressive fate restriction (Desai and McConnell, 2000; Frantz and McConnell, 1996; Luskin et al., 1988; McConnell and Kaczynski, 1991; Price and Thrulow, 1988; Shen et al., 2006; Tan et al., 1998; Walsh and Cepko, 1988); additional studies, such as systematic and precise clonal/lineage analysis of individual progenitor cells (e.g., Cux2 expressing versus non-Cux2 expressing), are required to resolve this discrepancy and to better understand the progenitor heterogeneity with regards to the generation of diverse neuron types in the neocortex.

In the mature neocortex, L1 mainly contains distal tufts of pyramidal cell apical dendrites, axon terminations, Cajal-Retzius cells and some GABAergic cells, but lacks excitatory neurons. L2/3 predominantly consists of callosal projection neurons, which project their axon collaterals across the corpus callosum and mediate the communication between the two cerebral hemispheres, in addition to participating in local circuits. In primary sensory areas, L4 contains spiny stellate cells, which form an important population for thalamic innervation. L5/6 contains largely corticofugal projection neurons that provide major cortical output to the thalamus, midbrain, spinal cord and other brain regions, as well as a small population of callosal projection neurons (Molyneaux et al., 2007; Fame et al., 2011). Importantly, the corticofugal projection neurons in L5 are morphologically and physiologically heterogeneous, depending on their long-range projection targets (Hatton and Nelson, 2007). In fact, the projection identity of L5 neurons is regulated by a network of transcription factors (Srinivasan et al., 2012). L6 neurons are also diverse, with at least two broad categories based on their morphology and physiological properties (Thomson and Lamy, 2007) (Fig. 1C, right). Interneurons are present in all layers and mainly contribute to local circuitry (Markram et al., 2004).

In 1989, in trying to understand the rules that govern the synaptic connections between different neuronal types in different layers of the neocortex, Douglas and Martin developed the model of a ‘canonical cortical microcircuit’ based on electrophysiological and modeling studies in the cat visual cortex. Their model contains three groups of neurons: superficial pyramidal cells, deep pyramidal cells and a common pool of inhibitory cells. All three groups are interconnected, while thalamic input mainly targets superficial pyramidal cells and inhibitory cells. These connections allow the circuit to amplify transient thalamic input while maintaining the balance of excitation and inhibition (Douglas and Martin, 1991; Douglas et al., 1989). Subsequent studies demonstrated that the most frequently connected cells were located in the same layer, whereas interlaminar connections are dominated by feedforward connections from L4 to L3 and from L3 to L5 (Douglas et al., 2002; reviewed by Bastos et al., 2012). Based on substantial studies, we now know that in the canonical microcircuit of the neocortex, thalamic relay cells provide input into the cortex and mainly target L4, although they also form synapses with neurons in other layers. This input is relatively weak, and is amplified by recurrent excitation of L4 excitatory neurons. Recurrent excitation can be potentially detrimental in leading to hyper-excitation of the circuit; inhibition is therefore required to modulate this excitation. Within all layers, excitatory and inhibitory neurons form recurrent connections. Between cortical layers, information flow has a strong directional tendency: from L4 up to L2/3 and then down to L5/6. There is also a weaker projection from L4 directly down to L5/6. The principles of the canonical microcircuit can be applied to other cortical areas, such as the motor cortex, suggesting that they may reflect the underlying organization of the entire neocortex (Douglas and Martin, 2007). This model has greatly advanced our understanding of the wiring principle for cortical circuits. Nonetheless, recent studies also suggest that there may be some variations in circuit organization in certain cortical areas, e.g., the rodent somatosensory cortex (Bruno and Sakmann, 2006; Meyer et al., 2010; Oberlaender et al., 2012; Wimmer et al., 2010). In summary, substantial work has demonstrated a precise orchestration of neuronal production and migration leads to the formation of distinct cortical lamina, each of which contains unique populations of neurons poised for the assembly of highly organized circuits.

The neocortical column

The second fundamental feature of the neocortex is the functional column. The concept of the ‘neocortical functional column’ was first introduced by Mountcastle in 1957 and has proven invaluable in understanding the functional organization of the neocortex. When recording the somatosensory cortex of cats and monkeys, Mountcastle and colleagues discovered that neurons sharing common functional properties, including corresponding peripheral receptors, receptive fields and firing latencies, were located in a radial column extending from pial surface to white matter (Mountcastle, 1957; Powell and Mountcastle, 1959). The diameter
of columns is of approximately same size in both cats and monkeys. The functional properties of neurons are similar within a column, but significantly differ between adjacent columns (Mountcastle, 1997).

Seminal work by Hubel and Wiesel in the 1960s and 1970s then triggered tremendous interest in studying the neocortical column. Echoing Mountcastle’s observation in the somatosensory cortex, they found that, in the visual cortex of cats, neurons with a similar orientation selectivity were located in a single radial penetration from pia to white matter (termed ‘orientation columns’) (Hubel and Wiesel, 1962; Hubel and Wiesel, 1963). Later, they found that the two eyes differentially activated cortical neurons: neurons with similar eye preference were also grouped into columns (termed ‘ocular dominance columns’), and left and right eye-dominated columns alternated across the cortex (Wiesel and Hubel, 1963). The relationship between orientation columns and ocular dominance columns was summarized in their classic ice-cube diagram, in which thin orientation slabs cut the coarser ocular dominance columns at right angles (Hubel and Wiesel, 1977).

Similar functional columns were also discovered in the cat primary auditory cortex (Abeles and Goldstein, 1970) and in many other cortical areas (Mountcastle, 1997). These observations prompted a deep thought that ‘the cells behave as though they shared certain connections among themselves, but not with cells of neighboring columns, and in this sense a single group of cells is looked upon as a more or less autonomous functional unit’ (Hubel and Wiesel, 1974). Importantly, however, columns are not isolated; in fact, extensive studies have demonstrated horizontal connections between columns, especially between those with similar functional properties (Bosking et al., 1997; Gilbert and Wiesel, 1983; Gilbert and Wiesel, 1989; Katz et al., 1989; McGuire et al., 1991; Ts’o et al., 1986).

Despite the long history of successful identification of neocortical columns with electrophysiological recordings, especially in mammals such as cats and monkeys, the anatomical basis of these columns has remained elusive. Minicolumns have been proposed to be the basic unit of the neocortex, which are composed of chains of neurons (typically ~80-120 in primates) spanning cortical layers whose cell bodies are vertically aligned within a diameter of ~40-50 μm; about ~50-80 minicolumns link together to form the structural basis of functional columns (Mountcastle, 2003). Another candidate is a related structure referred to as ‘bundles’, which mainly comprise closely associated apical dendrites of pyramidal cells whose cell bodies are located in different layers (Peters and Kara, 1987; Peters and Setthares, 1996; Peters and Walsh, 1972; Peters et al., 1997). However, both views have met ample criticism (Rockland and Ichinohe, 2004; da Costa and Martin, 2010). Whether there is a structural correlate of the functional columns remains controversial. One obvious challenge is that functional columns are defined based on the functional properties of neurons, which may not be simply reflected anatomically. A more effective search for the structural correlate of functional columns requires a precise characterization of the functional properties of individual neurons. Recent advances in the field of in vivo Ca²⁺ imaging provide a powerful route to reveal the functional properties of large ensembles of neurons (Ohki et al., 2005; Bock et al., 2011; Chen et al., 2011; Ko et al., 2011), effectively bridging the gap between structure and function (Li et al., 2012).

**Lineage-dependent circuit assembly of neocortical excitatory neurons**

In the rodent visual cortex, the equivalent of the functional columns observed in higher mammals has proven difficult to find. This may be largely attributed to the fact that, in the horizontal dimension (i.e. within the same cortical layers), neurons with different orientation preferences are intermingled spatially in a ‘salt-and-pepper’ fashion (Ohki and Reid, 2007; Ohki et al., 2005). Similar fine-scale heterogeneity in the functional properties of neurons has been observed in the somatosensory and auditory cortices (Rothschild et al., 2010; Sato et al., 2007). These findings imply that if functional columns exist in rodents, they may be built at a single-cell resolution. Interestingly, by combining in vivo two-photon Ca²⁺ imaging and ex vivo patch-clamp recording techniques, Ko et al. recently demonstrated that, in L2/3 of mouse visual cortex, neurons that share orientation preference response properties are more likely to be synaptically connected, highlighting the presence of fine-scale subnetworks dedicated to processing related sensory information (Ko et al., 2011). A remaining mystery is how these highly connected subnetworks are constructed. One possibility is that neurons with similar functional properties fire at the same stimuli repeatedly and therefore gradually develop strong specific connections, as suggested by the Hebbian learning rule (see Glossary, Box 1). Alternatively, it is possible that there are other rules that govern their connectivity even before their functional properties fully emerge.

How do functional columns emerge in the developing neocortex? In 1988, Rakic proposed the ‘radial unit hypothesis’ (Rakic, 1988). According to this hypothesis, neurons derived from the same proliferative unit in the VZ migrate along the radial glial fiber(s) and form ‘ontogenetic/embryonic’ columns, which are the building blocks for the cerebral cortex. Based on the similarity of a vertical organization of neurons, it was postulated that ontogenetic columns might relate to functional columns. However, this hypothesis has only recently been tested experimentally. Yu et al. injected EGFP-expressing retroviruses intraventricularly into developing mouse embryos at embryonic days E12-E13 to label individual asymmetrically dividing RGCs, which give rise to ontogenetic columns composed of four to six vertically aligned sister excitatory neurons spanning different cortical layers. Multiple-electrode whole-cell patch clamp recordings at postnatal stages (P10-P21) revealed that sister neurons are preferentially connected by chemical synapses when compared with nearby non-sister neurons. Interestingly, the direction of interlaminar connectivity among sister neurons in an ontogenetic column resembles that observed in the mature cortex, suggesting that these ontogenetic columns could lead to the formation of functional columns in the cortex (Yu et al., 2009). Tracing this back to even earlier in development, sister neurons preferentially form transient electrical synapses with each other (peaking at ~P1-P2, largely disappearing after P6), which allow for selective electrical communication and promote action potential generation/synchronous firing between sister neurons (Yu et al., 2012). Although these gap junctions largely disappear before functional chemical synapses can be detected, they are necessary for the formation of specific chemical synapses between sister neurons. Blockade of electrical communication impaired the subsequent assembly of lineagelated sister excitatory neurons into specific microcircuits (Yu et al., 2012). This line of studies not only demonstrates a new principle of circuit assembly that depends on the lineage relationship of neurons (i.e. on their specific developmental history), but also suggests that ontogenetic columns may contribute to the emergence of the functional columns in the neocortex (Fig. 2A).

To test this directly, Li et al. used the same retrovirus-labeling technique to label sister excitatory neurons derived from the same
RGCs at E15-E17 in the mouse visual cortex, and performed in vivo two-photon Ca²⁺ imaging to assess their orientation tuning response properties at P12-P17 (Li et al., 2012). They found that sister neurons have similar orientation preferences compared with those of nearby non-sister neurons (Fig. 2B). Interestingly, in line with the findings of Yu et al., blockade of electrical coupling between sister neurons abolished the functional similarity between sister neurons, highlighting the significance of early gap junction-mediated electrical communication in establishing specific connections between sister neurons and their shared functional properties (Li et al., 2012).

How far does the lineage relationship go in shaping neocortical circuitry? In another recent study, Ohtsuki et al. used a different approach to label lineage-related neurons (Ohtsuki et al., 2012). They used a transgenic mouse line in which Cre recombinase is expressed sparsely in progenitor cells early in forebrain development, generating individual clones containing 600-800 fluorescently labeled neurons derived from the same progenitors. They then used in vivo two-photon Ca²⁺ imaging to examine the orientation tuning response properties of clonally related neurons and nearby non-clonally related neurons. Interestingly, even in such a large population of neurons, the lineage relationship of which is much further away compared with that of the sister neurons labeled by Li et al., orientation preferences among clonally related neurons were still more similar than those among unrelated neurons (Fig. 2B). However, there was considerable diversity within the large clones, such that nearly one half of all neuronal pairs had preferred orientations with a difference greater than 30°, and one quarter of them exhibited a difference greater than 60° (Ohtsuki et al., 2012). One plausible explanation is that remote lineage relationship, although still influential, is not as strong as close lineage relationship (i.e. neurons derived from asymmetrically dividing RGCs) in predicting shared functional similarities among neurons. However, there might be other factors that contribute to the observed diversity. Ohtsuki et al. performed the experiments in relatively mature (P49-P62) mice whose visual system was well developed, whereas Li et al. conducted the experiments in young mice (P12-P17), close to the time of eye opening. As it is well established that neuronal activity as well as visual experience exert tremendous influence...
on circuit development (Cang et al., 2005; Caporale and Dan, 2008; Hubel and Wiesel, 1965; Katz and Crowley, 2002; Katz and Shatz, 1996), it is possible that lineage relationship instructs the formation of the initial neocortical circuit, which is then further modified by experience. In this regard, it will be interesting to test whether remote lineage-related neurons behave more similarly when tested in younger animals or, vice versa, whether the shared functional similarities between closely related ‘sister neurons’ persist into adulthood.

Together, these studies strongly suggest that, in the mouse, lineage plays a crucial role in organizing neocortical excitatory neuron circuits. Ontogenetic columns formed by clonally related neurons could be the basic structural and functional unit that constitutes the neocortex. One important issue is whether these lineage-related specific circuits also exist in other mammalian species, especially higher mammals such as cats and monkeys. The tremendous interspecies difference in the organization of orientation preference maps from rodents to higher mammals could be due to the differences in patterns of neurogenesis and the layout of ontogenetic columns (Fig. 3). It has been proposed that the extensive proliferation capacity of progenitor cells in the SVZ underlies cortical expansion from rodents to higher mammals, including ferrets, cats and primates (Kriegstein et al., 2006), which presumably would give rise to ontogenetic columns that are much larger in size and with many more horizontal features. Recent discoveries of OSVZ progenitors with increased amplification capability support this hypothesis (Lui et al., 2011). Interestingly, computational modeling based on the ‘wire length minimization principle’ predicts that a strong horizontal connection pattern would lead to smooth varying maps, such as those discovered in cats and monkeys. By comparison, the proliferation potential of IPCs in the SVZ of rodents is much more limited, and lack of specific horizontal connections is predicted to produce an apparent salt-and-pepper organization of maps (Koulakov and Chklovskii, 2001). It will be interesting to test whether ontogenetic columns are the long-awaited structural basis of functional columns in higher mammals. In addition, two recent studies demonstrated that neurons with certain molecular identities are arranged in a periodic manner in L5 of mouse and human neocortex (Kwan et al., 2012; Maruoka et al., 2011). It will be interesting to understand the cellular events responsible for the formation of these ‘molecular minicolumns’ and how they relate to functional columns. It is also important to note that the cellular organization of the thalamus that relays information to the neocortex appears different between rodents and higher mammals; for example, the lateral geniculate complex is distinctly laminated in cats, monkeys and humans, but not in mice and rats, and this difference may also fundamentally influence the functional organization of the cortex.

### Lineage-related production and organization of inhibitory neurons

Neocortical inhibitory neurons exhibit an incredibly rich diversity of subtypes and are distributed throughout the neocortex in a stereotypical manner. Extensive studies have suggested that, as for excitatory neurons, lineage and/or the developmental history (i.e. place and time of birth) of cortical interneurons underlies their subtype specification and distribution in the mature neocortex, thereby contributing to their proper circuit assembly and function. It is important to note that, unlike excitatory neurons, very little is known about the lineage of interneurons at the single progenitor level, and most previous work has focused on interneuron lineage at the level of populations of progenitors. Nonetheless, the advent of optogenetic tools, in combination with mouse genetics, allows researchers to dissect the functional output of different classes of interneurons within cortical circuits in vivo (Cardin et al., 2009; Lee et al., 2012; Sohal et al., 2009; Wilson et al., 2012), and offers an unprecedented opportunity to understand how early developmental events and distinct lineages of interneurons contribute to the assembly of precise circuits.

### Cortical inhibitory neuron development

Unlike dorsally derived excitatory neurons, GABAergic inhibitory neurons are born in transient ventral telencephalic regions known as the ganglionic eminences (GEs) (Anderson et al., 1997; Anderson et al., 2002; Butt et al., 2005; Gelman and Marin, 2010; Valcanis and Tan, 2003; Wichterle et al., 1999; Wichterle et al., 2001; Xu et al., 2004; Xu et al., 2006; Xu et al., 2008) (Fig. 1B). The GE is further subdivided into three distinct domains, namely the lateral, medial and caudal GEs (LGE, MGE and CGE, respectively). Numerous fate-mapping studies have demonstrated that the MGE and the CGE are the predominant sources of cortical interneurons (Fogarty et al., 2007; Nery et al., 2002; Xu et al., 2008). More specifically, MGE progenitor cells produce ~70% of...
neocortical interneurons (Fogarty et al., 2007; Lavdas et al., 1999; Xu et al., 2008) and CGE progenitor cells give rise to the remaining 30% (Butt et al., 2005; Miyoshi et al., 2010; Nery et al., 2002). In addition, a small subpopulation (fewer than 3%) of cortical interneurons is derived from the embryonic preoptic area, which is situated in the telencephalic stalk (Gelman et al., 2009). MGE progenitors also give rise to interneurons of the striatum (see Glossary, Box 1) and are characterized by the expression of a transcription factor, the homeobox protein Nkx-2.1 (Marin et al., 2000; Sussel et al., 1999). Notably, it has been suggested that, in humans as well as non-human primates, besides the GE of the ventral telencephalon, the VZ and/or SVZ of the dorsal telencephalon also produce a substantial population of cortical GABAergic neurons (Furtuzinhos et al., 2009; Jakovcevski et al., 2011; Letinic et al., 2002; Petanjek et al., 2009). However, additional studies are needed to explore this further (Hansen et al., 2010).

In rodents, interneurons destined for the cortex must undergo a long and tortuous journey from their subpallial origins to reach the cortex. In order to avoid the developing striatum, interneurons migrate superficially or deep relative to the striatal mantle (Marin et al., 2001). Two main migratory streams of interneurons follow tangentially oriented paths to enter the cortex: a superficially migrating early cohort migrates through the MZ of the cortex (Lavdas et al., 1999); a deeply migrating second and more prominent cohort migrates predominantly through the lower intermediate zone and SVZ (Wichterle et al., 2001). Upon reaching the cortex, interneurons adopt a radial trajectory to settle into their final laminar position within the CP (Ang et al., 2003; Havern et al., 2004; Polleux et al., 2002; Tanaka et al., 2003) (Fig. 1B).

Lineage and subtype specification
One of the most striking features of interneurons in the adult neocortex is the incredibly rich diversity they display in their morphology, biochemical marker expression, electrophysiological properties and synaptic connectivity patterns. Axons of inhibitory neurons are highly selective in the postsynaptic neuronal compartment (i.e. soma, dendritic tree or axon initial segment) that they target. The distinct subcellular targeting and firing patterns of each subtype allows populations of interneurons to exert their inhibitory influence on surrounding neurons in numerous ways, thereby shaping circuit output dynamically and allowing for a wide range of neuronal computations. For example, perisomatic inhibition controls the output of the postsynaptic neuron, and is primarily mediated by parvalbumin (PV)-containing basket cells. Dendritic inhibition, by contrast, sculpts the local input of the postsynaptic neuron, and is mainly mediated by somatostatin (SST)-expressing interneurons, which are mostly Martinotti cells (McGarry et al., 2010). PV-positive chandelier (also referred to as axo-axonic) cells exclusively target the axon initial segment of pyramidal cells and play an enigmatic role in the cortical circuit as negative biopolar cells that display a fast-adapting firing pattern (Lee et al., 2010; Miyoshi et al., 2010; Nery et al., 2002). Within the MGE, there seems to be additional spatial bias in the specification of SST-positive and PV-positive interneurons (Flames et al., 2007; Fogarty et al., 2007; Wonders et al., 2008). A comprehensive analysis of expression patterns of several transcription factors in the VZ of the developing mouse GE by Flames et al. revealed that expression of Nkx-6.2 in the dorsal MGE underlies specification of SST-positive interneurons, whereas PV-positive interneurons are derived from Nkx-2.1-expressing progenitors located more ventrally within the MGE, suggesting that anatomically defined subpallial regions can be further divided into subdomains that give rise to functionally distinct interneuron subtypes (Flames et al., 2007) and that lineage plays an important role in generating diverse interneuron populations that are characteristic of the mature neocortex.

In addition to the presence of spatially distinct progenitor domains, a temporal bias in neurogenesis is thought to contribute to interneuron subtype specification. Fate mapping of MGE-derived interneurons showed that these progenitors undergo temporal changes in fate such that they progress from generating mainly SST-expressing neurons to making mainly PV-positive interneurons (Miyoshi et al., 2007). Recent transplantation and lineage-tracing experiments elegantly demonstrated that chandelier cells in the mouse neocortex are selectively born in the MGE at late stages of embryonic development (Inan et al., 2012; Taniguchi et al., 2013).

Although the spatiotemporal dynamics of neurogenesis evidently contribute to diversification of neocortical interneurons, it remains unclear whether interneuron subtype specification occurs at the population or single progenitor level. Using mouse genetics in combination with in utero retroviral labeling, Brown et al. were the first to conduct a clonal analysis of the MGE at the single progenitor level to show that individual RGCs within the MGE are able to generate clones of cortical interneurons that share the same neurochemical markers, as well as clones that contain interneurons expressing different neurochemical markers (Brown et al., 2011). More extensive characterization of interneuron subtypes within clonal clusters using morphological and physiological analysis will help elucidate how many subtypes a single progenitor can generate and in what combinations, as well as the early developmental principles that generate interneuron diversity.

Lineage and spatial distribution
Similar to excitatory neurons, interneurons are distributed throughout the cortex in a laminar fashion. Classic birthdating studies and transplantation experiments have demonstrated that interneurons born at different times in the MGE populate specific layers of the neocortex in an inside-out order (Nery et al., 2002; Valcanis and Tan, 2003). This is also apparent in the case of CGE-derived interneurons that are born relatively late during embryonic neurogenesis and tend to occupy more superficial layers of the cortex in comparison with most MGE-derived interneurons (Miyoshi et al., 2010). Interestingly, cortical interneurons and projection neurons born at roughly the same time tend to occupy
the same cortical layer (Valcanis and Tan, 2003). In attempting to understand mechanisms that direct interneurons to position themselves precisely within specific cortical layers, Lodato et al. replaced subcerebral projection neurons with collosal projection neurons and found that this led to abnormal lamination of interneurons. In addition, artificial introduction of corticofugal or collosal projection neurons below the cortex was sufficient to recruit cortical interneurons to these ectopic locations. This study demonstrated that different populations of projection neurons can influence the laminar fate of interneurons (Lodato et al., 2011). Therefore, although the temporal dynamics of neurogenesis correlate with the laminar fate of both projection neurons and interneurons, there may be additional rules that govern their positioning within the neocortex.

Interestingly, Brown et al. showed that neocortical interneurons are produced as spatially organized clonal units in the MGE; in the adult neocortex, these clonally related interneurons are organized into spatially isolated clusters (Brown et al., 2011). Lineage, or clonal relationship, therefore, plays a pivotal role in the production and spatial organization of neocortical interneurons. So how far do lineage/clonal relationships go in establishing functional interneuron microcircuitry? Although some interneurons form and receive non-specific synaptic connections (Fino and Yuste, 2011; Hofer et al., 2011; Packer and Yuste, 2011), several studies have shown that inhibitory interneurons in the neocortex exhibit highly specific synaptic connections within functional circuits (Jiang et al., 2013; Otsuka and Kawaguchi, 2009; Thomson and Lamy, 2007; Yoshimura and Callaway, 2005). Moreover, the synaptic connectivity between local interneurons and excitatory neurons also shows a stereotypic spatial pattern across different regions of the neocortex (Kätzel et al., 2011), suggesting a high degree of specificity in the functional organization of neocortical interneurons. Given the ventral origin and long tangential migration of neocortical interneurons, how stereotypic inhibitory circuits form in the neocortex is an unresolved issue. The spatial organization of clonally related sister inhibitory interneurons raises an intriguing possibility of a lineage-dependent functional organization (electrical- and/or chemical synapse-based) of interneurons that may contribute to specific inhibitory circuits in the mammalian neocortex.

Conclusions
Here, we have reviewed recent findings on how early developmental processes, such as neurogenesis and neuronal migration, instruct circuit assembly for both excitatory and inhibitory neurons in the neocortex. One emerging theme is that, lineage, or the developmental history of a neuron, strongly influences its connectivity. For excitatory neurons, sister neurons derived from the same progenitors preferentially form synaptic connections with each other and process related sensory information. In the case of inhibitory interneurons, lineage appears to play a crucial role in their production and organization. With the recent surge of tools that enable labeling and manipulation of circuit components, we are optimistic that many new insights will be revealed along this line in the near future. For example, does lineage-related circuit assembly only apply to the neocortex, or does it also play a role in circuit formation in other brain regions? How is lineage-related circuitry regulated by experience? How does this basic structural and functional unit evolve from mouse to human? Is it affected in neurological diseases? Answers to these questions would greatly advance our knowledge about the fundamental principles that nature uses to construct functional brain circuits.

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