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Axon Guidance Events in the Wiring of the Mammalian Olfactory System

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Abstract The detection of odorant signals from the environment and the generation of appropriate behavioral outputs in response to these signals rely on the olfactory system. Olfactory sensory neurons (OSNs) of the olfactory epithelium are located in the nasal cavity and project axons that synapse onto dendrites of second-order neurons in the olfactory bulb (OB) that in turn relay the information gathered to higher order regions of the brain. The connections formed are remarkably accurate such that axons of OSNs expressing the same olfactory receptor innervate specific glomeruli within the complex three-dimensional structure that represents the OB. The molecular determinants that control this complex process are beginning to be identified. In this review, we discuss the role of various families of axon guidance cues and of recently characterized families of adhesion molecules in the formation of stereotypic connections in the olfactory system of mice.

Keywords Olfactory system · Olfactory receptor · Axon guidance · Cell adhesion molecules.

Introduction

In the mouse, olfactory information is relayed to the central nervous system through axons of olfactory sensory neurons (OSNs) that make connections with second-order neurons within the olfactory bulb (OB) in spherical neuropil structures termed glomeruli. Each OSN expresses one of over 1,000 functional olfactory receptors (ORs) that can bind a multitude of odorant molecules [1–3]. In order to ensure an accurate representation of the activation state of such a large number of ORs, OSNs expressing a single OR project their axons to two symmetrically bilateral glomeruli out of an estimated 1,800 glomeruli in the olfactory bulb [4–6]. Since OSNs expressing a specific receptor are randomly dispersed across one of four partially overlapping regions of the olfactory epithelium (OE), rather than strictly limited to, axonal convergence of axons expressing the same OR must take place at the level of the OB. How do OSN axons select their target glomeruli in such a complex three-dimensional target field? The target choice of OSN axons appears to rely on a combination of molecular determinants that first promote segregation of axons into broad regions of the OB to form a crude topography and then favor their sorting and convergence into specific glomeruli. While some families of non-classical axonal guidance cues, such as Wnts, have been implicated in promoting the growth of OSN axons towards the OB, we will restrict our discussion to the molecules that regulate glomerular targeting of OSN axons in the OB [7–10]. Furthermore, the role of transcription factors in the development of olfactory projections will not be covered in this review.

Axonal Projections in the Dorso-ventral Axis of the OB

OSN axons projecting along the surface of the OB must accurately innervate glomeruli that are positioned at specific coordinates in the dorso-ventral, antero-posterior, and medio-lateral axes of the OB. There is good evidence

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supporting a direct spatial relationship between the location of an OSN cell body inside the OE and the dorso-ventral position of the glomerulus it innervates in the OB.

Early in situ hybridization experiments suggested that each OR gene is expressed in a limited number of OSNs that are located in one of four defined regions of the OE termed zones [5, 6, 11, 12]. These zones were proposed to be spatially restricted and to span the olfactory epithelium from the dorso-medial (zone I) to the ventro-lateral (zone IV) region of the OE (Fig. 1b) [13]. More recent in situ hybridization experiments suggest that OR gene expression is continuous and overlapping in certain regions across the dorso-medial to ventro-lateral axis of the OE [14–16]. Dye-tracing experiments revealed a correlation between the location of OSNs in the OE and their projections in the dorso-ventral axis of the OB [15, 17, 18]. The concept of zone-to-zone axonal projections is also supported by immunohistochemistry and in situ hybridization experiments demonstrating that OSNs located in the dorso-medial region of the OE project their axons to the dorsal aspect of the OB [19, 20]. Hence, the correlation observed between glomerular positioning in the dorso-ventral axis of the OB and the location of OSN cell bodies in the dorso-medial to ventro-lateral axis of the OE suggest that spatial information may be provided through the differential expression of molecules that regulate the growth and targeting of axons.

The concept of zone-to-zone axonal projections raises the possibility that OSNs located in the four regions of the OE may express different molecular determinants that allow their axons to segregate in the dorso-ventral axis of the OB. However, recent in situ hybridization experiments suggest that OR gene expression is continuous and overlapping across the dorso-medial to ventro-lateral axis of the OE rather than strictly restricted to four specific zones [15, 19, 20]. This observation raises the possibility that graded expression of a receptor across the dorso-medial to ventro-lateral axis of the OE may direct the dorso-ventral targeting of axons in the OB. To examine this possibility, we evaluated the pattern of expression of various axon guidance receptors in the OE and observed that Robo-2, a receptor for Slit chemorepellents, is expressed in a high dorso-medial to low ventro-lateral gradient in the OE throughout development and in the adult. Interestingly, subsets of axons that normally project to the dorsal region of the OB are misrouted to the ventral part of the OB in Robo-2 mutant mice [21]. Furthermore, the chemorepellent Slit-1 is expressed in the ventral region of the OB and ablation of its expression leads to axonal targeting defects similar to the ones observed in Robo-2 mutant mice. These results suggest that Slit-1–Robo-2 interactions prevent the entrance of dorsally targeting axons into the ventral region of the OB and therefore promote the dorso-ventral segregation of axons in the OB (Fig. 2a).

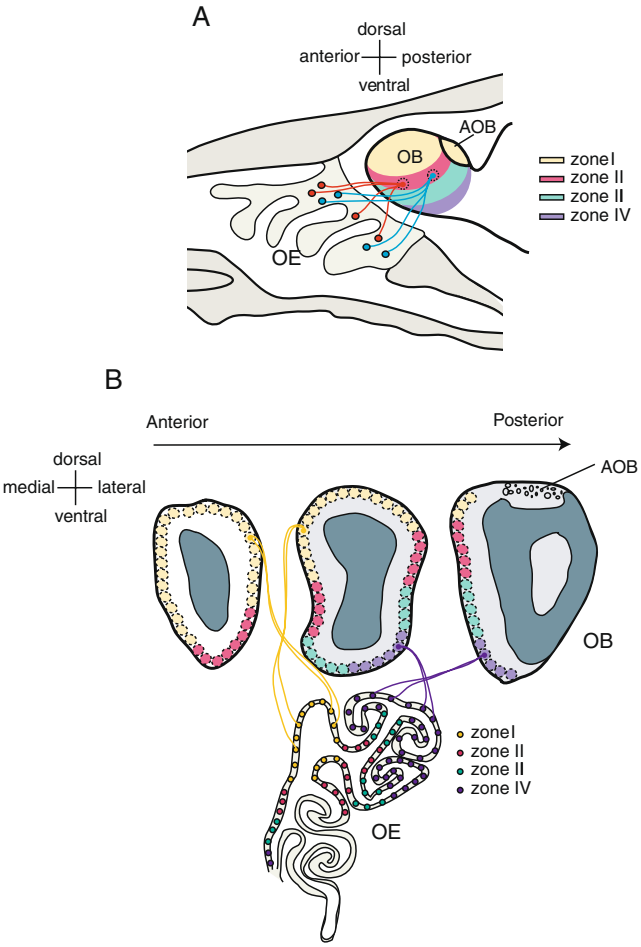

Fig. 1 Axonal projections in the main olfactory system. **a** OSNs (*closed red and blue circles*) located throughout the OE extend axons that synapse with dendrites of second-order neurons inside neuropil structures termed glomeruli (*dashed circles*) within the OB. Axons of OSNs expressing the same ORs (*red* or *blue*) converge to innervate specific glomeruli in the OB along the antero-posterior axis. The approximate positions of zones in the dorso-ventral axis of the OB are represented with different colors. *AOB* accessory olfactory bulb. **b** OSNs expressing the same OR are scattered within spatially defined but partially overlapping zones of the OE. Schematic representations of a coronal section through the OE and of sections through the OB at three different antero-posterior positions are shown. For simplicity, the zones are shown with distinct colors and overlap is not represented. OSNs located in the dorso-medial region of the OE (*yellow circles*) (zone I) innervate glomeruli (*dashed circle*) in the dorsal aspect of the OB while OSNs located in the ventro-lateral region of the OE (*purple circles*) (zone IV) project their axons to glomeruli in the ventral aspect of the OB. OSNs located in zones II (*red circles*) and zone III (*green circles*) project their axons to corresponding zones in the OB. The numbering of zones is according to the nomenclature in Sullivan et al. [13]. OSNs expressing the same OR (*yellow* or *purple*) innervate bisymmetrically located glomeruli on the medial and lateral sides of the OB at two different positions along the antero-posterior axis

It remains to be determined whether Robo-2 plays a role in the targeting of axons that project to other zones located in more ventral regions of the OB. Nonetheless, based on the observation that only a subset of dorsally projecting

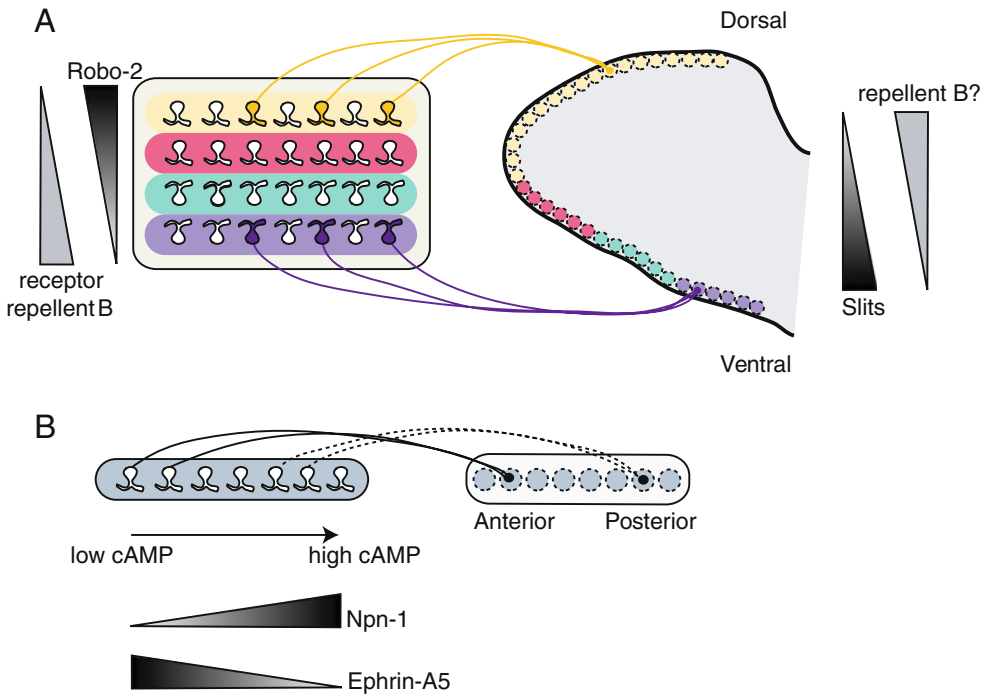

Fig. 2 Control of OSN axonal targeting by axon guidance cues. **a** Axonal targeting within the dorso-ventral axis of the OB is controlled by the graded expression of axon guidance cues and their receptors. The graded expression of the Slit receptor Robo-2 in a high dorso-medial (*yellow*) to low ventro-lateral (*purple*) gradient within the OE is required to segregate axons emanating from OSNs in the dorso-medial region of the OE to the dorsal aspect of the OB. Expression of Slit-1 in the ventral region of the OB prevents Robo-2-expressing axons from entering the ventral region of the OB. It is likely that other axon guidance receptors may be expressed in an opposite gradient to Robo-2 in the OE and serve to regulate the targeting of ventrally projecting axons in response to the graded expression of other repellents in the OB. **b** Axonal targeting in the antero-posterior axis of the OB is controlled through the differential expression of axon guidance molecules in OSNs. OR-mediated signaling modulates the levels of cAMP present in OSNs, which controls expression of specific axon guidance molecules such as Npn-1 and Ephrin-A5 through a transcriptional mechanism. OSNs containing high levels of cAMP express high levels of Npn-1 and low levels of Ephrin-A5 and as a result innervate glomeruli in more posterior regions of the OB within a specific zone. In contrast, OSNs containing low levels of cAMP express low levels of Npn-1 and high levels of Ephrin-A5 and project their axons to glomeruli located in the anterior part of the OB. The exact mechanism whereby differential expression of Npn-1 and Ephrin-A5 controls the anterior–posterior targeting of axons remains to be determined

axons are misrouted in Slit-1 and Robo-2 mutant animals, it is likely that other factors act in combination with Slit-mediated Robo repulsion to promote the dorso-ventral targeting of axons in the OB. For example, it remains possible that an attractive cue may be expressed in the dorsal aspect of the bulb that acts in combination with Slit-mediated repulsion to promote the growth of axons in the dorsal region of the OB. It is also likely that expression of a different repellent in a high dorsal to low ventral gradient in the OB may serve to segregate axons projecting from OSNs located in the ventral part of the OE to ventral regions in the OB. Interestingly, the secreted semaphorin receptor, Npn-2, is expressed in a high to low gradient in the ventral to dorsal axis of the OE and could therefore regulate targeting of axons to the ventral part of the OB [16]. Npn-2 and its ligand, Sema3F, have been implicated in the pruning of overshooting axons into the external plexiform layer but have yet to be shown to regulate the targeting of OSN axons to specific glomeruli [22, 23]. Hence, further identification of cues with graded expression in the dorso-ventral axis of the OB may reveal complementary mechanisms to promote the zonal segregation of axons in the OB.

Axonal Projections in the Medio-lateral Axis of the OB

In addition to targeting to the correct zone within the OB, OSN axons expressing the same OR innervate two symmetrically positioned glomeruli on either the medial or lateral side of the OB. Retrograde labeling experiments suggest that there is a loose correlation between the location of an OSN within each zone of the OE and the position of the glomerulus it innervates in the medio-lateral axis of the OB [24]. For example, OSNs projecting axons to a medial glomerulus are grouped within subregions of a specific OE zone. It is therefore possible that differential expression of axon guidance cues can also regulate the medio-lateral segregation of these axons.

The secreted chemorepellent Sema3A has been shown to direct the medio-lateral targeting of OSN axons. Sema3A is expressed in the nerve layer in the ventral aspect of the OB as well as in mitral cells of the OB [25, 26]. The Sema3A receptor, Npn-1, is expressed in subsets of OSNs and Npn-1-positive axons segregate to either the medial or lateral regions of the OB according to their targeting position along the antero-posterior axis. In Sema3A mutant mice, the medio-lateral segregation of Npn-1-positive axons is lost leading to changes in the formation of the olfactory map [26–28].

The insulin growth factors, IGF-1 and IGF-2, have also been implicated in the control of medio-lateral targeting in the OB [29]. Genetic ablation of IGFs or IGF-1R leads to a reduction in innervation of the lateral region of the OB. Furthermore, in IGF-1R mutant mice, the subset of axons expressing the P2 OR that normally target to a glomerulus located on the lateral side of the OB innervate a more ventro-medial region of the bulb. Interestingly, the subset of P2-expressing axons that innervate a glomerulus on the medial side of the bulb is unaffected in these mice suggesting that IGF signaling may only be required to guide laterally projecting axons. The exact mechanism through which IGFs control the targeting of these axons is not fully understood. Early in development, IGF-1 is expressed in a low medial to high lateral gradient at the anterior part of the bulb and IGF-1 can attract growing OSN axons in vitro. These observations suggest that IGF-1 could act as an attractant to promote the growth of axons into the lateral region of the bulb. However, while IGF-1R is expressed in OSNs, no gradient of expression that could explain how medially and laterally projecting axons respond differentially has been detected. Hence, the differential responsiveness of OSN axons to IGF may be modulated through yet unidentified mechanisms.

Axonal Projections in the Antero-posterior Axis of the OB

In contrast to dorso-ventral and medio-lateral targeting of OSN axons, the position of an innervated glomerulus in the antero-posterior axis of the OB does not appear to correlate with the position of OSN cell bodies within the OE. Changes in the levels of cAMP induced by signaling downstream of OR receptors have been proposed to control the expression of molecules that direct the targeting of OSN axons in the antero-posterior axis of the OB.

ORs are G-protein coupled receptors with seven transmembrane domains that transduce odorant-evoked signals into neuronal activity via activation of adenylyl cyclase type III leading to production of cAMP and opening of cyclic nucleotide gated channels [30]. ORs were proposed to play an instructive role in the targeting of OSN axons based on the observations that ORs are expressed on axon termini and that ablation of their expression leads to defects in OSN axonal convergence in the OB [31–33]. Furthermore, the genetic ablation of several important components in the odor-evoked signaling pathway does not grossly affect the projections of OSN axons to the OB. For example, mice homozygous for a null mutation in G_{olf} are anosmic but retain a normal map of sensory projections in the OB [34]. Mice bearing a targeted mutation in the alpha subunit of the olfactory cyclic nucleotide gated (CNGA2) ion channel show no defects in the pattern of convergence of olfactory axons for the majority of ORs examined [35, 36].

ORs were proposed to regulate axonal targeting by favoring the coalescence of axons expressing the same ORs [37, 38]. New insights into how ORs participate in the targeting of OSN axons were provided by experiments in which G-protein signaling and cAMP levels were altered in OSNs [39, 40]. Expression in OSNs of a mutated OR that does not interact with G proteins or of a non-functional tagged OR leads to a lack of axonal convergence. Interestingly, expression of a constitutively active G_s protein rescues these defects and can also promote axonal convergence, suggesting that G-protein signaling in OSNs is sufficient to direct growing axons in the OB [39, 40]. Partial rescue of the wiring defects was also observed by expressing a constitutively active form of PKA suggesting that the position of glomeruli in the antero–posterior axis of the OB may be regulated by the strength of cAMP/PKA signaling [35]. An important role for cAMP in axonal targeting was further supported by the observations that genetic ablation of adenylyl cyclase III perturbs the formation of glomeruli and the accurate convergence of OR-specific axons in the antero–posterior axis of the OB [41, 42].

How do changes in cAMP levels contribute to the targeting of axons in specific glomeruli within the OB? Different levels of cAMP/PKA signaling in OSNs may affect transcription and expression of axon guidance molecules at the surface of growing axons. RT-PCR analyses of single OSNs in which cAMP levels have been genetically manipulated revealed the differential expression of several genes including Npn-1 and PlexinA3, two well-characterized receptors for secreted semaphorins [39]. In addition, Npn-1 expression is reduced in adenylyl cyclase III mutant mice, further supporting the possibility that cAMP signaling controls expression of axon guidance receptors [41]. The expression of other classical axons guidance molecules is also regulated through OR-signaling dependent control of transcription. EphA5 and Ephrin-A5 are expressed in non-overlapping populations of OSNs in the OE [43, 44]. In OSNs lacking CNGA2, higher levels of Ephrin-A5 are observed while expression of EphA5 is

decreased [44]. The regulation of the levels of expression of EphrinAs on OSN axons plays an important role in the targeting of axons as ablation of expression of both Ephrin-A5 and Ephrin-A3 leads to a shift in innervated glomeruli towards the posterior aspect of the OB [43]. Hence, based on these studies, it is conceivable that OSNs expressing specific ORs could contain different levels of cAMP, which in turn affects the expression of different axon guidance molecules that regulate axonal targeting in the antero–posterior axis of the OB (Fig. 2b).

Compartmentalization of Axons in the Olfactory Bulb by Glycans

In addition to classical axon guidance molecules, glycans have been proposed to promote the gross compartmentalization of large populations of axons in the OB. In early studies characterizing immunoreactivity to monoclonal antibodies recognizing unique carbohydrate moieties, OSNs were found to express lactosamines [45, 46]. While low expression of lactosamines was observed throughout the OE, higher levels of lactosamines were detected in subsets of OSNs located in the dorso-medial regions of the OE [45–47]. The differential expression of lactosamine-containing glycans may therefore influence the segregation of subsets of axons in the OB.

The potential role of lactosamines in the patterning of the olfactory system was examined by disrupting their synthesis in mouse. β1,3-*N*-acetylglucosaminyltransferase I (β3GnTI) belongs to the β2GnT family that is responsible for the initiation and addition of *N*-acetylglucosamine to growing carbohydrate chains in many tissues [48, 49]. In β3GnTI-deficient mice, early projections of lactosamine-rich OSNs expressing P2 or I7 ORs were unable to properly innervate glomeruli in the OB and remained in the nerve layer adjacent to where they normally target [48]. In addition to these axon guidance defects, P2 and I7-expressing OSNs were progressively lost from the OE early in development, suggesting that lactosamines are also required for their survival. Interestingly, expression of lactosamines in a subset of axons may also be required to direct the growth of lactosamine-negative axons in the olfactory bulb. Indeed, ablation of β3GnTI expression leads to the formation of multiple glomeruli by axons expressing the OR M72, which do not normally express lactosamines [50].

How do lactosamines control the targeting of axons in the OB? It has been suggested that some members of the galectin family of carbohydrate binding proteins are involved in sorting carbohydrate-expressing axons as they grow towards the OB [51]. Galectin-1 is expressed in the ECM in the pathway taken by OSNs from the nasal cavity to the OB at embryonic ages when axons are projecting through this region [52, 53]. These ECM molecules are expressed in a highly restricted manner such that channel-like regions are formed at the boundaries between areas of high and low expression. Galectin-1 may therefore promote fasciculation of olfactory nerves by restricting the pathway of projection and by promoting axonal adhesion of lactosamine-expressing axons to the ECM [52, 54].

The observation that the targeting of lactosamine-negative populations of axons is also affected in β3GnTI raises the possibility that lactosamine-poor OSNs could express lactosamine-interacting molecules which help direct them to their appropriate positions in the OB. After loss of lactosamine expression, these OSN populations would lose crucial cell–cell interactions, potentially explaining why heterogeneous glomerular innervation occurs. Another intriguing possibility is that binding of lactosamines to lactosamine-interacting molecules modulates the responsiveness of axons to guidance cues expressed in the OB and ECM. The transmembrane protein EVA-1 that contains two predicted galactose-binding ectodomains was shown to modulate the responsiveness of axons to slit in *Caenorhabditis elegans* likely through an interaction with the Robo receptor homolog SAX-3 [55]. The direct binding of SLT-1, the slit *C. elegans* homolog, to EVA-1 may promote SAX-3 signaling. Human and mouse EVA-1 homologs have been identified but whether these proteins are expressed in the developing olfactory system remains to be established. Hence, it is possible that expression of lactosamines on subsets of OSN axons could modulate the response of these axons or of surrounding axons to Slits that are expressed in the OB.

Axon–Axon Interactions in the Formation of Glomeruli

The establishment of a coarse map is followed by the formation of specific glomeruli whereby axons of OSNs expressing the same OR converge into one to two glomeruli on each side of the OB. How do like axons sort into specific glomeruli? One possible mechanism of sorting would involve homotypic interactions between axons expressing the same receptor. While elegant genetic analyses have linked ORs to the establishment of axonal identity, several families of adhesion molecules have recently been implicated in the sorting of like axons to specific glomeruli in the OB. The identification of these molecules has led to a model whereby the sorting of axons may be regulated through the combinatorial expression of several families of cell adhesion molecules. Axons expressing the same set of cell adhesion molecules may fasciculate together and different strengths of interaction may be generated through combinatorial expression of cell adhesion molecules (Fig. 3a).

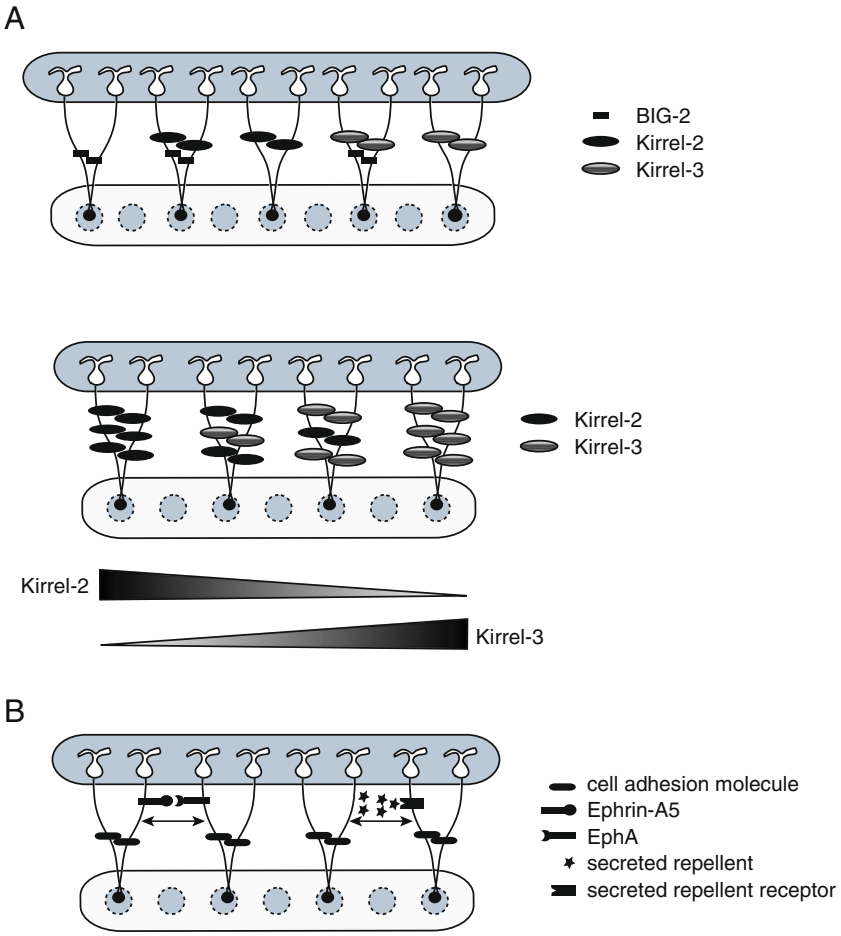

Fig. 3 Sorting of OSN axons to specific glomeruli through a combination of attractive and repulsive forces between axons. **a** OSNs expressing the same OR converge into specific glomeruli. The sorting of OR-like axons may be achieved through the combinatorial expression of cell adhesion molecules on axons. Kirrel-2, Kirrel-3, and BIG-2 are differentially expressed in OSNs. The strength of adhesive forces between axons may be directly related to the combination of receptors expressed. For example, while axons expressing BIG-2 alone fasciculate together, adhesive force between axons expressing both BIG-2 and Kirrel-2 are likely to be more robust (*top diagram*). Furthermore, the graded expression of Kirrel family members in OSNs may also contribute to the differential adhesive properties of their axons (*bottom diagram*). **b** In addition to adhesive forces that favor the fasciculation of like axons, repulsive forces between axons may promote the segregation of axons expressing different ORs into different bundles. Ephrin-A5 is expressed at various levels in different OSNs and may promote repulsion of axons expressing EphA5 receptors. It is also possible that chemorepellents secreted by OSN axons may promote the segregation of axons expressing different ORs within large bundles of axons

The Kirrel family of Ig domain containing molecules were first implicated in olfactory axonal pathfinding when some members were shown to be differentially expressed in OSNs using serial analysis of gene expression (SAGE) [44]. Kirrel-2 and -3 are expressed in subsets of OSNs in a roughly complementary fashion. OSNs expressing high levels of Kirrel-2 express low levels of Kirrel-3 and vice versa. Furthermore, both Kirrel-2 and -3 can undergo homophilic interactions but do not interact with each other. Glomeruli innervated by Kirrel-2- and Kirrel-3-positive axons are intermingled in the OB. Overexpression of Kirrel-2 in half of the OSNs expressing a specific OR leads to the splitting of axons into two adjacent glomeruli.

Another cell adhesion molecule reported to be involved in the convergence of axons in glomeruli of the OB is a member of the contactin subfamily of the Ig superfamily, BIG-2. BIG-2, or contactin-4, is a glycosylphosphatidylinositol anchored glycoprotein expressed in a subpopulation of OSNs and on axon termini [56]. Immunostaining analyses on the OB have revealed mosaic expression of BIG-2 on axon termini with some glomeruli expressing high or low levels of BIG-2 while others are BIG-2-negative. Furthermore, BIG-2 expression correlates with OR gene expression in OSNs. Ablation of BIG-2 expression in mice leads to the formation of numerous ectopic glomeruli suggesting that BIG-2 plays a role in the

establishment of the glomerular map. Hence the OR-correlated expression of cell adhesion molecules in different classes of OSNs may contribute to the sorting and convergence of like axons to specific glomeruli. In addition to Kirrels and BIG-2, the protocadherin-α (Pcdh-α) has been implicated in the accurate targeting of OSN axons [57]. Pcdh-α is highly expressed in OSNs and ablation of its expression leads to the disruption of convergence into specific glomeruli. In Pcdh-α null mice, OSN expressing the M71 and MOR23 ORs innervate multiple glomeruli. However, it is unclear how Pcdh-α contributes to the convergence of axons since it is expressed evenly among OSNs.

What mechanism regulates the expression of Kirrels and BIG-2 in different populations of OSNs? An important role for neuronal activity has emerged in the OR-correlated expression of Kirrels and Big-2. In mice with the CNGA2 mutation, random X-inactivation leads to the generation of two populations of OSNs in heterozygous female mice. A subset of OSNs retains expression of CNGA2 while others do not [58]. In these mice, CNGA-2-positive and CNGA-2-negative axons expressing the same OR segregate into two glomeruli in the OB. Immunohistochemical analyses of CNGA2-positive and negative glomeruli revealed lower levels of expression of Kirrel-2 and BIG-2 in CNGA2-negative glomeruli, suggesting that neural activity positively regulates their expression [44, 56]. In contrast, levels of Kirrel-3 expression were increased in CNGA2-negative glomeruli. Thus neuronal activity may regulate the expression of cell adhesion molecules and of other axon guidance molecules involved in controlling the fasciculation and targeting of OSNs. In keeping with this possibility, the levels of expression of Ephrin-A5 on subsets of olfactory axons is positively regulated by activity [44].

Is the combinatorial expression of cell adhesion molecules sufficient to sort OSN axons and promote their convergence into specific glomeruli? In addition to positive homophilic interactions between OSNs, it is likely that repulsive forces may also contribute to the segregation of axons expressing different ORs (Fig. 3b). Indeed, Ephrin-A5 and its receptor EphA5 are expressed in non-overlapping subsets of OSNs [44]. OSNs expressing high levels of EphA5 express low levels of Ephrin-A5 and vice versa. This observation raises the intriguing possibility that populations of axons expressing either Ephrin-A5 or EphA5 repel each other thereby promoting their segregation. In addition to contact-mediated repulsion, it is possible that controlled secretion of chemorepellents by subsets of axons could also contribute to this process. In combination with adhesive forces, these repulsive forces could serve to promote the segregation and sorting of axons. It remains to be determined whether Ephrin-A5–EphA5 interactions are required to segregate large populations of OSNs within bundles and whether other families of repulsive molecules also contribute to this process.

Refinement of the Sensory Map

Despite the multitude of mechanisms that have evolved to ensure the accurate targeting of axons in the OB, mistargeting of axons and formation of heterogeneous glomeruli innervated by axons expressing different ORs are observed in early post-natal animals. As the animal matures, these glomeruli are refined to form homogenous glomeruli innervated by axons expressing a single OR. This refinement process is dependent on olfactory sensory experience. Unilateral naris closure, which decreases both spontaneous and odorant-evoked activity, leads to the persistent presence of heterogeneous glomeruli in adult mice [59, 60]. The refinement of the map and the disappearance of heterogeneous glomeruli through activity-dependent mechanisms occur during a critical period that varies for different subsets of OSNs and that can be accelerated by odorant exposure [61, 62]. In addition to playing a role in the refinement of the sensory map, neuronal activity is important for the maintenance of OSN connections in the OB [58, 63].

Conclusions

The accurate wiring of the olfactory system relies on a stepwise process that includes the gross segregation of axons to specific regions of the OB, the convergence of axons into specific glomeruli, and the refinement of connections to establish the mature olfactory map. The recent progress made in our understanding of these processes has revealed that neuronal activity plays an important role not only in the refinement of the olfactory map but also in the guidance of axons and in their convergence into glomeruli. The activity-dependent control of expression of receptors at the surface of growing axons may be an efficient way to modulate the responsiveness of axons to adhesion and guidance molecules expressed on axons, glia, ECM, and within the OB. Considering the complexity of the glomerular map generated in the olfactory system, it is likely that other families of molecules capable of regulating OSN axon targeting will be identified in the years to come.

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