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Review

Emerging aspects of membrane traffic in neuronal dendrite growth

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Abstract

Polarized growth of the neuron would logically require some form of membrane traffic to the tip of the growth cone, regulated in conjunction with other trafficking processes that are common to both neuronal and non-neuronal cells. Unlike axons, dendrites are endowed with membranous organelles of the exocytic pathway extending from the cell soma, including both rough and smooth endoplasmic reticulum (ER) and the ER–Golgi intermediate compartment (ERGIC). Dendrites also have satellite Golgi-like cisternal stacks known as Golgi outposts that have no membranous connections with the somatic Golgi. Golgi outposts presumably serve both general and specific local trafficking needs, and could mediate membrane traffic required for polarized dendritic growth during neuronal differentiation. Recent findings suggest that dendritic growth, but apparently not axonal growth, relies very much on classical exocytic traffic, and is affected by defects in components of both the early and late secretory pathways. Within dendrites, localized processes of recycling endosome-based exocytosis regulate the growth of dendritic spines and postsynaptic compartments. Emerging membrane traffic processes and components that contribute specifically to dendritic growth are discussed.

Keywords: Dendrite; Golgi apparatus; Golgi outpost; Neuron; Sar 1; Syntaxin 16

1. Introduction

Neurons are highly specialized cells with elaborate junction connections with one another and with target organs. Polarized membrane traffic in neurons is necessary for both the establishment and the maintenance of the axonal and somato-dendritic plasma membrane domains [1–3]. Neuronal polarity is established during early development, as neural progenitor cells differentiate and generate processes that would become axons and dendrites. Axons typically function in neurotransmitter release in response to an action potential generated by an integration of dendritic input. Dendrites house, at their synaptic termini, receptors that serve to transduce chemical signals from several other neurons. It is clear that these plasma membrane domains must have a distinct set of proteins, and the composition of these proteins needs to be continuously maintained as such, as postmitotic neurons serve out the life span of the organism.

In general, polarized targeting to different neuronal surface domains can be achieved in several ways. Although neurons lack an equivalent of tight junctions, cytoskeletal-membrane protein complexes constituting the initial segment fence found at the axon hillock [4] could prevent free movement of membrane proteins to axonal domains. Selective, directed targeting of membrane cargoes in specific domain bound carriers has been observed for dendritic proteins [5,6]. On the other hand, some membrane cargo-ferrying carriers are not specifically targeted, and could find their way into the associated cytoplasmic region of both axons and dendrites. This is particularly the case for those utilizing plus-end-directed microtubule motors, which are present in both axon and dendrite [7]. For these, selective membrane docking/fusion is presumably required for eventual domain specific incorporation of cargo [8].

Polarized surface transport of membrane materials is required in the maintenance of adult neuron function, and is also acutely needed in polarized neurite growth during development and differentiation. How are these processes connected to, and regulated in conjunction with, the regular vesicular membrane traffic in neurons? Biosynthetic transport of membrane proteins and lipids along the exocytic pathway provides a continuous supply of membrane materials to the surface of all eukaryotic cells. Membrane proteins and secretory

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cargoes are first targeted to the endoplasmic reticulum, and then routed to the cell surface through multiple rounds of vesicle budding and fusion processes [9,10]. These processes are mediated by an evolutionarily conserved set of coat proteins that regulated vesicle budding [11] as well as Rab family GTPases, tethering complexes (such as the exocyst complex) and soluble *N*-ethylmaleimide sensitive factor (NSF) attachment protein receptors (SNARE) for vesicle docking and fusion [12]. Other than exocytosis, another way whereby membrane proteins could be added to the cell surface is through the recycling of internalized proteins via recycling endosomes [13,14]. Polarized membrane traffic in neurons could therefore be mediated by the same cellular machineries functioning along the paths and routes of exocytotic trafficking and/or endosomal recycling, but specifically regulated in some temporal and spatially unique manner [15]. There are some evidence for Rab8 [16,17], and the exocyst complex [18] being involved in exocytosis-mediated neurite extension. However, evidence for an absolute requirement for exocytic traffic in general and specific key components of the exocytic machinery, such as coat proteins and SNAREs (except TI-VAMP, which is more associated with lysosomal traffic in non-neuronal cells), are scarce. On the other hand, the requirement of for Rab11 [19] and syntaxin 13 [20]-mediated processes have implicated recycling endosome function in neurite outgrowth.

Neuronal dendrites are in most cases fairly elaborate structures with multiple branches. It is therefore conceivable that supply of nascent membrane proteins for its growth and maintenance from the cell body could be problematic. The problem may be partially alleviated by having mRNAs targeted [21] to local protein translational machinery of dendrites [22,23]. The Golgi apparatus is the penultimate membranous organelle within the cell soma where proteins destined for the secretory pathway traverse before reaching the plasma membrane. It is interesting to note that polarization in many eukaryotic cells is often associated with some morphological rearrangement of Golgi positioning, so as to facilitate directional targeting of membrane traffic to specialized plasma membrane domains [3]. However, is axonal and dendritic growth truly dependent on secretory traffic, and if so, how is membrane traffic from the cell body directed towards the growing neurites? Recent findings have revealed that growth of neuronal dendrites, unlike axons, is in fact very much dependent on the classical secretory pathway and its associated membranous compartments for its growth and function.

2. The dendrite is endowed with a localized exocytic apparatus

It has become clear over the past years that the neuronal dendrite is endowed with satellite endomembrane systems that modulate more localized membrane traffic. It was known earlier that local protein synthesis occurs in dendrites [24]. Interestingly, dendrites seemed to also possess the enzymes for N-linked glycosylation reactions that are usually associated with the Golgi apparatus [25]. Early immunocytochemical studies also suggest that membrane structures and markers of the rough and smooth endoplasmic reticulum (ER), ER–Golgi intermediate compartment (ERGIC) [26] and the Golgi apparatus could extend at least into the proximal part of major dendritic branches [27]. A large number of dendritic spines are known to contain a specialized smooth ER membrane extension known as the spine apparatus [28,29].

Ultrastructural studies by McCarthy et al. with immunogold labeling demonstrated that the ER translocon component Sec61α, ribosomes and lumenal ER proteins with the KDEL retention signal were all found associated with dendrites and dendritic spines [30]. There are also morphological evidence for the presence of ERGIC and Golgi/*trans*-Golgi network (TGN) markers within dendritic spines and distal dendrites [31]. Aridor et al. showed that ER export sites, marked by the small GTPase Sar1 and components of coat complex II (COPII), are assembled regularly throughout the dendritic tree [32]. *N*-methyl D-aspartate (NMDA) receptors were shown to be recruited to these ER export sites upon activation of metabotropic glutamate receptors, indicating that the formation of these sites could be regulated by neuronal activity [32].

With advances in live-imaging techniques, trafficking of fluorescent protein-labeled mobile transporters to both axons and dendrites could be visualized [8,33]. Horton and Ehlers [34] examined mobile carriers containing the antegrograde membrane cargo green fluorescent protein-labeled vesicular stomatitis virus G (GFP-VSVG) protein. Upon ER exit, these moves bidirectionally, and fused with both the somatic Golgi as well as some relatively static and long-live punctuated structures in the dendrite (but not the axon). The latter structures have Golgi-like membrane identities, as they are positively labeled for both the *cis*-Golgi marker GM130 [35] and the *trans*-Golgi marker galactosyltransferase [36]. Ehlers et al. further showed that these Golgi outposts are most prominently found in the longer and thicker apical dendrites. Golgi outposts appear to play a role in dendritic growth during morphological differentiation of hippocampal pyramidal neurons [37]. During dendritic growth, the Golgi apparatus becomes physically oriented towards the longest dendrite. This morphological change in Golgi polarity precedes the asymmetric dendrite elongation. Disruption of the Golgi apparatus by over-expressing GRASP65, a protein which facilitates Golgi cisternae stacking [38], abolished this asymmetry in Golgi orientation observed during dendritic growth, resulting in a marked reduction in dendritic polarity. Furthermore, a kinase-dead mutant of the TGN-localized protein kinase D (PKD-KD) [39], which blocks anterograde TGN-plasma membrane transport, had a specific and quick suppressive effect on dendritic over axonal outgrowth.

These important findings revealed that membrane traffic in growing dendrites are facilitated by discrete, satellite secretory apparatus distinct from that at the cell body. The existence of Golgi outposts poses several interesting questions, perhaps the most important of which pertains to their origin and function. Although the mode and mechanism of Golgi outpost biogenesis has remained largely unknown, several possibilities have been postulated [40]. The presence of ERGICs in dendrites [32,34] suggests that Golgi elements could be generated *de novo* by ER exocytosis at these sites remote from the cell body.

Alternatively, dendritic Golgi elements could arise from fragmentation and subsequent dispersion from the somatic Golgi, perhaps utilizing mechanisms of regulated phosphorylation that are related to mitotic Golgi breakdown. The dispersion process could be guided by microtubule-based transport [41], with reorganization of the Golgi stack at dendritic sites aided by local cytoskeletal modeling and interaction with dendritic ER/ERGIC membrane matrixes.

3. A TGN component for dendritic Golgi outposts

Another interesting question is whether Golgi outposts are complete versions of the somatic Golgi (albeit smaller), with a functionally polarized *cis*- and *trans*-face. A related question is whether dendritic Golgi outposts could function to receive both anterograde (i.e. from the local ER) as well as retrograde (i.e. from the plasma membrane and endosomes) traffic. Furthermore, it is unclear as to where cargo proteins transiting dendritic Golgi outpost would be subsequently directed to. This point is important because it is conceivable that Golgi outposts are not more than merely stripped-down versions of the somatic Golgi, evolved for specific anterograde delivery of proteins important for the function of the postsynaptic compartment.

To address these questions it would be important to confirm that dendritic Golgi outposts have an associated TGN. The TGN functions in both the exocytic and endocytic pathways, mediating the sorting of hydrolytic enzymes to lysosomes and anterograde targeting of other cargoes to the plasma membrane. The TGN also receives retrograde traffic from the late and recycling endosomes. In general, there are two retrograde transport routes from the endosome to the TGN, known from studies based on non-neuronal cells [42]. One major route entails transport from the late endosome to TGN. A prominent membrane cargo being transported through this route is the mannose 6-phosphate receptor [43]. The retrograde transport of proteins such as the B-subunit of Shiga toxin, furin and TGN38 occurs directly from the early or recycling endosome to the TGN [44,45]. This pathway is proposed to be important as it may allow signaling molecules such as CD19, interferon α/β receptors or GPI-anchored receptors such as CD14 to avoid degradation by late endosomes to reach sites where they can interact with their intracellular targets [45].

It is unclear if Golgi outposts have any retrograde transport function. For that matter, it was not particularly clear whether the Golgi outposts have a TGN. Early work has revealed TGN38 staining in membranous dendritic structures [31], but morphological tracking of the Golgi outposts were largely done using markers that are not specifically TGN-localized, such as the *cis*-Golgi marker GM130 [35]. We have recently found that the brain-enriched TGN SNARE syntaxin 16 (Syx16) [46,47] is specifically enriched in neuronal dendrites and found at Golgi outposts [48], thus confirming that Golgi outposts are endowed with a *trans*-Golgi network (TGN) component. Syx16 has a known role in the early/recycling endosome—TGN retrograde transport [45]. It is therefore conceivable that dendritic Golgi outposts may also receive cargo from dendritic plasma membranes or dendritic early/recycling endosomes.

4. Molecular components regulating dendritic growth

The molecular mechanisms underlying the establishment of neuronal polarity during development are complex, and involve the integration of many different signaling pathways [49]. Dendritic growth during development occurs in a manner that is spatially and temporally distinct from axonal growth. Dendritic arbors, once formed, are largely maintained throughout the neuronal lifetime, with morphological refinements occurring as a result of neuronal activity. On the other hand, dendritic spines, which are protrusions from dendritic shafts which form the immediate postsynaptic compartment in synapses, are much more dynamic structures. These could grow and shrink in size with the nature (inhibitory or excitatory), pattern and strength of presynaptic inputs. In regulatory terms, dendritic growth is expected to share features and components with axonal growth. Thus, several members of the key families of proteins that functions in axonal guidance and pathfinding (e.g. semaphorins, Slit/Robo, netrin/DCC and ephrin/Eph) also provide extrinsic modulation to dendritic growth during development [50,51]. Expectedly, the Rho family GTPases and their modulators and effectors, acting downstream of these extrinsic factors, play critical roles in cytoskeletal changes accompanying growth and arborization of dendrite [52–54], as they do for axons.

Factors that differentially modulate dendritic versus axonal growth had emerged over the recent years, with several interesting reports appearing over the past year alone. The earlier ones identified include the transcription factors NeuroD [55] and calcium-responsive transactivator (CREST) [56], as well as signaling molecules such as dendrite arborization and synapse maturation-1 (Dasm-1) [57] and bone morphogenetic protein-7 (BMP-7) [58,59]. It is known that Rac and Rho function antagonistically, with activation of the earlier promoting while the latter inhibiting neurite growth. Interestingly, it was recently shown that silencing of Rac1 in mouse hippocampal neurons rather specifically inhibited dendritic but not axonal growth [60]. A specific role for Rac1 in activity-dependent dendritic spine development has been implicated earlier by findings made on the Rac1-guanine nucleotide exchange factor (GEF), *T*-cell lymphoma *i*nvasion *a*nd *m*etastasis *1* (Tiam1) [61]. The latter is present in dendrites and spines of hippocampal neurons and is phosphorylated in a calcium-dependent manner in response to NMDA receptor activation. It was also recently shown that activation of EphB receptor by the ephrinB ligands induces phosphorylation and recruitment of Tiam1 to EphB complexes containing NMDA receptors [62]. This provided a mechanistic explanation for EphB's role in spine development [63].

Another factor upstream of Rac that appears to mediate *b*rain-*d*erived *n*eurotrophic *f*actor (BDNF)-induced dendritic growth in cortical neurons is the lipid raft-associated Ca^{2+}/cal*m*odulin-dependent protein *k*inase (CaMK)Iγ [64]. CaMKIγ is modified by a sequential prenylation followed by a kinase-activity-regulated palmitoylation, which is important for its targeting and anchorage into lipid rafts. Raft insertion of CaMKIγ specifically promoted dendritogenesis, acting upstream of another Rac GEF, the *S*IF and *T*iam 1-like *e*xchange

*f*actor (STEF) and Rac. Small GTPases regulating dendritic outgrowth is however not limited to members of the Rho family. Activation of Rit, a small GTPase belonging to a Ras subgroup, was recently shown to promote axonal growth but inhibits dendritic growth in both hippocampal and sympathetic neurons [65]. Rit appears to act downstream of several signaling pathways, including that of BMP-7, and affects neurite growth through activation of the *e*xtracellular signal-*r*egulated *k*inases 1 and 2 (ERK 1/2) [65].

In spite of the impressive array of molecules now known to regulate axonal and dendritic growth, particularly in terms of signaling and cytoskeletal dynamics, the links to exocytic (or secretory) traffic in these processes remain unclear. The presence of Golgi outposts in dendrites but not axons suggests that there could be fundamental differences in the way these cellular protrusions are connected to secretory membrane traffic at the cell body. The fact that a mutant of TGN-localized PKD (functioning in the fission of TGN-derived cargo containers) PKD-KD, had a more immediate suppressive effect on dendritic rather than axonal growth, also suggests that the earlier may be more dependent on classical secretory traffic [37].

5. Dendritic growth is mediated by Golgi outposts and is dependent on components of both the early and late secretory pathway

In a genetic screen to identify *Drosophila* mutants that are defective in axonal or dendritic development, Jan and colleagues [66] had isolated some interesting mutants with defects in dendritic but not axonal growth. These fell into eight different complementation groups, and amongst the genes mutated are those encoding *Drosophila* homologues of *Sec23*, *Sar1* and *Rab1*, all of which function in the early secretory pathway, more specifically in ER–Golgi transport. One of these mutated genes, *dar3*, codes for the *Drosophila* Sar1 orthologue (which functions in mediating vesicle budding from the ER). Interestingly, not only does *dar3* mutation affect dendritic (but not axonal) growth of *Drosophila* larva class IV da neurons, *Sar1* silencing by siRNA affected cultured mammalian hippocampal neurons in a similar manner. *F*luorescence *r*ecovery *a*fter *p*hotobleaching (FRAP) analysis showed that delivery of a surface membrane marker CD8-EGFP to dendrite (but not axon) was specifically affected by *Sar1* silencing. The authors also noted that Golgi outpost dynamics of the *Drosophila* class IV da neurons correlated well with dendritic branch dynamics (in terms of branch retraction and extension). In attempts to address whether Golgi outposts have a direct role in dendritic growth, the authors illuminated these structures with an intense pulse of laser, which markedly reduced dendritic dynamics compared to "control" illuminations targeted against regions without Golgi outposts. Furthermore, reasoning that the positioning of Golgi outposts could be perturbed by genetic manipulations of a *Drosophila* Golgin molecule Lava lamp (Lva) [67] (a cytoskeletal component which mediates interactions between the Golgi and the dynein/dynactin complex), the authors examined the effect of expressing dominant-negative forms of Lva and inducible RNAi. Both manipulations changed the distribution of Golgi outposts in class IV da neurons without affecting the somatic Golgi. As a result, branching in distal dendrites and total dendritic length was markedly reduced.

These studies are important because they involve manipulations that are targeted to dendritic Golgi outposts without affecting somatic Golgi. The data provided the first firm functional association between these dendritic satellite components with localized and polarized dendritic surface membrane growth. Specific disruptions of dendritic growth by mutations in components of the early secretory pathway, such as the ER–Golgi transport modulators Sar1 and Rab1, are themselves interesting. In the case of Rab1, this may in fact reflect a specialized function of Rab1-containing ER–Golgi intermediate compartment in neurons or neuron-like cells. It has been shown that in polarized PC12 cells induced to sprout neurite-like processes, the morphological differentiation involves expansion of the intermediate compartment and movement of Rab1-containing tubules to the neuritic growth cones [68]. This occurs without an associated movement of other ER and Golgi membranes marked by p58 and COPI, and interestingly, also excludes the known Rab1-associated tether proteins p115 and GM130. Whether this phenomena occurs in primary neurons undergoing morphological differentiation is yet unclear.

The TGN-localized Syx16 also appears to have a role in dendritic outgrowth [48]. Over-expression of wild type Syx16 moderately stimulates neurite outgrowth in both mouse Neuro-2a cells and primary mouse cortical neurons. A mutant Syx16-ΔNt (with the first 50 amino acids of Syx16 deleted), and siRNA mediated silencing of Syx16, significantly retards MAP2-labeled dendritic growth of primary cortical neurons, but had no significant effect on axonal extensions. As mentioned, Syx16 is known to be involved in early/recycling endosome to TGN transport [45], and in non-neuronal systems plays a role in the Golgi-endosomal trafficking of molecules such as the insulin-responsive glucose transporter GLUT4 [69,70]. Its role in anterograde traffic is however unclear, and its silencing by siRNA had no significant effect on surface expression of VSVG in non-neuronal HeLa cells [71]. Although the route and direction regulated by Syx16 in dendrites has yet to be delineated, its dendritic enrichment and presence in Golgi outposts allows some speculations outlined in the section below. It should be noted that dendrite to TGN transport has been clearly demonstrated, for example for the activated somatostatin type 2 receptors [72]. Upon agonist stimulation, dendritically localized receptors are retrogradely transported through a microtubule-dependent mechanism to a *trans*-Golgi domain that are enriched in the TGN markers syntaxin 6 and TGN38.

It should also be noted that membrane traffic associated with dendritic and axonal growth may share critical components. One such known example is another SNARE molecule, TI-VAMP (VAMP7) [73,74]. Over-expression of the isolated N-terminal Longin domain of TI-VAMP inhibits both axonal and dendritic outgrowth, while that of TI-VAMP without the Longin domain enhances growth [73]. In non-neuronal cells, TI-VAMP functions mainly in the late endosome/lysosome transport and membrane fusion [75], and also plays a role in phagocytosis [76]. Both of these are retrograde transport

processes. TI-VAMP's rather specific localization to vesicular structures in axons and dendrites [77] that exhibit anterograde, exocytic-like movements reflects a possibly adapted neuron specific function.

6. Modulation of dendritic membrane dynamics by Golgi-mediated and recycling endosome-mediated exocytosis

Another dendritic membrane traffic route that has relevance to those discussed above involves dendritically localized recycling endosomes (RE) [78]. In particular, membrane trafficking from RE is required for the growth and maintenance of dendritic spines. Ehlers et al. [79] have recently showed that REs labeled with transferrin are present in dendritic shafts and spines, and are frequently found positioned at the base of spines. A long term potentiation (LTP)-inducing stimulus could mobilize RE and associated vesicles into spines. Inhibiting such RE transport using a cell permeable, transmembrane domain-deleted mutant of the endosomal SNARE syntaxin 13 abolishes LTP-induced spine formation. In fact, maintenance of spines appear to require continuous transport from REs. Importantly, the authors showed that exocytosis from REs occur locally in spines, and is triggered by activation of NMDA receptors. This localized RE exocytosis results in LTP-associated plasticity, which occurs concurrently with activity-induced spine enlargement. Dendritic spine-associated RE therefore not only serve as a cytoplasmic reservoir for the ionotropic α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) or NMDA receptors in modulating synaptic plasticity [80,81], but also provides other membrane components for activity-dependent spine growth.

It would appear that REs are rather essential for spine growth in mature neurons. Could these also be important for the earlier stages of polarized dendritic and axonal growth during morphological development? RE-based transport contributes to the surface subdomain targeting of certain proteins, which occurs in both neuronal and non-neuronal cells [82]. RE-based exocytosis is also involved in the growth of the cleavage furrow during cytokinesis [83]. During morphological differentiation of hippocampal neurons, endosomes appears to cluster together with centrosomes and the Golgi apparatus close to the area where the first neurite would appear [84]. As mentioned earlier, both RE-associated membrane components Rab11 and syntaxin 13 have been implicated in the modulation of neurite outgrowth [19,20]. Schmidt and Haucke [85] have indeed speculated that REs may play major roles in polarized growth during neuronal differentiation. This notion of RE-mediated membrane component recycling playing multiple roles in the growth of dendritic surfaces is interesting, but requires further experimental verification. Syx16 mediates RE-TGN transport, and possibly partitions between RE and TGN. Its known function and

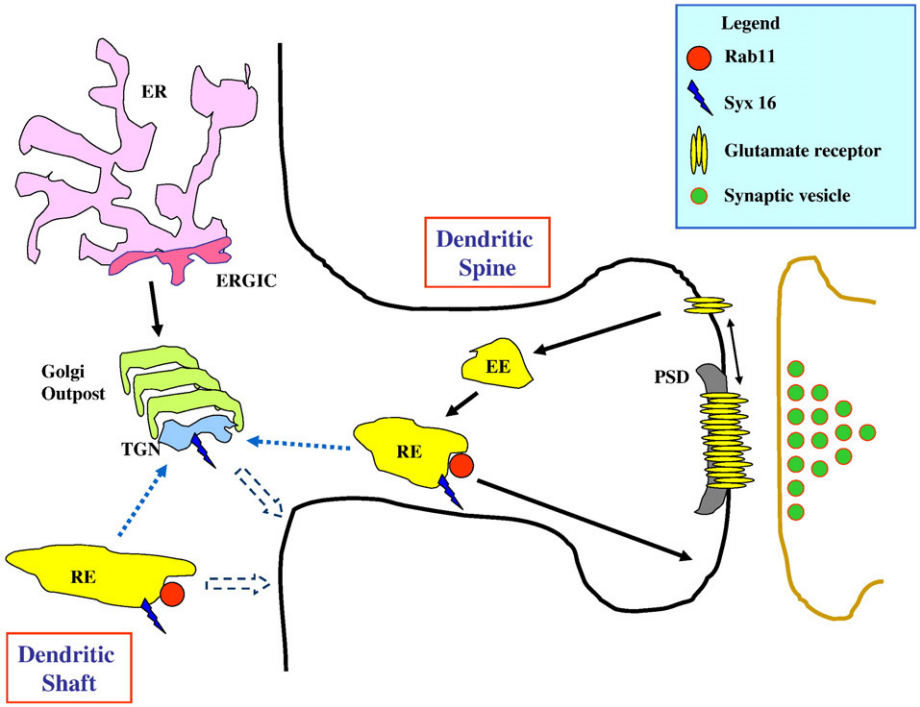

Fig. 1. A highly simplified schematic diagram showing the exo- and endocytic organelles at a portion of the dendrite shaft and dendritic spine. The membrane spanning structures denotes glutamate receptors, and membrane traffic components Syx 16 and Rab11 are depicted as indicated in the legend. It should be noted that the presence of Syx 16 on recycling endosomes (RE) is not yet confirmed. Apposing the postsynaptic compartment is an axon terminus from another neuron, which harbors synaptic vesicles (ER—endoplasmic reticulum, ERGIC—ER–Golgi intermediate compartment, TGN—*trans*-Golgi network, RE—recycling endosome, EE—early endosome, PSD—postsynaptic density). Known transport routes are marked with black arrows. Retrograde transport between dendritic REs (those at the spine and those at the shaft) and Golgi outposts have not yet been demonstrated (dotted line blue arrows). The potential contribution of Golgi outposts-based exocytosis and RE-based exocytosis on dendritic surface growth are marked by dotted block arrows.

localization are therefore in line with its apparent role in dendritic growth.

7. Conclusion and perspectives

The recent findings highlighted above have provided some novel and important insights into the phenomenon of neurite outgrowth. While it has always been known that axonal sprouting and dendrite growth happens at different times, how these processes themselves are differentially regulated is not exactly clear. We now realize that one important difference lies in their reliance on the continuous background of exocytosis, and perhaps also endocytosis. Many interesting questions arise from the fact that dendritic growth is specifically dependent on the components of the classical exocytic and endosomal recycling pathways. Dendritic growth could result from classical but localized ER–Golgi mediated exocytosis, as well as potentially from dendritic recycling endosomes (see Fig. 1). Other than locally translated proteins, how exactly do biosynthetic membrane components that are synthesized at the cell body reach the tips of a growing dendrite? How much of these are contributed by localized exit from dendritic ERGIC? Is there cargo exchange between somatic Golgi and the dendritic Golgi outposts? What is the role of REs in dendritic (or for that matter, axonal) growth during morphological differentiation? In view of Syx16's role in RE-TGN retrograde traffic, it would also be interesting to see if Golgi outposts serve a specific function in dendritic retrograde traffic.

Endocytosis occurs from both axonal and dendritic domains, and components of the elaborate neuronal endocytic machineries are likely involved in modulating neuronal morphogenesis. Mutations of the *Drosophila* Shrub (an evolutionarily conserved key component in the *e*ndosomal *s*orting *c*omplex *r*equired for *t*ransport (ESCRT-III)), for example, resulted in ectopic dendritic and axonal branching [86]. There is likely an overlap in the use of certain molecular components for developmental dendritic growth and dendritic spine plasticity. Defining the exact functions of these in the two different settings would be an important pursuit.

The axonal and dendritic domains of a neuron are also connected by both dendro-axonal (e.g. transferrin receptor) [87] and axonal-dendritic (e.g. neurotrophins) [88] transcytotic processes [89]. Bidirectional neuronal transcytosis is likely to be differentially modulated during axonal and dendritic outgrowth, and it would of great interest to investigate how the growth of the different neurites are connected to (or indeed in some ways coordinated by) transcytotic traffic.

More definitive answers to some of the questions posed above must wait for technical advances that allow more specific micromanipulations of the dendritic satellite exocytic/recycling apparatus. Jan and colleagues had cautiously noted that although their laser illumination of fluorescent protein marked Golgi outposts appeared to reduce dendrite dynamics, the exact extent of structural and functional ablation was unclear. Future identification of local dendritic components that functions specifically in dendritic growth (and not non-polarized surface growth that is mediated through the somatic Golgi) would also be useful. Meanwhile, live-imaging analysis of the relative dynamics of multiple markers, coupled to confirmation of their interactions by advances in fluorescence resonance energy transfer (FRET) during dendritic growth, would hopefully yield more immediate information.

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