STRUCTURAL AND FUNCTIONAL ORGANIZATION OF THE SYNAPSE

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Cover illustration: The cover illustration shows an immunofluorsecence micrograph of a hippocampal pyramidal neuron at two weeks in culture. The neuron was stained for the abundant calcium and calmodulin-dependent protein kinase CaMKII (green) and the presynaptic protein synapsin (red). The pictures was provided by Y. Chen and J. W. Hell, University of Iowa.

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Diversity in Synapse Structure and Composition

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1 Introduction

This chapter describes diversity in the structure and composition of synapses at the resolution of serial section transmission electron microscopy (ssTEM). Section 1 introduces the synapse. Section 2 describes the structure and composition of presynaptic axons. Section 3 elucidates postsynaptic structure and composition. Section 4 discusses the impact of perisynaptic astroglial processes on synapses. Throughout all sections an effort is made to understand regularities in the relationships among these features that might contribute to the diversity in synapse size and number in systematic ways.

A synapse has a presynaptic component, usually an axon but sometimes a dendrite, and a postsynaptic component, usually part of a dendrite, cell soma, or axonal initial segment and occasionally an astroglial process. Perisynaptic astroglia represent a third component that occurs at some synapses. When perisynaptic astroglia are present, the structural complex has been referred to as a 'tripartite synapse' (45). Figure 1 illustrates an electron micrograph of an ultrathin (50 nm) section through dendrites, axons, astroglial processes an synapses in the rat hippocampus. This picture was chosen to open this chapter because it nicely illustrates the diversity in the composition of even a tiny segment of the neuropil.

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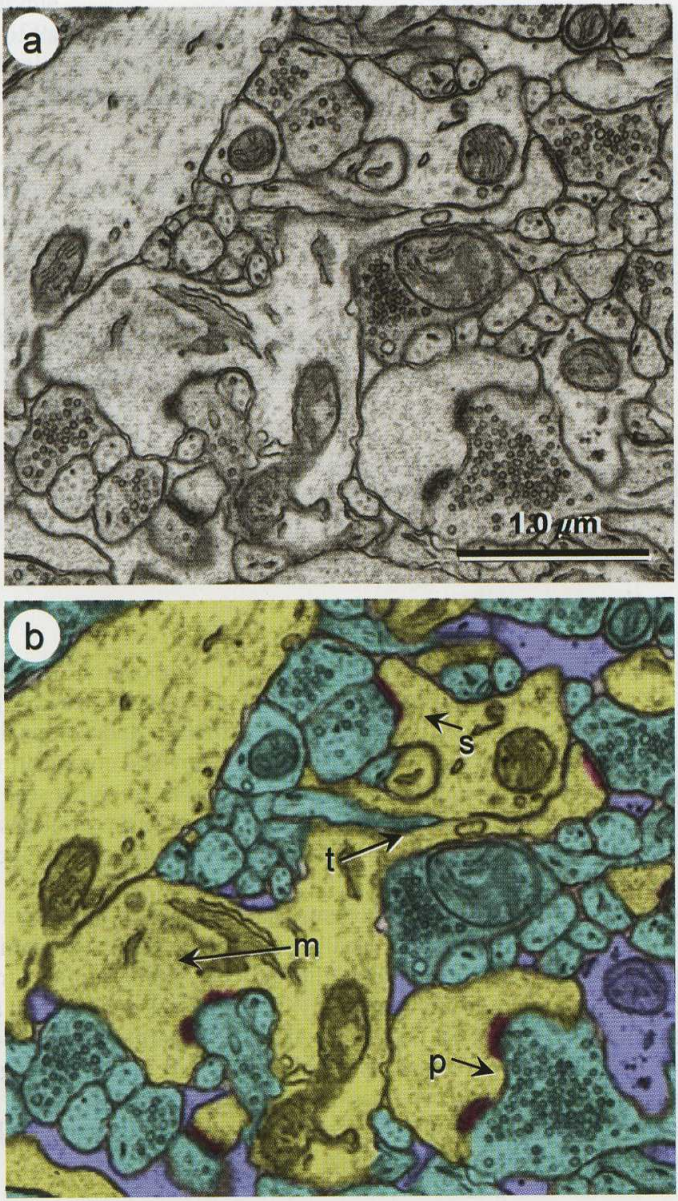

Fig. 1. A single thin section spanning approximately 10 μm^2 in the middle of the apical dendritic arbors of hippocampal area CA1 pyramidal cells. (**a**) *Gray* scale image obtained at the electron microscope. (**b**) Same section colorized to illustrate dendrites (*yellow*), axons and vesicle-filled axonal boutons (*green*), asymmetric postsynaptic densities of synapses located on diversely shaped dendritic spines (*red*), and astroglial processes (*lavender*). Stubby (s), thin (t) and mushroom (m) spines can be seen in longitudinal section emerging from dendrites. A large mushroom spine head has a perforated (p) postsynaptic density.

2 Presynaptic Structures and Composition

Three dimensional reconstructions of individual presynaptic axons that pass through the complex hippocampal neuropil show the diversity in their local trajectories (Fig. 2a). A three-dimensional reconstruction of a hippocampal dendritic segment that is approximately 10 microns long illustrates the more than 10 fold variation in the dimensions of neighboring dendritic spines and synapses (Fig. 2b).

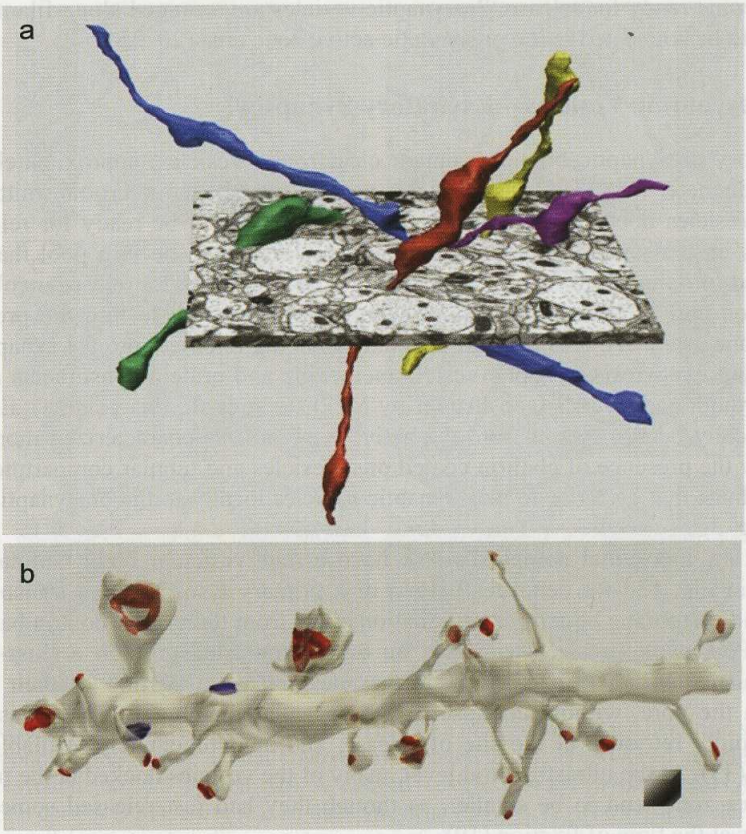

Fig. 2. Diversity in the trajectory of presynaptic axons and shapes of postsynaptic dendritic spines. (**a**) Three-dimensional reconstructions of a subset of axons passing through a single electron micrograph. (Adapted from (36)). (**b**) Three-dimensional reconstruction of a single dendritic segment, illustrating the diversity in dendritic spine shapes and their postsynaptic densities (*red*). Inhibitory shaft synapses are colorized in *blue*. (This is a recently surfaced image of dendrite 21 from (18) available at http://synapses.clm.utexas.edu/). Scale is approximately 0.5 μm3 for both reconstructions.

2.1 Presynaptic Active Zone

A presynaptic axonal bouton contains vesicles with a variety of shapes and sizes. These vesicles contain excitatory or inhibitory neurotransmitters, neuromodulatory peptides, proteins required to concentrate neurotransmitters, and a variety of proteins involved in the vesicle cycle or that are destined for the presynaptic active zone (11). The presynaptic active zone is a specialized region variously described as 'dense projections', the 'presynaptic grid' or mini-active zones (34) where vesicles dock and become ready for release. Presynaptic vesicles are arranged along filaments that appear to be connected to the presynaptic active zone area (20, 25).

2.2 Presynaptic Vesicles – Excitatory Synapses

Excitatory presynaptic boutons contain clear round vesicles, approximately 35–50 nm in diameter (Fig. 3). These vesicles usually contain the neurotransmitter glutamate. Vesicles docked at the active zones are thought to be ready for release and vesicles located away from the membrane are thought to be in a pool that can be recruited for later release. One mechanism for neurotransmitter release involves pore formation and subsequent collapse of the presynaptic vesicle into the presynaptic membrane at the active zone with the contents being released into the synaptic cleft. Following exocytosis, synaptic vesicle membrane and protein constituents are recycled through endocytosis (see chapter by McPherson et al., this volume). Endocytosis typically occurs distant from the active zone, and is characterized morphologically by the presence of clathrin coated pits, vesicles and tubular compartments with coated buds that give rise to new synaptic vesicles locally in the presynaptic bouton (e.g. Fig. 3g).

Sorting endosomal complexes also have a multivesicular body (similar to that shown in Fig. 12d for dendrites below) or a primary lysosome that transports proteins and membrane bound for degradation away from the axonal bouton back to the soma. Presynaptic vesicles can also be rapidly recycled through a 'kiss-and-run' mechanism. During kiss-and-run, the vesicles release a portion of their contents through the pore, without collapse of the vesicular membrane. These vesicles are then rapidly retrieved at the site of release, and are immediately available for re-release (33). At the ultrastructural level, many of the vesicles docked at the presynaptic active zone tend to be smaller, as though they had just released some of their contents at the time of fixation (19).

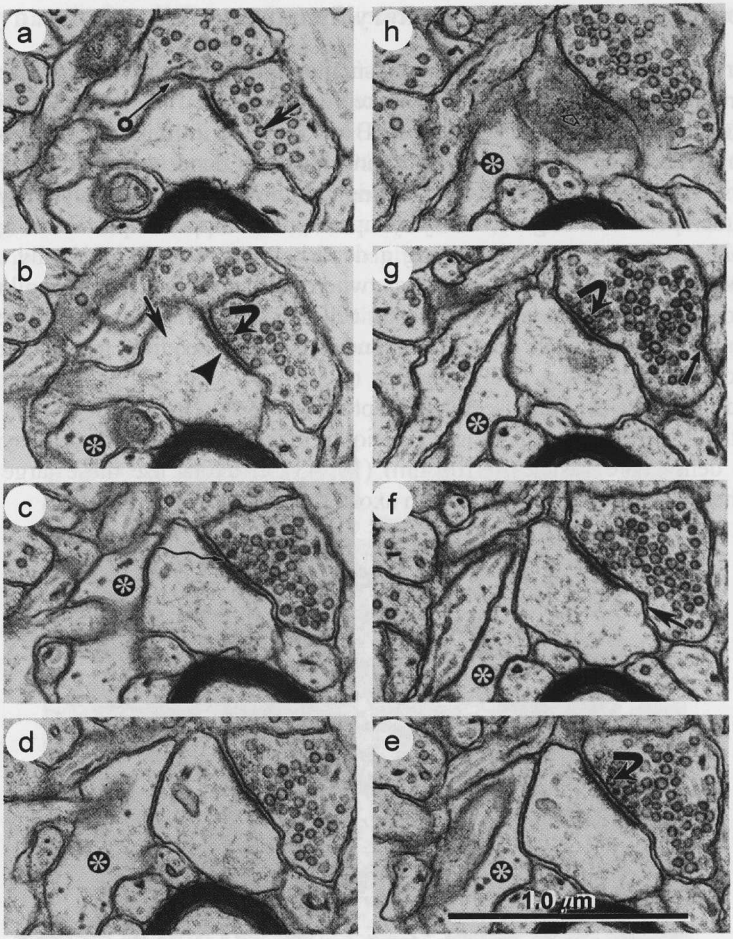

Fig. 3. Excitatory synapse revealed through ssTEM of a mushroom-shaped dendritic spine and its corresponding presynaptic axonal bouton. (**a**) A characteristic non-docked vesicle (*arrow*) in the presynaptic axonal bouton. (**b**) A docked vesicle (*curved arrow*) across from the postsynaptic density (PSD, chevron); on the spine head (*arrow*). (**c**) The synaptic cleft (*wiggly arrow*) located between the plasma membranes of the spine head and presynaptic bouton contains dense staining material, presumably composed of adhesion molecules and portions of receptors. (**d**) Serial section between (c) and (**e**) illustrates astroglial process (*) at the base of the spine head. (e) Curved arrow illustrates another docked vesicle. (**f**) Extracellular space (*arrow*) does not contain the dense-staining material found in the synaptic cleft. (**g**) Coated pit (*straight arrow*) at a site of endocytosis on side of the bouton away from the active zone; docked vesicle (*curved arrow*). (**h**) *Gray* surface of the plasma membrane (*open arrow*) viewed en face where it caps the head of the dendritic spine. In (a) the astroglial process labeled (°) is near the synaptic cleft where it might detect and control spillout of neurotransmitter; in all other sections the perisynaptic astroglial process is labeled with (*). (Adapted from (19)).

2.3 Presynaptic Vesicles – Inhibitory Synapses

Inhibitory presynaptic boutons contain smaller, pleiomorphic vesicles having both round and flattened shapes in aldehyde-fixed tissue (Fig. 4). The pleiomorphic vesicles usually contain the neurotransmitters GABA or glycine. Inhibitory synapses are most abundant at the neuronal soma (Fig. 4a) and proximal dendritic zones (see chapter by Arancibia-Carcamo, Triller and Kittler, this volume). In addition, inhibitory synapses can be interspersed among excitatory synapses (in the hippocampus about 1 inhibitory per 10–20 excitatory synapses) along a dendrite (Fig. 4b). Occasionally, inhibitory synapses are located at the axonal hillock, where activation of one or a small number of inhibitory synapses can regulate neuronal cell firing at their axons. In some brain regions an inhibitory synapse occurs on the necks of some dendritic spines, whether they veto excitatory activation likely depends on their frequency and the specific circuit involved (12, 24, 46). Neurosecretory peptides and some neurotransmitters are localized to the cytoplasm surrounding the pleiomorphic vesicles of inhibitory synapses, or in large dense core vesicles (~100 nm) (10, 43). If axons use these large secretory DCVs then more of them occur in each axonal bouton.

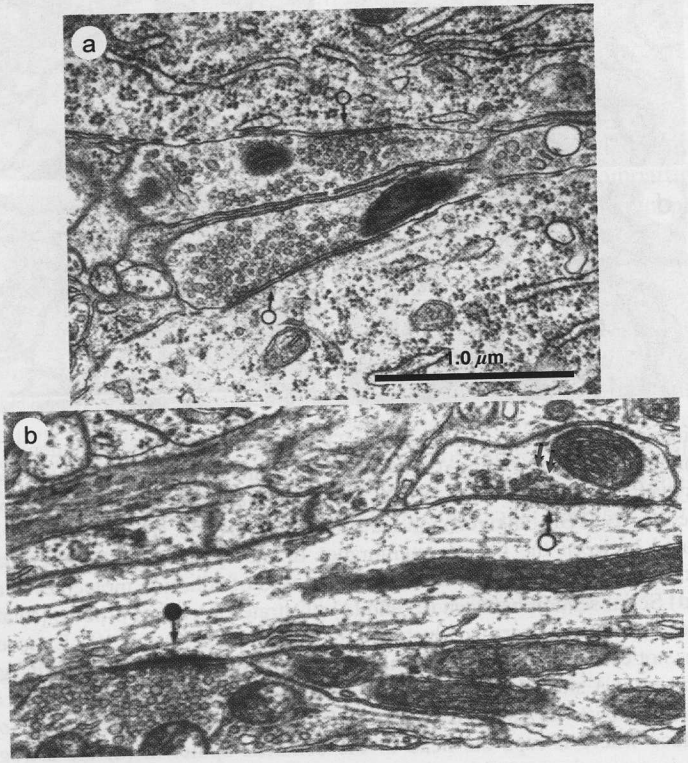

Fig. 4. Inhibitory symmetric synapses in mature hippocampus with thin pre- and postsynaptic densities and pleiomorphic vesicles. (**a**) Two inhibitory synapses on the pyramidal cell bodies (*open circles*). (**b**) Symmetric synapse (*open circle*) with flattened vesicles (*red arrows*) and asymmetric synapse (*closed circle*) with thicker PSD and larger rounder presynaptic vesicles. These synapses are located directly on the dendritic shaft of a nonspiny interneuron in mature hippocampal area CA1. (Modified from (16)).

2.4 Presynaptic Small Dense Core Vesicles as Active Zone Transporters

Small dense core vesicles (~80 nm) are distinct from large DCVs both in size and frequency (Fig. 5). Small DCVs are present in only about 20% of mature presynaptic axons, and when present, only 1–10 vesicles occur in a fully reconstructed axonal bouton. The outer membranes of small DCVs label with antibodies to proteins located at the presynaptic active zone, such as piccolo and bassoon, and they are prevalent along axons in the developing nervous system; hence the small DCVs are thought to be a local source of new presynaptic active zones (1, 50) (see chapter by Shen and Garner, this volume). Recent work has shown that there are fewer small DCVs during rapid synapse formation in the mature hippocampus in further support of their role in local delivery of presynaptic active-zones (39).

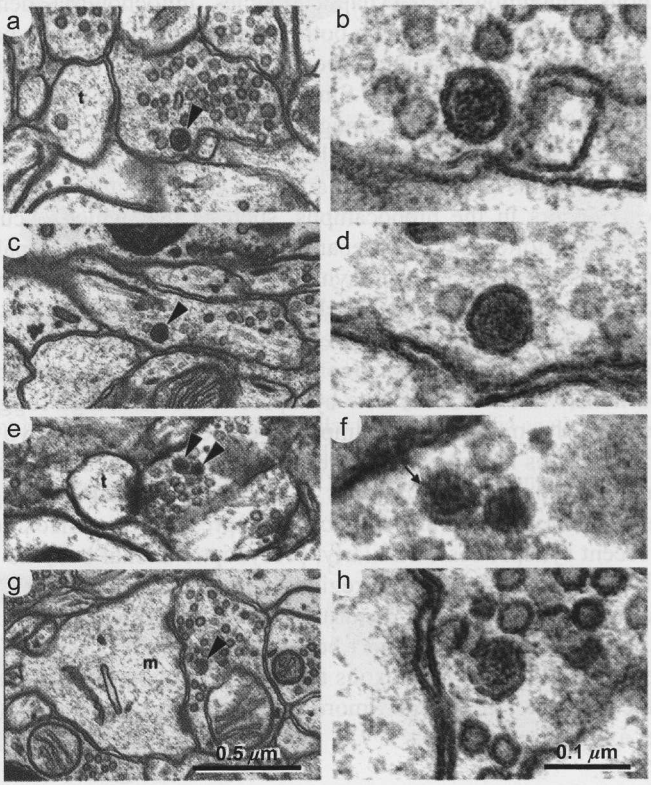

Fig. 5. Small dense core vesicles (*arrowheads*) at excitatory synapses in the apical dendritic field of mature hippocampus (CA1). (**a, b**) Low and higher power views of a small dense core vesicle (dcv) in typical location near plasma membrane but away from active zone. (**c, d**) Small DCV located near the beginning of an inter-varicosity region, suggesting it might be in transit. (**e, f**) Two small DCVs in a presynaptic axonal bouton. One of these vesicles clearly illustrates small 'spicules' emanating from its surface. (**g, h**) Small DCV located within one vesicle diameter of an active zone. DCVs are rarely located this close to an active zone, but show that they might also be involved in synapse enlargement, not just new synapse formation. Scale bar in (g) is for (a, c, and e). Scale bar in (h) is also for (b, d, and f). (Adapted from (38)).

2.5 Local Protein Synthesis in Presynaptic Boutons?

Although polyribosomes are not a prominent component of presynaptic axonal boutons in the central nervous system, mRNAs have been localized to them (32). In squid giant axons, local protein synthesis machinery appears to derive from the ensheathing glia (9, 14). Detailed three-dimensional reconstructions will be required to learn whether isolated polyribosomes are directed into vertebrate presynaptic axonal boutons to allow for local protein synthesis, similar to that observed in dendritic spines (see below).

2.6 Nonsynaptic, Single Synaptic and Multisynaptic Axonal Boutons

Axonal boutons containing clear synaptic vesicles, mitochondria, dense core vesicles, and multivesicular bodies occur both with and without postsynaptic partners (Fig. 6). In the mature hippocampus, about 96% of the vesicle-containing boutons have at least one postsynaptic partner. Single-synapse boutons predominate comprising about 75% of all vesicle-containing axonal boutons. About 21% of vesicle-containing boutons are multi-synaptic while about 4% are non-synaptic boutons in the mature hippocampus (36, 37). There are more multisynaptic boutons when rapid synaptogenesis occurs in the hippocampus, such as that which occurs during the estrus cycle (49) or following the preparation of mature hippocampal slices under ice-cold conditions (22, 23, 31). Multisynaptic dendritic spines, receiving input from more than one presynaptic axon occur relatively frequently during development and under conditions of synaptogenesis in the mature hippocampus. The axonal segment in Fig. 6c was from a hippocampal slice that had been prepared under ice-cold dissection conditions, which induces new synapses. These findings suggest that rapid synaptogenesis in the mature hippocampus does not require de novo formation of presynaptic axons. It is not known whether the nonsynaptic boutons also constitute a source of available presynaptic boutons to accommodate rapid synaptogenesis, if they represent vesicle clusters in transit between presynaptic varicosities, or if thery are sites of recent synapse loss. Similarly, presynaptic axonal boutons vary in structure along the axons from other brain regions such as cerebellar cortex (Fig. 7, (48)). Parallel fiber axons synapse with dendritic spines of Purkinje cell spiny branchlets (Fig. 7a–c; see also Figs. 8d, 11c and f below for further discussion of these postsynaptic spines). The climbing fiber axons that synapse along the proximal dendrite of the Purkinje cells have much larger, more irregularly shaped boutons (Fig. 7d) than parallel fiber axons.

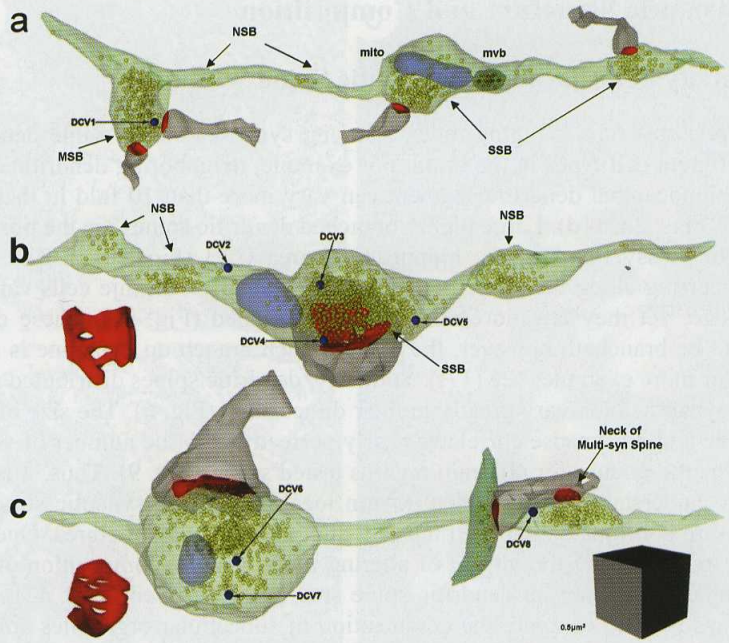

Fig. 6. Axonal segments from mature hippocampus (CA1). (**a**) This segment is 7.8 μm long with 2 single-synapse boutons (SSB), 2 nonsynaptic boutons (NSB), and 1 multiple synapse bouton (MSB) shared by 2 postsynaptic spines from different dendrites. (**b**) This axonal segment is 5.7 μm long with 1 SSB containing 3 small DCVs and a mitochondrion. It is surrounded by 3 NSBs. (**c**) This segment is 5.5 μm long with 1 SSB similar to that in (b) and another SSB in an unusual position along the neck of a multi-synaptic dendritic spine. (Axons – *green*, vesicles – *yellow*, mitochondria – *light blue*, DCVs – *dark blue*, PSDs – *red*; Adapted from (38)).

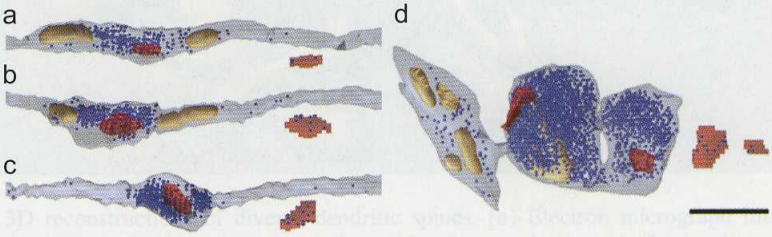

Fig. 7. Reconstructed axons from rat cerebellar cortex. (**a–c**) Parallel fiber axons. (**d**) Climbing fiber axon. Axons (*translucent light blue*); PSDs (*red*); vesicles (*dark blue*); mitochondria (*beige*). In all images, the locations of docked vesicles are superimposed on the 'enface' red PSD reconstructions to the right side of each axon. The scale bar is 1 micron for all 4 reconstructions. (Adapted from (48)).

3 Postsynaptic Structure and Composition

3.1 Diversity in Postsynaptic Dendritic Spine Structure

Postsynaptic structure is highly diverse among synapses on the same dendrite and across different cell types in the brain. For example, neighboring dendritic spines on a single hippocampal dendritic segment can vary more than 10 fold in their dimensions (e.g. Figs. 2b, 11d). Large highly branched dendritic spines are the postsynaptic partners of mossy fibers in the hippocampal area CA3 (Fig. 8a–c). The dendritic spines occurring along the spiny branchlets of cerebellar Purkinje cells vary widely in their size, yet they are more uniformly club-shaped (Fig. 8d). These cerebellar spines can be branched; however, the head of each branch on the spine is also club shaped (for more examples see (17)). Similarly, dendritic spines distributed along the same presynaptic axon vary greatly in their dimensions (Fig. 6). The size of the dendritic spine and its synapse correlates nearly perfectly with the number of vesicles in the presynaptic bouton for all brain regions tested so far (Fig. 9). Thus, it is of great interest to understand the rules that govern local changes in synaptic structure and how they are coordinated between the pre- and postsynaptic structures. One strategy has been to investigate the impact of altering the molecular composition of neurons to determine the impact on dendritic spine structure (reviewed in (4, 42)). Another strategy has been to compare the composition of subcellular organelles among dendritic spines and synapses of differing morphologies, during different stages of development, and during synaptic plasticity. Our focus in this chapter is on this second strategy.

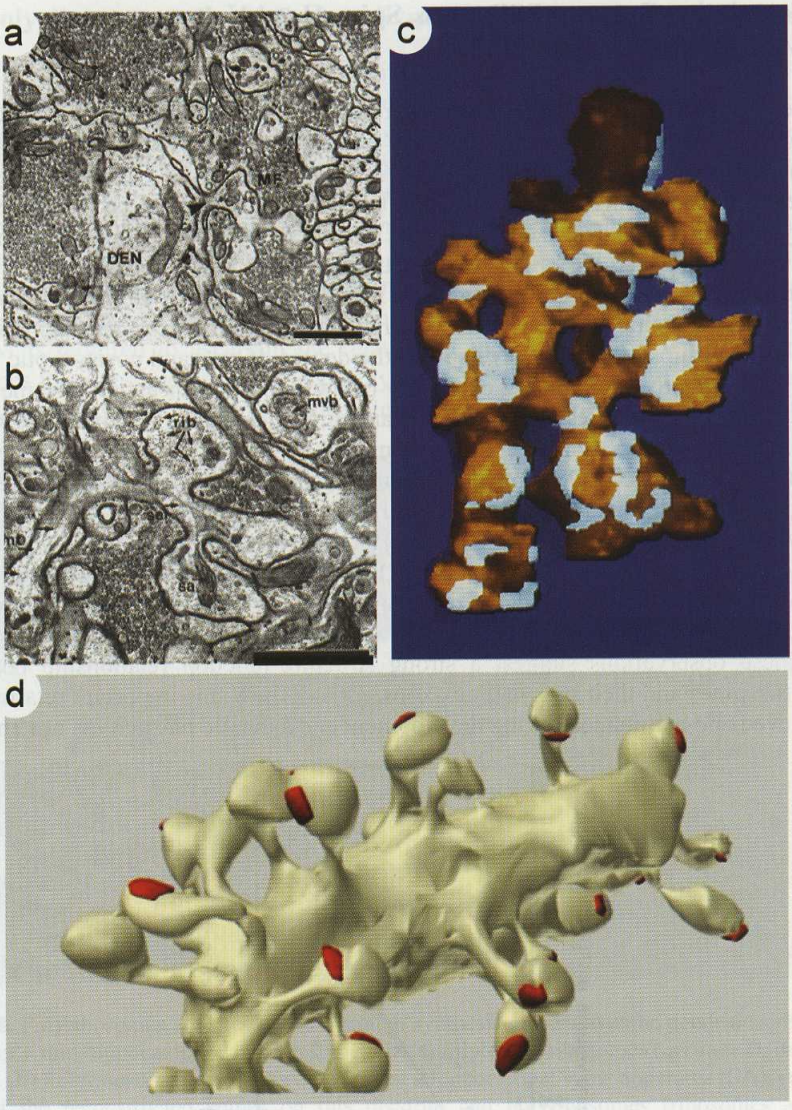

Fig. 8. 3D reconstructions of diverse dendritic spines. (**a**) Electron micrograph illustrating CA3 pyramidal cell dendritic shaft (den), the origin of a complex branched spine (*arrow head*) and mossy fiber (MF) bouton that synapses with spine heads from this and other dendrites. Scale = 1 μm. (**b**) CA3 spine which contains microtubules (mt), polyribosomes (rib), and a tubule of SER that becomes connected to a spine apparatus (sa). The fourth head connected to this spine on a different serial section, has a multivesicular body (mvb). (**c**) CA3 spine (*gold*) with multiple PSDs (*light blue*) distributed across its many heads. (**d**) Segment of spiney branchlet from a cerebellar Purkinje cell (PSDs *red*, dendrite and spines *beige*). Scale in (b) is 1 μm for (b, c, and d). (a–c) are Adapted from (5); and (d) is Adapted from http://synapses.clm. utexas.edu, courtesy J. Spacek.

3.2 Correlation Between PSD Area, Spine Head Volume, and Vesicles in Presynaptic Axon

The postsynaptic density (PSD) contains a host of important receptors, scaffolding proteins, and signaling complexes that vary from synapse to synapse (21). The diversity in molecular composition from synapse to synapse is great, with hundreds of different proteins having been identified in the PSD. In aldehyde-fixed tissue, excitatory synapses are characterized by a thick PSD relative to the thinner presynaptic active zone, and hence are called 'asymmetric synapses' (Figs. 1, 3, 5, 8, 10, 11, 12, 14, see also (15, 30)). Inhibitory synapses usually have equally thin PSDs and presynaptic active zones and hence are referred to as 'symmetric synapses' (Fig. 4). These relationships are highly diverse and the degree of pre- and postsynaptic thickening varies greatly even among excitatory or inhibitory synapses in the same brain region ((6), http://synapses.clm.utexas.edu/anatomy/chemical/colh.htm). Importantly, PSD thickness is also sensitive to experimental manipulations that are known to impact the abundance of PSD molecules, such as the calcium calmodulin dependent protein kinase type II (CaMKII), which are highly enriched in the PSD (28).

The reconstructed surface area of the PSD correlates nearly perfectly with the volume and surface area of the spine head (Fig. 9, (18, 26)). The PSD area and spine head volume also correlate nearly perfectly with the total number of presynaptic vesicles and the number of vesicles docked at the presynaptic active zone (18, 19, 26, 35). These observations suggest a strong structure-function relationship between dendritic spines and their presynaptic axons. Exactly how this important relationship is achieved and maintained during development and synaptic plasticity is not known.

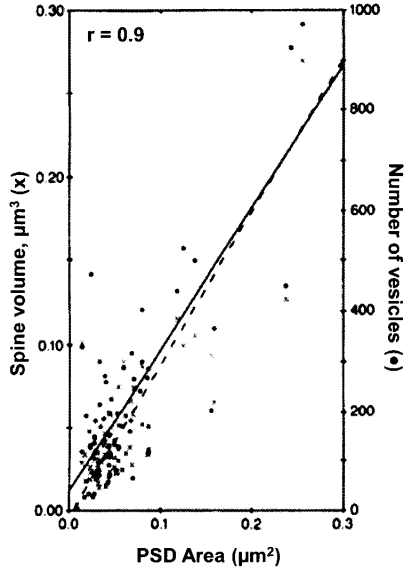

Fig. 9. Strong correlation between PSD area, spine volume, and t number of presynaptic vesicles. This dataset is from hippocampal area CA1, but these relationships hold true across all brain regions evaluated so far, including synapses in hippocampal area CA3, cerebellar cortex, striatum, and neocortex. (Adapted from (18, 26)).

3.3 Polyribosomes – Local Protein Synthesis in Dendrites and Spines

Polyribosomes occur in some, but certainly not all dendritic spines (41). They are sites where translation could have been occurring at the time of tissue fixation. Ribosomes are identified in dendrites and spines as 10–25 nm electron dense spheres surrounded by a gray halo; and polyribosomes are identified in ssTEM as having at least 3 ribosomes (Fig. 10ab). Free polyribosomes synthesize cytoplasmic proteins, like those in the PSD and have been found to increase in frequency during plasticity such as LTP (27). Bound polyribosomes are associated with endoplasmic reticulum and synthesize integral proteins, such as receptors. Occasionally polyribosomes have been detected in the vicinity of a spine apparatus (Fig. 10b, 11a). Quantitative ssTEM is required to ascertain the relative frequencies of free and bound polyribosomes in spines and at other synapse types in the developing and mature brain and during plasticity such as learning and memory which requires local protein synthesis.

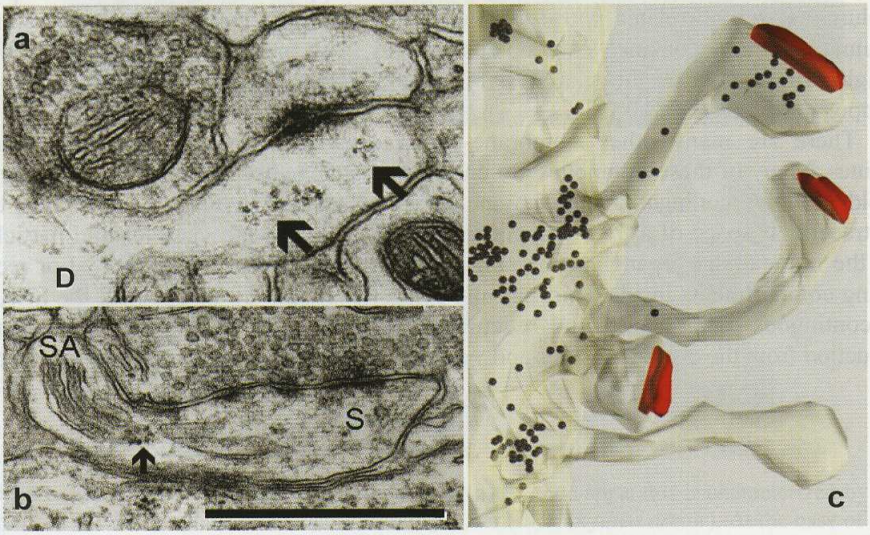

Fig. 10. Polyribosomes are sites of local protein synthesis present in some dendritic spines. (**a**) Head of a thin hippocampal dendritic spine with multiple polyribosomes (*arrows*; D, dendritic shaft). (**b**) Polyribosome (*arrow*) located amidst a hippocampal spine apparatus (SA). (**c**) Only one of the reconstructed spines on this dendrite had polyribosomes (ribosomes – *black spheres*, PSDs – *red*). (Adapted from http://synapses.clm.utexas.edu/anatomy/ribosome/ribo.stm courtesy J. Spacek). Scale in (b) is about 0.5 μm for all three images.

3.4 Diversity in Composition of Smooth Endoplasmic Reticulum and Endosomal Compartments in Dendrites and Spines

Some dendritic spines contain smooth endoplasmic reticulum (SER), which functions as an internal calcium store. In the hippocampus, only about 15% of all spines contain SER, and typically only the largest spines contain SER (40). In contrast, nearly all cerebellar dendritic spines contain SER (17). The spine apparatus is an enigmatic organelle variously thought to be involved in the regulation of calcium, synthesis of proteins, and post-translational modification of proteins like the Golgi apparatus (Fig. 11a–d). It contains SER-like membranes arranged in laminae separated by dense-staining bars that contain the actin-binding protein synaptopodin which is necessary for maintaining its laminar structure and for positioning it into spines (13). Only large hippocampal and cortical spines contain a spine apparatus; in contrast the SER of cerebellar spines forms a reticular membranous network without the dense-staining laminations found in the spine apparatus (Fig. 11ef). Total spine volume is well-correlated with the volume of SER, suggesting a functional role in maintaining ionic balance of cerebellar spines. The presence or absence of SER might also account for the relative stability of cerebellar spines, and plasticity among hippocampal spines (3).

There are many other membrane-bound organelles present in dendrites and spines. Many of these organelles are endosomes as revealed by tracking internalized gold particles conjugated with bovine serum albumin that were injected into the extracellular space of a PN21 hippocampal slice (7). The presence of gold particles in the intracellular organelles unambiguously identifies them as endosomal or recycling compartments inside dendrites and spines (Fig. 12a–d). In contrast, the SER is a continuous membrane-bound reticulum that is easily distinguished upon reconstruction from the discrete tubules and vesicles of endosomal compartments (Fig. 12e).

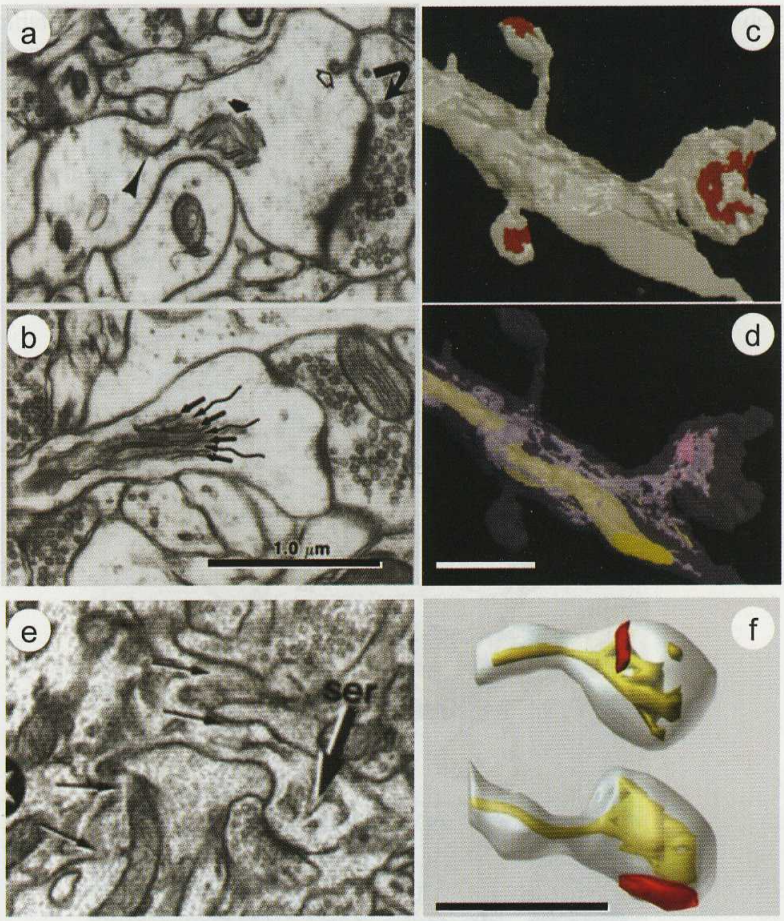

Fig. 11. Mushroom spines with spine apparatus. (**a**) A spine apparatus is characterized by the presence of SER laminated with dense staining material (*thick filled arrow*). At the base of this spine apparatus is rough endoplasmic reticulum (*thin arrowhead*). At the edge of the PSD there are reciprocal coated structures. The curved arrow indicates a double-walled coated vesicle, containing a small bit of the postsynaptic spine, or 'spinule', projecting from the spine into the presynaptic axon. The open *short arrow* indicates a coated pit, endocytosing into the postsynaptic dendritic spine. (**b**) Longitudinal section through a highly laminated spine apparatus. *Straight arrows* indicate SER; *wiggly arrows* indicate dense-staining material. (**c**) Reconstruction of a short hippocampal CA1 dendritic segment with 2 small thin spine and 1 large mushroom spine with surfaced PSDs (*red*). (**d**) Translucent surface of the dendrite and spine reveals the SER (*lavender*) and mitochondrion (*yellow*) in the dendritic shaft. Only the large mushroom spine has SER, which forms a spine apparatus (*pink*) in this example. (**e**) EM image of SER in dendrite and spines of a spiney branchlet from a cerebellar Purkinje cell. Other spine origins emerging from this dendrite are indicated with thin arrows on the left. (**f**) Reconstruction of cerebellar spines and their reticulum of SER inside. PSDs are *red* dendrite and spines are *gray* and ser is *yellow* in these spines. ((a–d) adapted from (40); (e) is adapted from (17); and (f) is from http://synapses.clm.utexas.edu/anatomy/compare/compare.stmcourtesy of J. Spacek).

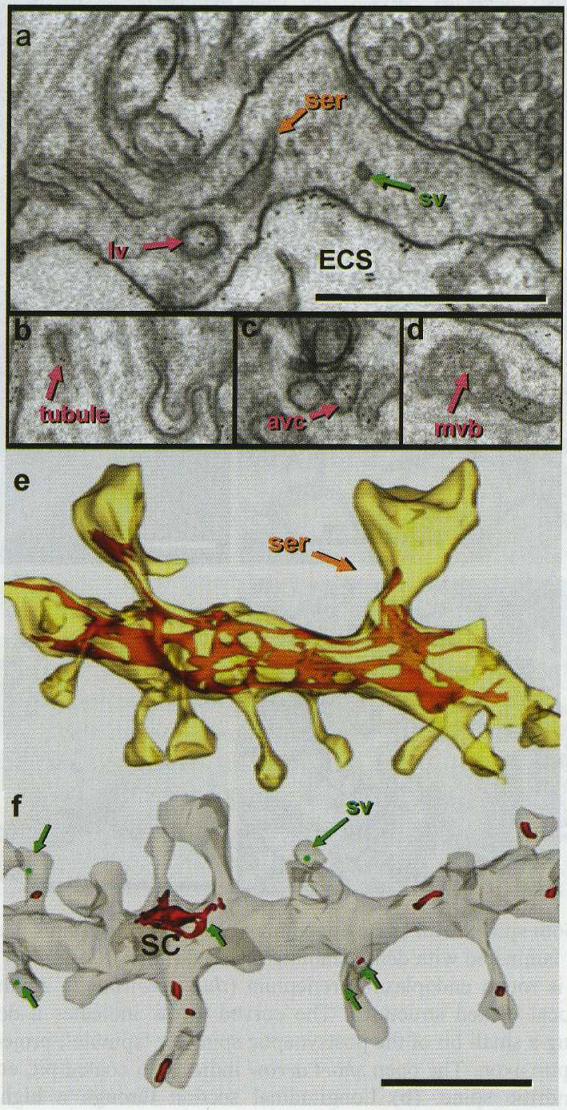

Fig. 12. Diversity among spines in composition of SER and endosomal compartments. (**a–d**) Large vesicles (lv), tubules with coated buds, amorphous vesicular clumps (avc) and multivesicular bodies with tubules all contained gold particles, endocytosed from the extracellular space. Smooth endoplasmic reticulum (ser) never contained gold particles, indicating that these two intracellular membranous compartments are not connected. (**e**) *Top*, reconstruction of SER in a dendrite and associated dendritic spines; about 14% of hippocampal dendritic spines contained SER. *Bottom*, reconstruction of a PN21 dendrite (*gray*) with endosomal structures (*red*) including a sorting complex (SC) which contains tubules, vesicles, a multivesicular body, and coated buds. *Green spheres* represent the locations of the smaller, smooth vesicles, presumably exocytic in function. (Adapted from (7)) Scale in (a) is 0.5 μm for (a–d), and in (**f**) it is 1 um for (e) and (f).

Quantitative work shows that about 50 percent of normal hippocampal dendritic spines from perfusion fixed brain contain no membrane bound organelles; and a distinction among spines that contain the different types of organelles (Fig. 13a). Interestingly, the amount of SER also varies along the shaft of the parent dendritic shaft (Fig. 13b). Recent work shows endosomal compartments to be dynamically regulated during synaptic plasticity (29). It will be interesting to learn whether the presence or absence of SER is also dynamically regulated in spines during synaptic plasticity.

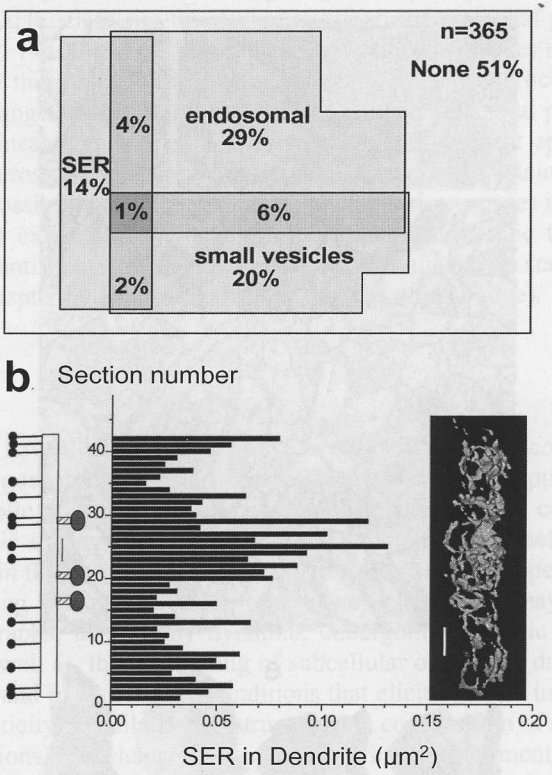

Fig. 13. Variation in the organization of SER and other membrane bound organelles in hippocampal dendrites. (**a**) Venn diagram showing the relative frequency of membrane-bound organelles inside hippocampal dendritic spines. About 49% of dendritic spines contain one or more membranous organelles. It represents an average of all of the dendritic spines that were reconstructed from PN15, PN21, and Adult area CA1 of the rat hippocampus. This relationship is about the same at all three ages in perfusion fixed brain, *in vivo*. (**b**) Relative volume of SER in the dendritic shaft varies by type and number of dendritic spines along the length. At the left is a schematic diagram of this dendrite with the location of thin spine origins along the dendritic segment illustrated to the left as small lollipops and mushroom spine locations illustrated by larger lollipops pointing to the right. The graph illustrates the cross-sectional area of SER in each section along the length of the dendrite. The three-dimensional reconstruction of the SER in *gray* also illustrates its non-uniform distribution in the dendritic shaft. (Diagram in (a) is Modified from (6); and in (b) from (40)).

4 Perisynaptic Astroglial Processes

In the cerebellum, nearly all synapses are enveloped by perisynaptic astroglial processes (Fig. 14ab). Astroglial processes are identified by their light cytoplasm and the presence of dark glycogen granules and astrocytic fibrils in larger processes. They end in thin or flattened processes that easily interdigitate among axons and dendrites throughout the brain. In contrast, less than 50% of neocortical and hippocampal dendritic spine synapses have astroglial processes at their perimeters (Fig. 14cd).

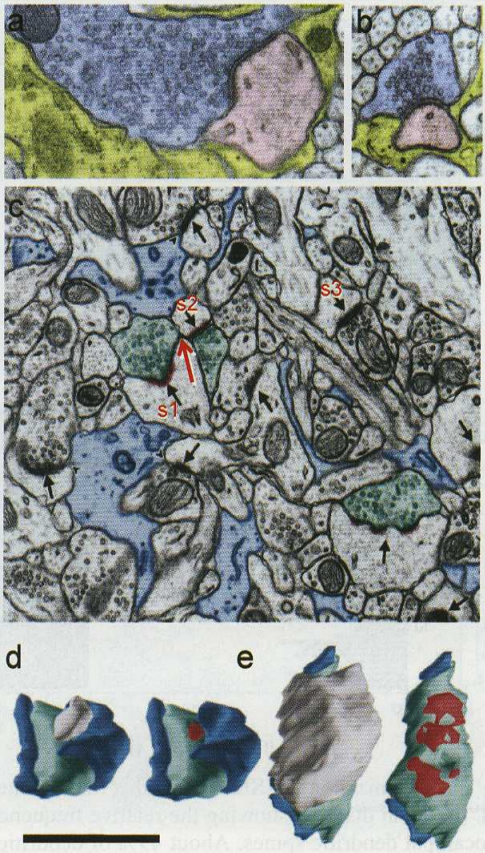

Fig. 14. Perisynaptic astroglial processes. In cerebellar cortex, astroglial processes surround the synaptic cleft of nearly all synapses made by (**a**) climbing fibers or (**b**) parallel fibers. In contrast, only about half of all hippocampal synapses have astroglial processes at the cleft, even between neighboring synapses (e.g. s1→ s2 *Red arrow*; s3 has no perisynaptic astroglia at the presynaptic bouton, the postsynaptic spine, or the perisynaptic cleft region. Scale bar = 1 um for all images). (a and b color schema: presynaptic axon (*blue*), postsynaptic spine (*pink*), perisynaptic astroglia (*yellow*); (**c**) and (**d**) color scheme: presynaptic axon (*green*); postsynaptic density (*red*); astroglia (*blue*)). (a, b adapted from (48)); (c, d adapted from (44)).

This differential arrangement of perisynaptic astroglia suggests that some synapses are highly regulated and supported by this important partner in the tripartite synapse, whereas other synapses are either more selective, unable to attract astroglial processes, or actively repel them (44). Exactly how this relationship is regulated by local synaptic plasticity remains to be determined, however, attraction to high concentrations of extracellular glutamate is a likely candidate mechanism (8). In the mature hippocampus, those synapses that have a perisynaptic astroglial process at its perimeter, are on average significantly larger than synapses without (47). Importantly, size is associated with the presence of an astroglial process juxtaposed to the postsynaptic spine and/or synaptic cleft; not the degree to which the astroglial process surrounds the synapse (Fig. 14de). In fact, very large hippocampal synapses might have only a small fraction of their perimeters surrounded by an astroglial process (compare Fig. 14de). These arrangements suggest that the perisynaptic astroglial processes do more than simply delineate boundaries between synapses to prevent spillover of neurotransmitter and crosstalk and are likely crucial partners in sustaining synapses (2). One intriguing possibility, small, new, relatively unstable synapses in hippocampus or cortex, share the extracellular products released during synaptic transmission from other synapses, until they grow big enough to attract their own stabilizing astroglial processes (see chapter by Eroglu, Barres and Stevens, this volume).

Summary

In this chapter I have described the highly diverse structure and composition of presynaptic axons, postsynaptic spines and dendrites, and perisynaptic astroglial processes. Postsynaptic size is proportional to presynaptic vesicle content and larger synaptic dendritic spines contain more of the subcellular organelles needed to remodel and sustain them. Perisynaptic astroglial processes also appear to be key players in determining synapse size. Although synapse structure may appear static in electron micrographs, it is highly dynamic, undergoing dramatic changes in gross morphology, as well as, the positioning of subcellular organelles during basic synaptic transmission and in response to conditions that elicit changes in synapse function or 'synaptic plasticity'. Similarly, the structure and composition of synapses can vary across brain regions, and change dynamically during development, maturation, normal aging, neurological disorders and trauma. Careful consideration of this structural diversity and plasticity is leading to new understanding about synapse formation, growth, maintenance and elimination.

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