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# Probing synaptic signaling with quantum dots

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**Synapses are the principal substrates of neuronal communication in the brain. Neuroscientists are trying to understand how the remodeling of synapses at the molecular level leads to changes in learning and memory. The lateral movement of neurotransmitter receptors is emerging as an important mechanism in the control of synaptic transmission. However, our understanding of the spatial dynamics of membrane receptors at synapses has been limited largely because of a lack of appropriate tools to resolve single receptors in living cells. Fluorescent quantum dots represent promising probes to monitor individual synaptic receptors in living neural circuits. Bats and colleagues (Bats, Groc, and Choquet, *Neuron* 53, 719, 2007) used quantum dots to track the ins and outs of glutamate receptors at synapses, showing that the receptors bring company as they diffuse in the synapse to become trapped via their partner's local connections. Their study sheds new light on the mechanisms used by synapses to change their efficacy, which may impact on our understanding of the cellular and molecular basis of learning and memory. [DOI: 10.2976/1.2735016]**

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One of the greatest scientific challenges of our current century will be to understand how the human brain works. The task is daunting due to the enormous complexity of the brain. A hundred billion neurons and an order of magnitude more of non-neuronal cells make up the brain. Each neuron is interconnected by thousands of specialized, micron-size structures termed synapses, which are the principal substrates of neuronal communication. In a volume as little as a cubic millimeter or a microliter of brain tissue, tens of millions of synapses can be found. These numbers testify not only to the complexity of the brain, but also to the magnitude of the challenge of studying how neural circuits develop and adapt.

Indeed, changes in the biochemical, structural, and physiological properties of synapses are thought to support the development of neural circuits. In addition, they support changes in circuit properties that are believed to underlie the expression of learning and memory. At the biochemical level, synapses exchange information through neurotransmitter molecules released from one neuron (presynaptic side) and neurotransmitter receptor activation on another neuron (postsynaptic side) (Fig. 1). The binding of neurotransmitters to receptors then triggers to a variety of signaling events at synapses, leading to changes in membrane potentials that propagate throughout the postsynaptic neuron and neural circuits. The efficiency of individual synapses, which eventually determine the summated changes in membrane potential, can be controlled by the amount of neurotransmitter that is released or the amount of neurotransmitter receptors that are activated.

Neuroscientists have been actively investigating the mechanisms that control the amount of neurotransmitters and their receptors at synapses. At excitatory synapses, a major neurotransmitter is glutamate. One class of glutamate receptors, termed AMPA receptors, mediate most of the excitatory postsynaptic potentials that are eventually summated with those from many other synapses to trigger ac-

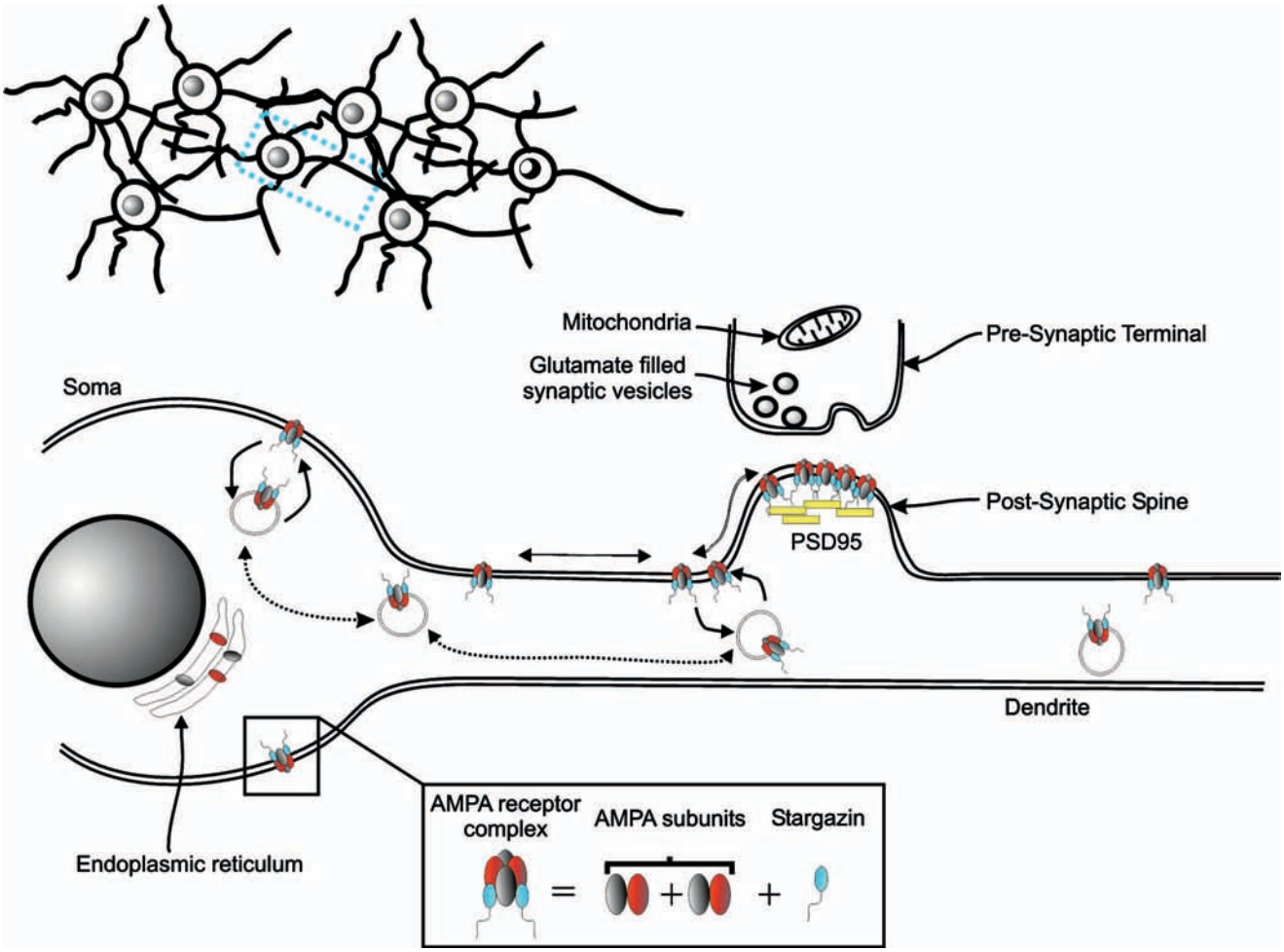


**Figure 1. Expression and targeting of AMPA receptors and stargazin to neuronal plasma membrane and synapses.**

tion potential firing in the postsynaptic neuron, leading to the spread of activity throughout circuits. Thus, the amount of AMPA receptors present at individual synapses is crucial for the efficiency of glutamatergic synapses and brain function. Furthermore, strong evidence suggests that changes in the number of synaptic AMPA receptors underlie activity-dependent changes in synaptic strength that are considered mechanistic underpinning of learning and memory (reviewed in Malinow and Malenka, 2002; Collingridge *et al.*, 2004).

But how are receptors recruited and maintained at synapses? Both constitutive and regulated receptor trafficking modes are important. Constant delivery and recycling of neurotransmitter receptors at synapses should support stability in transmission. Trapping mechanisms of receptors inside the synapse might ensure further stability. In contrast, rapid arrival and departure of receptors could be important for fast adaptation of synapses to presynaptic stimuli or postsynaptic excitability. Two principal modes of receptor delivery to synapses are being considered (reviewed in Choquet and Triller, 2003; Groc and Choquet, 2006): (1) exocytosis of synaptic receptor-containing vesicles at or near synaptic sites, and (2) trapping of receptors that arrived by diffusion from plasma membrane compartments located outside of the synapse. Both mechanisms likely work in concert, such that vesicles fuse near but outside of the synapse to which receptors diffuse and become trapped (Fig. 1). The necessity of vesicle fusion to supply synapses with AMPA receptors was demonstrated, for instance, by the introduction of toxins that block vesicle fusion (Lledo *et al.*, 1998). Evidence that receptors diffuse in and out of synapses within the plasma membrane is trickier to demonstrate. It requires the combination of sensitive imaging approaches as well as nifty probes.

## PROBING RECEPTOR DELIVERY WITH QUANTUM DOTS

Two groups from France have pioneered the use of fluorescent quantum dots (QDs) as probes to label the extracellular domain of individual glutamate or glycine receptors on the surface of neurons in culture (Dahan *et al.*, 2003; Groc *et al.*, 2004). QDs are direct-bandgap semiconductor nanocrystals that exhibit strong fluorescence at energies which depend

strongly on their size. QDs built with cadmium selenide (CdSe) exhibit tunable emission from approximately 480 to 610 nm as the diameter ranges from 2 to 8 nm, whereas CdTe QDs emit from 600 to 750 nm over the same size range (Michalet *et al.*, 2005). Longer wavelengths can be reached by using InP, InAs, or PbSe QDs. This wide range of emission spectra is attractive for multicolor imaging techniques. A number of reviews on the use of QDs for biological applications are available (e.g., Fu *et al.*, 2005; Alivisatos *et al.*, 2005). The major advantage of QDs for single molecule imaging is their remarkable photostability, allowing investigators to monitor a single QD for several minutes without significant photobleaching (Dahan *et al.*, 2003; Mansson *et al.*, 2004). However, for biological applications, QDs need to be rendered water soluble and biocompatible through various surface-coating methods. Two pioneering procedures appeared in 1998. One group coated QDs with a layer of silica to which biochemicals were subsequently bound (Bruchez *et al.*, 1998), while another group exchanged the initial organic ligands with thiolated carboxylic acids to which proteins were attached using a peptide bond (Chan and Nie, 1998). Since then, many groups have investigated other water-solubilizing strategies (Michalet *et al.*, 2005). Examples include using amphiphilic polymers (Wu *et al.*, 2003), dendrimers (Guo *et al.*, 2003), poly(ethylene) glycol (Billancia *et al.*, 2001), water-soluble phosphines (Kim and Bawendi, 2003), phospholipids (Dubertret *et al.*, 2002), peptides (Pinaud *et al.*, 2004), and a variety of thiolated compounds (Aldana *et al.*, 2001; Mattoussi *et al.*, 2000; Pathak *et al.*, 2001; Reiss *et al.*, 2001). Dubertret *et al.* (2002) were the first to show that QDs, via encapsulation in phospholipid micelles, could be introduced in cells for several days without detrimental toxicity effects.

QDs are now commercially available, the most common of which consist of a multilayer scheme of a CdSe or CdTe core, a ZnS protective shell, organic ligands, amphiphilic polymers and either a layer of streptavidin proteins (to which biotinylated biomolecules can be attached) or a layer of antibodies (to which antigens or other antibodies can be attached). To label individual receptors, they are first targeted with a primary antibody and then QDs bound to secondary antibodies are subsequently exposed to them. This method of labeling is less direct than the more common method of transfecting and overexpressing green fluorescent protein (GFP)-tagged proteins. It also leads to a larger tag than an organic fluorophore, such as an alexa or a cyanine. However, it has major advantages, in that it provides a higher signal and far longer observation times—both essential for single molecule tracking. Furthermore, it probes the native receptors, not an overexpressed form. A compromised approach, developed by Ting and colleagues (Howarth *et al.*, 2005), combines the use of transfection of the receptor of interest with more direct QD tagging. The approach involves the addition of a 15 aminoacid peptide on the extracellular portion of the receptor, which is then biotinylated by Escherichia coli biotin ligase added to the culture media. Biotinylated receptors are then probed with QDs coated with streptadivin. The development of mutant streptavidin that binds only one biotin (Howarth *et al.*, 2006) should reduce the possibility that individual QDs are linked to multiple receptors.

The advantages of QDs for monitoring synaptic receptor delivery was first demonstrated by Dahan and coworkers (Dahan *et al.*, 2003). They followed the diffusion of single glycine receptors in various synaptic domains (synaptic, perisynaptic, and extrasynaptic) of cultured neurons. The commercial streptavidin-QDs used in that study were labeled to glycine receptors by a primary antibody and biotinylated Fab fragments. Remarkably, individual QDs were observed inside the synaptic cleft by electron microscopy (being electron dense is another advantage of QDs), suggesting that despite their large size, it was possible for them to penetrate the cleft.

## TRAPPING AMPA RECEPTORS AT SYNAPSES

In a recent issue of *Neuron*, Bats *et al.* (2007) elegantly used tracking of QDs attached to AMPA receptors via an antibody to further our understanding of the trapping mechanisms of AMPA receptors at synapses. Two important proteins were known to play a major role in the synaptic accumulation of AMPA receptors, namely, PSD-95 and stargazin, but the underlying mechanisms were unknown. Stargazin is a member of the TARP family of proteins (for transmembrane AMPA receptor regulatory proteins). This protein is spontaneously mutated in stargazer mice that show absence epilepsy and cerebellar ataxia, probably because they lack sufficient AMPA receptors in the plasma membrane (Chen *et al.*, 2000). PSD-95 is a postsynaptic density (PSD) scaffolding protein at excitatory synapses, which associates with several synaptic components via specific binding motifs termed PDZ (reviewed in Kennedy, 2000).

Stargazin-AMPA receptor interactions were first shown to be required for proper packaging and delivery of the receptor to the plasma membrane (reviewed in Ziff, 2007). Bats *et al.* (2007) now show that stargazin is also important for trapping AMPA receptors into the synapse. By tracking QDs that were coated with secondary antimouse Fab fragments, which then bound to primary antibodies targeted to the AMPA receptor subunit GLUR1 (Fig. 2), they showed that receptor diffusion is reduced at synaptic sites compared to extrasynaptic sites. Synaptic sites can be monitored simultaneously by different means, such as with FM dyes, which load presynaptic terminals, or *Mitotracker* (Fig. 2), a marker for mitochondria that are particularly enriched in presynaptic terminals (Dahan *et al.*, 2003; Groc *et al.*, 2004). Here, the authors cotransfected GFP-tagged PSD95, which is largely concentrated postsynaptically, and showed that QD-tagged GLUR1 became less mobile when they reached and colocalized with a PSD95-GFP cluster. To show that stargazin is in-

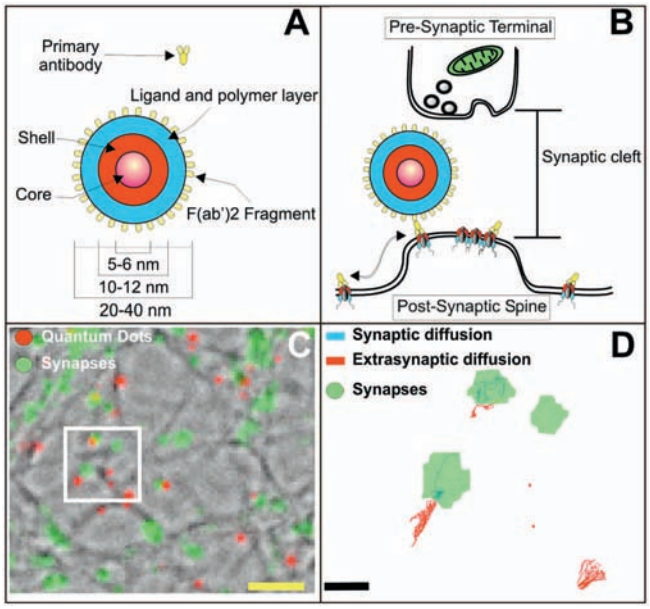


**Figure 2. Use of quantum dots to tag AMPA receptors.** (A) Typical functionalized quantum dot structure with polymer and antibody coating. (B) Labeling strategy: quantum dot is bound to primary antibodies attached to the extracellular domain of AMPA receptors. The size of the functionalized quantum dot is similar in magnitude to the synaptic cleft. The synapse can be labeled using multiple methods, including mitotracker green, which labels mitochondria. (C) Overlay of a light microscopy image of neuronal axons and dendrites grown in culture (Hudmon *et al.*, 2005), mitotracker green label of presynaptic terminals, and quantum dots targeted to AMPA receptors. Scale: 5 $\mu$m. (D) Tracking of quantum dots seen in the white box in (C). Some dots move in and out of synapses (S.L. and P.D.K. unpublished data). Scale 1 $\mu$m.

volved in this process, they used a stargazin mutant that lacks the last four amino acids of its intracellular tail. By overexpressing that mutant in neurons, they showed that AMPA receptor mobility was increased both synaptically and extrasynaptically, resulting in the disappearance of spontaneous synaptic currents (Chen *et al.*, 2000). In fact, they showed that even in non-neuronal cells, wild-type stargazin, but not the mutant, could cluster AMPA receptors with PSD95.

Why the last four amino acids? Because they encode a PDZ binding domain that would presumably attach to PDZ-domain-containing proteins, such as PSD95. To demonstrate that this interaction does indeed trap AMPA receptors at synaptic junctions, Bats *et al.* (2007) used a clever combination, first designed by Schnell and colleagues (Schnell *et al.*, 2002), of a stargazin mutant (T321F) that does not bind wild-type PSD95 and a PSD95 compensatory mutant that does bind to stargazin T321F. They could then show that AMPA receptor mobility was increased in neurons expressing stargazin T321F, but not when the compensatory PSD95 mutant was cotransfected.

AMPA receptors are generally formed by coassembly of multiple subunits, such as GLUR1 and GLUR2. GLUR2 also has a PDZ binding domain. Why then the need for the PDZ binding domain on stargazin to cluster AMPA receptors at synapses? The authors (Bats *et al.*, 2007) addressed this issue and showed that the GLUR2 C-terminal PDZ domain has a role in promoting surface expression of GLUR2, but not its stabilization at synaptic sites. Thus stargazin, through its PDZ-binding domain, can serve to stabilize AMPA receptors at synapses via an interaction with PSD95. This finding raised the question of whether AMPA receptors coming into the synapse by lateral diffusion are already tied to stargazin. The answer seems to be yes, since the authors showed that stargazin and AMPA receptors diffuse as complexes in the neuronal membrane. This was demonstrated by monitoring transfected HA-tagged stargazin with anti-HA-QDs and showing that their mobility was reduced at synaptic sites. More importantly, they showed that cross-linking transfected tagged GLUR2 with antibodies reduced the mobility of stargazin-GFP, assessed using fluorescence recovery after photobleaching (FRAP) techniques. Thus, it would seem that AMPA receptor delivery to synapses requires that auxiliary protein stargazin accompanies the receptor not only from the somatic factory (Ziff, 2007), but also from extra-synaptic membrane to the inside of the synaptic cleft.

## THE FUTURE OF QUANTUM DOT IMAGING FOR THE STUDY OF SYNAPTIC SIGNALING AND REMODELING

The study presented by Bats *et al.* convincingly showed that a lateral diffusion and trapping mechanisms can support the recruitment of AMPA receptors at synapses. It remains to be determined, however, whether this mechanism is involved in constitutive or regulated delivery of receptors to the synapse. For instance, could the increased transmission at glutamatergic synapses that is associated with long-term plasticity, learning, and memory be mediated by an increase in the synaptic trapping of diffusing receptors? In order to address the role of lateral diffusion and the trapping of receptors on synaptic plasticity, a major hurdle must be overcome: monitoring receptor dynamics in brain slice preparation. Indeed, much of our understanding of synaptic plasticity stems from work in slice preparations, where neural circuits resemble the native context and from which numerous models of long- and short-term plasticity have been developed. In addition, the local synaptic environment differs in a fresh brain slice compared to dissociated cultures. Thus, the diffusion of receptors in and out of synapses may be reduced or suppressed by elements surrounding the synapses, such as extracellular matrix components or astroglial feet, which might be missing in dissociated cultures.

Whether QDs will be the probe of choice for single receptor imaging studies in brain slice remains to be determined. To date, most neural cell labeling has taken advantage of commercial QDs. Their multilayer water-stabilizing structure, and coating with either secondary antibodies or strepta-

vidin results in a total size comparable to the size of the synaptic cleft (~20 nm, Fig. 2), or even larger. This large label, which then binds to receptors that have a primary antibody bound to their extracellular domain, may complicate quantitative analysis of diffusion and clustering in the confined space of the synaptic cleft. Even in dissociated culture, Groc and colleagues showed that receptor diffusion inside the synaptic cleft was significantly slowed down by QDs compared to organic probes (Groc *et al.*, 2004). To overcome this, smaller QDs, directly conjugated to primary antibodies, must be developed. However, smaller layers tend to result in less chemically stable QDs (Aldana *et al.*, 2005; Aldana *et al.*, 2001), thus limiting commercial availability. Collaborations between QD chemists and neurobiologists would drive the production of tailor-made QDs to overcome these size limitations and improve resolutions of synaptic protein dynamics.

Another limitation in using single QDs for tracking analyses is that, under continuous illumination, their fluorescence emission is intermittent, i.e., they blink on and off over a wide range of time scales (Bachir *et al.*, 2006; Heyes *et al.*, 2007; Kobitski *et al.*, 2004; Nirmal *et al.*, 1996; Shimizu *et al.*, 2001). Thus, when the QD is "off" it cannot be tracked. Sometimes, the on state can last up to several seconds, but patience is needed to wait for these events. Several researchers are investigating methods to reduce the blinking, but how they can be applied in live cells remains a challenge. On the other hand, the blinking properties of QDs offer an interesting advantage; it allows the experimentalist to recognize that the measured signal indeed comes from a single QD and not from a cluster of QDs, since it is statistically unlikely that a cluster would completely "turn off."

Therefore, the development of analytical approaches to overcome the problem of blinking is a worthwhile avenue. For instance, Bachir *et al.* (2006) developed an analytical method that does not follow QDs one by one, but instead measures correlations in the intensity fluctuations within images both temporally and spatially. Image correlation methods have been used to follow clustering of cell surface receptors and to extract their diffusion times and flow speeds (Hebert *et al.*, 2005; Wiseman *et al.*, 2004). Recent advances in the image correlation techniques have also involved the development of a new reciprocal space approach called kICS (Kolin *et al.*, 2006). The advantage of kICS is that it effectively separates the intensity fluctuations due to photophysics (such as blinking and photobleaching) from those due to transport and hence provides an unbiased measure of diffusion or flow for the labeled complex. Adapting these types of analytical approaches to the complex dimensions of neurons and synapses will constitute an important development.

## CONCLUDING REMARKS

The study of Bats *et al.* (2007) has paved the way to an exciting path in synaptic signaling research, which may reveal fundamental mechanisms of synaptic plasticity. Indeed, the dynamic exchange between diffusing extrasynaptic neurotransmitter receptors and their trapping at synaptic sites constitutes an appealing mechanism for gating rapid changes in synaptic transmission. Furthermore, their study reinforces that in order to understand cellular function, it is critical that we do not restrict ourselves at quantifying how much of a protein is expressed in a cell, but also precisely where in the cell and for how long. The use of QDs as probes for monitoring protein dynamics in cells is still a young approach. Chemists and physicists have demonstrated a strong interest in the development of QDs as improved probes or sensors of chemical or biological mechanisms. For neuroscientists, concerned with resolving molecular mechanisms at the subcellular level all the way down to synapses, QDs represent very promising tools, which have already proven successful in some studies. Collaborative work is needed, however, between chemists, physicists, and biologists to push the limits of what QDs can offer to answer biological questions. Indeed, as the surface chemistry of QDs improves, we might expect that cellular biologists will use them to tag all sorts of targets, both outside and inside the cell. As such, we should appreciate further the spatial and temporal dynamics of molecular signals in cells.

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## REFERENCES

Aldana, J, Lavelle, N, Wang, Y, and Peng, X (2005). "Size-dependent dissociation pH of thiolate ligands from cadmium chalcogenide nanocrystals." *J. Am. Chem. Soc.* **127**, 2496–2504.

Aldana, J, Wang, YA, and Peng, X (2001). "Photochemical instability of CdSe nanocrystals coated by hydrophilic thiols." *J. Am. Chem. Soc.* **123**, 8844–8850.

Alivisatos, AP, Gu, W, and Larabell, C (2005). "Quantum dots as cellular probes." *Annu. Rev. Biomed. Eng.* **7**, 55–76.

Bachir, A, Durisic, NBH, Grutter, P, and Wiseman, PW (2006). "Characterization of blinking dynamics in quantum dot ensembles using image correlation spectroscopy." *J. Appl. Phys.* **99**, 064503.

Bats, C, Groc, L, and Choquet, D (2007). "The interaction between stargazin and PSD-95 regulates AMPA receptor surface trafficking." *Neuron* **53**, 719–734.

Billancia, G, Wouters, D, Precup, AA, and Schubert, US (2001). "Towards functionalized nanoparticles." *Polym. Mater. Sci. Eng.* **85**, 508–509.

Bruchez, M, Moronne, M, Gin, P, Weiss, S, and Alivisatos, AP (1998). "Semiconductor nanocrystals as fluorescent biological labels." *Science* **281**, 2013–2016.

Chan, WC, and Nie, S (1998). "Quantum dot bioconjugates for ultrasensitive nonisotopic detection." *Science* **281**, 2016–2018.

Chen, L, Chetkovich, DM, Petralia, RS, Sweeney, NT, Kawasaki, Y, Wenthold, RJ, Bredt, DS, and Nicoll, RA (2000). "Stargazin regulates synaptic targeting of AMPA receptors by two distinct mechanisms." *Nature (London)* **408**, 936–943.

Choquet, D, and Triller, A (2003). "The role of receptor diffusion in the

organization of the postsynaptic membrane." *Nat. Rev. Neurosci.* **4**, 251–265.
Collingridge, GL, Isaac, JT, and Wang, YT (2004). "Receptor trafficking and synaptic plasticity." *Nat. Rev. Neurosci.* **5**, 952–962.
Dahan, M, Lévi, S, Luccardini, C, Rostaing, P, Riveau, B, and Triller, A (2003). "Diffusion dynamics of glycine receptors revealed by single-quantum dot tracking." *Science* **302**, 442–445.
Dubertret, B, Skourides, P, Norris, DJ, Noireaux, V, Brivanlou, AH, and Libchaber, A (2002). "In vivo imaging of quantum dots encapsulated in phospholipid micelles." *Science* **298**, 1759–1762.
Fu, A, Gu, W, Larabell, C, and Alivisatos, AP (2005). "Semiconductor nanocrystals for biological imaging." *Curr. Opin. Neurobiol.* **15**, 568–575.
Groc, L, and Choquet, D (2006). "AMPA and NMDA glutamate receptor trafficking: multiple roads for reaching and leaving the synapse." *Cell Tissue Res.* **326**, 423–438.
Groc, L, Heine, M, Cognet, L, Brickley, K, Stephenson, FA, Lounis, B, and Choquet, D (2004). "Differential activity-dependent regulation of the lateral mobilities of AMPA and NMDA receptors." *Nat. Neurosci.* **7**, 695–696.
Guo, W, Li, JJ, Wang, YA, and Peng, X (2003). "Conjugation chemistry and bioapplications of semiconductor box nanocrystals prepared via dendrimer bridging." *Chem. Mater.* **15**, 3125–3133.
Hebert, B, Costantino, S, and Wiseman, PW (2005). "Spatiotemporal image correlation spectroscopy (STICS) theory, verification, and application to protein velocity mapping in living CHO cells." *Biophys. J.* **88**, 3601–3614.
Heyes, CD, Kobitski, AYu, Breus, VV, and Nienhaus, GU (2007). "Effect of the shell on blinking statistics in single core-shell quantum dots—a single particle fluorescence study." *Phys. Rev. B* **75**, 125431.
Howarth, M, Chinnapen, DJ, Gerrow, K, Dorrestein, PC, Grandy, MR, Kelleher, NL, El Husseini, A, and Ting, AY (2006). "A monovalent streptavidin with a single femtomolar biotin binding site." *Nat. Methods* **3**, 267–273.
Howarth, M, Takao, K, Hayashi, Y, and Ting, AY (2005). "Targeting quantum dots to surface proteins in living cells with biotin ligase." *Proc. Natl. Acad. Sci. U.S.A.* **102**, 7583–7588.
Hudmon, A, Lebel, E, Roy, H, Sik, A, Schulman, H, Waxham, MN, and De Koninck, P (2005). "A mechanism for $Ca^{2+}$/calmodulin-dependent protein kinase II clustering at synaptic and nonsynaptic sites based on self-association." *J. Neurosci.* **25**, 6971–6983.
Kennedy, MB (2000). "Signal-processing machines at the postsynaptic density." *Science* **290**, 750–754.
Kim, S, and Bawendi, MG (2003). "Oligomeric ligands for luminescent and stable nanocrystal quantum dots." *J. Am. Chem. Soc.* **125**, 14652–14653.
Kobitski, AYu, Heyes, CD, and Nienhaus, GU (2004). "Total internal reflection fluorescence microscopy—a powerful tool to study single quantum dots." *Appl. Surf. Sci.* **234**, 86–92.
Kolin, DL, Ronis, D, and Wiseman, PW (2006). "*k*-Space image correlation spectroscopy (kICS): A method for accurate transport measurements independent of fluorophore photophysics." *Biophys. J.* **91**, 3061–3075.
Lledo, PM, Zhang, X, Sudhof, TC, Malenka, RC, and Nicoll, RA (1998). "Postsynaptic membrane fusion and long-term potentiation." *Science* **279**, 399–403.
Malinow, R, and Malenka, RC (2002). "AMPA receptor trafficking and synaptic plasticity." *Annu. Rev. Neurosci.* **25**, 103–126.
Mansson, A, Sundberg, M, Balaz, M, Bunk, R, Nicholls, IA, Omling, P, Tagerud, S, and Montelius, L (2004). "In vitro sliding of actin filaments labelled with single quantum dots." *Biochem. Biophys. Res. Commun.* **314**, 529–534.
Mattoussi, H, Mauro, JM, Goldman, ER, Anderson, GP, Sundar, VC, Mikulec, FV, and Bawendi, MG (2000). "Self-assembly of CdSe-ZnS quantum dot bioconjugates using an engineered recombinant protein." *J. Am. Chem. Soc.* **122**, 12142–12150.
Michalet, X, Pinaud, FF, Bentolila, LA, Tsay, JM, Doose, S, Li, JJ, Sundaresan, G, Wu, AM, Gambhir, SS, and Weiss, S (2005). "Quantum dots for live cells, in vivo imaging, and diagnostics." *Science* **307**, 538–544.
Nirmal, M, Dabbousi, BO, Bawendi, MG, Maklin, JJ, Trautman, JK, Harris, TD, and Brus, LE (1996). "Fluorescence intermittency in single cadmium selenide nanocrystals." *Nature (London)* **383**, 802–804.
Pathak, S, Choi, S-K, Arnheim, N, and Thompson, ME (2001). "Hydroxylated quantum dots as luminescent probes for in situ hybridization." *J. Am. Chem. Soc.* **123**, 4103–4104.
Pinaud, F, King, D, Moore, H-P, and Weiss, S (2004). "Bioactivation and cell targeting of semiconductor Cdse/ZnS nanocrystals with phytochelatin-related peptides." *J. Am. Chem. Soc.* **126**, 6115–6123.
Reiss, P, Bleuse, J, and Pron, A (2001). "Highly luminescent CdSe/ZnSe core/shell nanocrystals of low size dispersion." *Nano Lett.* **2**, 781–784.
Schnell, E, Sizemore, M, Karimzadegan, S, Chen, L, Bredt, DS, and Nicoll, RA (2002). "Direct interactions between PSD-95 and stargazin control synaptic AMPA receptor number." *Proc. Natl. Acad. Sci. U.S.A.* **99**, 13902–13907.
Shimizu, KT, Neuhauser, RG, Leatherdale, CA, Empedocles, SA, Woo, WK, and Bawendi, MG (2001). "Blinking statistics in single semiconductor nanocrystal quantum dots." *Phys. Rev. B* **63**, 205316.
Wiseman, PW, Brown, CM, Webb, DJ, Hebert, B, Johnson, NL, Squier, JA, Ellisman, MH, and Horwitz, AR (2004). "Spatial mapping of integrin interactions and dynamics during cell migration by image correlation microscopy." *J. Cell. Sci.* **117**, 5521–5534.
Wu, XY, Liu, HJ, Liu, JQ, Haley, KN, Treadway, JA, Larson, JP, Ge, NF, Peale, F, and Bruchez, MP (2003). "Immunofluorescent labeling of cancer marker Her2 and other cellular targets with semiconductor quantum dots." *Nat. Biotechnol.* **21**, 41–46.
Ziff, EB (2007). "TARPs and the AMPA receptor trafficking paradox." *Neuron* **53**, 627–633.