Rapid progress has been made in understanding the mechanisms of fast transmission mediated by the AMPA type of glutamate channel at central synapse. However, several important aspects of this process remain unclear, including the spatial distribution of receptors activated by a single vesicle release, the interactions that occur when multiple vesicles are released and the functional role of desensitization. Previous models have relied on the kinetic schemes matched to recordings of mEPSCs in CA3 mossy fiber inputs (Jonas et al. 1993). Since then, technically superior dendritic recordings of mEPSCs evoked by sucrose application near the recording electrode have been obtained (Magee and Cook 2000). Remarkably, the rise time of these EPSCs can be 50 μs, ~10 times faster than found earlier. This is a very important point, since the rise time is one of the major data constraints on models of mEPSCs. We study the origin of this rapid rise time using a simple analytic model of the receptor that is valid at early times.

To understand the detailed events in the cleft and to build a framework for understanding experimental data, we constructed a new model of AMPA receptors. Our kinetic model for AMPA receptors is based on recent evidence that channel opening depends cooperatively on the four subunits of the channel (Rosenmund et al. 1998, Smith and Howe, 1999): the higher the number of subunits that bind glutamate, the larger the average open channel conductance and the faster the transition from the closed to the open state. We adjusted the rate constants in our model to optimally fit data obtained by the controlled application of glutamate to excised patches (Andrasfalvy and Magee, 2001). We then constructed a biophysically realistic model of a CA1 hippocampal synapse using known structural constraints. To predict the waveform of the mEPSC, we simulated diffusion of a quantum of glutamate (2000 molecules) out of the vesicle, its spread within the synaptic cleft and the resulting activation of AMPA receptors using a Monte Carlo model. The simulated mEPSC matches the amplitude and rise-time of the experimentally recorded responses.

At the peak of the response, open channels have mostly 3 or 4 glutamate molecules bound and occur in a small hotspot having dimensions similar to the glutamate spike (80% of the current is carried by channels in a 250 nm diameter region). This is quite small compared to many CA1 synapses, which can have diameters of 400-1000 nm. Given measured densities of AMPA receptors on CA1 mushroom spines (~ 600 channels/μm^2), we estimate that the hot-spot contains about 35 channels of which about half (i.e. 15-20 channels) are open at the peak. This confirms the findings that AMPA receptors are not saturated by the release of glutamate from single vesicles. The peak mEPSC is Gaussian distributed with a CV of 0.18 which agrees with experimental observations.

We investigated other factors that determine the size of the response. The response scaled almost linearly with channel density and with the number of glutamate molecules contained in the vesicles. On the other hand, adding channels far from the site of vesicle release had very little effect on the size of the mEPSC. We then studied the role of desensitization of the AMPA receptor in producing spatially restricted receptor activation by simulating the action of cyclothiazide, which blocks desensitization. We found that desensitization blocks the opening of channels that are far from the site of vesicle release, terminating the response and reducing the variability. Thus, we were able

to account for experimental observations where blocking desensitization has relatively little effect on early currents while more strongly increasing the falling phase to which spatially diffuse receptors contribute. We were also able to account for another effect of blocking desensitization: increasing the apparent probability of response at synapses with low probability of response without an increase in the actual probability of vesicle release. If synapses are "partially silent", i.e. contain regions with and without AMPA channels, vesicles released at regions lacking AMPA channels will normally activate distant AMPA channels so weakly that the response is below recording noise; however, after addition of drug, distant activation is much more effective, making the response detectable.

It is also of interest to consider how responses to two vesicles interact since there is increasing evidence for multivesicular release at many types of CNS synapses including CA1 synapses. Amplitude histograms of evoked responses recorded from one or a small number of synapses often have evenly spaced peaks, indicating linear summation of quantal events, but it has been unclear whether linear summation could occur at single synapses, given the nonlinear activation of AMPA channels. To explore this issue, we simulated two simultaneous vesicle fusion events (200 nm separation) at the same synapse. The summation was virtually linear because the channels activated by each vesicle were almost completely non-overlapping. The linearity of responses to multiple vesicles accounts for several additional properties of quantal responses. The implication is that large synapses act as if they have multiple, independent modules of AMPA-mediated transmission. We also modeled the activation of NMDA channels to fit the amplitude and noise of the EPSC (Liu et al. 1999) in zero Mg and then studied the effect of single and multivesicular release on the AMPA/NMDA ratio.

These results have important implications for models of synaptic plasticity and quantal analysis. Because of the localization of transmitter action, additional AMPA channels can be added to a synapse without substantially affecting quantal size. Quantal size, thus, does not reflect the total number of AMPA channels, but rather reflects the local density. This requires a revision of our understanding of how synaptic strength is determined and measured. A further implication is that synapses can have a wide dynamic range. The response to multiple vesicles can summate linearly, even though the receptors themselves are highly nonlinear. Recent work shows that AMPA responses generated at different dendritic locations on CA1 pyramidal neurons summate linearly and our results now suggest that this linearity extends even to multiple vesicles released at a single synapse.