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Synaptic Plasticity: Close Encounters of the Tonic and Phasic Kind

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Neuronal circuits have the capacity to maintain relatively constant activity in the face of perturbations that alter excitability or synaptic properties. A new study demonstrates that different classes of neurons co-innervating the same postsynaptic target express homeostatic plasticity with unique presynaptic features.

Neurons can change their connection strength with partner cells using a host of mechanisms that modulate synaptic communication. Some forms of plasticity strengthen or weaken synaptic connections to alter specific routes of information transfer across circuits to support learning and behavioral adaptation. In contrast, homeostatic synaptic plasticity ensures a global stabilization of neuronal communication by offsetting changes to synaptic strength that maintains equilibrium across the network. For example, reductions in postsynaptic receptor function can be offset by increasing presynaptic output through a process known as presynaptic homeostatic potentiation (PHP). PHP is induced by the affected postsynaptic cell through a retrograde signal that activates presynaptic mechanisms to increase synaptic vesicle fusion and offset the reduction in postsynaptic response. It is unclear if an individual postsynaptic cell can differentially induce PHP across potentially hundreds of distinct presynaptic partners. In a new study reported in this issue of *Current Biology*, Genç and Davis tested whether PHP in *Drosophila* is differentially expressed by two distinct classes of presynaptic neurons that innervate the same postsynaptic cell [1].

The *Drosophila* neuromuscular junction (NMJ) is an established system for studying synapse biology [2,3], including acute and chronic forms of PHP. The wasp venom philanthotoxin (PhTx) blocks postsynaptic glutamate receptors, significantly reducing the response to released neurotransmitters. Within minutes, the reduction in postsynaptic response to a single synaptic vesicle (quantal size) is offset by a presynaptic increase in the number of synaptic vesicles released per action potential (quantal content) such that overall synaptic strength returns to its original level [4]. This homeostatic increase in quantal content can also be expressed chronically. Mutations in the glutamate receptor subunit GluRIIA reduce quantal size throughout development, and the presynaptic terminal compensates by increasing quantal content [4,5].

Most *Drosophila* muscles are innervated by two motor neuron classes with different release properties — a tonic MN-1b neuron and a phasic MN-1s neuron. Genç and Davis used MN-1s- and MN-1b-specific GAL4 drivers to individually manipulate the two motor neurons [6]. By driving channelrhodopsin in a cell-type specific manner, the authors were able to use light to induce action potentials in each of the motor neurons individually and record the size of the postsynaptic response. They found that MN-1s has greater presynaptic output than MN-1b at low external Ca^{2+} (0.3 mM), but generates a similar response at higher Ca^{2+} (3 mM). These observations indicate that the MN-1s connection has higher Ca^{2+} sensitivity than MN-1b (Figure 1A), consistent with previous studies [7,8]. Next, the authors tested whether acute PhTx-induced PHP is expressed at MN-1b and MN-1s terminals. In low Ca^{2+} conditions, the MN-1s neuron robustly expressed PHP, while MN-1b weakly responded. When the authors raised the Ca^{2+} to 3 mM,

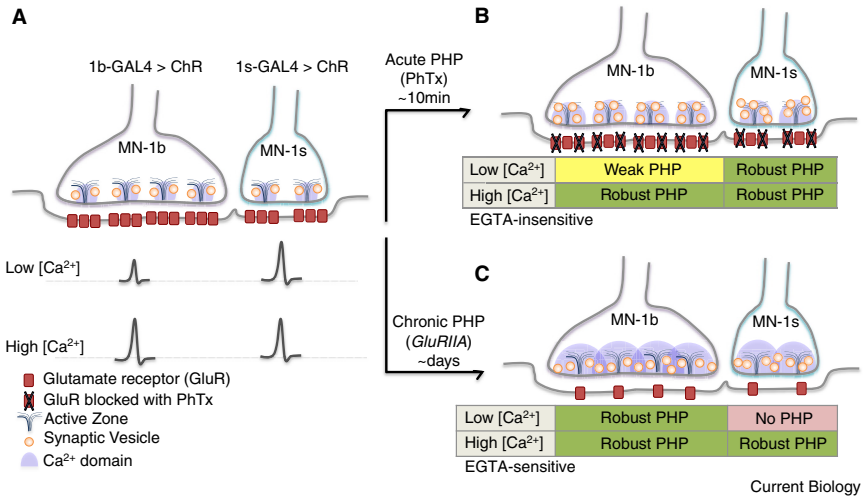

Figure 1. Tonic and phasic motor neurons differentially contribute to synaptic strength and homeostatic plasticity.

(A) Motor neurons of the MN-1b and MN-1s class co-innervate peripheral muscles. GAL4 drivers specific for each class allow independent activation using channelrhodopsin (ChR). Electrophysiological recordings from postsynaptic muscles reveal differential strength and Ca^{2+} sensitivity between the two connections.

(B) Acute presynaptic homeostatic potentiation (PHP) is expressed in presynaptic motor neurons within 10 minutes after application of the glutamate receptor blocker PhTx. In low extracellular Ca^{2+}, the 1s neuron dominates the homeostatic response, with minor contributions from 1b. In elevated Ca^{2+}, both terminals robustly express PHP and are EGTA-insensitive.

(C) Chronic PHP is expressed presynaptically to offset the reduction in quantal size in animals with mutated GluRIIA. The 1b neuron robustly expresses PHP across the range of extracellular Ca^{2+}, but the 1s neuron only expresses PHP in high Ca^{2+} conditions. Presynaptic release in GluRIIA mutants is EGTA-sensitive, suggesting active zone organization is altered in neurons following chronic PHP.

both terminals robustly expressed PHP (Figure 1B), potentially reflecting differences in Ca^{2+} sensitivity of release between the two neurons.

To determine whether both motor neurons participate in chronic PHP, the authors measured release from MN-1s and MN-1b in animals carrying a mutation in the postsynaptic GluRIIA receptor. In low external Ca^{2+}, the MN-1s motor neuron did not participate in chronic expression of PHP, while MN-1b displayed robust PHP, consistent with prior Ca^{2+} imaging data [8]. However, when the authors assayed release in high Ca^{2+}, both MN-1b and MN-1s robustly expressed PHP (Figure 1C). The participation of MN-1s in chronic PHP only in high Ca^{2+} cannot be easily explained by differences in baseline release properties, given MN-1s participates in acute PHP in both low and high Ca^{2+} conditions. These findings indicate PHP can be differentially expressed between neurons co-innervating a target cell depending on the method of PHP induction and external Ca^{2+}.

The observation that acute and chronic PHP are expressed differentially at MN-1s and MN-1b in a Ca^{2+}-dependent manner motivated the authors to probe the architecture of presynaptic Ca^{2+} usage during both forms of plasticity. One mechanism to increase presynaptic release is to change the coupling distance between synaptic vesicles and the source of Ca^{2+} influx [9]. To test whether PHP involves a change in this coupling distance, the authors used the slow Ca^{2+} chelator EGTA to reduce Ca^{2+} available to loosely coupled vesicles. Terminals expressing acute PHP were insensitive to EGTA, indicating that fast changes in vesicle–Ca^{2+} channel coupling are not playing a role in this form of plasticity. In contrast, both MN-1s and MN-1b synapses were EGTA-sensitive during chronic PHP, suggesting an additional pool of loosely coupled vesicles is recruited during the expression of chronic PHP. One presynaptic mechanism contributing to PHP is the rapid insertion of new epithelial Na^{+} channels (PPKs) that depolarize the membrane, enhancing the amount of Ca^{2+} influx into the presynaptic terminal [10]. In chronic PHP, pharmacological inhibition of PPKs in the presence of EGTA did not further reduce release, suggesting that the increased Ca^{2+} influx promoted by PPK channels is involved in supplying Ca^{2+} to loosely coupled vesicles.

In summary, these data indicate that acute forms of homeostatic plasticity are more robust at phasic synapses, especially at lower Ca^{2+}, at *Drosophila* NMJs. In contrast, chronic homeostatic plasticity induced by genetic deletion of a glutamate receptor subunit is more robust at tonic synapses. The differential sensitivity to EGTA argues that the transition from acute to chronic homeostatic plasticity is likely to involve an alteration in the morphology, structure or spacing of active zones that leads to more loosely-coupled synaptic vesicles available for release. Active zone remodeling is supported by recent studies that have identified structural changes in *Drosophila* active zones during PHP [11,12]. The failure of MN-1s neurons to express chronic PHP in low Ca^{2+} conditions could reflect differences in the nano-architecture of active zones between MN-1b and MN-1s that allow differential recruitment of loosely coupled vesicles. Many interesting questions remain, including the nature of homeostatic retrograde signals. Likewise, whether the specificity of PHP expression can be driven by compartmentalization in the release of postsynaptic retrograde signals or only by differences in the unique presynaptic responses mediated by different neuronal classes innervating the same postsynaptic partner is unclear.

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