Coexistence of Muscarinic Long-Term Depression With Electrically Induced Long-Term Potentiation and Depression at CA3–CA1 Synapses

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INTRODUCTION

Cholinergic input is known to be critical for normal functioning of hippocampal synapses because hippocampal-dependent learning and memory are impaired in animals with cholinergic lesions (Berger-Sweeney 2001; Gold 2003; Wrenn et al. 1999), and degeneration of cholinergic input from the medial septum to hippocampus is one of the hallmarks of Alzheimer’s disease (Collerton 1986; Kasa 1997; McKinney and Jacksonville 2005; Schliebs 2005). However, much remains to be understood regarding the mechanisms by which acetylcholine and muscarinic receptors modify hippocampal function.

Our laboratory recently reported that a form of long-term depression (LTD) at CA3–CA1 synapses is induced by activation of M1 muscarinic acetylcholine receptors (mAChRs) and is thus termed muscarinic LTD (mLTD). mLTD is both activity and N-methyl-D-aspartate receptor (NMDAR) dependent (Scheiderer et al. 2006), characteristics shared by forms of synaptic plasticity believed to be substrates of learning and memory at the cellular level, including long-term potentiation induced by high-frequency stimulation (HFS-LTP) and long-term depression induced by low-frequency stimulation (LFS-LTD) at CA3–CA1 synapses (Braunewell and Manahan-Vaughan 2001; Malenka 1999; Tonegawa et al. 1996). These electrically induced forms of synaptic plasticity can also occur sequentially over time (Bear 2003; Dudek and Bear 1993; Mulkey and Malenka 1992). This is important because for synapses to remain plastic and allow learning to occur, they must be alternately potentiated and depressed, likely by different mechanisms (Abraham and Bear 1996; Abraham and Tate 1997; Bear 2003). Although mLTD is activity and NMDAR dependent, it remains unclear whether mLTD can participate in the dynamic interplay of synaptic potentiation and depression believed to mediate learning and memory at the cellular level. Therefore we tested whether mLTD can depotentiate LTP and whether mLTD expression will interfere with subsequent induction of LTD. In addition, we investigated the interplay between mLTD and LFS-LTD to determine whether induction of one form of NMDAR-dependent LTD will influence the ability of synapses to undergo induction and expression of another form of LTD also dependent on NMDAR activation.

METHODS

Slice preparation and electrophysiology

All experiments in this study were conducted with an approved protocol from the University of Alabama at Birmingham Institutional Animal Care and Use Committee, in compliance with National Institutes of Health guidelines. Hippocampal slices (400 μm) were prepared from 3- to 4-week-old Sprague–Dawley rats using standard methods (Scheiderer et al. 2004). Rats were anesthetized with halothane or isoflurane and decapitated. Their brains were removed and placed in ice-cold “high-sucrose” artificial cerebrospinal fluid (aCSF) containing (in mM): NaCl 85; KCl 2.5; MgSO4 4; CaCl2 0.5; NaH2PO4 1; NaHCO3 26; and glucose 10, kynurenic acid 2; and ascorbate 0.5. This aCSF was used during the slicing procedure because its low Na+ and Ca2+ and high-sucrose content promote neuronal survival. Coronal slices of the dorsal hippocampus were cut with a vibratome (Vibratome, St. Louis, MO). Hippocampal slices were incubated for 30 min in high-sucrose aCSF and then for 30 min in a standard aCSF containing (in mM): NaCl 119; KCl 2.5; CaCl2 2.5; MgSO4 1.3; NaH2PO4 1; NaHCO3 26; and glucose 10, kynurenic acid 2 saturated with 95% O2-5%CO2 (pH 7.4). To make recordings,
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Richard Mooney, Duke University). The fEPSP slope was measured and plotted versus time, with each point representing the average of five raw data points. Only experiments with <5% change in the fEPSP slope during the baseline were included in the averages. Student’s t-test or one-way ANOVA was used to assess statistical significance between groups, with the significance level set at $P < 0.05$. Data are presented as means ± SE. LTP and LTD were measured at 30 min after washout of the agonist or application of electrical stimulation.

RESULTS

We recently reported (Scheiderer et al. 2006), and illustrate in Fig. 1A, that application of the cholinergic agonist CCh (50 μM, 10 min) causes an acute depression of hippocampal CA3–CA1 field excitatory postsynaptic potentials (fEPSPs), which is followed by a long-term depression on washout of agonist (mLTD, 80 ± 3% of baseline fEPSP slope, $n = 4, P < 0.01$). This form of LTD, which is also observed in visual (Kirkwood et al. 1999) and perirhinal cortex (Massey et al. 2001), is activity and NMDAR dependent and requires activation of M1 mACHRs (Scheiderer et al. 2006). In contrast, the acute depression that occurs during CCh application results from activation of some other subtype of mACHR, likely M3 (Scheiderer et al. 2006).

Prior expression of mLTD does not alter the magnitude of LTP

To determine whether mLTD expression interferes with subsequent induction of LTP, we applied HFS after 40 min of mLTD expression and found that the fEPSP can be potentiated (Fig. 1A, mLTD: 80 ± 3% of baseline fEPSP slope, $n = 4, P < 0.01$; LTP: 138 ± 8% of fEPSP slope just before HFS, $n = 4, P < 0.01$). Importantly, the potentiation induced returned the fEPSP slope to the original baseline level. In these experiments, because HFS was applied after a prolonged period of recording, it was important to investigate whether a general decline in slice health could influence the magnitude of LTP independent of any effect caused by application of CCh to induce mLTD. Thus to be able to compare the magnitude of LTP elicited in naïve slices (not exposed to CCh) with that elicited after mLTD expression, we performed control experiments in which a 70-min baseline was obtained before application of HFS (four 0.5-s trains of 100-Hz stimuli separated by 10 s). In these experiments, LTP was successfully induced (Fig. 1B, 124 ± 3% potentiation of baseline fEPSP slope, $n = 3, P < 0.02$). In fact, there was no statistical difference in the magnitude of LTP expressed in slices after expression of mLTD compared with that induced in naïve control slices (Fig. 1C, $P > 0.4$). These data demonstrate that LTP can be induced in hippocampal slices expressing mLTD.

mLTD can be induced at potentiated synapses

Next, we tested whether mLTD can be induced after expression of LTP. In this set of experiments, two independent synaptic pathways were stimulated within the same slice so that we could compare the magnitude of mLTD induced in the pathway receiving HFS with that induced in an untetanized pathway (Fig. 1D, tetanized pathway: 145 ± 7% of baseline fEPSP slope, $n = 4, P < 0.01$; control pathway: 97 ± 4%, $n =
Forty minutes after HFS application, CCh was applied to induce mLTD. When compared with the level of transmission just before CCh application, the amount of mLTD in the tetanized pathway did not differ significantly from that in the untetanized control pathway (Fig. 1, D and E; tetanized pathway: 67 ± 11% of fEPSP just before CCh application, n = 4 vs. control pathway: 72 ± 13% of baseline fEPSP slope, n = 4, P > 0.4), indicating that irrespective of whether synapses are potentiated, mLTD of equal magnitude is induced. Notably, the fEPSP slope was returned to the original baseline when mLTD was induced in the pathway with prior LTP expression.

An important consideration in these experiments is that mLTD may have been induced only at synapses in the tetanized pathway that failed to undergo LTP induction. Therefore as a further test of whether potentiated synapses can be depotentiated by mLTD, we applied two rounds of HFS to ensure that LTP was saturated; saturation occurred after the first tetanus (Fig. 1F, 158 ± 6% of baseline fEPSP slope after second tetanus, n = 4, P > 0.09). After saturating LTP, mLTD could still be induced (Fig. 1F, 68 ± 4% of fEPSP slope just before CCh application, n = 4, P < 0.01). As before, induction of mLTD returned transmission to a level not significantly different from baseline (P > 0.8), demonstrating that mLTD can depotentiate synapses expressing LTP. Remarkably, the magnitude of mLTD induced very soon after saturating LTP (Fig. 1F, CCh application 6 min after the second tetanus) does not differ from that induced at a time point when LTP should be consolidated (Fig. 1D, CCh application 40 min after tetanus, P > 0.8), and when it is difficult to induce depotentiation with LFS because LFS-induced depotentiation becomes less effective as the time from tetanus increases (Barrionuevo et al. 1980; Fujii et al. 1991; O’Dell and Kandel 1994; Staubli and Chun 1996). Importantly, Fig. 1F also shows that a third tetanus applied during mLTD expression reinstates the LTP to the previously saturated level (Fig. 1F, third tetanus: 140 ± 13% of the fEPSP slope just before HFS; second tetanus: 149 ± 8% of baseline fEPSP slope, P > 0.2). Even though mLTD is not saturated, the fact that mLTD returns transmission to the original baseline and that the third tetanus returns potentiation to the saturated level of LTP suggests that at least some fraction of the synapses is undergoing bidirectional plasticity and that mLTD is reversible.

After mLTD saturation, LFS-LTD is both NMDAR and mGluR independent

Our next experiments were designed to investigate how mLTD interacts with LFS-LTD. We hypothesized that because both forms of LTD are activity and NMDAR dependent, they might be mechanistically similar and thus occlude one another. Before testing the idea that prior saturation of LFS-LTD would prevent further induction of mLTD, it was first determined in separate experiments that LFS-LTD is saturated by three rounds of LFS applied at 20-min intervals. After saturation of LFS-LTD, CCh (50 μM) was applied to induce mLTD. As expected, prior saturation of LFS-LTD occludes induction of mLTD (Fig. 2A, 56 ± 5% of baseline fEPSP slope after third application of LFS vs. 54 ± 5% of baseline fEPSP slope after CCh application, n = 4, P > 0.4). We performed the converse experiment by repeatedly applying CCh (50 μM, 10 min; three to seven applications spaced 20 min apart) until mLTD was saturated and no further depression could be elicited by CCh. The maximum level of mLTD that could be induced varied across experiments, such that some slices required more applications of CCh than others to saturate the depression. Thus mLTD was considered saturated (and the experiment included in the final analysis) only if the amount of depression after the
Because LFS-LTD induced at synapses that previously expressed saturated levels of mLTD is no longer dependent on NMDARs, its induction must require some alternate mechanism. Activation of mGluRs was an obvious possibility because mGluRs can also mediate depression at CA3–CA1 synapses, even LTD induced by certain protocols of LFS (Bolshakov and Siegelbaum 1994; Huber et al. 2001; Oliet et al. 1997; Palmer et al. 1997). Furthermore, a change in the mGluR and NMDAR dependence of electrically induced LTD may occur during development (Kemp and Bashir 1997, 1999; Kemp et al. 2000), and activation of mGluRs can induce metaplasticity that alters subsequent electrically induced plasticity (Bortolotto et al. 1994; Cohen et al. 1998; van Dam et al. 2004). The control experiments shown in Fig. 2D as well as previously published reports using similar LFS protocols (Dudek and Bear 1992; Selig et al. 1995) indicate that LFS induces NMDAR-dependent LTD; this was previously shown to be completely independent of mGluR activation (Dudek and Bear 1992; Selig et al. 1995). Nevertheless, we performed control experiments to ensure that the LFS-LTD elicited by our experimental protocol in naïve slices (which had not been exposed to CCh or expressed mLTD) was not mGluR dependent. As expected, the LFS-LTD in naïve slices does not require mGluR activation (Fig. 3A, 60 ± 2% of baseline fEPSP slope in aCSF, n = 6, vs. 67 ± 3% in interleaved slices in MCPG, n = 5, P > 0.09). To test whether the LFS-LTD induced after mLTD saturation is mGluR dependent, mLTD was saturated (Fig. 3B, 38 ± 3% of fEPSP slope after next to last CCh application and 33 ± 3% of fEPSP slope after final CCh application, P > 0.10) and LFS was applied in the presence of the Group I/II mGluR antagonist MCPG (500 μM). Significant depression was still elicited (Fig. 3B, 77 ± 6% of fEPSP slope after mLTD saturation, n = 4, P < 0.02), indicating that the depression does not require activation of Group I/II mGluRs. Taken together, these data indicate that after mLTD saturation, LFS-LTD is both NMDAR and mGluR independent.

When we compared the magnitude of LFS-LTD elicited in naïve slices with that elicited after mLTD saturation, we found that although mLTD saturation did not occlude LFS-LTD, it did significantly reduce it, irrespective of whether antagonists were present (Fig. 3C; mLTD in naïve slices vs. mLTD saturated (Fig. 2C, 66 ± 6% of baseline fEPSP slope after next to last CCh application vs. 61 ± 5% of baseline fEPSP slope after final CCh application, n = 4, P > 0.09) and d,L-APV (100 μM) was applied to block NMDARs, yet LFS still resulted in roughly 28% further depression (Fig. 2C, LFS-LTD 72 ± 2% of fEPSP slope after mLTD saturation, n = 4, P < 0.01). The amount of LFS-LTD elicited after mLTD saturation was the same irrespective of whether d,L-APV was present (75 ± 5% of fEPSP slope after mLTD saturation in control aCSF, n = 5 vs. 72 ± 2% in d,L-APV, n = 4, P > 0.8). Control experiments were performed to confirm that the LFS-LTD elicited under our experimental conditions was indeed NMDAR dependent, and could be blocked by the NMDAR antagonist d,L-APV in naïve slices in which mLTD had not been elicited before LFS. As expected, LFS-LTD is prevented by d,L-APV (100 μM) in naïve slices (Fig. 2D, 98 ± 6% of baseline fEPSP slope, n = 5, P > 0.7). These data suggest that some mechanistic switch occurs during saturation of mLTD whereby LFS-LTD, which is NMDAR dependence in naïve slices, can occur independently of NMDAR activation.

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saturated in control aCSF, in the presence of D,L-APV, or in the presence of MCPG. ANOVA, \( P < 0.01 \). This is not surprising, however, considering that synapses already depressed by saturation of mLTD may be unable to depress to the same extent as synapses in naïve slices.

**LFS-LTD becomes NMDAR independent after a single application of CCh**

Although we were initially interested in whether saturation of mLTD could occlude LFS-LTD, the unexpected finding that LFS-LTD becomes NMDAR independent led us to ask whether the switch in the induction mechanism of LFS-LTD might occur after a single occurrence of mLTD. Thus we induced LFS-LTD in either control aCSF or in the presence of D,L-APV (100 \( \mu M \)) after only one application of carbachol to induce mLTD. As shown in Fig. 4A, a single occurrence of mLTD is sufficient to render subsequent LFS-LTD NMDAR independent because D,L-APV does not block the depression (Fig. 4A, LFS-LTD 65 \( \pm 7\% \) of fEPSP slope just before LFS in control aCSF vs. 76 \( \pm 3\% \) in D,L-APV, \( n = 5, P > 0.10 \)).

Next, we sought to determine whether the NMDAR independence of LFS-LTD requires prior expression of mLTD or whether it can be induced by CCh application alone. To do this, CCh was applied, but stimulation was turned off during its application (and 3 min after to ensure CCh washout) to prevent mLTD induction (because mLTD is activity dependent; see Scheiderer et al. 2006). Interestingly, the same magnitude of LFS-LTD could be elicited 40 min after CCh washout in the presence or absence of D,L-APV in interleaved slices (Fig. 4B, LFS-LTD 72 \( \pm 5\% \) of fEPSP slope just before LFS in control aCSF, \( n = 5 \) vs. 73 \( \pm 4\% \) in D,L-APV, \( n = 4, P > 0.6 \)), even though mLTD was not induced. These data indicate that the NMDAR independence of LFS-LTD induction arises from some cholinergic mechanism activated by CCh application, rather than being a result of prior expression of mLTD at these synapses. Future studies are needed to clearly define which cholinergic receptor subtype is responsible.

To rule out the possibility that the NMDAR independence of LFS-LTD after CCh is a result of slices becoming resistant to 3118 MCCUTCHEN, SCHEIDERER, DOBRUNZ, AND McMAHON

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**FIG. 3.** LFS-LTD remains metabotropic glutamate receptor (mGluR) independent after mLTD saturation. A: LFS-LTD is unaffected by the Group I/II mGluR antagonist (RS)-\( \alpha \)-methyl-4-carboxyphenylglycine (MCPG, 500 \( \mu M \)), \( n = 6 \) in control artificial cerebrospinal fluid (aCSF, \( n = 5 \) in MCPG). B: LFS-LTD that follows mLTD saturation is not blocked by MCPG, \( n = 4 \). C: bar chart illustrates that the magnitude of LFS-LTD in naïve slices is greater than LFS-LTD induced after mLTD saturation in all conditions (in control aCSF, in D,L-APV, or in MCPG).

**FIG. 4.** LFS-LTD becomes NMDAR independent after a single CCh application. A: D,L-APV (100 \( \mu M \)) fails to block LFS-LTD after a single induction of mLTD, \( n = 5 \) in each group. B: D,L-APV fails to block LFS-LTD after a single application of CCh when induction of mLTD is prevented by discontinuing stimulation during CCh application (and for 3 min after to ensure CCh washout), \( n = 5 \) in control aCSF, \( n = 4 \) in D,L-APV. C: D,L-APV (100 \( \mu M \)) blocks LFS-LTD after a prolonged baseline, \( n = 5 \) in each group. D: combined application of D,L-APV (100 \( \mu M \)) and MCPG (500 \( \mu M \)) fails to block LFS-LTD after a single application of CCh when induction of mLTD is prevented by discontinuing stimulation, \( n = 3 \) in each group. E: bar chart illustrates that the paired-pulse facilitation (PPF) ratio does not change during the NMDAR-independent LFS-LTD (induced in the presence of D,L-APV) that follows CCh application, indicating that its expression remains postsynaptic. Waveform traces are averages of 20 paired-pulse events 10 min before and 30 min after application of LFS, from experiments shown in Fig. 5A (in the presence of D,L-APV). Scale bar: 0.5 mV, 10 ms.
D,L-APV during the lengthy period of recording before LFS application, control experiments were performed where baseline transmission was recorded for a period of time equal to that which had elapsed in previous experiments in which D,L-APV failed to block LFS-LTD (70 min in Fig. 4B). As shown in Fig. 4C, LFS-LTD is NMDAR dependent in naïve slices not exposed to CCh, even after lengthy baseline recording (98 ± 3% of baseline fEPSP slope in D,L-APV, \( n = 5 \), vs. 71 ± 7% of baseline fEPSP slope in interleaved control slices, \( n = 5 \), \( P < 0.02 \)).

Because neither D,L-APV nor MCPG alone blocks LFS-LTD induced after cholinergic stimulation, we next tested whether blockade might be achieved by the combination of the two inducers after CCh application, we next tested whether D,L-APV (100 \( \mu \)M) also failed to block the LFS-LTD induced after CCh application (in the absence of stimulation to prevent mLTD) (Fig. 4D, 72 ± 1% of fEPSP slope just before LFS in D,L-APV and MCPG vs. 68 ± 6% in interleaved control slices, \( n = 3 \), \( P > 0.6 \)).

Despite the switch to NMDAR independence, this LFS-LTD still appears to be expressed postsynaptically, as is the LFS-LTD that is NMDAR dependent (Carroll et al. 1999; Man et al. 2000). Analysis of the paired-pulse facilitation (PPF) ratio, an indirect measure of presynaptic release probability (Dobrunz and Stevens 1997), is unchanged during expression of NMDAR-dependent LFS-LTD (PPF ratio of 1.4 ± 0.1 before LFS and PPF ratio of 1.5 ± 0.2 during LFS-LTD expression, \( n = 5 \), \( P > 0.08 \); data not shown) as well as during expression of the NMDAR-independent LFS-LTD that is induced after CCh application (Fig. 4E, PPF ratio of 1.5 ± 0.1 during mLTD expression before LFS and PPF ratio of 1.5 ± 0.2 during LFS-LTD expression, \( n = 5 \), \( P > 0.5 \)).

The mechanistic switch in LFS-LTD induced by CCh might affect hippocampal synapses specifically, or it could be a general mechanism by which the cholinergic system modifies synaptic plasticity across different brain regions. This was tested by recording from glutamate synapses in visual cortex, where CCh also induces activity-dependent mLTD (Kirkwood et al. 1999). Layer IV was stimulated and recordings were made from layer II/III. As in hippocampus, CCh was applied to visual cortex in the absence of stimulation to prevent mLTD. LFS was applied 20 min after washout of agonist. Unlike in hippocampus, LFS-LTD remained NMDAR dependent after CCh application (Fig. 5, LFS-LTD 82 ± 5% of fEPSP slope just before LFS in interleaved control slices, \( n = 5 \), vs. 99 ± 3% in D,L-APV, \( n = 4 \), \( P < 0.4 \)), indicating that the effect of CCh on subsequent LFS-LTD induction is specific to hippocampus. Importantly, these data also provide further evidence that the inability to block hippocampal LFS-LTD after CCh exposure is not merely a result of the long recording period before LFS causing either a general degradation of slice health or drug insensitivity.

**DISCUSSION**

Here we have examined the interplay of mLTD, a recently characterized form of activity and NMDAR-dependent LTD at CA3–CA1 synapses (Scheiderer et al. 2006), and the well-characterized electrically induced forms of LTP and LTD at these synapses. With regard to LTP, our data demonstrate that induction of mLTD can depress synapses expressing LTP and vice versa or “depotentiate” transmission back to its baseline level. Other forms of depotentiation were described previously, i.e., NMDAR-dependent depotentiation (induced by LFS) (Dudek and Bear 1993; O’Dell and Kandel 1994; Staubli and Lynch 1990) and mGlur-dependent depotentiation (induced with either paired-pulse LFS or the mGlur agonist DHPG) (Basir and Collingridge 1994; Bashir et al. 1993; Palmer et al. 1997; Zho et al. 2002). Although NMDAR-dependent depotentiation is mediated by dephosphorylation and internalization of \( \alpha \)-amino-3-hydroxy-5-methyl-4-isoxazolopropionic acid (AMPA) receptors (AMPARs) (Huang et al. 2001; Lee et al. 2000), and some evidence suggests the same is true for mGlur-mediated depotentiation (Delgado and O’Dell 2005; Oliet et al. 1997; Zho et al. 2002; but see Delgado and O’Dell 2005; Oliet et al. 1997), further experiments are necessary to determine how mLTD depotentiation is mediated intracellularly. Nevertheless, our results demonstrate a novel form of hippocampal depotentiation and may suggest some mechanistic similarity between mLTD and LFS-LTD.

We also demonstrate here that hippocampal slices expressing mLTD can be potentiated, which suggests that mLTD expression may be reversible. However, because mLTD was not saturated before LTP induction, the possibility exists that only the synapses in the test pathway that were not depressed were selectively potentiated. This issue is minimized by data shown in Fig. 1F. Even though mLTD expression was not saturated before the tetanus, the fact that mLTD returned transmission to the original baseline and that the subsequent tetanus returned potentiation to the saturated level of LTP is consistent with the interpretation that bidirectional plasticity is occurring at the level of individual synapses. Furthermore, the reinstatement of LTP (to the previously saturated level) after mLTD expression suggests that mLTD is reversible.

We demonstrate that saturation of LFS-LTD occludes mLTD, as might be predicted from these two plasticities’ shared activity and NMDAR dependence. However, mLTD saturation does not occlude LFS-LTD, although it does significantly reduce it, likely because some synapses are already depressed before LFS. Further experiments demonstrated that CCh application causes a mechanistic switch in LFS-LTD such that it becomes NMDAR independent. We determined that this switch does not involve Group I/II mGlurRs, and expression of the resulting LTD does not occur through a presynaptic mechanism, as indicated by a lack of change in the PPF ratio. However, after discovering that the switch was caused by...
application of CCh itself rather than expression of mLTD, we did not pursue the switch in mechanism further because the goal of this study was to examine how electrically induced LTP and LTD interact with mLTD specifically. Understanding this switch will require determining not only what alternate signaling event(s) mediate LFS-LTD after the switch occurs, but also which cholinergic receptor(s) and signaling molecules are activated during CCh application to induce it. One possibility is that the switch is mediated by internalization of mAChRs because mLTD is induced by application of muscarinic agonist, which has been shown to induce mAChR internalization (Ferguson 2001; Maloteaux and Hermans 1994). This idea is supported by a recent report that at mossy fiber–CA3 interneuron synapses, a single application of mGluR agonist induces internalization of mGluRs, which causes a molecular switch that alters subsequent electrically induced LTD (Pelkey et al. 2005). Interestingly, the switch in LFS-LTD does not occur in visual cortex, suggesting that this may be a cholinergic mechanism specific to hippocampal synapses. These data may indicate that there is more flexibility in the induction mechanisms available to mediate LTD in hippocampus.

mLTD is a cholinergic-dependent depression of glutamate transmission. Studies of both animals (Berger-Sweeney 2001; Gold 2003; Wrenn et al. 1999) and humans (Collerton 1986; Kasa 1997; McKinney and Jacksonville 2005; Schliebs 2005) with cholinergic lesions have established that cholinergic input to the hippocampus is critical for normal learning and memory. That mLTD can occur sequentially with forms of synaptic plasticity thought to be relevant to learning and memory supports the hypothesis that mLTD can function as one of the mechanisms by which the cholinergic system modifies hippocampal function. In addition, data presented here demonstrate that activation of the cholinergic system can also modify the induction mechanism of the best characterized form of hippocampal LTD.

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