Neurobiology of Learning and Memory 138 (2017) 173–181

Contents lists available at ScienceDirect

Neurobiology of Learning and Memory

journal homepage: www.elsevier.com/locate/ynlme

Bi-directional interplay between proximal and distal inputs to CA2 pyramidal neurons

Kaoutsar Nasrallah, Rebecca Ann Piskorowski, Vivien Chevaleyre *

CNRS UMR8118, Team Synaptic Plasticity and Neural Networks, FR3636, Université Paris Descartes, Sorbonne Paris Cité, 45 rue des Saints-Pères, 75006 Paris, France

ARTICLE INFO

Article history:
Received 21 April 2016
Revised 22 June 2016
Accepted 25 June 2016
Available online 25 June 2016

Keywords:
Hippocampus
Area CA2
Long-term depression
Delta-opioid receptor
Disinhibition

ABSTRACT

Hippocampal area CA2 is emerging as a critical region for memory formation. Excitatory Scaffer collateral (SC) inputs from CA3 do not express activity-dependent plasticity at SC-CA2 synapses, and are governed by a large feed-forward inhibition that prevents them from engaging CA2 pyramidal neurons. However, long-term depression at inhibitory synapses evoked by stimulation of SC inputs highly increases the excitatory/inhibitory balance coming from CA3 and allows the recruitment of CA2 pyramidal neurons. In contrast, distal excitatory inputs in stratum lacunosum moleculare (SLM) can drive action potential firing in CA2 pyramidal neurons and also express a long-term potentiation. However, it is unknown whether stimulation of distal inputs can also evoke plasticity at inhibitory synapses and if so, whether this plasticity can control the strength of excitatory inputs. Here we show that stimulation in SLM evokes a long-term depression at inhibitory synapses. This plasticity strongly increases the excitatory drive of both proximal and distal inputs and allows CA3 to recruit CA2 pyramidal neurons. These data reveal a bi-directional interplay between proximal and distal inputs to CA2 pyramidal neurons that is likely to play an important role in information transfer through the hippocampus.

1. Introduction

Activity-dependent changes in synaptic efficacy, such as long-term potentiation and depression (LTP and LTD), are crucial to experience-driven refinement of neural connections in the mammalian brain and are an important component of the cellular mechanisms underlying learning and memory formation (Martin, Grimwood, & Morris, 2000; Mayford, Siegelbaum, & Kandel, 2012). Such forms of synaptic plasticity have been extensively studied in the hippocampus, a brain region critically involved in memory formation. In the hippocampus, the pyramidal cells in areas CA1, CA2 and CA3 receive distinct sets of inputs at discrete regions in their dendritic arbor. Much less is known about how synaptic plasticity at one set of synaptic inputs can modulate the synapses at a separate set of inputs that converge onto the same postsynaptic neuron. Such hetero-synaptic forms of plasticity are of great interest because they enable different neuronal circuits to perform a wide range of mnemonic processing (Abbott & Regehr, 2004; Spruston, 2008).

Area CA2 of the hippocampus has been shown to play a crucial role in social memory (Hitti & Siegelbaum, 2014; Stevenson & Caldwell, 2014) and aggressive behavior (Pagani et al., 2015). Furthermore, this region has recently been reported to be the basis of a hippocampal network that encodes location during immobility and sleep (Kay et al., 2016). Given the importance of these newly-discovered roles, a better understanding of the physiology of the often-overlooked area CA2 is necessary. CA2 pyramidal neurons (PNs) have distinct biophysical, molecular and anatomical properties that clearly distinguish them from their CA1 and CA3 neighbors (recently reviewed in (Dudek, Alexander, & Farris, 2016). For instance, they are more hyperpolarized, have lower input resistance, larger membrane capacitance less of a sag in response to hyperpolarizing current. At a molecular level, they uniquely express numerous proteins such as vasopressin 1b receptors, adenosine receptors and the regulator of G-protein signaling protein, RGS14. CA2 PNs receive glutamatergic inputs from CA3 PNs via the Schaffer collateral (SC) pathway onto their proximal apical dendrites located in *stratum radiatum* (SR) and from the entorhinal cortex onto their distal apical dendrites located in *stratum lacunosum-moleculare* (SLM) (Kohara et al., 2013). Unlike SC-CA1 excitatory synapses, the SC-CA2 glutamatergic synapses do not express activity-dependent long-term potentiation (LTP) (Zhao, Choi, Obrietan, & Dudek, 2007), potentially because of the unique hippocampal expression of postsynaptic signaling molecules and potential calcium binding proteins (Lee et al.,

* Corresponding author.
E-mail address: vivien.chevaleyre@parisdescartes.fr (V. Chevaleyre).

http://dx.doi.org/10.1016/j.nlm.2016.06.024
1074-7427/

2010; Simons, Escobedo, Yasuda, & Dudek, 2009). Moreover, the Schaffer collateral (SC) input to CA2 PNs is dominated by a powerful feed-forward inhibition (FFI) that tightly controls the excitatory postsynaptic potential (EPSP) and prevents action potential firing of CA2 PNs, while SC inputs strongly excite CA1 PNs (Chevaleyre & Siegelbaum, 2010; Piskorowski & Chevaleyre, 2013). In contrast to SC-CA2 excitatory synapses, inhibitory transmission from parvalbumin-expressing (PV+) interneurons in area CA2 is highly plastic, undergoing a long-term depression (iLTD) that is mediated by delta opioid receptor (DOR) activation following stimulation of SC inputs or activation of DORs with agonist application (Piskorowski & Chevaleyre, 2013). This DOR-mediated iLTD allows a lasting increase of both proximal and distal input excitatory drive onto CA2 PNs (Nasrallah, Piskorowski, & Chevaleyre, 2015). Therefore, we wondered whether the crosstalk between proximal and distal inputs could be bi-directional, i.e. whether stimulation of distal inputs could also trigger an iLTD of inhibitory transmission and a dis-inhibitory increase in proximal and distal excitatory inputs.

Using electrophysiology and selective pharmacology in adult mouse hippocampal slices, we show that high frequency stimulation (HFS) of distal inputs induces both homo-synaptic and hetero-synaptic iLTD in area CA2. We found that this dis-inhibition produces an activity-dependent increase in the excitatory/inhibitory (E/I) ratio at both proximal and distal inputs in a DOR- and GABA-dependent manner. Moreover, induction of iLTD with HFS of distal inputs increases action potential firing of CA2 PNs in response to both distal and proximal input stimulation.

2. Results

2.1. HFS in SLM induces iLTD and alters the excitatory/inhibitory balance at distal inputs of CA2 PNs

We have recently shown that inhibitory synapses recruited in SLM express an activity-dependent hetero-synaptic LTD following HFS of proximal inputs (Nasrallah et al., 2015). We investigated whether iLTD of distal CA2 inputs could also be induced homo-synapticaly by HFS in SLM. To address this question, we performed whole-cell voltage clamp recordings of CA2 PNs and evoked inhibitory postsynaptic currents (IPSCs) by electrical stimulation of distal inputs. We monitored the IPSC amplitude and applied a HFS (100 pulses at 100 Hz, repeated twice) after a stable baseline. In the first experiment, we placed the glass pipette stimulating electrode in CA1 SLM close to area CA2 as shown in Fig. 1A (~150 μm away from CA2 in CA1), and performed the recordings in the continuous presence of NBQX (10 μM) and D-APV (50 μM) to block glutamate ionotropic receptors. In these conditions, we found that HFS in SLM induced a lasting decrease in the IPSC amplitude (Fig. 1B: 83.0 ± 4.6%, p = 0.023, n = 5). In addition, this iLTD was accompanied by a significant increase in the paired-pulse ratio (PPR), consistent with a pre-synaptic decrease in GABA release (Fig. 1C: from 0.638 ± 0.038 to 0.684 ± 0.0374, p = 0.0189). In this experimental configuration, the IPSCs likely resulted from direct stimulation of inhibitory interneurons by the stimulating electrode. To examine the circuit in more physiological conditions, i.e. to recruit inhibitory neurons in a feed-forward manner by stimulating excitatory inputs in SLM, we placed the stimulating electrode in the middle of CA1 SLM, far from CA2 as shown in Fig. 1D (700–800 μm from CA2 in CA1). The cells were voltage-clamped at +10 mV (close to the reversal potential for excitatory currents) in order to isolate the inhibitory responses. In this configuration, we found that a HFS triggered iLTD when excitation was kept intact (Fig. 1E: 83.6 ± 3.7%, p = 0.007, n = 6), but not when excitatory transmission was blocked with NBQX and D-APV (Fig. 1E: 97.7 ± 5.6%, p = 0.50, n = 6). This data suggest that the interneurons that express and/or trigger iLTD are not directly recruited by the stimulating electrode when stimulation is far from CA2, but they are recruited either directly when stimulation is close or indirectly in a feed-forward manner when excitatory transmission is left intact.

We have recently shown that a HFS in SR induces an iLTD in area CA2 that results in a large increase in the depolarizing component of the PSP amplitude evoked by SC stimulation (Nasrallah et al., 2015). We asked whether the plasticity of inhibitory transmission that we can induce with a distal input HFS could be sufficient to modulate the level of excitatory drive at SLM–CA2 synapses. To examine further, we performed whole-cell current clamp recordings of CA2 PNs in response to electrical stimulation of distal input in CA1 SLM close to CA2, either in the presence or absence of $GABA_A$ and $GABA_B$ receptor antagonists (1 μM SR 95531 and 2 μM CGP 55845). Because a HFS is known to induce a NMDA-dependent LTP at the SLM-CA2 excitatory synapses (Chevaleyre & Siegelbaum, 2010), we applied D-APV (50 μM) to block NDMA receptors during the HFS (from 10 min before to 5 min after HFS). We found that EPSPs recorded in presence of $GABA_A$ and $GABA_B$ receptor antagonists are not potentiated by a HFS when NMDA receptors were blocked (Fig. 2A: filled circles: 103.9 ± 11.3%, p = 0.75, n = 5), confirming that distal inputs to CA2 PNs do not express LTP when NMDA receptors are blocked. We then performed the same experiment with inhibitory transmission intact. In these conditions we found that a HFS induced a lasting increase in the PSP amplitude (Fig. 2A: open circles: 127.5 ± 10.0%, p = 0.033, n = 7). To ensure that this change in PSP amplitude was not an artifact of directly stimulating interneurons with the stimulating electrode, we performed the same experiment with the stimulating electrode in CA1 SLM far from CA2. In these conditions, HFS application in SLM still induced a long-term increase in the PSP amplitude when inhibitory transmission was intact (Fig. 2B: white circles: 170.8 ± 21.9%, p = 0.032, n = 5) but not in the continuous presence of $GABA_A$ and $GABA_B$ receptor antagonists (Fig. 2B: filled circles: 106.3 ± 8.6%, p = 0.65, n = 5, p = 0.026 with the interleaved controls).

In order to test whether the HFS-induced increase in PSP amplitude occurs when both intracellular [Cl^-] and membrane potential are unaltered by the intracellular recording solution, and to determine whether inhibitory synapses that express iLTD exert a local control in CA2 SLM, we performed extracellular recordings of field PSPs (fPSP) by placing a recording pipette in CA2 SLM. The fPSPs were evoked with a stimulating pipette placed in CA1 SLM far from CA2. We found that HFS induced a lasting increase in the fPSP amplitude in absence (Fig. 2C: opened circles: 119.9 ± 3.8%, p = 0.007, n = 5) but not in presence of $GABA_A$ and $GABA_B$ receptor antagonists (Fig. 2C: filled circles: 100.4 ± 2.2%, p = 0.43, n = 5, p = 0.002 with the interleaved controls). Altogether, these data indicate that HFS of distal inputs is capable of increasing distal PSP amplitude via a dis-inhibitory mechanism, and part of this dis-inhibition occurs locally in SLM.

2.2. Stimulation in SLM induces a hetero-synaptic iLTD and increases proximal excitatory drive onto CA2 PNs

In area CA2, a HFS of proximal SC inputs can trigger a hetero-synaptic iLTD of distally evoked IPSCs and a hetero-synaptic dis-inhibition of distal excitatory inputs (Nasrallah et al., 2015). Could this interplay be bi-directional? Can a HFS in SLM also trigger iLTD and a dis-inhibition of proximal excitatory inputs? To answer this question, we first recorded IPSCs in CA2 PNs evoked by stimulation in SR in the continuous presence of NBQX (10 μM) and D-APV (50 μM). After a stable baseline period, we applied a HFS in SLM with a stimulation pipette near CA2. We found that this resulted