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SYNAPTIC PLASTICITY IN THE MORMYRID ELECTROSENSORY LOBE

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Summary

The mormyrid electrosensory lateral line lobe (ELL) is one of several different sensory structures in fish that behave as adaptive sensory processors. These structures generate negative images of predictable features in the sensory inflow which are added to the actual inflow to minimize the effects of predictable sensory features. The negative images are generated through a process of association between centrally originating predictive signals and sensory inputs from the periphery. *In vitro* studies in the mormyrid ELL show that pairing of parallel fiber input with Na^+ spikes in postsynaptic cells results in synaptic depression at the parallel fiber synapses. The synaptic plasticity observed at the cellular level and the associative process of generating negative images of predicted sensory input at the systems level share a number of properties. Both are rapidly established, anti-Hebbian, reversible, input-specific and tightly restricted in time. These common properties argue strongly that associative depression at the parallel fiber synapse contributes to the adaptive generation of negative images in the mormyrid ELL.

Key words: electric fish, mormyrid, synaptic plasticity, corollary discharge, expectation, electrosensory, sensory processing, cerebellum.

Introduction

This paper is concerned with synaptic plasticity in the electrosensory lateral line lobe (ELL) of mormyrid electric fish. The mormyrid ELL is one member of a group of cerebellum-like sensory structures found in fish, amphibians and mammals (Fig. 1). These structures possess a molecular layer, an underlying sensory input layer and an intervening layer of large cells. The large cells have apical dendrites that receive input from parallel fibers, and basilar dendrites that receive input from the periphery, either directly from primary afferent fibers or indirectly *via* interneurons. The large cell layer contains the output neurons that convey sensory information to higher stages of the system. The parallel fibers of the molecular layer arise from an external granule cell mass that receives a rich variety of information, including corollary discharge signals associated with motor commands, descending signals from higher stages of the same sensory modality and signals from other sensory modalities such as proprioception (Montgomery et al., 1995; Fig. 2).

Several of these structures in fish have been shown to be adaptive sensory processors and to generate negative images of predictable features in the sensory inflow. Addition of such negative images to the actual or concurrent sensory input minimizes the predictable features and allows novel or unexpected inputs to stand out more clearly (Bell, 1982; Montgomery and Bodznick, 1994; Bodznick, 1993; Bastian, 1995; for a review, see Bell et al., 1997a). The negative images are generated by a process of association between various types of predictive signals and particular patterns of sensory input. The predictive signals that are effective are the same as those that are conveyed by parallel fibers, i.e. corollary discharge signals, descending signals from higher levels of the same sensory system and proprioceptive signals conveying information about body or fin movements. Recent work with mormyrids (Bell et al., 1997c), gymnotids (Bastian, 1996b; Wang and Maler, 1997) and elasmobranchs (Bodznick et al., 1996) indicates that the generation of negative images in these fish is largely due to plasticity at the synapses made by parallel fibers and other types of descending input to the molecular layer.

This paper is concerned with mormyrid fish. The following sections present work on the plasticity as studied with natural sensory and motor signals in these fish, on the functional circuitry of the mormyrid ELL and on the phenomenology and mechanism of plasticity at the cellular level.

Plasticity in the mormyrid ELL using natural sensory and motor signals

All the work in mormyrid fish has been carried out using corollary discharge signals linked to the motor command that elicits the electric organ discharge (EOD). These electric organ corollary discharge (EOCD) signals are prominent in the

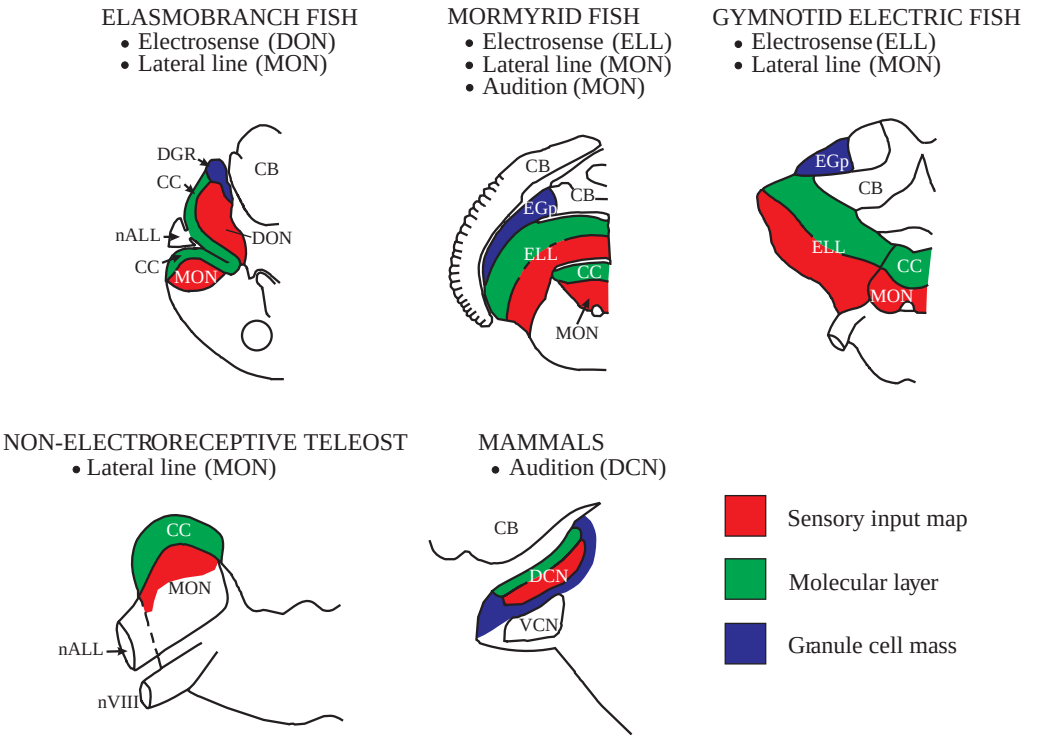

Fig. 1. Cerebellum-like sensory structures associated with octavolateral systems in different taxa. Filled circles below the name of the taxon indicate the sensory systems and associated structures in each group. The granule cell mass is located anterior to the cerebellum-like structure in non-electroreceptive teleosts and is therefore not included in the figure at lower right. CB, cerebellum; CC, cerebellar crest; DCN, dorsal cochlear nucleus; DGR, dorsal granular ridge; DON, dorsal octavolateral nucleus; EGp, eminentia granularis posterior; ELL, electrosensory lateral line lobe; MON, medial octavolateral nucleus; nALL, anterior lateral line nerve; nVIII, eighth nerve (adapted from Montgomery et al., 1995).

mormyrid ELL and in the external granule cell mass that gives rise to the parallel fibers (Bell, 1982; Bell et al., 1992). The experiments are performed using curarized fish in which the EOD is blocked but in which the EOD motor command continues to be emitted spontaneously at rates of 2–8 s^{-1} and in which all the sensory input that follows the command is under experimental control. In these experiments, the responses of ELL cells to the EOCD alone are examined first. An electrosensory input is then delivered after each EOCD for a period of a few minutes. Responses to the EOCD alone are then examined again after the pairing with an electrosensory input. After pairing, the EOCD responses of ELL cells become a surprisingly faithful negative image of the cell's response to the paired sensory input (Bell, 1982; Bell et al., 1997b).

A few seconds of pairing with a sensory stimulus will cause a change in the EOCD effect, but 5 or 6 min of pairing is necessary for the effect to reach a maximum (Bell, 1982). The time course of decay of the negative image at stimulus-off is similar to the time course of its establishment. The decay is not a passive disappearance of the negative image, but an active process of rematching the new sensory conditions that are present after stimulus-off (an absence of sensory input) due,

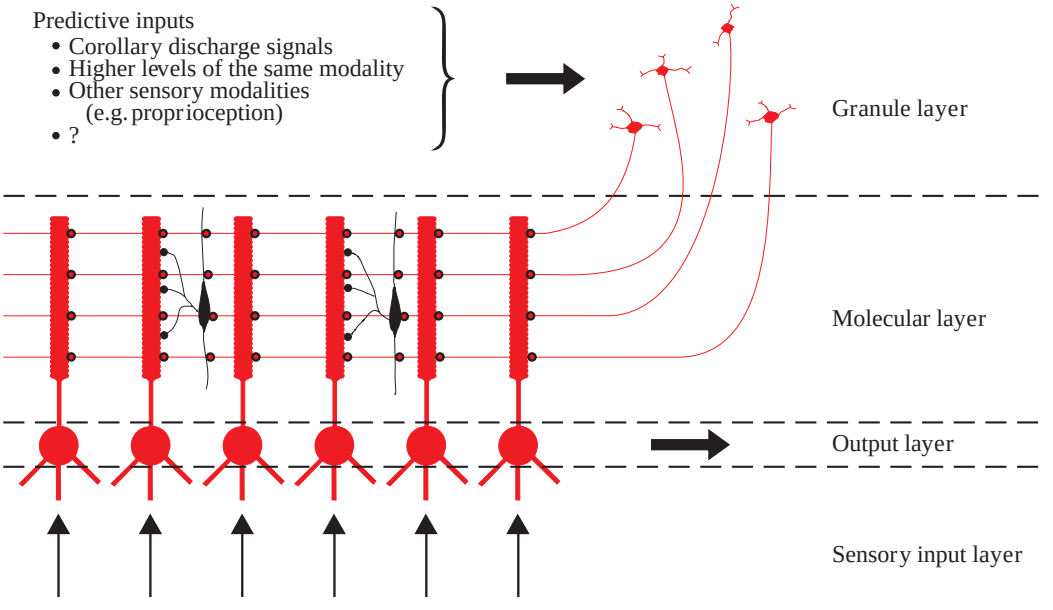

Fig. 2. Schematic drawing illustrating the basic circuitry of cerebellum-like sensory structures. Synaptic plasticity and the association between predictive inputs conveyed by parallel fibers and sensory inputs conveyed by primary afferent fibers lead to the parallel fibers evoking a negative image of the associated sensory input pattern. Inhibitory stellate cells of the molecular layer are shown in black (adapted from Bell et al., 1997a).

presumably, to changes in the synaptic effect of the EOCD (Bell, 1986). Such a process of active rematching has also been demonstrated in gymnotid (Bastian, 1996a) and elasmobranch (D. Bodznick, personal communication) fish.

The change in the EOCD response of ELL cells could be due to (a) plastic change in the response of ELL cells to an unchanging EOCD input; (b) plastic change in some distant structure that results in an altered EOCD input to ELL cells; or (c) both of these alternatives. This issue was investigated by pairing an intracellularly injected current pulse with the EOCD rather than with a sensory stimulus (Bell et al., 1993) on the basis that, if such pairing yielded a plastic change and if the intracellular current pulse only affected the recorded cell, then the plastic change would have had to have been initiated in the recorded cell and to have taken place at its input synapses. Pairing the EOCD with a depolarizing intracellular current pulse for several minutes did indeed result in a hyperpolarizing response to the EOCD at the delay that was appropriate for matching the previously paired current pulse. Moreover, in at least one major cell type, the medium ganglion cell (see below), the plastic change only took place if the depolarizing pulse evoked what is probably an apical dendritic spike, i.e. it only took place if the injected current evoked an event that was restricted to the recorded cell. These results show that the responses of ELL cells to EOCD signals are capable of plastic change, but do not exclude the possibility of additional change at distant sites that is relayed to the ELL by descending connections.

Pairing with depolarizing current pulses was effective, but pairing with hyperpolarizing current pulses was not. This is in contrast to pairing with sensory stimuli, where pairing with stimuli that inhibit the cell is as effective as pairing with stimuli that excite the cell (Bell, 1982). The ineffectiveness of pairing with injected hyperpolarizing current in the mormyrid ELL is also in contrast with the findings in the ELL of gymnotids (Bastian, 1996a) and the dorsal octavolateral nucleus of elasmobranchs (D. Bodznick, personal communication), where both polarities of injected current pulses appear to be equally effective.

Functional circuitry of the mormyrid ELL as examined *in vivo* and *in vitro*

Three cell types of the mormyrid ELL have apical dendrites extending throughout the molecular layer and basilar dendrites that extend for varying distances towards the sensory input layer: medium ganglion cells, large ganglion cells and large fusiform cells (Meek et al., 1996; Grant et al., 1996). The large ganglion and large fusiform cells are efferent cells with axons that project to higher stages of the system. Large fusiform cells are E-cells that are excited in the center of their receptive field by stimuli that excite primary afferent fibers, whereas large ganglion cells are I-cells that are inhibited by such stimuli (Bell et al., 1997b).

The medium ganglion cells appear to be of central importance for ELL function. They are GABAergic inhibitory interneurons that terminate locally on the large ganglion, on large fusiform cells and on each other. The number of medium ganglion cells is six times greater than the combined number of large ganglion and large fusiform cells, and each medium ganglion cell has approximately twice as many apical dendrites as either of the other two cell types (Meek et al., 1996). These cells are therefore the major postsynaptic component of the molecular layer. Two morphological classes of medium ganglion cells can be distinguished, MG1 and MG2 (Meek et al., 1996; Han et al., 1998). The morphological differences have suggested the hypotheses that the MG1 cell is an I-cell that inhibits large fusiform cells, whereas the MG2 cell is an E-cell that inhibits large ganglion cells (Han et al., 1998). These hypotheses about the functional circuitry are illustrated in Fig. 3, which also contains the additional hypothesis, for which there is as yet no evidence, that the two classes of medium ganglion cells are mutually inhibitory.

The proposed circuitry has two important functional consequences: (1) mutual inhibition between opponent types of medium ganglion cells would enhance contrast between the E- and I-cells of the circuit; and (2) such mutual inhibition could explain the finding that plasticity is present in these cells after pairing with inhibitory sensory stimuli but not after pairing with hyperpolarizing current pulses. Pairing the EOCD with sensory input that excites or depolarizes one type of medium ganglion cell will result, after pairing, in an EOCD-driven inhibition of that cell due to the plasticity. After such pairing, a medium ganglion cell of the opposite type, which

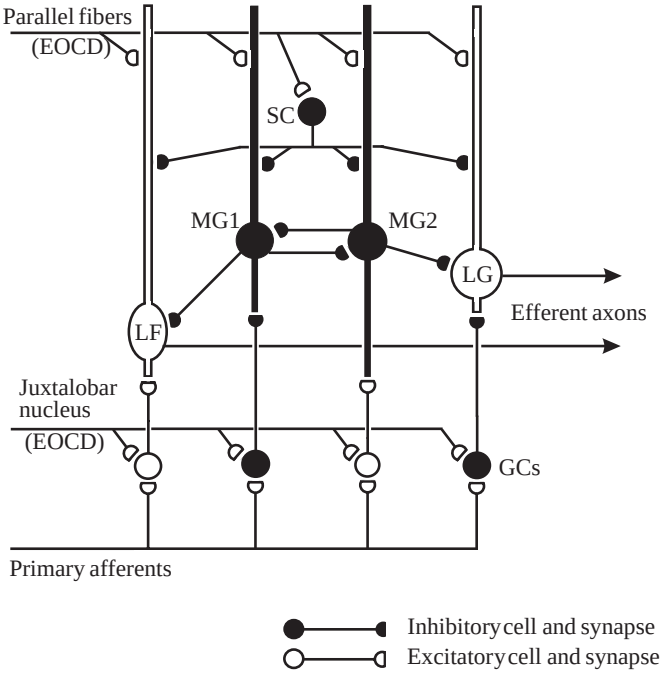

Fig. 3. Hypothesized functional circuit of the mormyrid electrosensory lateral line lobe (ELL). See text for a description of the circuit. EOCD, electric organ corollary discharge; LF, large fusiform cell; LG, large ganglion cell; GCs, granule cells; MG1 and MG2, medium ganglion cells: SC, stellate cell of the molecular layer (taken from Han et al., 1999, with permission).

had been inhibited or hyperpolarized during the pairing with the same sensory input, would be excited by the EOCD because of release from inhibition by the first cell and not because of any plastic change in its synaptic inputs.

Intracellular recordings from medium ganglion cells show two types of spike, a small narrow spike and a large broad spike (Bell et al., 1993; Grant et al., 1998). Both types are blocked by tetrodotoxin and are therefore Na^+ spikes (Bell et al., 1997c). The small narrow spike has a lower threshold, often occurs in isolation, and is probably an axon spike that does not invade the soma. Several observations suggest that the large broad spike is a dendritic or soma-dendritic spike that propagates into the apical dendrites of medium ganglion cells. First, such spikes may be recorded both extracellularly and intracellularly from the molecular layer. Second, synaptic activation by parallel fiber stimulation and synaptic or antidromic activation by stimulation in the deep layers of the ELL evokes a prominent extracellularly recorded negative potential that is initiated in the deep molecular layer and propagates out to the external border of the ELL (Grant et al., 1998). Similarities in latency and other common features strongly suggest that the negative field potential is due to propagation of the broad spike in the apical dendrites of medium ganglion cells.

Parallel fiber stimulation evokes an excitatory–inhibitory postsynaptic potential (EPSP–IPSP) sequence in medium ganglion, large ganglion and large fusiform cells (Fig. 4A,F; see Fig. 6C). The IPSPs are probably due to parallel fiber activation of molecular layer stellate cells or medium ganglion cells and are therefore disynaptic. The IPSPs are mediated by receptors of the GABA-A type since they are completely blocked by bicuculline. The parallel-fiber-evoked EPSPs are mediated by glutamate receptors of both the αAmino-3-hydroxy-5-methylisoxazole-4-proprionic acid (AMPA) and *N*-methyl-D-glutamate (NMDA) types, as shown by the effects of antagonists for these receptors (Fig. 4) and by recordings in Mg^{2+}-free Ringer's solution (Grant et al., 1998). The presence of NMDA receptors at the synapses between parallel fibers and apical dendrites of ELL cells was surprising since parallel-fiber-evoked EPSPs in the adult cerebellum are mediated exclusively by glutamate receptors of the AMPA type (Kano et al., 1988).

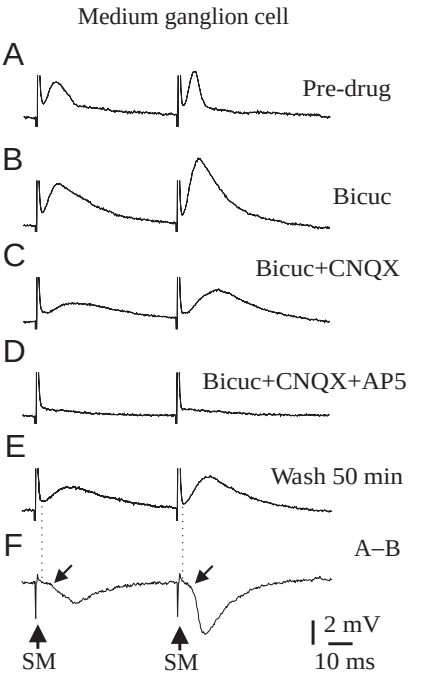

Fig. 4. Pharmacology of parallel-fiber-evoked synaptic responses in medium ganglion cells. Parallel fibers were stimulated in the molecular layer of an electrosensory lateral line lobe (ELL) slice preparation while recording intracellularly from a medium ganglion cell at the soma. Two identical stimuli were delivered with an interpulse interval of 60 ms and at an intensity below threshold for a spike. Blocking agents were bath-perfused. (A) Control. (B) Effect of the GABA-A receptor blocker bicuculline (Bicuc; 30 μmol l^{-1}). Note the marked paired pulse facilitation of the excitatory postsynaptic potential (EPSP). Subtraction of B from A reveals an inhibitory postsynaptic potential (IPSP) with a latency that is slightly greater than that of the EPSP and that also shows paired pulse facilitation (see F). (C) Effect of bicuculline plus 6cyano-7-nitroquinoxaline-2,3dione (CNQX; 25 μmol l^{-1}), a blocker of the AMPA type of glutamate receptor. (D) Effect of bicuculline plus CNQX plus D-2-amino-5-phosphonopentanoate (AP5; 50 μmol l^{-1}). The latter blocks glutamate receptors of the *N*-methyl-D-glutamate (NMDA) type. Note that the EPSP disappears in the presence of both blockers. (E) Responses after a 50 min wash in normal bathing solution. The response has not recovered to the level shown in B (before the addition of glutamate blockers) because CNQX takes a very long time to wash out of a slice. (F) Subtraction of trace B from trace A shows the approximate size and time course of the IPSP evoked by parallel fiber stimuli (SM). Note that the apparent onset of the IPSP (small arrows) follows the onset of the EPSP by several milliseconds. Note too the apparent paired pulse facilitation of the IPSP. This paired pulse facilitation could be due, however, to paired pulse facilitation at the excitatory synapse from parallel fiber to inhibitory interneuron (taken from Grant et al., 1998, with permission).

Phenomenology and mechanisms of the plasticity studied *in vitro*

The hypothesis for negative image generation in the mormyrid ELL and in other similar structures is as follows: the size of the EPSPs evoked by parallel fibers will depend on the history of association between the predictive signals conveyed by these fibers and membrane potential changes evoked in the same cells by sensory inputs to the basal dendrites. Such plasticity must have a number of properties if it is to explain the appearance of negative images after pairing of normal sensory and motor signals. (1) The plasticity must be anti-Hebbian (Hebb, 1949), i.e. pairing of parallel fiber input with depolarization of the postsynaptic cell by sensory input must lead to a reduction in the amplitude of the associated EPSP relative to other EPSPs and a consequent reduction in excitation or relative hyperpolarization at the time of the predictive input. (2) Changes in efficacy should only take place

at parallel fiber synapses that are active within a narrow temporal window of the sensory-evoked depolarization. This requirement arises from the close temporal match between the sensory-evoked response and the negative image that results from pairing with the response. In the mormyrid ELL, recordings of input fibers to the eminentia granularis posterior (EGp) suggest that different parallel fibers may convey spikes at different delays with respect to the motor command signal (Bell et al., 1992). Temporal specificity would arise if the only parallel fiber synapses that are changed are those that are active at the same delay as the sensory-evoked depolarization. (3) The plasticity should be input-specific. Pairing with one predictive signal should not affect the responses to other predictive signals. Pairing with a parallel fiber active at one delay should not affect the response to a parallel fiber active at another delay. (4) The changes should be readily reversible when the parallel fiber input is no longer paired with postsynaptic depolarization, just as the negative image disappears when the predictive signal is no longer paired with sensory input. (5) The time course of changes in the parallel-fiber-evoked EPSP should be similar to the time course of changes in the negative image.

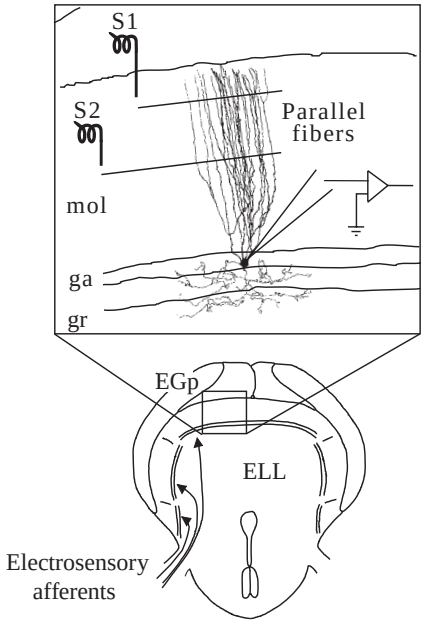

Fig. 5. Diagram of a transverse electrosensory lateral line lobe (ELL) slice to show the positions of the recording and stimulating electrodes for testing the effects of pairing on synaptic efficacy. The inset shows the experimental arrangement. Note the intracellular recording electrode in the soma and the two stimulating electrodes (S1 and S2) in the molecular layer. The use of two stimulating electrodes makes it possible to test whether the plastic change is specific to the paired input and to control for changes in the postsynaptic cell that are unrelated to the pairing. The cell is a reconstruction of a biocytin-filled medium ganglion cell. ga, ganglionic layer; gr, granular layer; mol, molecular layer; EGp, eminentia granularis posterior (taken from Bell et al., 1997c, with permission).

Synaptic plasticity with many of the required properties has now been observed at parallel fiber synapses in the mormyrid ELL using the *in vitro* slice preparation (Bell et al., 1997c; V. Han, Y. Sugawara, K. Grant and C. C. Bell, unpublished observations). The methods used were as follows. Transverse sections of the mormyrid ELL were prepared and maintained in an interface chamber (Fig. 5). Two stimulating electrodes, S1 and S2, were placed at different levels of the molecular layer to stimulate separate bundles of parallel fibers and to test for specificity of plastic change. Intracellular recordings were made from medium ganglion, large ganglion and large fusiform cells (for details of the methods, see Bell et al., 1997c). In the most common protocol, stimuli were delivered first to S1 and then to S2 with an interval between stimuli of 500 ms. The S1–S2 stimulus sequence was repeated at a rate of 0.1 Hz during the pre- and post-pairing test periods, but at 1.0 Hz during pairing. During pairing, a 20 ms intracellular current pulse was delivered at a fixed delay with respect to S1 and S2. Several pairings were usually carried out in the same cell, each with a different timing of the intracellular current pulse with respect to the parallel fiber stimuli. Pre-pairing, pairing and post-pairing periods were usually 6 min, although post-pairing periods as long as 30 min were also tested. Additional protocols were also sometimes used.

Parallel-fiber-evoked EPSPs in all three types of cell, medium ganglion, large ganglion and large fusiform cells, showed changes in EPSP size after pairing (V. Han, Y. Sugawara, K. Grant and C. C. Bell, unpublished observations), but the changes were more consistent and more marked in medium ganglion cells. Most of our studies have therefore focused on medium ganglion cells, and only the results with this cell type are described below. The broad spikes occur only in medium ganglion cells, and recordings from this cell type can thus be distinguished without the need for morphological identification.

The *in vitro* studies confirmed that EOCD plasticity in medium ganglion cells requires that the intracellular current pulse evoke a broad spike (Bell et al., 1993). Perhaps the most important finding was that the direction of change depended on the precise timing of the intracellularly evoked broad spike relative to the parallel-fiber-evoked EPSP during pairing. The cell shown in Fig. 6, for example, underwent six different pairings. The S1-evoked EPSP decreased only after pairings in which the broad spike was evoked just after the start of the EPSP (P2 and P6), but increased after all other pairings. Similarly, the S2-evoked EPSP decreased only after the one pairing in which the broad spike was evoked just after the start of the EPSP (P3). This was the general rule, as indicated in Fig. 7, which summarizes 188 pairings in 43 cells. Pairings in which the broad spike was evoked between 0 and 60 ms after EPSP onset resulted in EPSP decreases, whereas pairings at other delays yielded enhancements. Note that the EPSP evoked by one parallel fiber stimulus decreased when the broad spike was given at a short delay, whereas the other EPSP, which was evoked during the same pairing period but at a different delay, either increased (P2, P3) or remained the same (P6). Thus, input

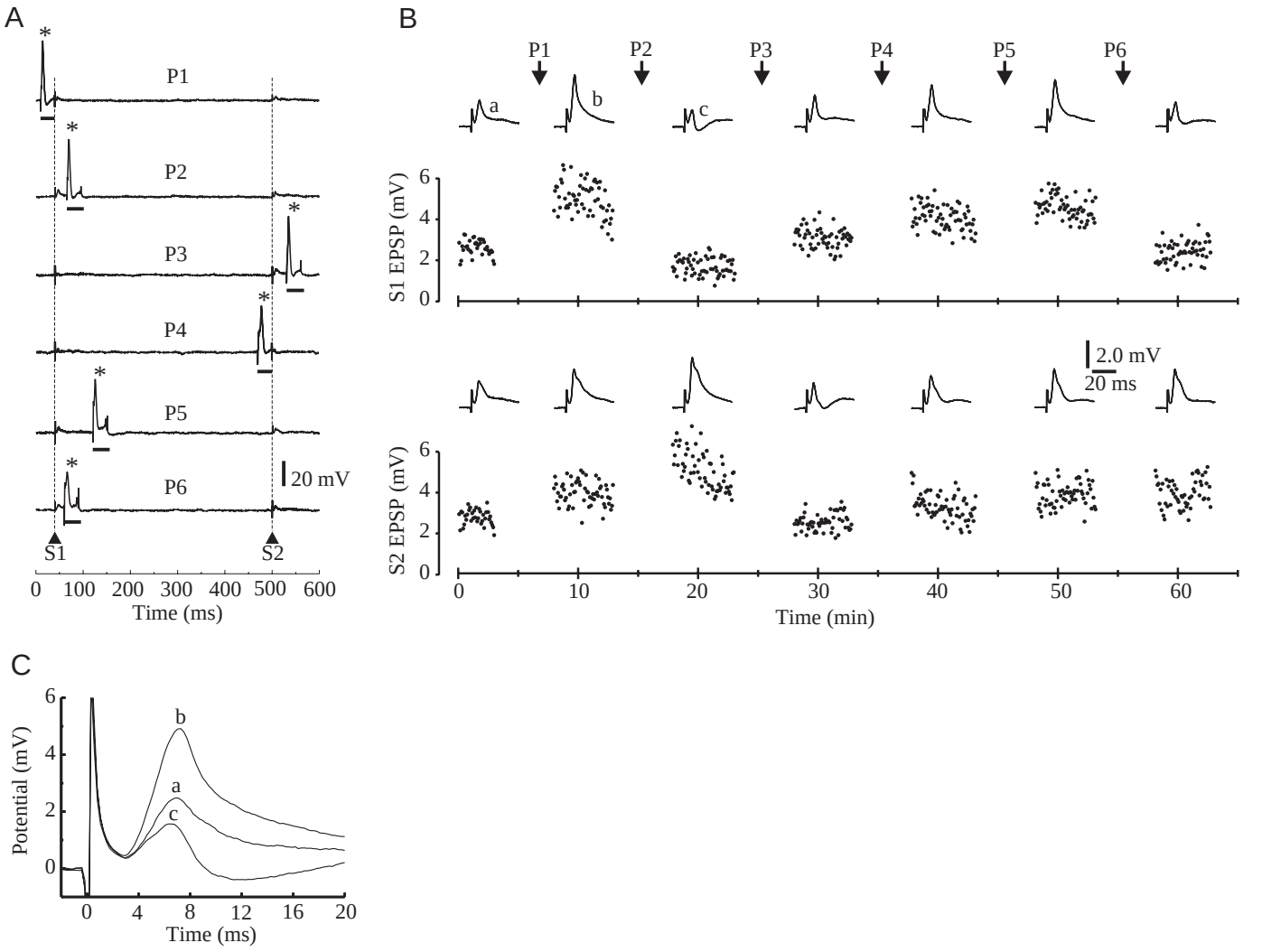

Fig. 6. Effects of different pairings in the same cell. Each pairing has a different timing of the broad spike (evoked by an intracellular current pulse) and the excitatory postsynaptic potentials (EPSPs) (evoked by stimulation of electrodes S1 and S2 as shown in Fig. 5). Pairing was carried out at 1 Hz, and testing of EPSP amplitude before and after pairing was carried out at 0.1 Hz. (A) Timing of EPSPs (vertical dashed lines) and broad spikes (asterisks) for pairings 1–6 (P1–P6). (B) Graphs showing the peak amplitudes of the S1 EPSP (top) and S2 EPSP (bottom) before and after pairings 1–6. Traces above the graphs show mean responses for each period. Traces a, b and c show the responses to S1 before pairing 1, after pairing 1 and after pairing 2, respectively. (C) Enlarged versions of traces a, b and c shown superimposed (taken from Bell et al., 1997c, with permission).

specificity is demonstrated and the decreases in EPSP size cannot be attributed to nonspecific changes in the condition of the cell or the recording. The pairing-induced EPSP depression could last 30 min, the longest post-pairing period tested.

Parallel fiber stimulation evokes an IPSP immediately following the initial EPSP with a latency that is sufficiently short to affect the peak of the EPSP (Fig. 4). However, the superimposed traces of Fig. 6C show that the initial slopes of the EPSPs are affected by pairing in the same manner as the peaks. This initial slope change indicates a change in the EPSP alone, because the EPSP is monosynaptic and the IPSP is disynaptic (note in Fig. 4A,B that the initial slope is not affected by the addition to the bath of bicuculline, a drug that blocks all parallel-fiber-evoked IPSPs, and that in Fig. 4F that the apparent onset of the IPSP follows the onset of the EPSP by several milliseconds). Associative changes in the EPSP alone were also demonstrated by carrying out the pairing experiments in the presence of bicuculline and obtaining the same results (Bell et al., 1997c). These findings demonstrate EPSP plasticity, but do not exclude the possibility of IPSP plasticity. Indeed, changes in parallel-fiber-evoked IPSPs that complement the changes in EPSPs are a strong possibility and are being investigated.

The finding that parallel-fiber-evoked EPSPs in the ELL are partly mediated by NMDA receptors and the demonstrated role for such receptors in the synaptic plasticity of other structures (Bliss and Collingridge, 1993; Bear et al., 1992) suggested the possibility of a role for NMDA receptors in ELL plasticity.

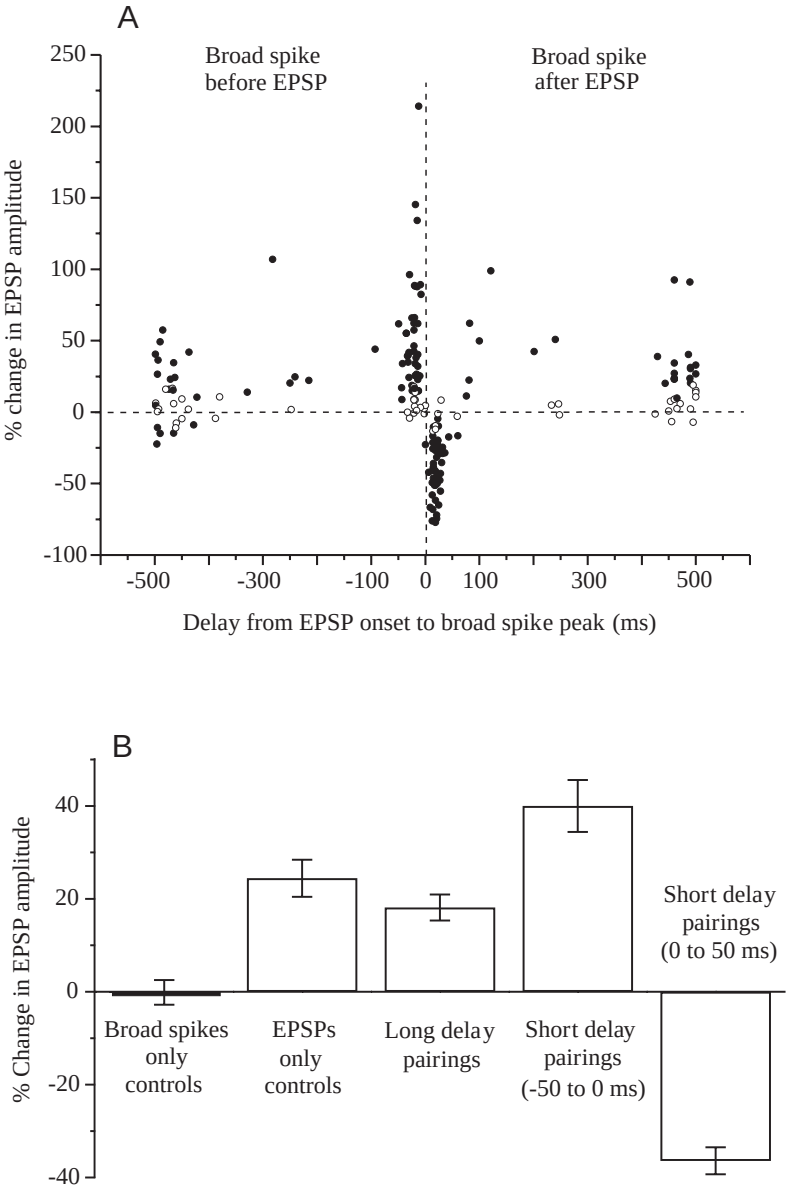

Fig. 7. (A) Percentage change in excitatory postsynaptic potential (EPSP) amplitude plotted against the delay between EPSP onset and the broad spike peak during pairing. A negative delay with regard to the previous spike and a positive delay with regard to the following spike were present for each pairing. The shorter of these two delays is plotted. Filled circles, significant changes; open circles, non-significant changes (at $P<0.01$). (B) Mean changes in EPSP amplitude following unpaired controls and pairings at different delays (bars show standard error; $N=188$ pairings in 43 cells). Long-delay pairings include both positive and negative delays between 300 and 500 ms (taken from Bell et al., 1997c, with permission).

Such a role was tested in 10 cells. Pairings with a broad spike at a short delay resulted in depression of the EPSP in these cells when carried out in normal Ringer, but resulted in no change or enhancement when carried out with the NMDA antagonist AP-5 in the bath (V. Han, Y. Sugawara, K. Grant and C. C. Bell, unpublished observations). Thus, activation of NMDA receptors appears to be essential for the associative depression observed in these cells.

The involvement of the NMDA receptor provides a ready explanation for the sharp dependence of associative depression on the relative timing of the EPSP and the broad spike. Opening of the NMDA channel requires both binding of glutamate to the receptor and significant postsynaptic depolarization to relieve the Mg^{2+} block. The binding of glutamate will occur for a brief period following EPSP onset. If significant postsynaptic depolarization, such as that provided by the broad spike, occurs during this period, then the channel will open, but the EPSP alone or the broad spike alone will not result in channel opening. Plasticity with a similarly sharp dependence on the relative timing of an EPSP and a postsynaptic spike has been demonstrated in both the cerebral cortex (Markram et al., 1997) and the developing optic tectum (Zhang et al., 1998). NMDA receptor activation was also required for plasticity at both these sites.

EPSP enhancement was generally seen with pairings in which the broad spike occurred at delays other than the brief period between 0 and 60 ms. The enhancement may include an associative component, i.e. a component that requires pairing of the EPSP and the broad spike, but such associativity has not yet been demonstrated. Control experiments in which the EPSP was given alone at 1 Hz without any broad spike yielded an enhancement that was not significantly different from pairings in which the broad spike was given just before EPSP onset or at long delays of 200–500 ms (Fig. 7B). These experiments were

confounded by the fact that testing was performed at 0.1 Hz, but pairings with the broad spike and the control in which EPSPs alone were delivered were both performed at 1.0 Hz. Accordingly, additional experiments were carried in which testing as well as pairing and controls were all performed at the same frequency of 0.5 Hz (V. Han, Y. Sugawara, K. Grant and C. C. Bell, unpublished observations). The difference between enhancement due to delivery of the EPSPs alone and enhancement due to pairings with the broad spike evoked at latencies just before the EPSP was greater using this protocol, but was still not significant. Thus, the issue of an enhancement due to an associative relationship between the broad spike and the EPSP is still open. It is quite possible that enhancement is not related to the broad spike and takes place whenever an EPSP occurs with no subsequent broad spikes during the 50 ms following EPSP onset.

Discussion

The associative plasticity observed *in vitro* has many of the same properties as the plasticity observed *in vivo* with normal sensory and motor signals. Both types of plasticity are anti-Hebbian, occur within a narrow temporal window, are input-specific and reverse in the absence of pairing at appropriate delays. The time courses of change for the two types of plasticity are also at least approximately similar. The EOCD is emitted *in vivo* at rates of 2–5 Hz, and plasticity takes approximately 5 min of pairing with sensory stimuli to reach a maximum and a similar period to disappear after pairing (Bell, 1982). Pairing frequency as a parameter has not yet been investigated in studies of synaptic plasticity *in vitro*, but the plastic change appears to reach a maximum after approximately 5 min of pairing at 1 Hz.

The role of synaptic plasticity at parallel fiber synapses has been investigated in two recent modeling studies. A modeling study of the elasmobranch dorsal octavolateral nucleus showed that the generation of negative images of predicted sensory input can be explained by plasticity at parallel fiber synapses (Nelson and Paulin, 1995). These results could be applied with only minor modification to the mormyrid ELL. A second modeling study, specific to the mormyrid ELL, further showed that an asymmetric learning rule, such as the one shown in Fig. 7A, results in the storage of accurate negative images of sensory input patterns, whereas other learning rules, such as symmetrical ones or ones with an opposite form of asymmetry, are much less effective (P. D. Roberts and C. C. Bell, unpublished observations). The latter study also showed that EPSP enhancement, outside the narrow temporal window in which EPSP depression occurs, is essential for the recovery or 'forgetting' that takes place when the sensory input is no longer paired with the predictive parallel fiber inputs. Non-associative enhancement was more suited to this task than any form of associative enhancement.

The shared properties of the *in vitro* and *in vivo* plasticity together with the modeling results argue strongly that synaptic plasticity at parallel fiber synapses makes an important contribution to the generation of negative images of predicted sensory input. Some additional factors must be investigated, however, before the relationship between plasticity at the cellular and systems levels becomes clear.

Plasticity at other sites besides parallel fiber synapses must be investigated. The possibility of plasticity at inhibitory synapses in the ELL has already been mentioned as a subject for future study. In addition, other inputs to the ELL besides parallel fibers may show plasticity. In this regard, it is particularly important to investigate the input to the deep molecular layer from the nucleus preeminentialis, the source of recurrent feedback from higher electrosensory centers. The input to ELL cells from the nucleus preeminentialis has been shown to be plastic in the gymnotid ELL (Bastian, 1996b; Wang and Maler, 1997), and the same could be true in the mormyrid ELL. Finally, the connection from the nucleus preeminentialis is the final link in a series of recurrent loops within the electrosensory system that include the lateral toral nucleus, the valvula cerebelli and the telencephalon. Plasticity probably also takes place at some of these higher levels, and the stored information could be returned to the ELL *via* the nucleus preeminentialis.

The physiological properties of EGp cells and parallel fibers must be established. Axons of EGp granule cells form the parallel fibers. We know something about the inputs to EGp cells, but we know nothing about the normal discharge patterns of EGp cells themselves or about their responsiveness to naturally occurring sensory and motor signals. Correlating the time course with which synaptic plasticity develops with pairing and reverses in the absence of pairing at the cellular level with similar measurements using normal sensory and motor signals will only be meaningful if the discharge properties of parallel fibers are known and simulated in studies at the cellular level. The problem is not unique to the mormyrid ELL. The normal discharge properties and sensory responsiveness of granule cells giving rise to parallel fibers have also not been established in other cerebellum-like structures or even in the cerebellum itself.

In conclusion, the ease with which sensory signals can be delivered and centrally originating predictive signals can be recorded or manipulated in these systems and the adaptive changes that occur when these two types of signals are associated suggest that further study of these systems will be instructive about the general issues of central processing of sensory information and the roles of adaptive mechanisms in such processing. The new findings on plasticity at the cellular level in the mormyrid ELL and in similar structures in other fish provide further support for this conclusion.

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