NEURAL CIRCUITS OF THE CEREBELLUM: HYPOTHESIS FOR FUNCTION

EGIDIO D’ANGELO
Department of Physiology, University of Pavia
Via Forlanini 6, I-27100, Pavia, Italy
and
Consorzio Interuniversitario per le Scienze Fisiche della Materia (CNISM), Via Bassi 6, I-27100 Pavia, Italy
and
Brain Connectivity Center
Istituto Neurologico IRCCS Fondazione C. Mondino
Via Mondino 2, I-27100 Pavia, Italy

Received 19 June 2011
Accepted 28 June 2011

The rapid growth of cerebellar research is going to clarify several aspects of cellular and circuit physiology. However, the concepts about cerebellar mechanisms of function are still largely related to clinical observations and to models elaborated before the last discoveries appeared. In this paper, the major issues are revisited, suggesting that previous concepts can now be refined and modified. The cerebellum is fundamentally involved in timing and in controlling the ordered and precise execution of motor sequences. The fast reaction of the cerebellum to the inputs is sustained by specific cellular mechanisms ensuring precision on the millisecond scale. These include burst-burst reconversion in the granular layer and instantaneous frequency modulation on the 100-Hz band in Purkinje and deep cerebellar nuclei cells. Precisely timed signals can be used for perceptron operations in Purkinje cells and to establish appropriate correlations with climbing fiber signals inducing learning at parallel fiber synapses. In the granular layer, plasticity turns out to be instrumental to timing, providing a conceptual solution to the discrepancy between cerebellar learning and timing. The granular layer subcircuit can be tuned by long-term synaptic plasticity and synaptic inhibition to delay the incoming signals over a 100-ms range. For longer sequences, large circuit sections can be entrained into coherent activity in 100-ms cycles. These dynamic aspects, which have not been accounted for by original theories, could in fact represent the essence of cerebellar functioning. It is suggested that the cerebellum can, in this way, operate the realignment of temporally incongruent signals, allowing their binding and pattern recognition in Purkinje cells. The demonstration of these principles, their behavioral relevance and their relationship with internal model theories represent a challenge for future cerebellar research.

Keywords: Cerebellum; motor control; ataxia; timing; LTP; LTD.

1. Introduction

The cerebellum (Fig. 1) represents one of the most intriguing parts of the brain. The cerebellum is present in the most primitive vertebrates and evolves through phylogeny reaching its maximum expansion in humans. Early functional hypothesis
on the cerebellum preceded and then coevolved with the histological work of Camillo Golgi and Ramón y Cajal [126, 39]. At the beginning of the 19th century, by using clinical observation and experimental ablation experiments, it was clear that the cerebellum was involved in motor control and coordination leading to the definition of the triad of symptoms still used for clinical diagnosis of cerebellar dysfunction: atonia, asthenia, and astasia. Thereafter, clinical observations from soldiers wounded in the cerebellum during the 1st World War and from patients with cerebellar tumors revealed the association with voluntary tremors and diskinesia, leading to the modern concept of ataxia. Thus, initially, knowledge on the cerebellum was exclusively bound to movement. Only later, the role of cerebellum in coordinating emotional and visceral functions, in making sensory predictions and in elaborating certain aspects of cognition has been supported [93, 70, 16, 157]. In addition, recently, dysfunctions of the cerebellum have also been correlated with cognitive disorders including autism, schizophrenia and dyslexia. Therefore, the
mechanisms of cerebellar functioning have to be viewed in an expanded context, of which motor control is just one important aspect [93, 70, 16, 19]. It is possible that the cerebellum operates through a common mechanism affecting different aspects of behavior in turn. In order to define this mechanism, it is useful to evaluate the most characteristic anatomical and functional properties of the cerebellum.

(1) The cerebellum is connected with the most important structures of the central nervous system, including brain-stem, spinal cord, basal ganglia, limbic system, thalamus and several specific and associative areas of the cerebral cortex. Given the homogeneity of the cellular and circuit organization, these new discoveries imply the existence of a basic cerebellar module (Fig. 1) implementing a "general cerebellar algorhythm". The variety of effects deriving from cerebellar functions has then to depend on the areas to which a certain cerebellar module is connected [7, 24, 73, 142, 189].

(2) The cerebellum is extremely fast and reacts to the inputs in about 10 ms, i.e., 1–2 orders of magnitude faster than the cerebral cortex (this is largely due to the feed-forward rather than recurrent organization of the cerebellar circuit; [17]). Moreover, cerebellar control on movements operates in the millisecond range, reaching a precision unattainable in cerebro-cortical circuits [182]. In this respect, and because of its ability to coordinate movement sequences, the cerebellum has been considered a timing machine [62, 61].

(3) The cerebellum is involved in learning implicit memory tasks but, different from the cerebral cortex, it does not retain explicit memory. The cerebellar circuits are capable of strong plastic modifications and can also redirect plasticity toward extracerebellar structures. For this reason, the cerebellum has been considered a learning machine [88–90].

(4) The cerebellum has a characteristic double connectivity with central and peripheral structures (Fig. 2). This would allow the cerebellum to compare cortical with sensory patterns, an operation leading to the sensory prediction process [93, 95, 94].

(5) The cerebellar cortex shows a complex topology determining multiple fractured somatotopy maps, differing from the more linear organization of body parts or sensory features characterizing specific areas of the cerebral cortex (though multiple representations, e.g., of muscles, characterize also the neocortex; [160]). Following the input stage, the signals are propagated through a double vertical and longitudinal organization of fibers. This architecture, together with specific aspects of local neuronal connectivity, is thought to represent the structural basis allowing to process, order and combine complex spatio-temporal sequences [182, 170, 169].

(6) The cerebellum contains about half of the neurons of the whole brain and more neurons than the cerebral cortex (3.6:1) [80] but, differently from the latter, it has little to do with consciousness. Similar to the cerebral cortex organized in columns, the cerebellum is organized in modules. However the cerebellar
modules have relatively weak recurrent connectivity and interconnectivity compared to cortical columns, a fact that has been invoked to explain the marginal implication of the cerebellum in conscious percepts [183, 23].

It is clear that these functional observations do not always find correspondence with symptoms caused by cerebellar dysfunction (which nonetheless have had large application and importance in clinical practice). A new approach to the cerebellum has to include, on one hand, a definition of the fundamental operations that the cerebellum performs and, on the other hand, a detailed investigation of the cellular properties of the circuit. In this paper, I will consider how the current concepts and
observations on cerebellar functions can be reconciled, integrated and extended through recent evidence on cerebellar neurons and network and on cerebellar connectivity.

2. The Cerebellum and Brain Activity

In order to interpret the complex set of neurophysiological and neuropathological data related to the cerebellum, a conceptual framework is required including the principles of cerebro-cerebellar interaction. Brain activity elaborated in the thalamo-cortical circuits, by exploiting internal memories, continuously generates an internal representation that is then compared with the objective world [95, 23, 71, 156, 143]. This operation is assisted by subcortical loops involving the cerebellum, which, by using its own internal memories and a broad set of afferent signals, informs the cortex in a feedback loop about the correctness of predictions [90]. These cortico-cerebellar loops, by improving the identification of errors and novelty (sensory prediction), can trigger automatic corrections, promote learning and redirect attention. This operation is probably part of a more general strategy. The cortex, given the slow elaboration times, the randomness affecting activity in recurrent neuronal assemblies and the incomplete predictability of controlled systems reactions [23], is thought to generate crude commands, which need to be refined, ordered and sorted. These operations are thought to require the precise timing and learning capabilities of the cerebellum [37]. Dysfunction in these mechanisms and loops would be the ultimate cause of several pathological states. In summary, the function of the cerebellum could be conceived not just for its motor consequences but, more generally, within the framework of high-level cognitive processing controlling the correctness of internal brain predictions with respect to the evolution of states in the environment and in the body and brain itself.

2.1. Cerebellar connections and functions

Different parts of the cerebellum contribute to distinct behavioral aspects. The vestibulo-cerebellum, constituted by the flocculo-nodular lobe, regulates equilibrium and vestibulo-ocular reflexes: it receives mostly vestibular and ocular inputs. The spino-cerebellum, including the vermis and the intermediate part of hemispheres, is involved in movement execution including feedback adjustments; it receives somatosensory, labyrinthine, visual and auditory input. The cerebro-cerebellum, represented by the lateral part of the cerebellar hemispheres, plays an important role in preparation, initiation and timing of motor acts via the dentate nuclei; its principal inputs arise from the premotor and posterior parietal cortex. Prominent among these connections, are the loops involving the cortico-cerebellar oculomotor system [73, 72]. Moreover, the association and limbic areas are largely connected with the posterior lobe of the cerebellum, in particular crus I and II.
Reciprocal connections appear to transmit information through the cortico-nuclear microcomplex to the deep cerebellar nuclei and from there to the thalamus and back to the cerebral cortex [99, 104, 145]. By maintaining a similar anatomic organization in all its parts, the cerebellum could organize and modulate cognition and emotion in a similar way as it organizes and modulates motor coordination and control. Accordingly, cerebellar alterations affecting the cerebello-cortical loops may lead not only to motor abnormalities but also to behavioral, cognitive and affective alterations. The cerebellum is fundamental for contextualizing specific stimuli and coordinating their spatio-temporal evolution, generating coherent ensemble activities [93, 95, 94]. Therefore, dysfunction of the cerebellar circuits and of information reentry toward the frontal and parietal cortex may contribute to preventing the formation of coherent and contextualized behaviors. Additionally, the cerebellum is critical for revealing differences (either error or novelty) between predictions elaborated by the cortex and the stimuli conveyed by the senses. Thus, dysfunction of the cortico-cerebellar circuits may prevent the detection of novelty and impair attention switching [93, 95, 94, 4, 5, 33, 132].

2.2. Cerebellar dysfunction

Cerebellar ataxia (from Greek α-τάξις, meaning “lack of order”) [36] is expressed through a variety of elementary neurological deficits, such as antagonist hypotonia, asynergy, dysmetria, dyschronometria, and dysdiadochokinesia. In a very general description, (1) dysfunction of the vestibulocerebellum impairs body balance (Romberg test) and the control of eye movements (saccade alterations, nystagmus), (2) dysfunction of the spinocerebellum impairs gait (wide-based “drunken sailor” gait, characterized by uncertain start and stop, lateral deviations, and unequal steps), (3) dysfunction of the cerebrocerebellum causes disturbances in carrying out voluntary, planned movements. These disturbances include intention tremor (coarse trembling, accentuated over the execution of voluntary movements, possibly involving the head and eyes as well as the limbs and torso), peculiar writing abnormalities (large, unequal letters, irregular underlining), a peculiar pattern of dysarthria (slurred speech, sometimes characterized by explosive variations in voice intensity despite a regular rhythm). Finally, a less known sign is the so called “dysmetria of thought”, which arises as a consequence of the damage of the cortico-cerebellar connections relating the cerebellum to prefrontal and limbic areas. This disturbance configures the cerebellar cognitive-affective syndrome (CCAS) [159]. CCAS is characterized by: (1) disturbances of executive functions, which include deficient planning, set-shifting, abstract reasoning, working memory, and decreased verbal fluency; (2) impaired spatial cognition, including visual-spatial disorganization and impaired visual-spatial memory; (3) personality change characterized by flattening or blunting of affect and disinhibited or inappropriate behavior; and (4) linguistic difficulties, including disprosodia, agrammatism and mild anomia. The net effect of these cognitive disturbances is a general lowering of overall intellectual performance resembling a prefrontal syndrome.
3. The Cerebellum and Motor Control: Theories and Models

The cerebellum has inspired some amongst the most renown theories and models of brain functioning. These have played an important role in fueling discussion and stimulating research on cerebellar mechanisms of function.

3.1. Early theoretical models

The first insight on how the cerebellum might work, came both from the realization that it is involved in motor control and from anatomical analysis and electrophysiological recordings in vivo. These showed that the cerebellum forms a large inhibitory loop controlling the deep cerebellar nuclei, in which excitation is conveyed to Purkinje cells through the mossy fiber–granule cell–parallel fiber pathway and the inferior olive–climbing fiber pathway (Fig. 2) [62, 137, 92, 60]. Then it was found that all the neurons in the cerebellar cortex are inhibitory, with the exception of the granule cells. The reaction times of the cellular elements to peripheral stimulation were reported [61]. These ideas were synthesized by two main theories: Eccles proposed the beam theory [61, 62] and Marr and Albus the motor learning theory [123, 3, 67]. Later on, Ito developed the forward controller theory [88, 90, 87], which considers the cerebellar function in the context of motor system control (see below). Since then, the view of how the cerebellum operates has been focused on three main concepts: (1) the cerebellum operates by decorrelating the inputs and controlling gain in the granular layer and by detecting known patterns in Purkinje cells; (2) pattern recognition in Purkinje cells is regulated by memory storage at the parallel fiber–Purkinje cell synapse under climbing-fiber control; (3) when unfamiliar patterns are detected, the Purkinje cells change their firing rate and regulate activity in the deep cerebellar nuclei, which then emits the correction signals required to control behavior. Under a different perspective, the parallel fiber timing hypothesis maintained that different delays in parallel fiber activity would be critical for explaining cerebellar functioning [20]. Although conduction delays are probably marginal, the timing issue is relevant and has been supported recently by electrophysiological data and cognitive experiments. Other more abstract approaches have also been developed. The mathematical foundations of the Marr-Albus-Ito hypothesis have been investigated through the adaptable filter theory (for a review, see [50]). The tensor network theory implies that the cerebellum acts as a metric tensor establishing a geometry for the central nervous system motor hyperspace [140].

3.2. The motor control models

From the engineering viewpoint, the cerebellum has been supposed to provide the key to resolve the problem of multi-dimensional nonlinear control required for regulating movement [89, 83, 82, 81, 103, 165, 168, 195]. Thus, understanding the cerebellum has been turned into the problem of controlling non-stiff-joint “plants”.
Motor control theories are based on the general connectivity of the cerebellum (Fig. 2) making broad assumptions on its internal microcircuits [166]. When the cortex sends signals to motoneurons in the brain stem and spinal cord, it also sends a copy of this message to the cerebellum (Fig. 2). In addition, information arrives to the cerebellum from muscle spindles, joints and tendons. Therefore, the cerebellum receives both motor commands and sensory signals. This allows extracting corrective models on the “plant” (the ensemble of joints, muscles, and forces) for accurate movements (internal models are dealt with in detail by Amir Karniel in this issue). Traditionally, feedback from an internal model of the “plant” has been used to generate a *forward model* able to predict the dynamics of muscles from the state of the system and to compute a controller output. The natural error signal for learning motor commands is the difference between actual and correct commands (motor error). By considering the possibility that the cerebellum adds corrective signals on the sensory rather than motor signals, it has been proposed a cerebellar control loop called *recurrent model*. In autonomous systems, the correct command is typically unknown. Only information about the sensory consequences of incorrect commands is available, which leads to an error representation based on sensory signals. Both the forward and recurrent models have been proposed as biologically plausible to explain cerebellar motor control functions and may indeed develop a complementary functionality. Experimental evidence locating forward models in the cerebellum has been provided using modeling, fMRI studies [82, 54] and TMS [32, 31].

3.3. From theoretical to computational models

Clearly, while theoretical models are appealing for their conceptual simplicity and can help resolve certain computational problems in motor control, they have very poor biological details and do not help in explaining how the cerebellar network operates. In the last decades, experimental data have suggested indeed that the cerebellum operates in a much more complex manner than predicted by available theories. One of the major issue is that of spatio-temporal dynamics, whose description based on anatomical observations proved insufficient. Schematically, (1) the mechanisms of the granular layer go far beyond simple decorrelation [38], (2) long-term synaptic plasticity does not occur only in the parallel fibers [38, 76], (3) the inferior olive operates as a complex timing system and not simply to control Purkinje cell plasticity [97]. Moreover, the cerebellar neurons without any exceptions, and especially the Purkinje cells and the deep cerebellar nuclei cells, (4) have operative states that go far beyond the concept of firing rate regulation [96] (see below). This means that knowledge on the functioning of neuronal networks of the cerebellum is insufficient at the very least, and that new experimental and computational tools are needed to investigate cerebellar network function and dynamics. Recently, a new generation of computational models has appeared. In these models, neuronal properties are explicitly implemented highlighting the contribution of single cell and synaptic dynamics [176, 127, 107, 118]. The gain of
this approach is realism in neuronal computation, the loss is a high complexity in
circuit functions: a further synthesis and abstraction process is then required to
reconcile computation with a general theoretical framework. A convenient approach
is provided by the Adaptable Filter Theory [51], which can incorporate salient
experimental properties of the network and reinterpret the results of the realistic
computational models.

4. The Neurons and Network of the Cerebellum

The cerebellar network can be subdivided into four main sections: the granular
layer, the molecular layer, the deep-cerebellar nuclei, and the inferior olive [88, 89,
137, 62, 8]. The molecular and granular layer form the cortical part of the cer-
ebellum. The deep cerebellar nuclei complex, which is part of the precerebellar
nuclei, represents the only output pathway of the cerebellar cortex. Signals are
then conveyed to various brain-stem nuclei (notably the reticular nucleus and the
red nucleus) and to the antero-lateral thalamus, through which signals are con-
veyed to the controlateral cortex. The inferior olive complex is the only source of
climbing fibers, while various precerebellar nuclei and spinal cord centers generate
the mossy fibers. The general organization of the cerebellar input–output
relationships is depicted in Fig. 2.

4.1. Structure and function of the granular layer

The granular layer is composed of three main types of neurons: the granule cells, the
Golgi cells and the Lugaro cells. In the vestibulo cerebellum, a fourth type is rep-
resented by the UBCs. The mossy fibers make excitatory glutamatergic synapses
with all these cell types (the connection with Lugaro cells is not yet well defined;
Michael Hausser, personal communication). The Golgi cells make inhibitory con-
nexions to granule cells (and UBCs) and the UBCs inhibit the Golgi cells. Con-
nections between climbing fibers and Golgi cells have also been reported on
morphological basis, although functional determinations are still poor (Galliano and
De Zeeuw, personal communication).

The mossy fibers provide the only excitatory input to granule cells (and to UBCs,
which activate other UBCs or granule cells). The granule cells, are the only neurons
sending excitatory outputs to the Purkinje cells and to the molecular layer inter-
neurons (the granule cells and UBCs are also the only excitatory neurons of the
cerebellar cortex as a whole). In turn, the Golgi cells provide the only inhibitory
input to the granule cells generating a complex combination of feed-forward, feed-
back and lateral inhibition effects (see [69] for review). Importantly, the Golgi cell
inhibitory axons extend far apart from their input field, causing a massive lateral
inhibition. This effect has been largely investigated and documented. Functionally,
the circuit seems designed to allow a massive spatio-temporal reconfiguration of the
input, with several control mechanisms provided by local connectivity, long-term synaptic plasticity, intrinsic excitability and neuromulatory systems.

This seemingly simple circuit contains about half of the neurons of the whole brain and has inspired several functional theories. The combinatorial organization of the granular layer [123] has inspired the hypothesis of signal decorrelation. This hypothesis originated from the observation that granule cells are much more numerous of the mossy fibers. Therefore, signals could diverge over many more lines than in the input, allowing decorrelation of common components.

The granular layer has been recently shown to transform incoming signals by making use of specific cellular mechanisms controlling excitation, inhibition, oscillation and plasticity [37, 65, 35]. The mossy fibers generate spike bursts following punctuate sensory stimulation [25, 100, 147]. The analysis of responses to such bursts has inspired numerous subsequent developments.

(i) By virtue of Golgi cell lateral inhibition, the granular layer response to mossy fiber bursts are spatially organized in center-surround, where excitation prevails in the center and by inhibition in the surrounding areas [120]. It has been recently shown that sensory stimuli (punctate facial stimulation) activate dense clusters of granule cells amounting to about 600 units, and that the clusters activated by sensory fibers and by cortico-pontine fibers are separated [56]. The percentage of spiking granule cells can vary from 2% to over 20% depending on the presence of LTD or LTP in the cluster. In response to the same inputs, localized activation of the Golgi cells can also be observed [56, 181].

(ii) By virtue of Golgi cell feed-forward inhibition, the granular layer generates a time-window effect limiting the duration and intensity of the output [37, 133].

(iii) By exploiting specific properties of NMDA and GABA receptors, the granular layer behaves as a high-pass filter allowing patterns over 50 Hz to be optimally transmitted [121].

(iv) By exploiting extended feedback inhibition through Golgi cells, the granular layer can sustain coherent oscillations [118] synchronizing large granule cell fields [139, 77]. Synchronous oscillations were also shown to exploit electrical junctions between Golgi cells [58]. The oscillations are enhanced when bursts are conveyed in the theta-frequency band due intrinsic cellular resonance [40, 174, 175].

(v) In response to specific burst patterns, the granular layer generates long-term synaptic plasticity at the mossy fiber — granule cell synapse. Since induction is regulated by synaptic inhibition (which controls membrane depolarization and therefore the level on NMDA channel unblock and calcium influx), LTP dominates in the center and LTD in the surround of the response fields, consolidating specific geometries of activity.

(vi) Finally, without requiring the burst activity regime, the granular layer can operate gain control operations exploiting tonic inhibition in the glomerulus [128].
But how do these mechanisms contribute to signal decorrelation and to help extend our understanding of cerebellar functioning? Using a realistic computational model of the granular layer [176], it has been possible to evaluate the impact of cellular and synaptic dynamics on circuit computations. Burst stimulation of a small mossy fiber bundle results in granule cell bursts delimited in time (time-windowing) and space (center-surround) by network inhibition. This burst–burst transmission shows marked frequency-dependence configuring a high-pass filter with a cutoff frequency around 100 Hz. The contrast between center and surround properties is regulated by the excitatory–inhibitory balance. The stronger excitation makes the center more responsive to 10–50 Hz input frequencies and enhances the granule cell output (with spike occurring earlier and with higher frequency and number) compared to the surround. Synaptic plasticity at the mossy fiber–granule cell relay, by exploiting changes in neurotransmitter cycling, can fine tune the transmission properties of the center-surround structure. The center, by generating LTP, reacts to spikes bursts over a broader input frequency range emitting new bursts with shorter delay and higher number of spikes compared to the surround (which is dominated by LTD). Interestingly, the plastic mechanisms revealed in vitro can explain LTP and LTD in vivo, concentrating the firing granule cells in the center and increasing inhibition in the surround [56].

An emerging novel hypothesis is that the granular layer network behaves as a complex set of filters operating in the space, time and frequency domains, and that this filter can be adapted through long-term synaptic plasticity and coordinated by theta-frequency oscillations. Thus, the original idea of input decorrelation may be extended to temporal dynamics of circuit activity and in particular to spike timing, an aspect that deserves future investigations.

4.2. Transmission of signals from granular to molecular layer

The signal generated by the granular layer consist of complex sequences of spikes in granule cells, which need then to be transmitted to the molecular layer. The parallel fibers, after dividing into two opposite branches originating from the ascending axon of granule cells, travel transversally for millimeters contacting numerous Purkinje cells. This characteristic organization has inspired the idea that signals generated by granule cells are conveyed along the parallel fibers activating beams of Purkinje cells (the “Beam Theory”: [62, 20]. Activation of beams is indeed observed using parallel fiber stimulation (e.g., see [190, 11]. However, punctuate stimulation in vivo causes a prominent vertical activation of Purkinje cells overlaying the active granular layer areas [19, 30, 117, 78, 152]. A possible explanation was that vertical activation could reflect either differential synaptic density or strength or differential spike delays along the ascending granule cell axon compared to parallel fiber synapses. It was originally assumed that the difference was not in strength but rather in the timing of the inputs in individual synapses of both types [113, 112]. This hypothesis was supported by the demonstration of the functional equivalence of the two inputs [191,
84], although differences in terms of long-term synaptic plasticity have been reported [171, 172]. Alternatively, differential properties of synaptic inhibition could be critical [63, 141, 129].

Clearly, solving the issue also depends on the development of appropriate recording and analysis methods. For instance, initially voltage-sensitive dye recordings did not allow one to determine the specific cellular components contributing to the signals (e.g., [30] while a more strict correspondence has recently been achieved with high resolution techniques and simultaneous imaging-patch clamp recordings in sagittal and coronal slices [121, 122]. These investigations have revealed that optimal responses to mossy fiber input trains occurred for input frequencies over ~50 Hz in the granular layer and over ~100 Hz in the overlying Purkinje cells. Conversely, Purkinje cell excitation along the parallel fibers occurred already at low frequency (<10 Hz) and did not improve with input frequency. The efficacy of GABA-A receptor-dependent inhibition increased passing from the granular to molecular layer and explained the different frequency-dependent responses in these subcircuits. Thus, while molecular layer mechanisms could enhance vertical transmission of high-frequency bursts, inhibitory systems in the molecular layer would prevent diffusion of such amplified responses along the parallel fibers. This result indicates that both spots and beams of excitation can coexist although with different dynamic properties. It should be noted that another mechanism proposed to regulate gain at the mossy fiber — granule cell relay is based on tonic inhibition in the glomerulus [128, 9]. This, however, may not be sensitive to rapid frequency changes between or during bursts (Fig. 3).

A related issue is that granule cells generate their spikes in the initial segment and these invade the whole ascending axon almost instantaneously, so that delay times are abolished in this circuit section [57]. Generation of spike delays in granule cells through long-term synaptic plasticity, Golgi cell inhibition and time-windowing is therefore the most relevant process controlling the temporal information flow toward Purkinje cells. It should be noted that different delays are expected in the different parts of the center-surround structures of the granular layer [56], thus determining the geometry of activation of the molecular layer.

4.2.1. The molecular layer and Purkinje cell firing dynamics

The Purkinje cells are the biggest and probably the most complex neurons of the cerebellar cortex. The Purkinje cells are critical for cerebellar functioning and have catalyzed scientific interest about their mechanisms. The Purkinje cells respond with precision of few milliseconds in relation to the initiation and termination of movement (e.g., a saccade; [134, 102, 136, 135]) and movement itself can be controlled on this same time scale [182]. The Purkinje cells collect signals generated from about 200,000 granule cells and are therefore suited for integrating information pre-ordered and pre-elaborated by the granular layer. In this sense, the Purkinje cells have been supposed to operate as perceptrons [22]. Moreover, the Purkinje cells receive input from a single climbing fiber relaying signals from the inferior olive. The
Purkinje cells has therefore two different kinds of inputs generating simple and complex spikes in response to parallel fiber and climbing fiber activity [61–64]. These neurons generate a rich repertoire of electroresponsive properties including pacemaking, bursting, rebounds, pauses and bistability [113, 112, 193, 179]. While these fundamental aspects have been revealed quite early, the mechanisms through which Purkinje cells process incoming signals have revealed unexpected complexity and are not yet fully clarified.

The Purkinje cells are composed of two main morpho-functional compartments. Parallel and climbing synaptic inputs, as well as those coming from molecular layer interneurons, impinge on a large dendritic tree generating local synaptic responses [110]. Both complex and simple spikes, in turn, have recently been shown to originate from the first node of Ranvier [6, 28, 42, 131, 138]. It is therefore critical to understand how these two cellular sections communicate their electrical signals.
Double-patch recordings and imaging techniques combined with computational modeling have revealed that, while synaptic potentials generated in the dendritic tree can easily reach the soma, spikes generated in the axon cannot travel efficiently into the dendritic tree because of the filtering properties of the structure. The two sections seem therefore to operate separately, with a dendritic portion working as an almost linear integrator without much interference from the spike generation mechanism [141]. The dendritic tree can, however, make use of climbing fiber signals to generate local calcium waves that have a relevant role for generation of synaptic plasticity [42]. Thus, different from cortical pyramidal cells, the firing state of the neuron cannot influence plasticity by itself, which is otherwise controlled (in particular parallel fiber — Purkinje cells LTD, see below) through an independent regulation of intracellular calcium in the dendrites by the climbing fibers [179, 129, 146].

The Purkinje cells express a complex set of ionic channels determining specific excitable properties. Some of the most recent and relevant observations are reported here. The Na\(^+\) channels, in addition to the transient and persistent current, generate a resurgent current favoring complex spike generation [105, 144]. The T-type Ca\(^{2+}\) channels regulate complex spike generation as well as controlling intracellular Ca\(^{2+}\) [198, 86] along with the P-type Ca\(^{2+}\) channels [184]. The role of the H-current in bistability [193, 6] has been recognized. Membrane potential can switch between two stable levels, UP and DOWN, and this transition is regulated by serotonergic modulation of the H-current [193]. Synaptic inputs (either excitatory from parallel and climbing fibers or inhibitory from molecular layer interneurons) can also bidirectionally shift the Purkinje cell states [153] and corticotropin releasing factor (CRF) has a significant impact on the ease and rapidity of the state transitions (Yarom, personal communication; see [91] for a review of neuromodulators affecting cerebellar functioning). Eventually, firing of simple spikes occurs exclusively during the depolarized state. Finally, Ca\(^{2+}\)-dependent K\(^+\) channels have been shown to play an important role in controlling the burst-pause behavior of the Purkinje cell [149, 26] along with inhibition coming from molecular layer interneurons. Attempts at explaining the combined effect of ionic channels unequally distributed over various compartments and of the interaction with synaptic inputs and intracellular Ca\(^{2+}\) concentration have been carried out by developing multicompartmental models [179, 45, 44, 177].

The activity of Purkinje cells is strongly influenced by molecular layer interneurons, the stellate and basket cells [17, 158, 18]. These neurons operate in feed-forward mode and can limit in time and space (through lateral inhibition) the response of the Purkinje cells. These neurons are almost equivalent for their firing pattern (which is also very similar to that of Golgi cells), but stellate cells lay in the higher part of the molecular layer contacting Purkinje cell dendrites with multiple small contacts, while basket cells lay in the lower part of the molecular layer surrounding the Purkinje cell soma with a large basket like synapse. Therefore, stellate cells have been thought to be more suitable for regulating dendritic integration while...
basket cell to efficiently regulate the spike output. In a model reconstruction including molecular layer interneuron connections to the Purkinje cell dendrites, beams of activity caused by parallel fibers activity occurred only when inhibition was blocked without remarkably affecting Purkinje cell excitability. Therefore, it has been suggested that feed-forward cortical inhibition can regulate the excitability of the Purkinje cell dendrite without directly influencing the Purkinje cell spiking output independent from synapse location [158]. This act is again a reflection of the electroresponsive organization of the Purkinje cell, since shunting synaptic current in the dendrite or in the soma does not make much difference, since in both cases these currents cannot reach the initial segment and activate spike generation therein.

Another relevant issue is that, while the synapse between parallel fibers and molecular layer interneuron is highly efficient and usually shows short-term depression, the synapse between parallel fibers and Purkinje cells is weaker and shows short-term facilitation [171, 172, 12, 173]. Therefore, single stimuli transmitted from granule cells at low frequency are likely to excite the molecular layer interneurons better than the Purkinje cells, which requires short bursts to transmit efficiently. These properties, combined with the spatial distribution of molecular layer interneuron inhibition, indicates that the molecular layer can exert profound filtering effects on incoming signals determining a large variety of patterns. Long-term synaptic plasticity in the molecular layer interneuron network could further design and stabilize these patterns (see below) sculpting the landscape of Purkinje cell activation.

In summary, available results provide a framework for a functional hypothesis on how the Purkinje cells and molecular layer operate. The Purkinje cells are continuously active because of their intrinsic pacemaking, whose average firing rate can be modulated by parallel fiber activity. Inputs from specific input channels of the granular layer can modify this activity state through impulsive inputs conveyed along the ascending axon synapses generating burst-pause responses [179]. Multiples of such inputs, once integrated over the dendritic tree, can appear as changes in spike variance, as indeed revealed in awake animals during specific behavioral tests [37, 48, 164]. Purkinje cells bistability [193, 116] could provide an additional element of control [164, 197], potentially extending the integration time window to longer times. The action of Purkinje cells is limited in time and space by activity in the molecular layer interneuron network, which can therefore substantially regulate the output of Purkinje cells [130]. The Purkinje cells and molecular layer interneuron synapses can be modified by long-term synaptic plasticity, as considered below.

4.2.2. The deep cerebellar nuclei and the cerebellar output

The cells of deep cerebellar nuclei finally convert the activity of microzones into the cerebellar output. The deep cerebellar nuclei are at a key location within the cerebellar network [194]. All of the afferent pathways to the cerebellar cortex make collateral connections on to neurons of the deep cerebellar nuclei (both mossy fiber
and to a lesser extent climbing fiber), while the main output of the cerebellum is formed by the deep cerebellar nuclei projection neurons [13, 186]. In the deep cerebellar nuclei cells, intrinsic dynamics generate subthreshold oscillations, silent pauses and possibly rebound excitation, producing alternating phases of activity [27, 111, 98, 180, 1, 106]. Despite this knowledge, the role of deep cerebellar nuclei in cerebellar computation is largely unclear or controversial [185].

A classical view was that the deep cerebellar nuclei simply acts as a “relay station” between cerebellar mossy fiber (MF) input and cerebellar output to premotor areas, either directly (“direct pathway”) or via the cerebellar cortex (“indirect pathway”). This concept has been challenged by three main observations. First, deep cerebellar nuclei neurons inhibit the inferior olive cells regulating their coupling [53] and can therefore take part in controlling the whole inferior olive-deep cerebellar nuclei—Purkinje cell loop [97]. The disruption of this system would be an important cause of ataxia [26, 109]. Secondly, collaterals of deep cerebellar nuclei axons can travel back to the granular layer and generate closed-loop oscillations [38, 107]. Rebound excitation could help maintaining activity in the theta-frequency range. Thirdly, the deep cerebellar nuclei may act as a substrate for motor memory storage [76, 2, 163]. The demonstrations of mechanisms which cause modification of synaptic strength and active membrane properties support this latter viewpoint (see below). The role of rebound excitation in driving plasticity and circuit oscillations is not yet fully resolved.

In addition to these possible roles in cerebellar memory, it appears likely that the neuronal network within the deep cerebellar nuclei processes sensory and motor information in “real-time” but very little is known about the computational functions of the deep cerebellar nuclei, particularly with respect to the role of specific cell types. Indeed, morphological and electrophysiological studies have revealed that the deep cerebellar nuclei consists of diverse neuronal populations with distinct integrative properties [185]. Thus, one can hypothesize that the synchronous oscillations in the Purkinje cell activities together with plasticity at the mossy fiber — deep cerebellar nuclei and the Purkinje cell — deep cerebellar nuclei synapses form the main mechanistic tools to control the activity in the deep cerebellar nuclei output neurons, and that different sets of neurons in the deep cerebellar nuclei are sensitive for oscillations at different frequency ranges [194].

4.2.3. The olivo-cerebellar loop

By considering the different anatomical origin of parallel fibers and climbing fibers and their double innervation of Purkinje cell, it was assumed that climbing fibers, originating from the inferior olive, play a fundamental role in motor learning [92, 123, 3]. This contrast between the convergence of a single climbing fiber per Purkinje cell and the massive number of parallel fiber inputs provided the sole basis for the Marr-Albus theory [88]. However, soon thereafter new insight arose from the investigation of neuronal dynamics leading to an alternative hypothesis: the olivo-cerebellar loop may work as a timer for motor activity [114, 192].
Recently this alternative hypothesis has been expanded, considering the olivo-cerebellar loop as a generator of temporal patterns [97, 96, 196, 115]. This hypothesis postulates that the temporal patterns are encoded in the complex-spike trains, and is based on three assumptions supported by experimental evidence: (1) Inferior olivary neurons form a network of electrically coupled cells, where the coupling is modulated by inhibitory input from the deep cerebellar nuclei. Only in a network configuration, the neurons generate propagating waves of subthreshold oscillations. (2) The electrical synapses in the inferior olive operate only in the absence of inhibitory input from the deep cerebellar nuclei. Hence, olivary networks are dynamically and continuously reassembled by the activity pattern of the GABAergic projection neurons of the deep cerebellar nuclei [96, 27, 53, 187, 125]. (3) Cerebellar Purkinje cells display complex dynamics of spiking activity, suggesting that a request for specific patterns delivered via the mossy fiber system is translated into patterns of olivary activity, which can in turn reorganize activity in specific sections of the cerebellar cortex by sending climbing fiber signals to Purkinje cell organized in sagittal bands [97, 196].

4.2.4. Cerebellar long-term synaptic plasticity

Marr [123] and Albus [3] predicted that cerebellar learning should occur with some form of plasticity between parallel fibers and Purkinje cells under control of climbing fibers. The climbing fibers originating from the inferior olive were assumed to play the role of a teacher, instructing the cerebellar cortex to modify its connectivity in order to cope with new motor demands. Parallel fiber–Purkinje cell “LTP” was predicted by Marr and reversed into “LTD” by Albus: LTD was in fact discovered more than a decade later by Ito [92]. The resonance of this discovery can be compared to that of LTP in the hippocampus [15], which followed Hebb’s postulate on brain plasticity [79].

In 1984, Eccles said: “For me the most significant property of the cerebellar circuitry would be its plastic ability, whereby it can participate in motor learning, that is the acquisition of skills. This immense neuronal machine with the double innervation of Purkinje cells begins to make sense if it plays a key role in motor learning... it could be optimistically predicted that the manner of operation of the cerebellum in movement and posture would soon be known in principle” (from the foreword to [88]). For more than a decade, the dominant idea had been that LTD was not just the most important but also probably the only relevant form of plasticity in the cerebellum. For instance, Marr [123] explicitly negated the possibility that mossy fiber–granule cell synaptic weights could be modified by activity. He noted that “sooner or later all weights would be saturated” and so plasticity would be useless. The Marr’s model does not include any mf-grc synaptic plasticity (nor any plasticity at other synapses), although the subsequent extension due to Albus [3] is more permissive. However, this view was challenged by new experimental facts.
Although parallel fiber LTD can be robustly reproduced in vitro, its demonstration in vivo has remained partial and its involvement in physiologically relevant behaviors has been questioned [163, 148, 43, 29, 49]. In turn, several novel forms of plasticity have been demonstrated. Taking the move from the consideration that NMDA receptors, the main responsible of LTP induction in the brain [14], are abundantly expressed in granule cells, experiments were undertaken demonstrating that LTP can indeed be generated by mossy fiber bursts at the mossy fiber–granule cell synapse [41, 10]. In addition to LTP, mossy fiber–granule cell LTD was also demonstrated. LTP and LTD were related through a bidirectional learning rule based on intracellular calcium concentration changes driven by NMDA receptors [68, 52, 150] and showed a reciprocal spatial distribution [120] preventing the detrimental “saturation” of plasticity predicted by Marr. Even more significantly, LTP and LTD could be demonstrated in the granular layer in vivo following patterned tactile stimulation [150] and their mechanism was shown to correspond to that expected from experiments in vitro [56]. Once appropriately implemented into a model based on the Marr-Albus-Ito theory, mossy fiber–granule cell LTP and LTD proved able to improve motor learning and performance [166, 108].

Mossy fiber–granule cell LTP was the first form of cerebellar long-term synaptic plasticity to be demonstrated following parallel fiber–Purkinje cell LTD. This finding was soon followed by numerous others revealing novel forms of plasticity at the parallel fiber–Purkinje cell synapse and at the Purkinje cell–deep cerebellar nuclei synapse (reviewed in [89, 76]). At variance from classical LTD, some forms of LTP are purely presynaptic, for others there is evidence for a bidirectional postsynaptic switch [163, 167]. For all of these, the experimental results are now sufficiently clear to formulate appropriate learning rules and gating processes (e.g., see [166, 29, 52, 178, 75]. Thus, the cerebellar network is plastic in a more extended sense than originally envisaged. The functional meaning of this extended plasticity in computational terms remains largely to be assessed.

The plasticity issue requires further comments. First of all, that just because a synapse can be shown to be plastic, does not necessarily mean that it is involved in “learning” in the classical sense. It is likely that at some level all synapses in the brain are plastic, the question is for what functional purpose. For example, the classical parallel fiber–Purkinje cell LTD is Hebbian and supervised in nature, while the aforementioned mossy fiber–granule cell LTP and LTD are Hebbian but unsupervised, with fundamentally different impact on learning and behavior. In fact, as far as we can understand, plasticity in the granular layer could tune the response timing of specific granule cells and therefore the activation patterns of Purkinje cells, opening questions about the meaning of “motor learning”. Secondly, some forms of LTD appear to be trans-synaptic. That makes cerebellar learning, explicitly not-Hebbian. The apparent fact that active parallel fiber synapses can influence the weights of inactive parallel fiber synapses is a fundamental problem for traditional theories of cerebellar Purkinje cell learning [76].
4.2.5. Cerebellar synchrony, oscillations and resonance

The first hints that the cerebellum might generate coordinated activities was raised by the discovery of gap junctions (the molecular correlate of the electrical synapses) in the inferior olive between olivary neurons and in the molecular layer between stellate cells [178, 119]. More recently, gap junctions have been observed in the granular layer between Golgi cells [58, 188]. It was then observed that, as well as other major systems of the brain (like the thalamo-cortical system), also the olivocerebellar system can generate rhythmic activities (reviewed in [35, 47]). In addition, intrinsic neuronal properties can contribute to generate oscillations: the inferior olivary neurons can generate theta-frequency oscillations, and recently a similar property has been found in Golgi cells [55, 66].

Low-frequency oscillations are fundamental for several neurophysiological processes, including sensory motor control, the formation of memories and sleep (for review see [23]. Sensory motor control is based on 6–9 Hz oscillations [74] spreading through a cerebro-cerebellar loop involving cerebral cortical areas (prefrontal cortex, PFC; premotor cortex, PMC; primary sensory motor cortex, S1-M1; posterior parietal cortex, PPC), cerebellum, thalamus and back to the cerebral cortex [161]. The fundamental operations controlled by cerebro-cerebellar loops are explained through the “timing hypothesis” (the temporal goal is a requisite component of the task representation [182]) and the “sensory prediction” hypothesis (match between the predicted and actual sensory outcome of motor commands [59]), [182, 95, 94]. The proposed functions of the cerebro-cerebellar loop include motor sequence generation, sensory motor control, switching of attention and decision making. Some circuit elements of the cerebro-cerebellar loop can intrinsically generate and sustain the rhythm while others are probably entrained by circuit activity. These two mechanisms, entraining and being entrained, are probably not disjoined because large-scale brain oscillations are collective processes, in which coalitions of neurons transiently reinforce their reciprocal interaction. Voluntary movement causes oscillatory activity in the prefrontal areas, which then propagates to the PMC, S1-M1, PPC and is then relayed to the cerebellum (through pontine nuclei). The cerebellum may therefore initially be entrained and then participate to reinforce theta-band oscillations in the cerebro-cerebellar loop.

Both the granular and molecular layer can be entrained into theta-frequency cycles driven by the cerebral cortex [34, 154, 124]. The granular layer spike patterns, once recognized by Purkinje cells, could set up theta-frequency oscillations in specific subsection of the inferior olive–Purkinje cell–deep cerebellar nuclei system (see above). Due to climbing fiber branching, this activity would affect specific groups of Purkinje cells located along the sagittal axis. The parallel fibers (which can reliably transmit at low-frequency, see above), could drive into the theta-frequency Purkinje cell beams located along the transverse axis. The final effect would be a positive interference of theta-frequency activity in Purkinje cells that lie at the intersection between the transverse (parallel fiber-driven) and sagittal (climbing
fiber-driven) groups. This mechanism could provide the necessary coherence for multiple inputs occurring in different regions of the cerebellum, extending the concepts of congruence of climbing and mossy fiber signals [107, 21].

5. Hypotheses and Open Issues for Cerebellar Functioning

The recent findings on cerebellar neurons and circuits reported above [39] suggest to revisit concepts on cerebellar network functioning in view of the spiking nature of neuronal outputs, of the specific dynamic processing that neurons and synapses impose to input signals, and of the intricate interconnectivity within the network. For simplicity of description, cerebellar operations can be divided into the following three steps:

(1) Spatio-temporal reconfiguration in the granular layer

This sub-circuit provides the gain and phase regulation required to reconfigure the input signals, eventually emitting spike patterns appropriate to control molecular layer interneuron and Purkinje cell activity [37]. Signals coming as bursts along the mossy fibers are reconverted into bursts and several processes control the granular layer output pattern (delay, duration, frequency, regularity), including synaptic plasticity, tonic and phasic inhibition and glomerular crosstalk. Feed-forward inhibition enforces the time-window mechanism and cause the emission of patterns consisting of short spike bursts (1–3 spikes) at high frequency maintaining high temporal precision (ms range). Presynaptic expression of long-term synaptic plasticity at specific mossy fibers–granule cell synapses and intrinsic excitability regulation in granule cells provide the basis for storing memory in the circuit (learning). Eventually, the architectural arrangement of circuit elements would allow to combine and sort different inputs and to distribute the output along many more lines (expansion recoding) with differential delays and gain determined through long-term synaptic plasticity. In this manner, the granular layer analyzes the signals and subdivides them into their multiple components (adaptable filtering) (Figs. 3 and 4). This mechanism can generate the spatio-temporal transformation required for ordering signal sequences in a complex input space. These include commands sequences emitted by the cerebral cortex or coming from different sensory systems (timing) and ensembles of cerebro-cortical commands and feedback sensory signals during voluntary movements (sensory prediction). This mechanisms operate on the 1–100-ms scale, and could account for learning elementary associative behaviors like eye-blink conditioning and conditioned responses in general. On a longer time scale, synchronization processes are needed. These can be provided by low-frequency oscillations, which can set the coherent temporal framework on the 100-ms scale for the synchronization of complex data-sets and repetition of activity cycles. In the vestibulo-cerebellum, the additional presence of UBCs could allow for further temporal storage required for implementing slow VOR reactions.
Signal synthesis in the molecular layer and deep cerebellar nuclei

The molecular layer elaborates the patterns generated by the granular layer. These patterns arrive along vertical lines organized in center-surround, so that activation of Purkinje cells and molecular layer interneurons laying above a specific granule cells cluster is privileged [17, 152, 18, 151]. Transmission along this line is almost instantaneous (<1 ms), consistent with the fact that delay regulation occurs in the granular layer [56]. Parallel fibers may not be important either to activate specific Purkinje cells or to generate remarkable delays, but rather to transmit low-frequency patterns and to set the background discharge of Purkinje cells. The Purkinje cells would operate as oscillators, whose frequency can be almost instantaneously increased or reduced by the contribution of their numerous inputs along the ascending brand and parallel fibers. The molecular layer interneurons can generate a pause just after Purkinje cells have discharged, accentuating the burst-pause pattern and maintaining the precision of response into the ms range (timing).

By collecting information from as many as 200,000 granule cells, the Purkinje cells are well suited to operate the synthesis of the multidimensional granular layer transformations. Plasticity at the mossy fiber–Purkinje cell synapses and at molecular layer interneuron–Purkinje cell synapses can store the patterns detected...
from the granular layer generating a more stable, reach and redundant representation of the input space. Inputs from the inferior olive can provide further synchronizing signals at low frequency, potentially integrating with the low-frequency oscillations conveyed from the granular layer, and could also favor LTD on parallel fibers activated in close time synchrony (Fig. 3). The UP/DOWN states could extend timing regulation in Purkinje cells over the second range.

Eventually, deep cerebellar nuclei neurons need to be able to perform a secondary synthesis by sampling several Purkinje cell contributions as well as inputs from the mossy fibers [13]. This operation needs also to occur with ms precision to account for the final precision of the cerebellar computation and of motor behavior as a whole.

(3) Modularity and synchronization in the deep cerebellar nuclei/Purkinje cell/inferior olive loop

The cerebellum is organized in a manner that allows the operation of multiple modules with partial interdependence. In such modules, the mossy fibers and the Golgi cells distribute their ramifications to the granule cells and the climbing fiber activate multiple Purkinje cells on the sagittal axis. The activity in the granular layer is organized in clusters, which provide a preliminary selection of active neuronal assemblies. Conversely, parallel fibers travel along the transverse axis allowing to merge the mossy fiber and climbing fiber pathways at the multiple intersections of the two afferent fiber systems. This organization could allow to organize the numerous granule cell clusters and Purkinje cells activated by the mossy fiber pathway into functional assemblies provided by climbing fibers following activity generated in the inferior olive. The inferior olive—Purkinje cell-deep cerebellar nuclei loop could also be able to generate low-frequency sequences based on the migrating depolarizing waves observed in the inferior olive and on the feedback control exerted through inhibitory deep cerebellar nuclei—inferior olive connections [97]. The synchronization of low-frequency activity in the deep cerebellar nuclei/inferior olive/Purkinje cell loop with the mossy fiber input may occur at the intersection of granular layer clusters and climbing fiber domains as well as though the influence of the climbing fibers on the granular layer (Fig. 3).

5.1. Learning and timing: An integrated hypothesis on cerebellar mechanisms

Learning seems to occur at multiple critical points in the cerebellar network [76, 163, 49]. Distributed learning would allow a maximization of storing capabilities and may have different meaning in different subcircuits. In the granular layer, LTP and LTD are translated into signal timing and determine a spatial reconfiguration of activity (Fig. 4). In Purkinje cells, LTP and LTD could have the role of revealing the appropriate coincidence detection of impulses coming from the parallel fibers and climbing fibers. In the deep cerebellar nuclei, LTP and LTD could have the role of revealing the appropriate coincidence detection of impulses coming from the mossy fibers and the Purkinje cells. Plasticity in the inhibitory loops provided by the Golgi
cells and molecular layer interneuron may be homeostatic, counterbalancing the modifications of strength along the main mossy fiber–granule cell–Purkinje cell axis. In all cases, various mechanisms seem suited for regulating different aspect of learning and memory and to translate it into circuit operations. However, eventually plasticity at multiple sites could have a reflection on gain and timing, so that distinguishing a specific role of plasticity for each type of synapse may be quite hard.

Since the cerebellum is thought to learn how to order input sequences (e.g., activation of muscles in a complex movement) and how to determine the proper matching between inputs of different origin (e.g., motor commands and sensory feedback), long-term synaptic plasticity could be primarily involved in setting the appropriate reaction times of the different elements in the network (see Figs. 3 and 4) [37, 38, 48]. Let suppose that signals come with different delay but need to be synchronized in order to allow their simultaneous processing. This case is relevant indeed, since the Purkinje cells have a limited integrative time window (about 10 ms) that is dictated by their membrane time constant and would therefore process two inputs as coincidental only if they fall within the same time window. This time window matches the spontaneous firing of the Purkinje cell: with a basal rate of 50–100 Hz, spike displacement would be meaningful on the scale of a few milliseconds. Now let assume that signals are channeled through two granule cell clusters (Fig. 4). If LTD delays the response of the first granule cell cluster and LTP anticipates the response of the second, signals coming from the two clusters would arrive simultaneously on the receiving Purkinje cells. In order to better exploit this mechanism, the granule cell clusters need to be aligned with the same Purkinje cells (related clusters lay indeed in close vicinity). The Purkinje cells should therefore be able to perform the comparison of the two signals through coincidence detection of the two stimuli [22]. Important to say, the perceptron operation can exploit a second alignment provided by feed-forward inhibition exerted by molecular layer interneurons [85].

In this way, a motor command could be aligned with its related sensory feedback arriving tens of milliseconds later. Or a conditioned and unconditioned stimulus could be associated over a 100-ms scale. Or, by an inverse mechanisms of decoupling, a global motor command (e.g., a reaching movement of the arm involving well-timed sequences of shoulder, elbow, wrist and fingers extensions) could be decomposed into an appropriate timed sequence of commands for the different muscles involved. Operations of logical association of exclusion could further ordinate cluster activity [121]. By regulating the parallel fiber synaptic strength appropriately, the Purkinje cell would then set the contribution of the different clusters to the combined signal. In response to the simultaneous activation through the two clusters, the Purkinje cells could eventually be able to generate a burst-pause response. One should then imagine to reiterate the process over many clusters and over many Purkinje cells. Eventually, the convergence of these many Purkinje cell signals on the deep cerebellar nuclei neurons would impart the correction to the mossy fiber inputs that these latter neurons receive.
Clearly, in order to have this mechanism working properly one needs to imagine local learning rules with the following properties: (1) The active cluster in the granular layer should influence each other, thus generating LTD and LTP in specific orders, (2) learning should be regulated by neuromodulators relating it to the attentional and global state of the subject, (3) plasticity should be reversible in case of changes in the motor strategy (e.g., inversion of a reflex) or osteo-muscular conditions (e.g., a change in load or an injury), (4) plasticity should be consolidated (e.g., through gene expression and protein synthesis) to ensure its long endurance, (5) mechanisms of saving should be enabled to recover previous memories after reconfiguration, (6) learning should be coordinated by low-frequency oscillations under the guidance of climbing fibers and the control of cortico-cerebellar dynamics. The identification of the corresponding cellular mechanisms is a challenge for future research.

The mechanism could be exploited in several contexts. In the eye-blink conditioning reflex, the conditioned and unconditioned stimuli have to be precisely associated through learning with precision of few milliseconds, implying mechanisms compatible with those considered here in the granular and molecular layer. In programming a saccade, the eye moves at high constant speed in a ballistic manner and the control of its end-point corresponds to the time required to reach it with a precision on millisecond scale. Again the present mechanisms could guarantee the association of the sensory and motor cues through memories appropriately pre-programming the execution times. In the vestibulo-ocular reflex, plasticity could account for adaptation of phase and gain of the reflex, and the longer execution times could require protracted excitation along UBC chains.

This timing/learning mechanisms could provide the substrate for coordinated signal processing in the 100-ms range with precision on the millisecond scale [46]. Longer sequences could be ordered on the basis of theta cycles, which repeat every about 100 ms (Fig. 3). This is indeed the dominant activity initiated in the cerebral cortex when a motor command is generated and into which the cerebellum is entrained [74, 161, 162]. In the Purkinje and deep cerebellar nuclei cells, climbing fibers could deliver signals correlating learning with the theta-frequency cycle.

6. Conclusions and Future Directions

This revision on functional mechanisms of the cerebellum shows that original postulates need to be profoundly revisited in view of the recent acquisitions of cellular and circuit neurophysiology and of computational modeling.

As far as the general function of the cerebellum is concerned, in addition to the well established role in sensory motor control, a role in cognition and emotion is also receiving support. Available observations suggest that this is not just used for improving motor control but also to generate a broad-range control of cerebral functioning. The proposal that the cerebellum makes use of a standard circuit structure to control multiple aspects of brain activity remains to be demonstrated.
The granular layer emerges with novel functions and with much more relevant and complex properties than previously thought for spatio-temporal reconfiguration of the inputs. The role and function of Purkinje cells and deep cerebellar nuclei cells in signal processing is under re-evaluation and crucial issues remain to be solved about the functional mechanisms of these neurons and their plastic modifications. Moreover, circuit architecture is not yet fully defined with open questions regarding the functional connectivity between the inferior olive system and the granular layer, between mossy fibers and Purkinje cell and between Purkinje cell and deep cerebellar nuclei. Beside this, it is not yet clear how the cerebellum forms its specific connectivity during ontogenesis [155], which presumably implies both programmed mechanisms and structural plasticity.

If understanding signal coding and morpho-functional connectivity still reveals unknown aspects, understanding the role of plasticity seems even harder. A critical and yet unresolved issue is not just about how the learning rules operate but also about how learning is controlled. Learning cannot occur indiscriminately but needs to be gated to allow its deposition at specific synapses, to prevent destructive interference with previously acquired memories, and to provide a simple and reliable mechanism to store information about multiple tasks. The role of teacher and instructor attributed to the climbing fibers by theory seems far too simple to cope with the complexity of synaptic processes revealed in the cerebellum and does not answer to the question of where motor errors are detected. A related paradox is that of “temporal credit assignment”, since the climbing fibers, which are supposed to convey the motor error, generate the calcium spike required for LTD before parallel fibers, which relay the signals to be learned. If the cerebellum has to detect the motor errors and process them in coincidence with the motor commands, it should therefore embed appropriate mechanisms to do so rather than be taught by an external entity. This external teacher has in fact never been identified but may not be the inferior olive, which rather works as a highly synchronized and specialized clock. It is therefore suggested that the learning rules and their control mechanisms will have to be identified within the cerebellar network itself (e.g., through mechanisms like that proposed in Figs. 3 and 4).

Beside these unresolved aspects, the cerebellar circuit appears well suited to regulate timing in the millisecond range, to order complex multidimensional sequences on the theta-band, to compare different sets of inputs and to regulate its internal dynamics on the basis of learned synaptic modifications. In its essence, the historical contrast between learning and timing could be solved by considering that synaptic plasticity is the substrate for tuning the timing mechanisms of the cerebellar circuit and that oscillations and resonance regulate plasticity (“plasticity is timing”). These mechanisms can in turn implement a general algorithm allowing to reconfigure incoming patterns in the huge multidimensional space of the mossy fiber input. This algorithm, by operating inside different brain loops, could determine the various functional and pathological consequences of cerebellar activity. Realistic modeling can provide a critical step toward the reconstruction and investigation of
these network properties (e.g., see [166, 176, 127, 107, 148, 101]). Casting these concepts into a well-defined theoretical framework (which, like the adaptive filter theory, may be able to incorporate learning rules and cell dynamics [51]), could provide new clues on cerebellar circuit functioning reconnecting biology to general theories of signal processing and eventually explaining how the cerebellum is actually operating.

Acknowledgments

This work was supported by the European Union (CEREBNET FP7-ITN238686, REALNET FP7-ICT270434) to ED. Particular thanks to Francesca Prestori for help in setting the final layout of the manuscript.

References


