Modeling cerebellar granular layer excitability and combinatorial computation with spikes

Chaitanya Medini, Sathyaa Subramaniyam, Bipin Nair, Shyam Diwakar*
School of Biotechnology, Amrita Vishwa Vidyapeetham (Amrita University)
Amritapuri, Clappana P.O., 690525
Kollam, Kerala, India
*e-mail: shyam@amrita.edu

***Abstract*—The cerebellum input stage has been known to perform combinatorial operations [1] [3] on input signals. In this paper, we developed a model to study information transmission and signal recoding in the cerebellar granular layer and to test observations like center-surround organization and time-window hypothesis [1] [2]. We also developed simple neuron models for abstracting timing phenomena in large networks. Detailed biophysical models were used to study synaptic plasticity and its effect in generation and modulation of spikes in the granular layer network. Our results indicated that spatio-temporal information transfer through the granular network is controlled by synaptic inhibition [1]. Spike amplitude and number of spikes were modulated by LTP and LTD. Both *in vitro* and *in vivo* simulations indicated that inhibitory input via Golgi cells acts as a modulator and regulates the post synaptic excitability.**

Keywords: Cerebellar granular layer, network, modelling, plasticity, inhibition.

I. INTRODUCTION

Cerebellar granular layer forms the input stage of the cerebellum in which information coming from the peripheral and central systems converge through the mossy fibers. The granular layer has by far the smallest (~ 5μm) and the most numerous neurons (~ 10^{11}) in humans. Understanding how the granular layer process information appears critical to understand the cerebellar function, since signals coming into upper cortical layers are provided by the granular layer. The granule cells form the largest neuronal population in the mammalian brain and regulate information transfer along the major afferent systems to the cerebellum. The granule layer receives excitatory input primarily from mossy fibers and inhibitory interneurons like Golgi cell. The Mossy fiber input excites both the granule cell and inhibitory interneurons like Golgi cell. The granule cell is a small neuron with three-five dendrites.

In this paper we focus on the impact of excitation on granule cells. The study also includes understanding the effect of inhibition on the granular layer circuitry, and ensemble activity seen in populations of granule cells in terms of combinatorial operations on granular layer network [3].

In order to estimate spiking behaviour, we tried using detailed and simple models of neurons in our network models. Using properties of the granule cell [4] and Purkinje cell [5], we developed simple spiking models [6] to represent the spiking behavior in a network. Estimates of spiking and reproducibility of spiking could be very useful for computationally efficient and large network models.

We also expanded the network to use biophysical models such as [5], [7], [8], [9] to build a biophysical realistic model. In the cerebellar network, another important study was to configure the complex spike activity observed experimentally in the Purkinje cell [5]. To complete Purkinje neuron model in terms of its biophysical relevance, we modeled complex spikes which were characteristic of Purkinje neurons. The complex spike is characterized by 2-3 spikes as a burst and a characteristic after-hyperpolarization (AHP). Modeling changes in calcium and potassium conductance, the effect caused by climbing fibre inputs were reproduced.

The characteristic study on our network model was the effect of inhibition. Our granular layer network model contained 1680 granule cells (GrC), 1 Golgi cell (GoC) excited by 218 Mossy-fiber (MF) rosettes, where 140 homogenous Mossy-fiber rosette exciting granule cells through 140 glomerular connections and the 78 homogenous MF rosettes for Golgi cell excitation [3][10]. Varying strengths of excitatory inputs from the center to the periphery were applied to the model to understand the concept of center-surround activation patterns. Another goal was to understand the effect of combinatorial operations on the granular layer network. Combinatorial operations included combined excitation and inhibition which forms the spatiotemporal pattern in granular layer network *in vitro* and *in vivo*. All modeling and simulation were made using the NEURON environment [11].

The paper reports the effect of inhibition on information transfer along the granular layer network. Spike properties of underlying cells and modulation of spikes were also studied. Variations due to synaptic plasticity in the granular network and modulation of LTP-LTD are also reported.

II. METHODS

The study carried out in this paper involved the use of computational models of neurons, modified ion channel properties for intrinsic excitability and analysis of simulation data.

A. Neuronal models

Mathematical neuron models of granule cell [4], [7], Purkinje cell [5] and Golgi cell model [8] [9] were used in this network study. A single compartmental model was used to represent granule cell and was adapted from [4] with 13-state sodium channel model from [7]. Modeling reliability for spiking models was based on the extensive characterization of membrane currents and the compact electrotonic structure of cerebellar granule cells [4] [7]. The model used AMPA and NMDA components as excitatory MF-GrC synapses and GABAergic synapses for the Golgi cell- GrC relay [7].

978-1-4244-6439-5/10/$26.00 ©2010 IEEE

B. *Granular layer network*

Granular layer spiking network model consisted of 140 homogenous Mossy fibers rosettes (MF), 1680 Granule cells (GrC) and 1 Golgi cell (GoC). In this network, about 48 GrC receive 1 excitatory input from the same mossy fiber and each granule cell receives four excitatory connections from four different mossy fibers. Along with these excitatory inputs given to GrC, mossy fibers also give excitatory input to GoC whose ratio is about 78:1 giving overall glomeruli connectivity pattern. The network topology is shown below in Fig. 1. The study have been made by varying the level of inhibition uniformly over the network of 1680 Granule cells and by varying the release probability of the GABAergic synapses from 0.1 to 0.8. *In vitro* like behaviors were studied by giving single spike as input. *In vivo* like behavior were characterized by burst. Short burst means 5 spikes per burst and long burst means 9 spikes per burst.

C. *Center-surround "spot" pattern*

Stimulating mossy fibers with an electrode at a particular point activates granule cells in the network in a center-surround activation pattern [3]. Within a 'spot', cells which are in close proximity to the electrode will receive high excitation and the periphery layer cells receive less excitation. Our model of the granular network shows the pattern activated as a 'spot'. The center-surround pattern showing the decreasing strengths of excitation can be noticed from the center to the periphery.

D. *Simulating LTP/LTD*

By modifying intrinsic excitability and release probability [12], we simulated plasticity in the granule cells. We modified intrinsic excitability by changing ionic current density or gating. We modified the on-off gating characteristics of sodium channel to modify sodium activation and inactivation parameters [13] for higher and lower intrinsic excitability.

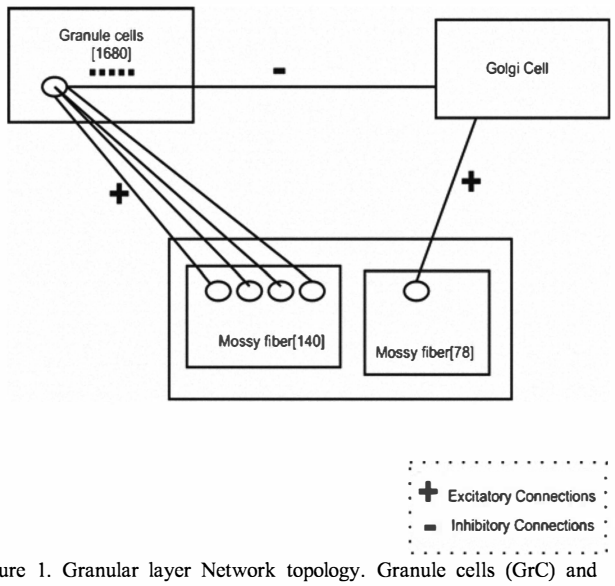

Figure 1. Granular layer Network topology. Granule cells (GrC) and Golgi cell (GoC) receive excitatory inputs from mossy fibers. GrC receive inhibitory input from a single GoC.

III. SPIKING BEHAVIOUR IN ARTIFICIAL MODELS

Large scale network contains many spiking neurons which needs a suitable model to represent their behavior effectively with the reasonable computational cost. We tried using simple neuron in network model. A good model should be feasible with Hodgkin-Huxley dynamics and be computationally efficient [6] to reproduce the firing behavior of bio-realistic model. The firing pattern of Granule cell [4] and Purkinje cell [5] were reproduced and these simplified models were used in Granular layer network.

Four parameters regulate the spiking and bursting behavior of these bio-realistic models [6] (Fig. 2 B1, B2). These parameters include time scale of the recovery variable (a), sensitivity of the recovery variable (b), after-spike reset of the membrane potential (c), after-spike reset of the recovery variable (d). Modifying the parameters in different ways result in various intrinsic firing patterns [6] (Fig. 2 C1, C2).

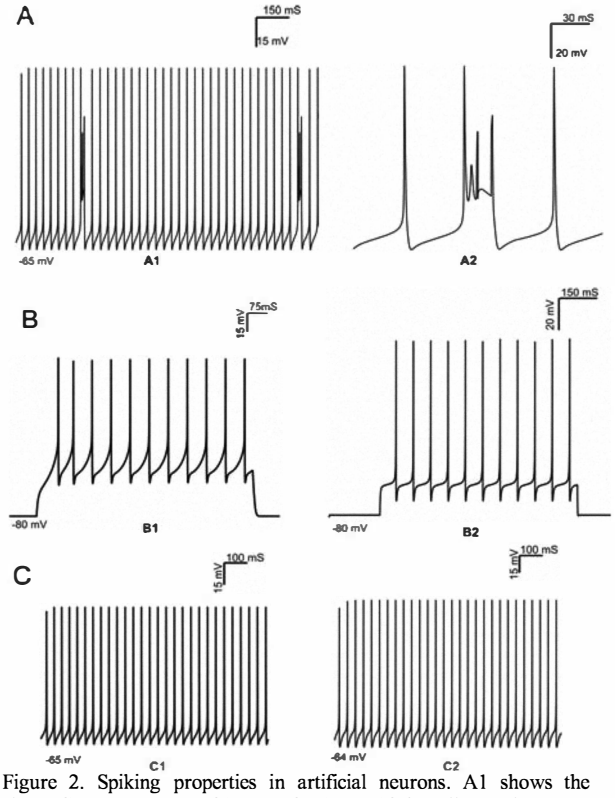

Figure 2. Spiking properties in artificial neurons. A1 shows the complex spike reproduced with 1 Hz in the Purkinje cell model [5]. A2 shows the complex spike reproduced by the model. B1 shows firing pattern of granule cell model [4] with a current injection of 12pA while B2 shows the simple spiking model. C1 shows simple spikes by Purkinje cell model [5] while C2 shows simple spiking in our adapted Izhikevich model [5].

TABLE I. ARTIFICIAL NEURON MODEL PARAMETERS

Bio-realistic models\Parameters	a	b	c	d	I (pA)
Granule cell model	1	0.26753	-65	1.55	12
Purkinje cell model	0.204	0.26478	-66	0	0

a. Parametric values used in artificial cell to reproduce the spiking behavior of granular and Purkinje cell models.

Different parameters used for representing the firing behavior of these bio-realistic models as given in Table I.

A. *Calcium and potassium conductance modulation plays key role in complex spike induction*

To complete the Purkinje cell, both simple and complex spikes needed to be reproduced. Complex spikes are thought to represent a critical signal for the operation of the cerebellar cortex, conveying both timing information [14] [15] and triggering synaptic plasticity [15] [16] [17] [18]. Simple spikes have a frequency of ~50 Hz [19] [20] and complex spikes have a frequency of ~1 Hz [19] [20] in *in vivo* condition. To regulate these kinds of spikes, Purkinje neuron contains morphology and ionic mechanisms [20]. Purkinje neuron model [5] contains channels which can modulate the mechanisms to reproduce the complex spike behavior in this model.

For complex spike behavior in our Purkinje neuron model [5], conductance of the channels like P-type calcium current, BK-type calcium-activated, slow TEA-insensitive potassium current were modified at different time intervals. At different time intervals, the conductance of these channels was varied to match the complex spike like behavior [15]. When a complex spike input is induced in the Purkinje neuron (Fig. 2 A1, A2) [5], the conductance of the above mentioned channels change with respect to the time interval (see Table II). Complex spike is a burst of small spikelets [21] and to model it, temporal alterations in depolarizing phase of the simple spike were done. To regulate the spike in the model, dynamic variation of conductance was performed (see Table II).

With a complex spike, the first 6.9ms was the first spikelet, another spikelet followed until 7.7ms and then an interval until 15.5ms and finally an AHP till 16.3ms. To model a spikelet calcium conductance was increased, calcium-dependent potassium conductance and K-slow conductance were decreased (Fig. 2 A2). AHP was obtained resetting the conductance values. After all changes, channels were restored to default values for normal spiking behavior of the model (Fig. 2 C1, 2 A1 and Table II).

IV. EXCITATION AND PLASTICITY IN GRANULAR LAYER NETWORK

LTP in granule cells [12] comprises of variation in release probability and intrinsic excitability. The network model was tested with high intrinsic excitability observed by changes to sodium channel properties and release probabilities of MF synapses, thereby simulating granule cell LTP. LTD was also simulated by combining low intrinsic excitability and low release probability. Without varying release probability of MF synapses and by varying intrinsic excitability of the model cell, the spike amplitude changed slightly (~ 6%) although the number of spikes remained constant.

A. *In vitro network excitability changes with varying excitatory release probabilities*

In the case of *in vitro* like behavior of network, with release probability 0.416 (Control) the cells with normal intrinsic excitability receiving 4 excitatory inputs produced doublet and cells receiving 3 excitatory inputs produced single spikes.

TABLE II. VALUES OF CONDUCTANCE

Conductances\time interval(TI)(ms)	desired ms - 6.9 ms	Till 7.7 ms	Till 15.5 ms	Till 16.3 ms	Till 30 ms
Cap (mho/cm^2)	6e-05	0.00055	0.05	0.007	6e-05
Bkpkj (mho/cm^2)	1e-05	1e-06	3.8	0.004	0.0004
K slow (mho/cm^2)	0.0001	0.004	2	1e-06	1e-06

a. Modified values for conductances to reproduce the complex spike in Purkinje cell model.

Cells receiving 2 excitatory and 1 excitatory input did not produce any spikes.

Decreasing MF synapse release probability from 0.3 to 0.1, all granule cells in the network did not generate spikes (Fig. 3A, B). With release probability of MF synapses = 0.3, the cells receiving 4 excitatory inputs produced single spike with increased first spike latency and decreased spike amplitude when compared to the control condition (Release probability of MF synapses U=0.416) (Fig. 3C) and there is a decrease in number of spiking cells (from 192 to 48). When we increase the release probability of MF synapses from 0.42 to 0.6, there is an increase in number of spikes with no significant change in first spike latency and spike amplitude where the number of spiking cells increased (from 192 to 432) (Fig. 3D, E).

With the higher release probabilities like 0.7, 0.8 of MF synapses, the number of spikes became saturated and the number of spiking cells remained the same (as seen in 0.6 release probability of MF synapses) (Fig. 3F, G).

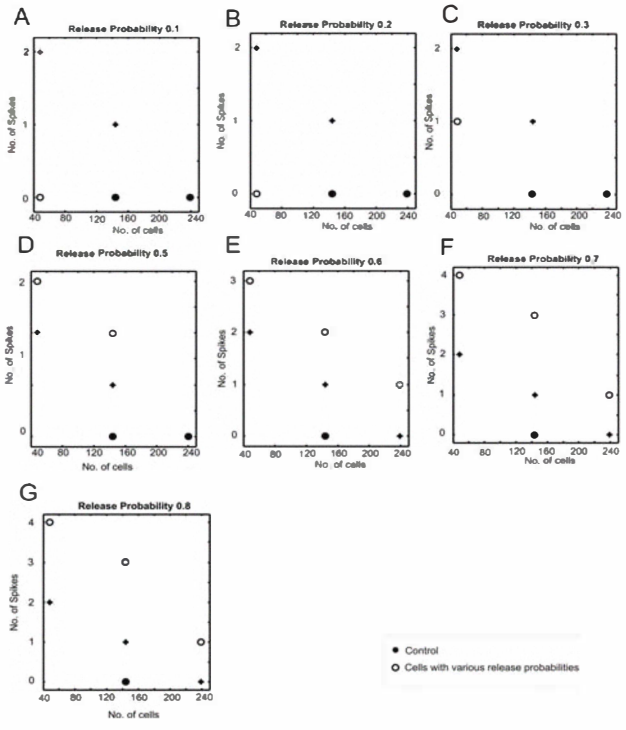

Figure 3. Variation of spiking cells with change in release probabilities. Plot shows number of spikes vs number of spiking cells at various release probabilities for combinations of excitatory inputs. A-G show number of spikes at different release probabilities from 0.1-0.8. Control is shown as a star. For example (say G), 48 cells in the network receive 4 excitatory inputs, in control (star) they generate 2 spikes, with U=0.8 those cells generate 4 spikes (circle). Note dark circles are over-plots of two conditions.

The modulation of intrinsic excitability from normal to higher excitability showed a significant increase in the spike amplitude (~6%) for all spiking cells and an increased number of spikes was observed only for the cells receiving 2 excitatory inputs with 0.5 release probability of MF synapses (Fig. 4A). Thus the number of spiking cells varied from 192 to 432 in the network of 1680 granule cells. The cells with high intrinsic excitability receiving 4 and 3 excitatory inputs showed increased spike amplitude, whereas number of spikes and first spike delay remains the same (Fig. 4 B-D).

With U = 0.3, release probability of MF synapses, the granule cells with low intrinsic excitability receiving 4 excitatory inputs produced a single spike with a significant decrease in spike amplitude and consistent first spike timing (Fig. 5C), whereas the cells with lesser excitatory inputs did not produced spikes (Fig. 5A, B). For the lesser release probability of MF synapses like U = 0.2 and 0.1, all the cells irrespective of their excitatory inputs produced the EPSPs alone. Thus the number of spiking cells with 0.3 release probabilities was 48 which decreased to zero, when the release probability of mossy fiber reduced to 0.2 and 0.1.

V. Inhibition and its effect on Granular layer population

On an average, each granule cell receives excitatory connections from 4-5 mossy fibers [22]. The strength of the inhibition depends on the number of inhibitory connections and synaptic release probability.

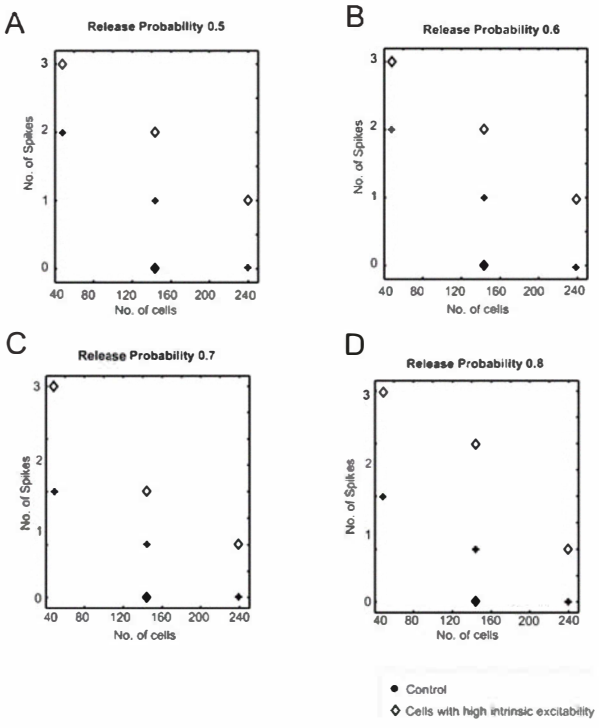

Figure 4. Variation of spiking cells with LTP. Variation in number of spikes with number of spiking granule cells in the network with high intrinsic excitability (diamond) against control condition (U=0.42, star) A show spiking cells at U=0.5, B shows U=0.6, C shows U=0.7 and D shows U=0.8. For example, 48 cells in the network receive 4 excitatory inputs, in control (star) they generate 2 spikes, with U=0.8 those cells generate 4 spikes (diamond). Note dark diamonds are over-plots of two conditions.

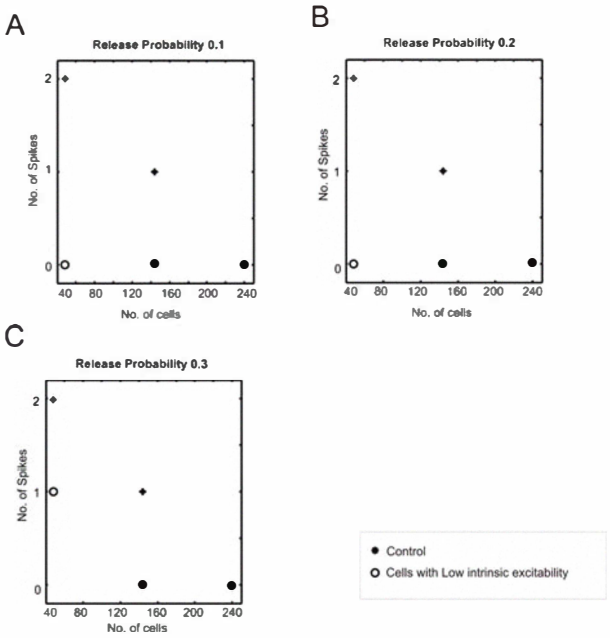

Figure 5. Variation of spiking cells with LTD. Number of spikes against number of spiking granule cells with low intrinsic excitability are compared to control condition (U=0.42, star) at different release probabilities of MF synapses (circle). A-C show number of spikes at different release probabilities from 0.1-0.3. For example, 48 cells in the network receive 4 excitatory inputs, in control (star) they generate 2 spikes, with U=0.3 those cells generate 1 spike (circle). Note dark circles are over-plots of two conditions.

The granule and Golgi cell will receive excitatory inputs from mossy fiber (MF) at the same time. The inhibitory input from Golgi cell reaches the granule cell with a delay of ~4ms compared to the Mossy fiber input through GABAergic synapses [2]. The inhibition-based time-windowing in granule cells allow one or more spikes and is seemingly regulated by varying inhibitory inputs.

A. Impact of inhibition on Granular layer network

Golgi cells converging through lateral connections onto some granule cell subsets could generate combined inhibition [2] [3].The impact of the inhibition on granular layer circuitry differs with respect to two different properties; amount of inhibitory connections and the GABAergic release probability. The effects of inhibition on the circuitry was tested on the *in vitro* model where one spike/burst in MF was used as stimulus and on the *in vivo* model where 5 spikes/burst at 500Hz was used and a longer *in vivo* burst model where 9 spikes/burst at 500Hz input were used as stimuli.

The increase in inhibitory connections to granule cells in the underlying network model decreased number of spikes (see spike count in Fig. 6A, 8A, 10A), spike amplitude (if the spike rises after the inhibitory signal reaches the Granule cell) and decreased spike latency. In the GABAergic release probability (U_{inh}) modulation, the decrease in U_{inh} from its control value (0.34) will result in increase in number of spikes. Increase in the value of GABAergic release probability U_{inh} contributes to a reduction of the doublet to a single spike as seen *in vitro*. Amplitude decreased gradually (for *in vitro* case) (see Table III) and no significant change with the first spike latency for varying GABAergic release probability U_{inh}.

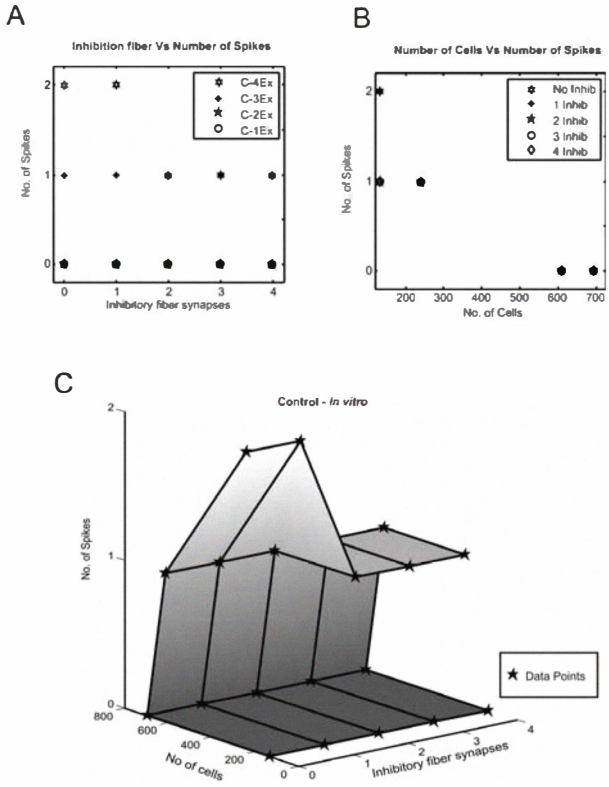

Figure 6. Variation in number of spikes and spiking cells with respect to increasing active inhibitory synapses (0-4). Inhibitory synaptic release probability was set at control value (U_{inh}=0.34). A shows the variation in number of spikes with respect to the number of active GABAergic synapses. B shows the variation in number of spikes with respect the number of cells with different activation pattern. C is a 3D-plot shows the variation of number of spikes in granule cells in network with respect to the increase in the number of active GABAergic synapses in *in vitro* like behavior.

B. In vitro GrC network

When we changed the value of inhibitory (GABAergic) synapse release probability U_{inh}, the change in the number of spikes was seen varying between 2 spikes for GABAergic release probability U_{inh} =0.1 and 1 spike when GABAergic release probability U_{inh} =0.8 (more inhibition) where all cells have 4 excitatory inputs. (Table IV) See Table IV for various combinations.

TABLE III. VARIATION OF U_{INH} AND SPIKE PROPERTIES.

Release probability	Spike Amplitude	First spike latency
0.1	12.6198	24.625
0.2	10.6924	24.625
0.3	8.57065	24.65
0.4	7.99669	24.65
0.5	7.31193	24.65
0.6	6.75183	24.65
0.7	6.28039	24.65
0.8	6.0447	24.65

a. Increase in release probability of GABAergic synapses show a significant decrease in the spike amplitude where the first spike latency remained the same. The data in the table are explained with respect to cells with I1E3 (1 active inhibitory synapse and 3 excitatory inputs) activation pattern, for *in vitro* like behaviour.

For neurons with 3 excitatory synapses, higher inhibitory release probability will have only a single spike, where the spike amplitude decreased by increasing the GABAergic release probability (0.1 – 0.8) (see Table III). A significant decrease in spike amplitude was observed with release probabilities 0.1 – Control (0.34), the decrease in spike amplitude become less significant (decrease in ~0.3 mV) (Fig. 7 A, B) with the higher release probabilities of GABAergic synapses U_{inh} >0.5 (see Table III). The changes in control condition and values of U_{inh} (Fig. 6C) with 0.4 and 0.5 were the same (Fig. 7 C, D). When the inhibitory release probability U_{inh} > 0.5, then the cells with 4 excitatory inputs with 1-4 inhibitory synapses allow single spike while other spike is suppressed (Fig. 7 E, F, G). First spike latency did not show significant changes.

The impact of synaptic excitatory inputs to generate a spike was 3 and any lesser number of inputs favoured generation of EPSP [7]. The number of spiking and Non-spiking cells remained unchanged at 852 and 828 respectively. The number of spiking cells (with excitatory release probability constant at control value) can vary between cells with single and double spikes on modulating the GABAergic release probability values (Fig. 6, 7).

The increase in inhibition decreased the spike amplitude and increase in first spike latency.

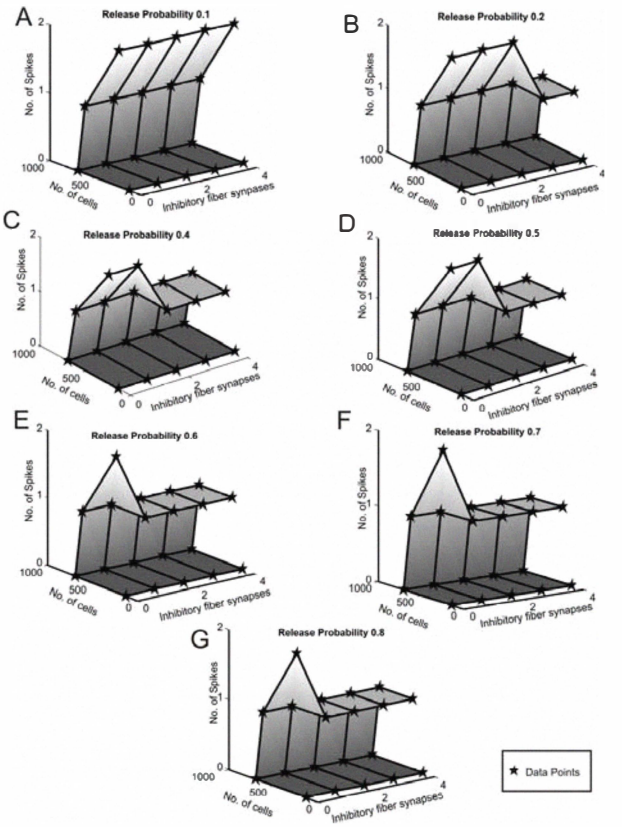

Figure 7. *In vitro* like behaviour, firing pattern seen in granule cell network by varying the GABAergic release probabilities. A-G shows the variation in number of spikes in the network granule cells (with different excitatory inputs) with respect to the increase in active inhibitory synapses at different GABAergic release probabilities like 0.1, 0.2, 0.4 – 0.8 respectively.

C. *In vivo network and spatial inhibition*

In the *in vivo* case, an increase in inhibition level decreased number of spikes (Fig. 8-11) under control condition (inhibitory release probability=0.34) (Fig. 8C, 10C). Variation in GABAergic release probability modulated the number of spikes, where the spike amplitude and first spike latency remained the same (see Table V). As inhibitory release probabilities increased, the number of spikes decreased for the same amount of excitation (see Table V and Fig. 8, 10).

When the GABAergic release probability and number of GABAergic synapses were increased, a decrease in number of spikes was observed.

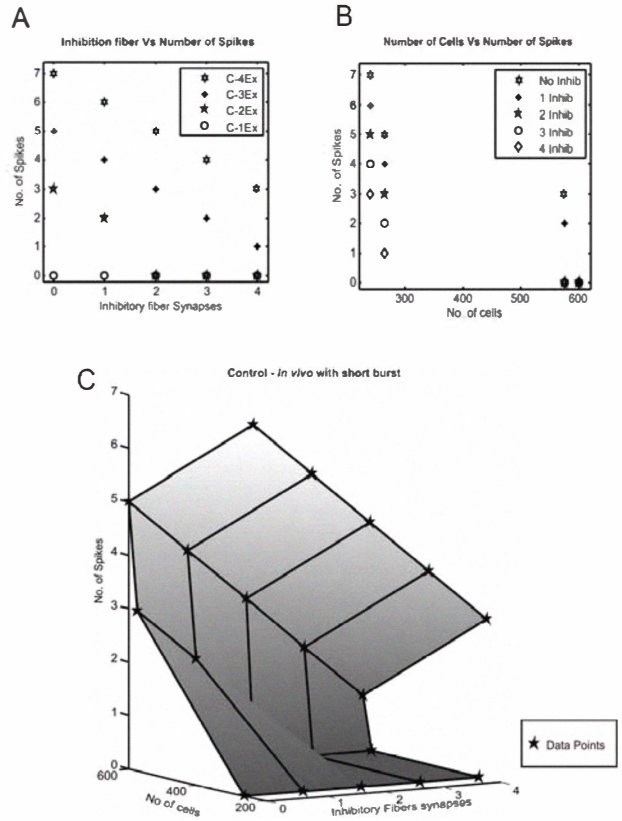

Figure 8. Variation in number of spikes and spiking cells with respect to increasing active inhibitory synapses (0-4) with release probability of GABAergic synapses as 0.34. A shows the variation in number of spikes with respect to the number of active GABAergic synapses. B shows the variation in number of spikes with respect the number of cells with different activation pattern. C is a 3D-plot showing the variation of number of spikes in granule cells in network with respect to the increase in the number of active GABAergic synapses *in vivo* (input given was 5 spikes/burst).

TABLE IV. EFFECT OF INHIBITION WITH INCREASING INHIBITORY RELEASE PROBABILITY OF CELLS WITH 4 EXCITATORY INPUTS.

GABAergic release probability	# Inhibitory synapses	# spikes observed in the cells with 4 excitatory inputs
0.1	-	All cells with doublet
0.2	3 and higher	1
Control, 0.4, 0.5	2 and higher	1
0.6 – 0.8	1 and higher	1

a. With the release probability of GABAergic synapses 0.2, the cells receiving 4 excitatory inputs produced doublet with 0, 1, 2 inhibitory synapse. The number of spikes reduced to a single spike when the number of active inhibitory synapses increase above 2.

The change in decrease of spikes is gradual with the change in the number of inhibitory synapses for lower inhibitory release probabilities (Fig. 9A). It was observed that there were minimum 2 spikes for those cells with 2 excitatory synapses and varying number of inhibitory synapses (inhibitory synapses 0-4). As inhibitory release probability was increased, the decrease in number of spikes with increasing number of inhibitory synapses became distinct (Fig. 9 A-G). Those cells with 2 excitatory synapses and with 4 inhibitory synapses did not produce spikes (Fig. 9B). Cells with 1 or 2 excitatory synapses and high inhibitory release probabilities did not produce spikes in the network (Fig. 6C-G). The number of spiking cells remained unchanged when GABAergic release probability $U_{inh} > 0.2$, with a variation in number of spikes.

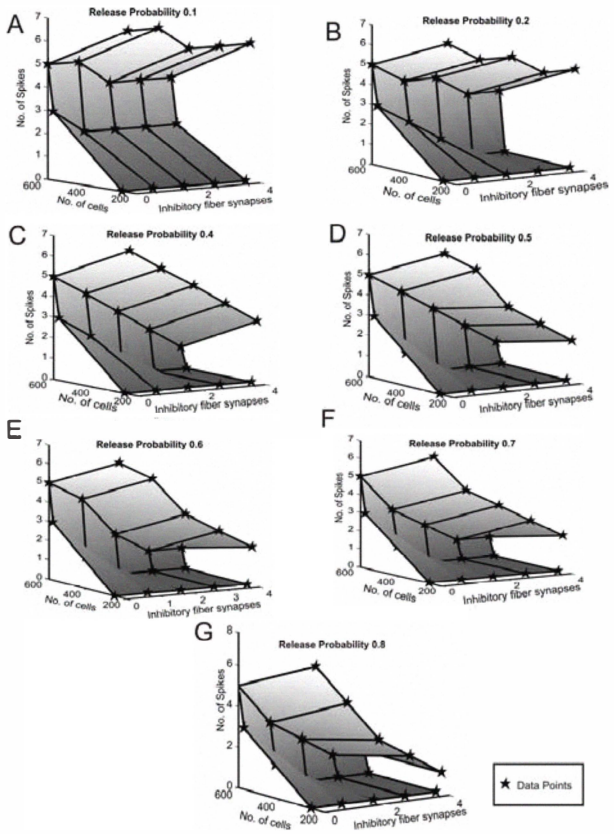

Figure 9. *In vivo* like behaviour (input given was 5 spikes/burst), firing pattern seen in granule cell network by varying the GABAergic release probabilities. A-G shows the variation in number of spikes in the network granule cells (with different excitatory inputs) with respect to the increase in active inhibitory synapses at different GABAergic release probabilities like 0.1, 0.2, 0.4 – 0.8 respectively.

TABLE V. VARIATION OF U_{INH}

Release probability	Spike amplitude	First spike latency	Number of spikes
0.1	14.83	24	5
Control (0.34)	14.83	24	4
0.8	14.83	24	3

a. Increase in release probability of GABAergic synapses did not show a significant change in the spike amplitude and first spike latency. The data in the table are explained with respect to cells with 1I1E3 (receiving 3 excitatory and 1 inhibitory inputs) activation pattern, for *in vivo* like behaviour.

To simulate conditions *in vivo*, we tried with two different burst lengths in mossy fiber synapses corresponding to T wave and C wave of the granular layer LFP [23]. The short burst had 5 spikes at 500 Hz through the MF synapses and the long burst corresponded to 9 spikes at 500 Hz through the MF synapses. The major observations for short and long burst inputs were: 1) Change in the firing frequency of spiking cells and 2) Level of inhibition (number of inhibitory synapses per cell) under which the cells with 2 excitatory inputs did not generate spikes.

When the release probability of GABAergic synapses $U_{inh} < 0.3$, the cells with 2 excitatory synapses produced spikes. But for inhibitory release probability U>0.2 and with 2 inhibitory synapses per cell, the cells did not produce spikes (Fig. 10, 11).

Among 1680 granule cells, 240 cells produced burst, where the burst length varied from 8 (with 1 inhibition and lesser GABAergic release probabilities) to 2 (with higher GABAergic release probability and 4 inhibitory connections), 600 cells produced 6-4 spikes as an output burst, with the lesser release probability of GABAergic synapses (0.1, 0.2) and burst reduced to single spike with the higher GABAergic release probability (0.5 - 0.6). With less active inhibitory synapses, less release probability and 2 excitatory synapses, 576 cells in our network produced spikes which were not seen as number of inhibitory synapses increased in higher release probabilities.

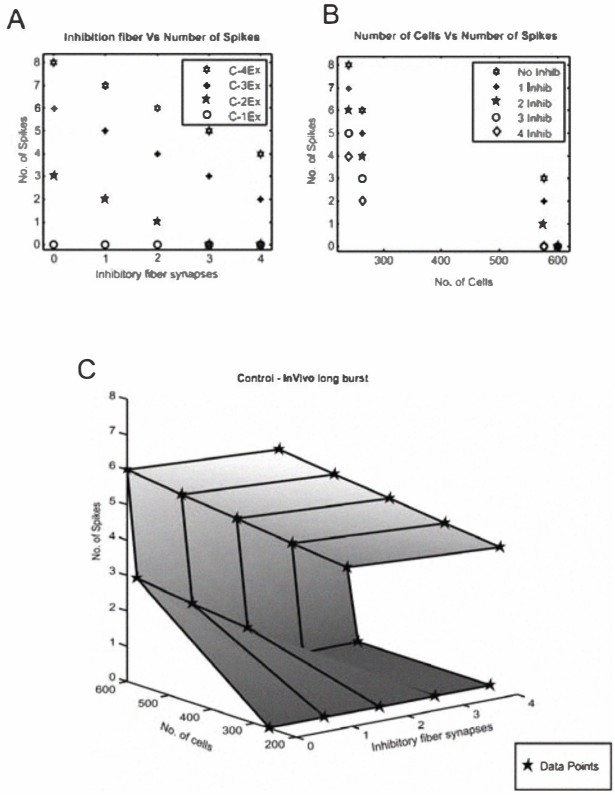

Figure 10. Variation in number of spikes and spiking cells with respect to increasing active inhibitory synapses (0-4) with release probability of GABAergic synapses as 0.34. A shows the variation in number of spikes with respect to the number of active GABAergic synapses. B shows the variation in number of spikes with respect the number of cells with different activation pattern. C is a 3D-plot shows the variation of number of spikes in granule cells in network with respect to the increase in the number of active GABAergic synapses in *in vivo* like behaviour (input given was 9 spikes/burst).

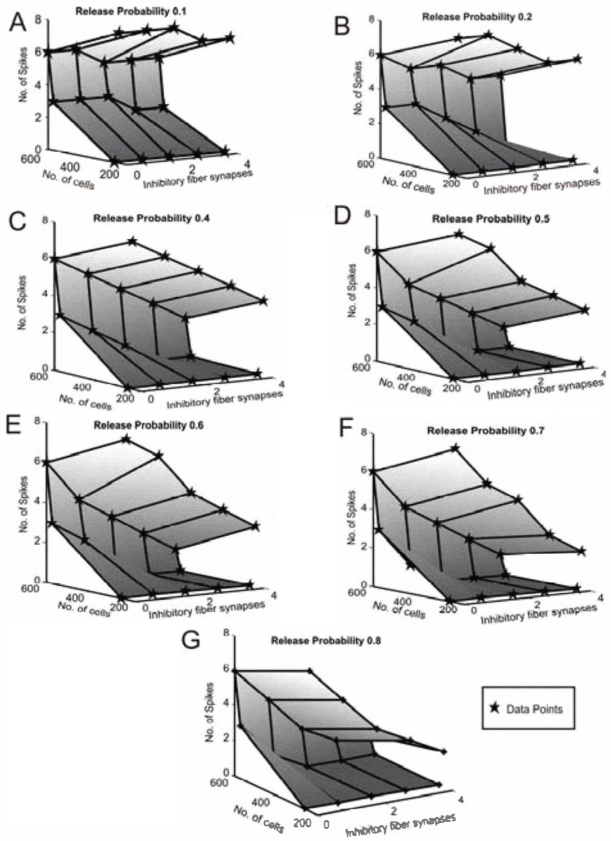

Figure 11. *In vivo* like behaviour (input given was 9 spikes/burst), firing pattern seen in granule cell network by varying the GABAergic release probabilities. A-G shows the variation in number of spikes in the network granule cells (with different excitatory inputs) with respect to the increase in active inhibitory synapses at different GABAergic release probabilities like 0.1, 0.2, 0.4 – 0.8 respectively.

The number of spiking cells varied based on the number of inhibitory synapses for the cells with 2 excitatory inputs (see Table VI at the bottom).

The increase in inhibition decreased number of spikes in the network. The inhibitory inputs modulated the spike count, spike amplitude and first spike latency although changes were distinct only with increased inhibition.

VI. CENTER-SURROUND EXCITATION IN POPULATIONS OF GRANULE CELLS

To understand combinatorial properties of the granular network layer and impacts of double mossy fiber bundle stimulation was simulated.

The '*spots*' are maps of excitatory activity as seen in the cerebellar granular layer [3] when MF rosettes were stimulated. In the model configuration, the center of the spot receives stronger excitatory inputs and the consecutive peripheral neurons receive weaker excitatory input thereby expressing a center-surround configuration (see Fig. 12 A).

In this model network, about 144 cells receive 4 excitatory inputs, 432 cells receive 3 excitatory inputs, 144 cells receive 2 excitatory inputs and 432 cells receive 1 excitatory input. The impacts on a spot by varying release probabilities and intrinsic excitability, thereby modeling LTP and LTD in both *in vitro* and *in vivo* cases were studied. The cells in the granular layer network receive GABAergic synaptic inputs with respect to the number of

excitatory inputs given to the cells in the granular layer network (see Table VII).

When all the inhibitory synapses of granule cells in the network (with *in vivo* like behaviour) were switched off, the cells receiving 4 excitatory inputs produced burst of 7 spikes, cells receiving 3 excitatory inputs produced burst of 5 spikes, cells with 2 excitatory inputs produced a short burst of 3 spikes, cells with 1 excitatory input produced EPSP (see Fig. 12B, Table VIII).

TABLE VII. ACTIVATION PATTERN GIVEN TO CELLS IN GRANULAR LAYER NETWORK.

# cells	# excitatory inputs	# inhibitory inputs
144	4	1
432	3	2
144	2	3
432	1	4

a. Number of cells contributing to the spot corresponding to activation patterns shown as combination of excitatory and inhibitory synapses

TABLE VIII. EFFECT OF INHIBITION OVER THE NUMBER OF SPIKES IN SPIKING CELLS OF NETWORK

No. of cells	# active MF synapses	# spikes	
		Network without inhibition	Network with inhibition
144	4	7 spikes/burst	6 spikes/burst
432	3	5 spikes/burst	3 spikes/burst
144	2	2 spikes/burst	EPSP
432	1	EPSP	EPSP

a. Cells with 4 excitatory inputs produced 7 spikes/burst when inhibitory synapse was switched off and produced 5 spikes/burst when it was switched on.

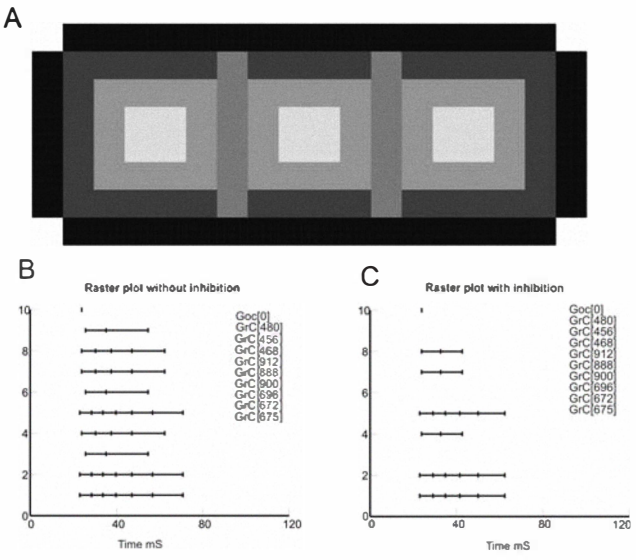

Figure 12. Center-surround "*spot*" activation. A. Varying intensities of grey color in the spot corresponds to varying levels of excitation as mossy fiber inputs to granule cells in the network. The white color in the middle of the spot form I layer, with the cells receiving 4 excitatory inputs. The light grey layer represents II layer cells which receive 3 excitatory inputs. The dark grey colored layer form the III layer cells receiving 1 excitatory input and the cells of III layer present in the overlapping region of the two spots received 2 excitatory inputs (represented in grey color). The black layer represents the outermost layer of the spot form the IV layer (no excitatory inputs given). Each spot contains 648 granule cells in which 96 granule cells (III, IV layer) are present in the overlapping region of the two spots. The spots are activated by double mossy fiber bundle stimulation [3]. B. Raster plot of the network with no active GABAergic synapses. C. Raster plot of the network with active GABAergic synapses.

The effects of inhibition on the circuitry were tested on *in vivo* model where 5 spikes/burst at 500Hz in MF was used as stimuli. Inhibitory synapses were activated in the network, where the number of active inhibitory synapses to a cell varied according to its excitatory inputs. Through the GABAergic inhibitory synapses, 1 spike was provided along with the excitatory stimuli. Different combinations of activation pattern to the cells in the network are listed in Table VII.

The number of spiking cells decreased from 720 to 576 after activating the GABAergic synapses. The cells with 2 excitatory inputs lost its spikes with 3 active inhibitory synapses (see Fig. 8C, 12C). In the cells with 4 and 3 excitatory synapses, decreased number of spikes was observed (see Table VIII).

VII. DISCUSSION

The study shows the modulatory impacts of varying inhibitory and excitatory release probabilities on the activities of granule cells in the granular layer network of the cerebellum. The paper also explores the effects of combined excitation and combined inhibition [3]. Both *in vitro* and *in vivo* simulations indicate inhibitory input cannot completely alter the excitation rather it acts as a modulator that regulates the post synaptic excitability.

The variations of excitatory inputs (without combination of inhibition) showed differences in number of spikes and spike amplitude and did not show variations in first-spike latency. The most promising outcome in variation of spikes and spike behaviour was with the induction of LTP/LTD where both intrinsic excitability and excitatory release probabilities change the nature of information flow.

Artificial neuron models can be tuned to function as biophysical models. However there are some limitations. Synaptic functions in spiking models are not very reliable. And artificial models have limitations unlike biophysical models for understanding certain population activities like generation of LFP etc.

This study on granule neuron excitation and inhibition is one of the first detailed simulation works where a model has been used to explore the parameter space and test plasticity. The presynaptic mechanism coexisted with postsynaptic regulation of ionic channels, which played a major role in determining the granule cell output firing frequency. Intrinsic bursting and modulatory effects of inhibition can be seen by mechanistic control of number of spikes in a granule cell.

Impact of excitation on single neurons affected network activity. With increased excitation, along with an increase in spikes, first-spike latency also decreased. During LTP, there was no significant change in the first spike latency (*in vitro*). With higher intrinsic excitability and with same release probability of excitatory synapses, spike amplitude was increased. This will also impact the local field potential and could probably explain the observations *in vitro* [24]. *C*ombining higher intrinsic excitability and with higher release probability, there was gradual increase in number of spikes. In both *in vitro* and *in vivo* simulations, the number of spikes was dependent on the release probability of the synapses while higher or lower intrinsic excitability caused slight change in spike amplitude.

The key role of local circuit inhibition for determining granular layer combinatorial operations was supported by several observations. Increasing active inhibitory connections saw lesser number of spikes in the network. *In vivo* bursts along mossy fibers combined with inhibitory input showed a consistent reduction of 1 spike as inhibition increased.

The studies on intensity of mossy fiber synapses and inhibitory synapses help to understand spatio-temporal operations [3]. Combining granule neurons, Golgi cell and Purkinje cell, this study will help to reveal coincidence detection properties and spatial pattern separation [25]. This work is a preliminary start in modeling to understanding long-sought spatiotemporal filtering predicted by the Motor learning theory [26].

ACKNOWLEDGMENT

This project derives direction and ideas from the Chancellor of Amrita University, Sri Mata Amritanandamayi Devi. This work is supported partially by the Sakshat project of National Mission on Education through ICT, Department of Higher Education, Ministry of Human Resource Department, Government of India.

REFERENCES

[1] Sergio Solinas, Thierry Nieus and Egidio D'Angelo, A realistic large-scale model of the cerebellum granular layer predicts circuit spatio-temporal filtering properties, Frontiers in Cellular Neuroscience volume 4, article 12, 2010.

[2] E. D' Angelo, Cl. De Zeeuw, Timing and plasticity in the cerebellum: focus on the granular layer. Trends in Neurosciences, Volume 32, Issue 1, 2009, 30-40.

[3] Jonathan Mapelli, Daniela Gandolfi, and Egidio D' Angelo, Combinatorial Responses Controlled by Synaptoc Inhibition in the cerebellum Granular layer, J Neurophysiol 103, 2010, 250-261.

[4] D' Angelo E., T. Nieus, A. Maffei, S. Armana, P. Rossi, V. Taglietti, A. Fontana, G. Naldi, Theta-frequency bursting and resonance in cerebellar granule cells: experimental evidence and modeling of a slow K+-dependent mechanism, J. Neurosci. 21, 2001, 759-770.

[5] M. Zayd Khaliq, W. Nathan. Gouwens, and Indira M. Raman, The Contribution of Resurgent Sodium Current to High-Frequency Firing in Purkinje Neurons: An Experimental and Modeling Study, The Journal of Neuroscience, 23(12), 2003, 4899-4912.

[6] M. Eugene, Izhikevich, Simple Model of Spiking Neurons, IEEE Transactions of Neural Networks, Vol 14, No. 6, 2003, 1569 - 1572.

[7] Shyam Diwakar, Jacopo Magistretti, Michell Goldfarb, Giovanni Naldi, Egidio D' Angelo, Axonal Na+ channels ensure fast spike activation and back-propagation in cerebellar granule cells, J Neurophysion., 101, 2009, 519-32.

[8] Sergio M. Solinas, Lia Forti, Elisabetta Cesana, Jonathan Mapelli, Erik De Schutter and Egidio D' Angelo, Fast-reset of pacemaking and theta-frequency resonance patterns in cerebellar Golgi cells: simulations of their impact in vivo, Front Cell. Neurosci., 2007, 1:4. doi:10.3389/neuro.03.004.2007

[9] Sergio M. Solinas, Lia Forti, Elisabetta Cesana, Jonathan Mapelli, Erik De Schutter and Egidio D' Angelo, Computational reconstruction of pacemaking and intrinsic electroresponsiveness in cerebellar Golgi cells, Front. Cell. Neurosci., 2007, 1:2. doi:10.3389/neuro.03.002.2007.

[10] Richard R. Carrillo, Eduardo Ros, Silvia Tolu, Thierry Nieus, Egidio D' Angelo, Event-driven simulation of cerebellar granule cells, Biosystems, 94 (1-2), 2008, 10-7.

[11] ML Hines, NT Carnevale, The NEURON simulation environement, Neural Comput 9: 1997, 1179-1209.

[12] Thierry Nieus, Elisabetta Sola, Jonathan Mapelli, Ellene Saftenku, Paola Rossi, and Egidio D' Angelo LTP Regulates Burst Initiation and Frequency at Mossy Fiber-Granule Cell Synapses of Rat Cerebellum: Experimental Observations and Theoretical Predictions, J Neurophysiol 95: 2006, 686-699.

[13] Shyam Diwakar et al., Unpublished.

[14] JP Welsh, R Llinas, Some organizing principles for the control of movement based on olivocerebellar physiologu, Prog Brain Res 114, 1997, 449-461.

[15] Jenny T. Davie, Beverley A. Clark and Michael Hausser, The origin of the Complex spike in cerebellar Purkinje cells, The Journal of Neuroscience, 28(30), 2008, 7599-7609.

[16] PF Gillbert, WT Thach, Purkine cell activity during motor learning, Brain Res 128, 1977, 309-328.

[17] C. Hansel, DJ. Linden, E. D' Angelo, Beyond parallel fiber LTD: the diversity of synaptic and non-synaptoc plasticity in the cerebellum, Nat Neurosci 4, 2001, 467-475.

[18] M Ito, Cerebellar long-term depression: Characterization, signal transduction, and functional roles, Physiol Rev 81, 2001, 1143-1195.

[19] S. L. Shin, F. E.Hoebeek, M. Schonewille., C. I. De Zeeuw, A. Aertsen, and E. De Schutter, Regular patterns in cerebellar Purkinje cell simple spike trains, 2007, PLoS ONE 2, e485.

[20] Sungho Hong and Erik De Schutter, Purkinje Neurons: What is the signal for complex spikes?, Current Biology Vol 18 No 20, 2008, DOI: 10.1016/j.cub.2008.08.056.

[21] A. Rasmussen, D. A. Jirenhed, and G. Hesslow, Simple and complex spike firing patterns in purkinje cells during classical conditioning, Cerebellum 7, 2008, 563-566.

[22] Anna D' Errico, Francesca Prestori and Egidio D' Angelo Differential induction of bidirectional long-term changes in neurotransmitter release by frequency-coded patterns at the cerebellar input, J Physiology, 2009, 1-15.

[23] Leda Roggeri, Bruno Rivieccio, Paola Rossi and Egidio D' Angelo Tactile stimulation evokes long-term synaptic plasticity in the granular layer of cerebellum, The Journal of Neuroscience, 28(25), 2008, 6354-6359.

[24] Jonathan Mapelli, Egidio D' Angelo, The Spatial Organization of Long-term synaptic Plasticity at the Input Stage of cerebellum, The Journal of Neuroscience, 27, 2007, 1285-1296.

[25] JS. Albus, A theory of cerebellar function, Math Biosci 10, 1971, 25-61 .

[26] M. Fujita, Adaptive filter model if the cerebellum, Biol. Cybern. 45, 1982, 195-206.

TABLE VI. MODULATION OF SIPIKING WITH VARYING INHIBITION.

Release probability	MF input – 5 spikes/burst		# inhibitory synapses at which cells with 2 active MF synapses lose it spikes (576 cells)	MF input – 9 spikes/burst		# inhibitory synapses at which cells with 2 active MF synapses lose it spikes (576 cells)
	# spiking cells	# Non Spiking cells		# spiking cells	# Non spiking cells	
0.1	1416	264	All cells with 2 MF are spiking	1416	264	All cells with 2 MF are spiking
0.2	1416-840	840-264	3 and higher	1416-840	840-264	4
Control, 0.4	1416-840	840-264	2 and higher	1416-840	840-264	3 and higher
0.5 - 0.8	1416-840	840-264	2 and higher	1416-840	840-264	2 and higher