E-Neuron Project
Constructing a Nerve Growth Cone model using E-CELL

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Abstract

Details of signal transduction information in nerve growth cone still remain unclear. Elucidating functions of nerve growth cone is necessary for pathological research. Though reliable computer simulations are essential for furthering neural research, there have not been any outstanding computer simulations. We developed a nerve growth cone model using E-CELL system, a generic simulator for biological systems. All of the data for this model is based on literature. From the simulation results, we reconfirm that the optimum concentration of Ca^{2+} in a cell is a key factor for tubulin polymerization. Unknown pathways were also estimated using CALI System on E-CELL.

Summary

Recently, researches of signal transductions in nerve growth cone have been one of the most attentive fields. Although there are some discussions and hypotheses, the whole signal transductions in nerve growth cone still remains unclear. Determination of the molecular basis and signal transduction network in nerve growth cone is an essential element to solve pathological problems. Revealing unknown signaling pathways only from the experimental system takes too much time. We suppose that experiments in silico will commit to the in vitro and in vivo.

First, we made a model of a signal transduction pathway in nerve growth cone considering any conventional hypotheses. That is to say, in this research, it has not aimed at carrying out a simulation of the biochemical signal transductions more correctly, but taking signal transductions as one system.

This signal transduction model is developed in order to contribute novel knowledge to biology. This study is important as a contribution to nerve network analysis which being under the necessity in pathology.

An advantage for computer simulation of nerve growth cone is as follows. Nerve growth cone is far from its cell body, and time-scale of signal transduction and gene expression occur in different order. Therefore, an influence of gene expression on nerve growth cone is supposed to be little, and in nerve growth cone, cell signal transduction is occurring mainly. Now, we can make a model of nerve growth cone without gene expressions.

We have developed a virtual pathway using literatures. The simulation of axon growth inspired by polymerization and depolymerization of tubulin is based on determinate data from many literatures. We used E-CELL system which is the generic cell-simulator currently developed by us.

The beginning of our pathway is the binding of basic fibroblast growth factor (bFGF) and fibroblast growth factor receptor (FGFR). bFGF stimulator phosphorylate phospholipase C (PLC) which hydrolyze phosphatidylinositol P2 (PIP2) into diacylglycerol (DAG) and inositol phospholipid 3 (IP3). Then they compose complex signaling network, including arachidonic acid (AA) degradated from DAG activate the Ca^{2+} channel, and finally tau protein and microtubule-associated protein 2 (MAP2) polymerize tubulin. The way of expression of the axon growth is an important problem for computer simulation. In this model, the axon growth is expressed with the microtubles' length extended by the polymerization of tubulin. As is mentioned before, it is demonstrated that Ca^{2+} is the key factor for the axon growth. We have confirmed our cascade works realistically reacting to continuous dosage of 0.8mM Ca^{2+}, which is followed by a result of tubulin polymerization by MAP2 and tau protein dephosphorylation. Also, MAP2 had the larger influence on tubulin polymerization than tau protein. Among many molecules which is influencing the MAP2 like microtubule-associated protein kinase (MAPK), calcineurin (CaN), calmodulin kinase II (CaMKII), cyclic guanosine monophosphate (cGMP), protein kinase G (PKG), CaN was the most effective dephosphatase for MAP2. And CaN cannot be activated continuously without activation by calmodulin (CaM). In NO synthesis, which start from activated CaM, the concentration of NO in NO synthesis pathway is low. GAP43 is associated with MAPK cascade which is a following cascade from upstream cascade including bFGF, PLCgamma, IP3, DAG, Ca^{2+}, AA, adenomatous

polyposis coli (APC). GAP43 was activated smoothly by PKC. In stead of a continuous Ca^{2+} stimulus of 0.8mM, we have examined tetanus stimuli of Ca^{2+}. By giving an abrupt stimulus like a Ca^{2+} transient, we observed remarkable dephosphorylation of tau protein and MAP2. We also examined the abrupt decrease of Ca^{2+}. And that leads to the phosphorylation of tau protein and MAP2, and then the depolymerization of tubulin. As well as Ca^{2+}, CaM is a key factor effecting CaN and CaMKII. From these results, we have confirmed the existence of optimum concentration of Ca^{2+}, and also Ca^{2+} stimulation influences each other in many ways through signal transduction pathway. In order to investigate the pathway more closely, we grouped all enzymes into three groups like fast, middle and slow enzymes according to the Km and Vmax. And almost the same enzyme activity was observed in our model.

Following to these experiments, we will undertake retraction of nerve growth cone. The signal transduction of the retraction in nerve growth cone being unclear, our prediction of unknown pathways can contribute to such a challenging field. Through this study, we emphasize that our attempt is the first kinetic simulation of nerve growth cone. And also, we can confirm these predicted pathway by Chromophone-assisted laser inactivation (CALI) on E-CELL. CALI system is a useful experimental methods which can inactivate the specific target molecules in the cell by irradiating laser beam at 620nm through dye-labeling. We will examine our predicted pathway with this CALI on E-CELL systems.