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Effect of K+Sequence with Different pH Combination on Cytoplasmic Streaming in PermeabilizedNitella Cell

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Abstract: ***The Cytoplasm keeps moving continuously as an unending process like rotating belt along a more or less in helical path. The rate and flow pattern of the Streaming are always constant and directional. The effect on Cytoplasmic Streaming of intact Nitella cell and Permeabilized cell were studied with different K+ sequence at different combination of pH because of the dependence of membrane potential on external K+ concentration and the phenomenon of oscillation in Cytoplasmic Streaming by change in Extracellular K+ and pH (Singh and Amin, 1989).***

Keywords: Cytoplasmic Streaming, K+, pH, Permeabilized, Nitella

1. Introduction

Some Organisms have no problem in randomizing their internal molecules since diffusion is a fast process over short distances. However, large cells need to mix their Components and they have developed internal machinery to get molecules and ions transferred from one place to another. This machinery is also used in big cells and in multicellular Organisms to position and move organenelles, change cell Shape and move themselves. A common example of this is the rapid Cytoplasmic Streaming seen prominently in some algal and higher plant Cells and also in many Protozoans in which it is called as Cyclosis.

Movement of all cells is achieved by mechno-chemical process in which chemical energy is converted into mechanical energy to move the cell or cell parts. In Prokaryotes, there are two types of movement;

i) gliding movement in some species of Bacteria and
ii) Flagellar movement

In Eukaryotes, three such kind of movements are known in which interaction of two Proteins produces force by Hydrolysis of ATP. These are as follows;

a) Interaction of tubulin and dynein, the basis of all ciliary and Flagellar movement.
b) Interaction of tubulin and kinetin, the basis for the transport of vescicles in the nerve cells.
c) Interaction of actin and myosin, the basis for the muscle contraction, the Cyclosis, the Cytoplasmic Streaming, the Cytokinesis and amoeboid movement.

Giant multinucleated algal Cells of Nitella and Charaare specially favourable for the study of Cytoplasmic Streaming. In Characeaninternodal cells it is a vivid and specialised phenomenon which can be observed even at considerable resolution as one internode (4to12cm in length) constitutes only onecell. The example of giant algal cell such as Nitellainternodal Cell is very interesting in this respect as here only actin exists in filamentous form and the myosin exists in free molecular form and their interaction causes Cytoplasmic Streaming instead of Contractile movement. The interconvertibility of the two that is free energy of ATP Hydrolysis and proton motive force (Mitchell, 1961) is best revealed in the Mitochondrial and thylakoid membranes (Nicholls, 1982). In giant algal Cells, the actin filaments are present in the sub cortical Cytoplasm attached to the inner surface of the Chloroplast running parallel to the length of the cell onto the direction of streaming. Here I will observe the effect of K+ sequence and different combination of pH on Cytoplasmic Streaming in intact cell and Permeabilized cell of internode of Nitella.

2. Materials and Methods

Nitella was procured from the local pond. Culture of Nitella is desirable both outdoor and indoor round the year to full fill the requirement of experimental material. In outdoor culture, Nitella can be grown in a pond having one metre depth and contains fine mud, rotting leaves, some compost of cow dung etc, . For indoor culture, Nitella were planted in big (20” high x 10” diameter) Jars containing 2.5” soil mixed with hummus and manure at the bottom. Jars were initially supplied with the natural pond water of thewater body from where the Nitella were collected since it contains all the biotic and abiotic Components required for normal growth. Internodal cells of diameter 1 mm and 4to 6 cm long were cut out from the plant and kept in Artificial Pond Water (APW) for a day or two before the experiment.

Permeabilization of Plasmalemma of Nitellainternodal Cell

There is absence of cortical layer outside the plasmalemma of Internodal cell in Nitella. Hence it can be done by Simple method as adopted by Shimmen and Tazawa (1983) that is electric Pulse. Due to the presence of cell wall, large molecules can not enter the Cytoplasm in Permeabilized cell but the integrity of the Cytoplasm is maintained since the Cytoplasm is sandwiched in between the tonoplast and the cell wall.

The present study on Cytoplasmic Streaming has been taken up in two Systems that is in intact cell and Permeabilizedinternodal Cell of Nitella. The parameters

selected here are the effects of K+ sequence and different combination of pH on the velocity of Cytoplasmic Streaming.

3. Result and Discussion

Effect on intact Nitella Cell; The effect on Cytoplasmic Streaming of intact Nitella cell were studied with different K ion sequence at different combination of pH because of the dependence of membrane potential on external K+ Concentration.

In my present experiment, streaming velocity was not appreciably affected by change in external K+ in the range of 0.01 to 3 mM KCl. At 10 mM KCl Concentration, the Streaming ceases. The recording of streaming velocity was made in each specimen for the whole sequence and for each sequence about ten specimen were used. In the Internodal cells studied by me, the Streaming velocity on an average was 50 mm to 60 mm per second. In all experiments I observed that in the range of about 2 units of pH around the neutral K+ concentration. The streaming velocity did show the change at both increasing and decreasing ranges of pH that is towards alkaline and acidic sides respectively. The streaming totally stopped when the pH was excessively acidic or alkaline. In different Internodal cells of Nitella the pH range at which the Streaming stopped, towards acidic side it was between 4.0 and 3.0 and on the alkaline side it was between 9.5 and 10.5. This was observed so when the same internode was treated with gradual change of pH of 0.25 units, but the Streaming stopped almost abruptly when the change by 0.5 unit was made both for the acidic and alkaline pH.

Recently, Shimmen (2010) has reported a unique cellular effect of bromoxynil, a herbicide, on Characean cells causing the inhibition of Cytoplasmic Streaming by Cytosol acidification. The decrease in electro chemical potential gradient for H+ across plasma membrane appears to result in disturbing the Intracellular homeostasis.

Effect on Permeabilized Cell

At neutral pH the higher K+ concentration has more pronounced effect in Permeabilized cell. The elevation of K+ concentration from 7mM to 9mM has effects almost irreversible. The effect of pH in different combination of K+ concentration when studied on the plasmalemma-Permeabilized Nitella Cell, it was found that ranges of acidic and alkaline pH have pronounced effect at the lower K+ Concentration that is at 2mM and 3 mM K+ Concentration.

4. Conclusion

It can be concluded that an increase in the external K+ concentration always depolarizes the membrane is in the line with the known high permeability of plasmalemma of living cells including algal Cells for this cation, since the depolarization of the Membrane potential for an increase in external K+ doesn't follow the Potassium Nernestpotential behaviour and thus influence the membrane potential and thereby the Streaming velocity. The influence of external pH on Cytoplasmic Streaming within the range of pH greater than 5.0 and less than 9.0 appears to be linear. In the Physiologically permissible range the fair dependence of Cytoplasmic Streaming on pH speaks for the involvement of electrogenic H+ pump. The situation of both high acidic and alkaline ranges finally lead to an abnormal pathological state of the algal cells leading to death of the cell.

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