كلية العلوم

PHYSICAL MODELLING OF SOME BIOLOGICAL SYSTEMS

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SUMMARY

The main objective of the present work is to simulate the patterns of behaviour of excitable cells and those having non-inactivating sodium channel in their plasma membrane. The cells were assumed to be space clamped and the relevant Hodgkin and Huxley equations, H-H model, were assumed to describe the electrical behaviour of the cells.

The present work is divided into four chapters :

***Chapter I** includes the review of literatures and the aim of work.*

***Chapter II** deals with the theoretical basis of the ionic theories of resting and action potentials and the electrical behaviour of the excitable tissues.*

***Chapter III** includes a detail description of the Voltage-clamp technique together with the Hodgkin-Huxley model.*

***Chapter IV** Concerns the obtained results and discussion. In this chapter Hodgkin-Huxley model has been modified to study the effect of changing :*

1- *The potassium permeability* (g_K).
2- *The sodium permeability* (g_{Na}).
3- *The simultaneous stochastic variations of potassium and sodium conductances* (g_K *and* g_{Na}).
4- *The current stimulus* (J_{stim}).
5- *The noninactivation of sodium channels (NI-NaC), on the electrical behaviour of the simulated cell such as the action potential (A.P) characteristics (amplitude, duration and frequency), sodium-potassium currents.* (J_k-J_{Na}) *and the (I - V) diagram.*

The results of the first part showed that : The amplitude and duration of the generated action potential are increased by the increase in the value of g_K *while the amplitudes of potassium and sodium current* (J_K, J_{Na}) *are decreased. At values of* $21.5 > \bar{g}_K > 3.5$ *the simulated cell generates a repetitive action potential whose frequency increases by the decrease in the value of* g_K.

The effect of increasing the value of g_{Na} (second part) showed an increase in the amplitude and duration of the action potential and also in the values of sodium and potassium currents J_{Na}, J_K. In addition a repetitive firing of action potential is obtained at values of g_{Na} in the range from 189 to 1400 mmho/cm^2. The frequency of action potential repetition is found to increase by the increase in the value of g_{Na}.

In part three the effect of the stochastic variations of g_K and g_{Na} on the frequency of the generated spike has studied. The obtained results showed that the frequency of the generated action potential takes a certain form with time which could indicate a certain kinds of informations. Accordingly one could conclude that the random variations in the frequency of the generated action potential represent an information, carried by the sensory cells, could changed either by changing in the values of g_K or g_{Na} or both in addition to the external stimuli.

The results of part four showed the effect of current strength, J_{stim}, on the electrical behaviour of the simulated cell at different values of g_K. This effect is dependant on the values of g_K except at g_K values in the range 3.5-21.5 mmho/cm^2.

The effect of non-inactivation of sodium channels studied in part five, showed that the characteristics of the generated action potential are dependant on the value of non-inactivation fraction and the values of g_{Na} and g_K.

CHAPTER 1

INTRODUCTION AND REVIEW OF LITERATURE

1.1 Introduction and Review of Literature

A new realm of physiological experimentation on the permeability of electrically excitable membranes was opened by the introduction of a radically new approach in which the membrane potential was set at a given value and made the independent variable in contrast to its usual role as dependent variable in classical electrophysiology. After the.introduction and use of this concept by Cole (1949), the experimental technique was improved by Hodgkin and Katz, (1947, 1949) and dramatically exploited in the study of the squid giant axon by Hodgkin and Huxley (1952a, b, c, d) presented in a new famous series of papers. The rational and procedure in the development of this novel and powerful approach have been reviewed in some detail by Cole (1955), Moore (1959a), Cole and Moore (1960) and Cole (1962). The kelvin cable equation, which seems certain to be both, necessary and sufficient to describe the more usual axon phenomenon, is a partial differential equation in time and distance and must be too complicated to be used as a tool for the analysis of experimental data. The short circuiting of longitudinal potential differences with internal and external electrodes first eliminated the spatial variation of the axon membrane potential-analytically, thus reducing the equation to an ordinary differential equation in time and preventing electrotonic spread and impulse propagation in the usual physiological sense. The next step was to change the membrane potential suddenly from its resting value to another potential in the direction taken by the normal action potential. The purpose was to eliminate current flow in or out of the membrane capacitance after an initial transient and to obtain ionic currents which were continuous functions of potential and time while preventing the " all-or- none " phenomena (Nastuk,1963)

These data gave an experimental basis for the usual excitation and propagation and for the rate of rise, amplitude, and recovery of the action potential. Hodgkin and Huxley 1952b then demonstrated that the membrane currents carried by the sodium and potassium ions could be separated from each other, and from the capacitive charging and discharging currents. They also further came to the very important conclusion that the permeabilities to both the ionic species were voltage rather than current dependent, and hence developed empirical equations to describe the changes of conductivity with voltage and time substituted them into the full equations describing the conditions of an uncontrolled membrane, and found that relatively normal action potentials could be computed. Other investigators have made computations with these equations using electronic digital (Cole, 1955, 1958, Huxley 1959, FitzHugh and Antosiewiez, 1959) and analog (Fitz Hugh, 1960) computers and have found that a variety of classical physiological data could be predicted. Also a computer simulation of the action potential propagation in septated nerve fibers has been studied by Barach et al (1987).

The power and effectiveness of this approach, called a " voltage clamp " by Hodgkin and Huxley, has become widely appreciated and a number of other investigators have applied it with various techniques. Not only has the work with the squid giant axon been extended but also it has been adapted for use on other preparations. Further studies with the voltage clamp in the squid axon have been reported by Tasaki and Hogiwara (1957), Tasaki and Bak (1957, 1958a) Tasaki and Spyropouls (1958a), by Shanes et al., (1959) and by laboratory (Moor, 1958,1959b, Cole and Moor, 1960, Taylor, 1959).

A voltage clamp technique has been developed for the isolated single node of myelinated nerve fibers by Dodge and Frankenhaeuser (1958, 1959), Tasaki and Bak (1958b) ; and del Castillo et al., (1957).

Motoneuron voltage clamp experiments have been reported by Frank et al., (1959). Marine nerve cell bodies have been clamped by Hogiwara and Saito (1959) and Frog muscle end-plates have been clamped Takeuchi and Takeuchi (1959, 1960).

The iron wire model for the nerve was studied under voltage clamp by Bartlett (1945) some years previous to the work on the squid axon.

The successful recording of membrane potential at the node of Ranvier led to the clever use of an extracellular insulating barrier for recording membrane potentials in whole axons or bundles of axons. Staimpfli 1953,first described the method, and it was developed considerably by Julion et al. (1962a,b.). Hodgkin (1964) reported that, for a single propagated impulse in the squid axon, the trans-location of sodium and potassium ions (3-4 PM /cm of membrane surface area) represents more than enough charge to alter the change on the membrane capacitance and to account for the amplitude of the action potential. Also the propagation of the action potential along the complex axonal trees was studied by Manor et al (1991). Hogiwara et al. (1964) showed that spikes of increasing height (rising from a resting membrane potential of 60 mV inside negative) were obtained by increasing the conductance-voltage curve along the voltage axis against changes in external calcium concentration. The cable theory has been applied quite extensively to predict some properties of irritable tissues using measured values for membrane resistance and capacitance and axoplasm resistance. The methods employed are well described by Taylor (1963) and were able to form lipid bilayers of controlled compositions between different electrolyte solutions and "dope " the resulting membrane with various current carriers.

Entry of Ca^{21} Ions during activity has been shown by Baker et al. (1971). Miyazaki et al., (1972) showed that the inward-current mechanism itself changes with egg cells which are precursors of the tunicate muscles have action potentials which are dependent on both sodium and calcium ions. The accumulation of potassium ions outside an active muscle fiber has been measured directly with K - sensitive electrodes by Hnik et al., 1972. Some measurement has been performed with snail neurons by Neher and Lux (1972, 1973). Gilbert and Ehrenstein (1969),

Mclaughin et al. (1971), Darrigo (1973), Brismar (1973), have suggested that calcium acts by forming a diffuse double layer outside the membrane, which can affect the trans membrane electric field. Ritchie and Rogart (1977) found the density of sodium channels for the single node of ranvier of 120,000 / m^2. A theoretical model accounts the properties of the inward rectification has been described by Cinani et al. (1978)

Matsummato and Kyung-skik, (1980) found some electrophysiological events observed at and around the state of spontaneous repetitive firing of action potentials which stimulated by the modified Hodgkin-Huxley equations. The events were ; 1. the frequency dependence of threshold to evoke action potentials when the mixture ratio of natural sea water and 550 mM NaCl as external fluid medium were parameters. 2. Linear dependence of the threshold up on the external Na concentration. 3. The power spectra of fluctuating membrane potentials when the mixture ratio of natural sea water and 550 mM NaCl was changed and 4. Effect of external sinusoidel current up on spontaneous repetitive spikes.Also the modelling of repetitive firing and bursting in a small unmyelinated nerve fiber was done by Scriven (1981) and the threshold for repetitive activity for a slow stimulus is studied by Rinzel et al(1988).

Celantano (1980) described an electronic clamp device which permits the differential measurement of the electrical potential difference across biological membrane and correction in the solution layers between the membrane and the voltage sensing electrodes.The voltage clamp technique has been also used by Garcia et al (1985) to study the capacitive transients of epithelia cells.

Meiri, (1981) studied the electrical membrane properties of exotomized and regenerating giant axons from the nerve cord of the cockroach, periplaneta americana, immediately after exotomy. There was a decrease in resting potential, input resistance and action potential amplitude near the cut end. This decrease was followed by the disappearance of the Na-dependent action potential, and an increase in the resting membrane conductance of a calcium dependent action potential.

Stochastic theory of singly occupied ion channels was described by Chiu et al (1989) . Ruben et al (1990) has shown that holding potential affects the apparent voltage sensitivity of sodium channel activation in cray fish giant axons . Gating current caused by charge movement in sodium and potassium channels has been studied in a number of works like Gilly et al (1980),Swenson (1983),Jurgen et al (1986) and Vandeuberg et al (1991).

1.2 Aim of Work

Normal voltage sodium channels can be induced to lose their ability to inactivate by treatment with proteolytic enzymes, with certain chemical reagents, or with toxins. The presence of non-inactivating sodium channels in the outer membrane of a cell is expected to profoundly modify the electrical properties of the cell because of the electrical depolarization of the cell and the opening of these channels reciprocally reinforce each other without intrinsic control. The normal resting state may thus be destabilized and new resting state at depolarized resting potentials may become possible (Steinberg -1988).

In this study, computer simulations were carried out to systematically explore the patterns of behavior of normal excitable cells and those having non-inactivating sodium channel (NI-NaC) in their plasma membrane. The cells were assumed to be space clamped and the relevant Hodgkin and Huxley equation ,H-H model, were assumed to describe the electrical behavior of the normal cells, and those having some or all of the sodium channels could not inactivate. The presence of NI-NaC in the membrane of a cell has the potential of profoundly modifying the electrical behavior of the cell. The activation of sodium channels depolarizes the cell and the depolarization in turn activates the channels. If the channels do not inactivate, the normal resting state may be destabilized and the cell may attain electrical stability also at depolarized membrane potentials. Accordingly the main aim of this work is to learn about the possible types of electrical behavior that may result from the presence of NI.Nac in the cell outer membrane. In this case H-H model should modified to study the effect of changing : 1-The