

STUDY OF SOME PHYSICAL PROPERTIES OF LIVING NERVOUS TISSUES

THESIS

Submitted as Partial Fulfillment

For

M. Sc. Degree in Physics

By

IBRAHIM HASSAN IBRAHIM MOHAMED

B. Sc. (Physics 1977) Ain Shams University

To

Department of Physics

Faculty of Science

Ain Shams University

1982

ACKNOWLEDGEMENT

I would like to express my deep gratitude to prof. Dr. A.A. Mohamed, Head of the Physics Department, for his valuable supervision and continuous encouragement.

Deep appreciation are also due to Dr. A.M. Sallam, for suggesting the point of research, encouragement, useful discussions throughout the progress of this work.

My thanks are also due to Dr. M. El-Moray for his kind supervision and useful help.

Finally, I would like to express my thanks to my colleagues in particularly Mrs. M.S. E. TALAT and Mrs. S. Sallam for their sincere cooperation.



CONTENTS

SUMMARY	Page
	i
CHAPTER I: INTRODUCTION.	
1- General Introduction	1
2- Electrophysiology as a tool of Research	6
3- Review of Literature.	9
4- Theoretical Background.	16
(a) Cable Theory	16
(b) Neuron Modeling.	20
The iron wire model	20
Threshold and accommodation model	23
The Hodgkin-Huxley model	25
CHAPTER II: MATERIALS AND ELECTROPHYSIOLOGICAL METHODS.	
1- Preparation and Solution	30
(a) Preparation	30
Sartorius muscle	
The sciatic nerve with the gastrocnemius muscle	
(b) Solution (Ringer Solution)	31
2- Types of Electrodes	33
(a) Metal Electrodes	33
(b) Glass Microelectrodes	36
Production and filling	36
Electrical properties.	39
3- Methods of Intra and Extra-cellular Recording	41
CHAPTER III: RESULTS AND DISCUSSION.	
1- Intracellular Recording	45
(a) Variation of Resting Potential in Frog	45
Sartorius Muscle	
Variation of resting potential with time	45
Measurement from a single fibre	46
Measurement from the whole muscle	48
Variation of resting potential with depth	51

(b) Variation of Resting potential with Time in Frog Sciatic Nerve	Page 56
2- Extracellular Recording	60
(a) Action potential Recorded from Sciatic Nerve	60
Effect of stimulus strength on Action Potential Amplitude	61
Effect of Distance on the Propagated wave of Excitation	66
Determination of the Length Constant	69
(b) Action Potential Recorded from Gastrocnemius Muscle	71
Stimulating the Nerve and Recording from the Muscle	71
Muscle Twitch	73
Stimulating and Recording from the Muscle	75
REFERENCES.	76

SUMMARY

SUMMARY

The present work used the two electrophysiological methods of recording "intra and extra-cellular methods" to study (1) the factors determining the resting and action potentials and (2) the structure and function of some living nerve and muscle fibres of the frog.

Stainless steel wire electrodes and glass micro-electrodes have a tip diameter less than 1 micron filled with 3-MK Cl were used for this purpose.

An electrophysiological unit CEPTU 812-81760-1 was used for stimulating and recording during this work.

The first chapter discussed the theoretical basis of electrophysiology as a tool of research, in addition to a complete review of literatures concerning the point of research.

The second chapter includes detailed explanation for the electrophysiological methods and materials used.

Chapter three indicates the results obtained and discussions about these results with a useful comparison with other results obtained by different authors.

It was found that the value of resting potential may be taken as an indication for the activity of living cell. The absolute value of resting potential recorded from an isolated sartorius muscle in normal Ringer solution was 85 ± 10 mV. The life time in this case was 4 hours approximately. The life time reduced to 2.5 hours in case of leaving the recording electrode inside the fibre during the experiment.

The profile of resting potential change during the advance of glass microelectrode through the muscle fibre was used for determining the fibre diameter and the muscle thickness.

Most muscles (sartorius muscle) showed a thickness ranged from 0.8-1.2 mm. and the muscle is composed of 2 or 3 fibre layers, each of which has a thickness ranging from 0.136 to 0.42 mm.

The life time of nerve (sciatic nerve) was longer than that of muscle fibres by nearly twice values.

The results showed that no noticable difference between the absolute values of resting potential for both nerve and muscle.

The results of action potential of the sciatic nerve indicated that the threshold value of electrical stimulus necessary for propagating action potential along the nerve was 0.5 V.

By increasing the stimulus strength above the threshold value, the value of action potential increases until it reach a full sized value depending on the distance between the cathodal stimulating electrode and the proximal recording electrode. In contrary, the duration of action potential showed a constant value independent to either the variation of stimulus strength or the distance between the stimulating and recording electrodes.

The variation of action potential amplitude with distance showed an exponential relationship in which at a distance of approximately 3 Cm, the action potential wave was blocked.

By studying the variation of action potential amplitude on the gastrocnemius muscle using different stimuli

- iii -

on the nerve connected it, two waves of action potential appeared, followed by the muscle twitch. The duration of the muscle contraction was about 11 msec. and the onset of the twitch is delayed by 4 m sec. After that of the action potential.

Typical biphasic action potential was recorded from the muscle by simultaneous stimulus and recording from it and no gradual change was observed in the amplitude of the action potential of both phases with the applied stimulus but a slight decrease in the amplitude of both phases noticed at higher stimulus strengths.

A mathematical expression deduced from the cable theory was used for determining the length constant of action potential. The relation between the calculated length constant and the stimulus strength showed an exponential relationship, indicated that the length constant has a value of nearly 3.5 Cm at the critical stimulus strength "0.5 V".

INTRODUCTION
CHAPTER I

INTRODUCTION

I.1. General Introduction:-

Biophysics is a form of applied physics. It is physics applied to a specific system composed of organic chemical materials organized into non homogeneous, non linear elements, the cells, and all parts of which do not follow exactly the same mathematical relationships. Academically there are no limits to biophysics, it could include all which is now described as "physical biochemistry", "general physiology" and it uses to the maximum extent possible physical models, scientific hypotheses, and physical instrumentation procedures.

From a practical point of view, physicists limit their attention to those fields whose bases are described by physics which could be regarded as encompassing three fields:-

- a. The use of physics to explain biological phenomena.
- b. Study of the effects of physical environmental agents on biological material.
- c. The use of physical instruments and techniques to study biological systems.

Electrophysiology is the branch of biophysical science which deals with electrical phenomena occurring in living matter. In general it is concerned with the study of bio-electrical phenomena.

Historically electrophysiology started as a field of research by the famous experiment of Galvani who explained the concept of animal electricity at the end of 18th century. The progress of research in this field seemed to be connected with the development of instruments and techniques used. This may be seen from the fact that, from the time of Galvani until the nineteenth century all the important research workers of bioelectrical phenomena were experimental physicists. In the second half of the last century electrophysiology of the living system marked a rapid development due to Meteucci (1845); Du Bois-Reymond (1841, 1849), Hermann (1879), Biedermann (1895), Bernstein (1912) and many others.

Du Bois-Reymond developed the technique of stimulation and recording using a galvanometer. In addition he demonstrated two fundamental types of bioelectrical potentials, the resting and action potentials.

Further knowledge of living matter increased with the development of electrophysiological techniques especially of recording apparatus such as the sensitive mirror galvanometer and Lippman capillary electrometer which permitted more detailed study for bioelectrical phenomena [Einthoven (1899)] invented the string galvanometer and used it to record the electrocardiography. Electrophysiology thus

moved from the laboratory, makes use of certain electrical manifestations during activity of some system for analysing this system itself.

After (1945) the latest phase of electrophysiology appeared by introducing microelectrode technique by Graham and Gerard (1946), Ling and Gerard (1949) used such electrodes for transmembrane intracellular recording of bioelectrical potentials.

The beginning of modern electrophysiology is marked by introducing the cathode-ray-oscilloscope [Erlanger and Gasser (1922)] which made possible the discovery of electroencephalogram. [Neminski (1925) and Berger (1929, 1930)].

During that period electrophysiology is frequently applied to the study of the nervous system, nervous activity and muscular contraction.

The study of an impulse wave propagated from its origin into the central nervous system had improved the methods used and made important discoveries such as epileptic rhythms in the electroencephalogram and localising tumors in the brain.

Application of intra and extra-cellular microelectrodes for measuring resting and action potentials resulted

in the formulation of ionic hypotheses [Hodgkin and Katz (1949), Hodgkin, Huxley and Katz (1949, 1952)] which considerably increased our knowledge about the chemical and electrical transmissions across the cell membrane and enlarged the list of structures available to electrophysiological experimentation.

Fundamental progress was also reached in electrophysiology by better understanding of the impulse transmission from the surface membrane to the intracellular structures [Huxley (1957 a)] and started the research into the mechanisms underlying this transmission

The ionic theory of nerve impulse [Hodgkin, Huxley (1952)] was in good agreement with the experimental finding and the use of digital computers [Cole (1955) and Huxley (1959)] helped for rapid progress in this field; computer modeling of networks of automata was successfully employed in research [Reschocko (1974) and Farlay (1976)]. The big amount of data obtained by simultaneous recording from many electrodes and the problems of detecting low signals from the noisy background caused the recent invasion of computers into electrophysiological research.

It can be seen that electrophysiology has been mainly employed in the study of the nervous system and it is most frequently used in research to study:

1. The relation ship between stimulus and response.
2. Correlation of spontaneous and evoked electrical activity in nerve structures.
3. Measurements of preperhal signals and their connections with the brain.
4. Physiological and pathological nerve processes.