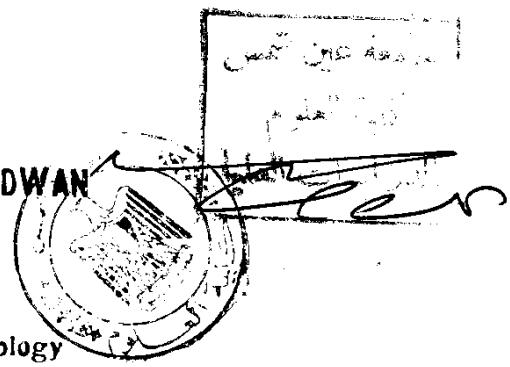
PHYSIOLOGICAL AND HISTOLOGICAL STUDIES ON THE RENAL-ADRENAL CORTEX ENDOCRINE SYSTEM IN THE MAMMAL ORYCTOLAGUS CUNICULUS

THESIS

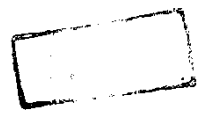
Submitted to the Faculty of Science

For the Degree of
MASTER OF SCIENCE



By
SHADIA ALI RADWAN

Department of Zoology
Faculty of Science
Ain Shams University
Cairo



1986

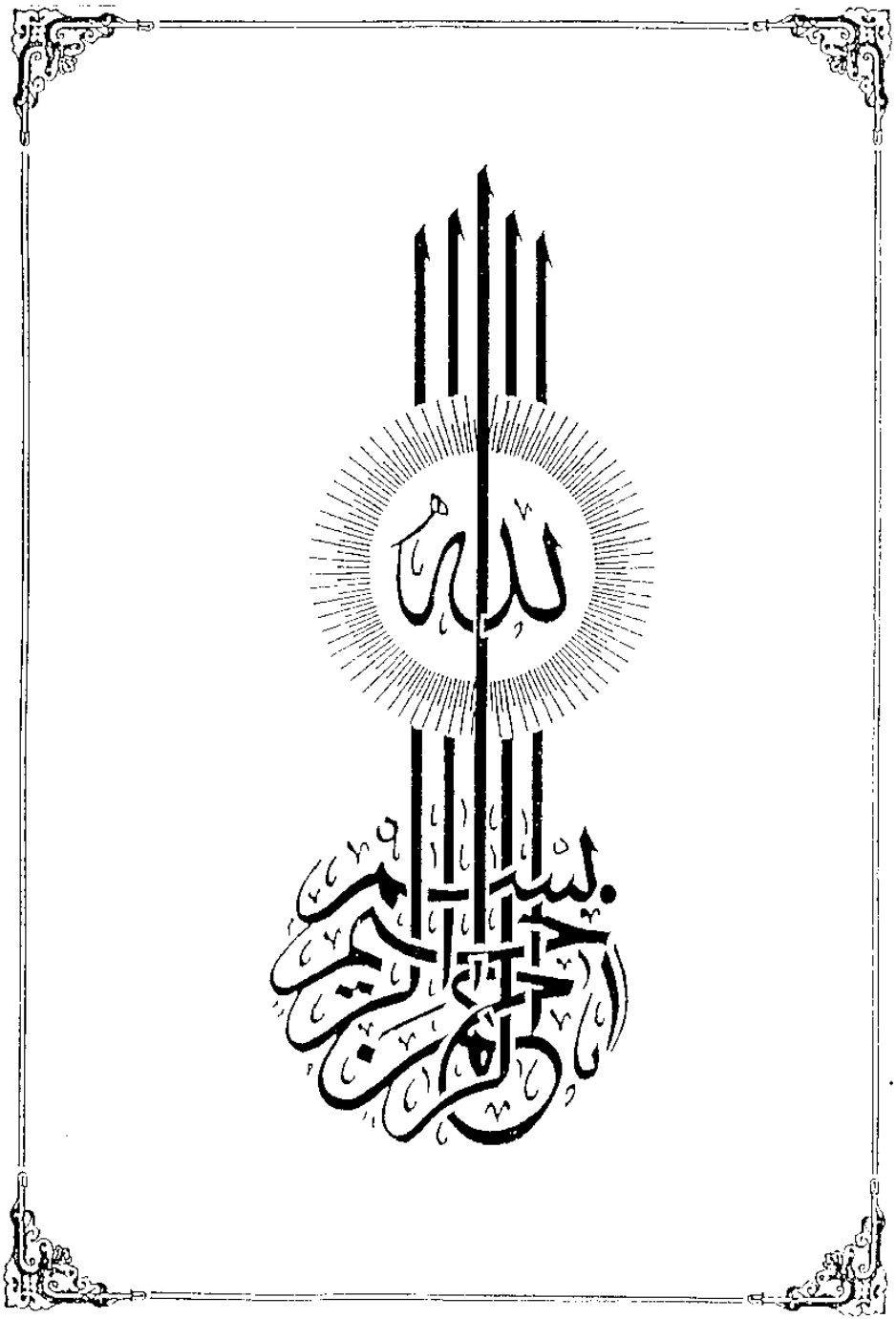
ACKNOWLEDGEMENTS

The author wishes to express her deep gratitude to late Dr. Tohamy A.Moussa, Professor of Cell Biology for suggesting the point of the present investigation and to Dr. Mahmoud A.El-Banhawy, Professor of Cytology and Histochemistry, Faculty of Science Ain Shams University and Dr. Amani A.H. EL-Hussaini Professor of Physiology, Department of Zoology, Faculty of Science, Ain Shams University for supervising the work.

Sincere thanks are also due to Dr. Ahmed R. Ezzat Lecturer of Physiology in the same Departmeht for his great efforts during the preparation of this thesis.

The writer is also indebted to the staff members of the Department of Biology Faculty of Education, Ain Shams University especially to Professor Dr. Amin Dowidar, Head of the Department of Natural History for the facilities put at my disposal for the present study.

The last and not the least, my deep appreciation to my colleagues, staff members and to Professor Dr. Mohemed A. Roshdy Head of the Department of Zoology, Faculty of Science Ain Shams University for their interest and continuous encouragement.



CONTENTS

Page

ACKNOWLEDGEMENTS

INTRODUCTION 2

PART I

HISTOLOGICAL AND HISTOCHEMICAL STUDIES

I- Previous Work 5

II- Materials and Methods 23

III- Observations 30

A- HISTOLOGICAL OBSERVATIONS

1- Normal histology of the adrenal gland of the male rabbit Oryctolagus cuniculus 30

2- Effects of ACTH-treatment..... 34

3- Effects of high-sodium diet..... 36

4- Effects of high-potassium diet..... 37

5- Normal histology of the juxtaglomerular region of the kidney 40

6- Effects of ACTH-treatment 41

7- Effects of high-sodium diet 42

8- Effects of high-potassium diet 42

B-HISTOCHEMICAL OBSERVATIONS

1- Total proteins..... 43

1.1- The adrenal gland of control rabbit 43

Page

1.2- Effects of ACTH-treatment 45

1.3- Effects of high-sodium diet..... 47

1.4- Effects of high-potassium diet 48

2- Ascorbic acid 49

2.1- The adrenal gland of control rabbits ... 49

2.2- Effects of ACTH-treatment 51

2.3- Effects of high-sodium diet..... 52

2.4- Effects of high-potassium diet 54

C-ULTRASTRUCTURAL OBSERVATIONS

1- The ultrastructure of the juxtaglomerular cells of control rabbits 56

2- Effects of ACTH-treatment 57

3- Effects of high-sodium diet 58

4- Effects of high-potassium diet..... 59

IV- Discussion 61

PART II

PHYSIOLOGICAL STUDIES

I- Previous Work 81

II- Materials and Methods..... 97

III- Results 105

Effects of ACTH , high-sodium diet, and high-potassium diet on:

1- Haematocrite value..... 105

2- pH of blood and urine 105

Page

3- Serum and urine sodium ion contents 109

4- Serum and urine potassium ion contents 111

5- Serum and urine calcium ion contents 111

6- Serum and urine magnesium ion contents 115

7- Serum albumin/globulin (A/G) ratio 120

8- Chromatographic separation pattern of aldosterone and cortisone 120

IV. Discussion. 125

SUMMARY AND CONCLUSION 148

BIBLIOGRAPHY..... 161

ARABIC SUMMARY .

PART I

INTRODUCTION

INTRODUCTION

The main function of the mammalian adrenal cortex in animals is to produce appropriate steroid hormones in adequate amounts. These hormones are essential for the continued existence of the animal. Steroids have a wide variety of activities including morphogenic, metabolic, and behavioral effects. Failure to produce the appropriate kinds or inadequate amounts of steroid hormones results in profound physiological disorders. In this concern, regulatory mechanismes, both within the adrenal cells and within the hypothalamic-pituitary-adrenal axis, normally coordinate to ensure the proper performance of the adrenal function (Gwynne and Ney, 1979; Tait et al., 1980 and Axelord and Reisine, 1984).

The regulation of the hypothalamic - hypophyseal adrenocortical axis has been studied in mammals (Temple and Liddle, 1970; Herbert et al., 1978 and Caselitz and Saeger, 1980). In this respect, it seems that the adrenocorticotrophic hormone (ACTH) secreted by the adenohypophysis is the most important factor controlling the secretion of the glucocorticoids (Jaanus et al.,1970 and Simpson et al.,1978) and, to some extent the androgens (Griffiths and Glick, 1966 and Grumback et al., 1978).

However, knowledge of the control of mineralocorticoid secretion, principally aldosterone, is not fully understood as that of glucocorticoids. Nevertheless, the nature of the control of aldosterone secretion has been lately one of the most intensive areas of endocrine research (Kenyon et al., 1984, Wingo, 1984 and Will et al., 1985).

It has been emerged from the current studies in this respect that the renin-angiotensin system (RAS) is a major regulator in the control of aldosterone in all the mammalian species along with the direct influence of potassium ion concentration (Bentley, 1971 and Mazzocchi et al.,1985). In this concern, it is worthy of mentioning that, in mammals, the main structural sites of the renin-angiotensin system are the juxtaglomerular cells and the macula densa located in the kidneys. The liver secretes renin substrate, angiotensinogen, into the blood where it is enzymatically converted to angiotensin I by the renin secreted by the juxtaglomerular cells. Angiotensin I is then transformed into angiotensin II by the converting enzymes located in the blood and the lung. Finally, the active hormone, angiotensin II acts to stimulate the secretion of dosterone by the adrenal cortex. ACTH is also regarded to have a permissive effect on aldosterone secretion because of its trophic action on the adrenal

cortex, but this effect is usually transit (Komor and Muller, 1979). According to Oelkers et al.,(1975) , aldosterone secretion is stimulated by salt depletion through the RAS. However, this point of view is still questionable and in need of further investigation.

In view of the above mentioned findings, the present investigation was planned to study the effects of the pituitary hormone (ACTH), high sodium, and high potassium diets on the structure and function of the adrenal cortex in the male rabbit Oryctolagus cuniculus. It was also hoped in this study to throw more light on the response of the renal-adrenal system of this mammal to the above mentioned treatments.

The rabbit was chosen as a model in this study because, inspite of the fact that this species is commonly used in laboratory experiments,yet the structure and function of its adrenal glands have been little and sporadically investigated (Nussdorfer, et al., 1978).

In brief, the present work comprises two main parts:

Part I, Includes histological and histochemical studies on the adrenal cortex in normal rabbits and in those subjected to the above mentioned treatments. The histochemical

investigation includes the study of both total proteins and ascorbic acid inclusions. The structural organization of the juxtaglomerular cells (JGC) of the kidney, as being integral part of the renin angiotension (RA), system was also investigated. Furthermore, the ultrastructure of the juxtaglomerular cells and their granular inclusions were also presented in this part.

Part II, Concerned with the physiological study which involves the haematocrit value hydrogen ion concentration of blood and urine, concentration of some electrolytes (Na, K, Ca, Mg) in serum and urine, electrophoretic seperations of serum proteins, and chromatographic separations of the serum aldosterone and cortisone in both normal and treated animals.

PREVIOUS WORK

PART I

PREVIOUS WORK

A- The Adrenal Cortex

The histological structure of the mammalian adrenal gland has been investigated by several authors (Deane and Greep, 1946 ; Chester jones, 1957 and El-Maraghy, 1976). According to these outhors, three distinct regions have been recognized in the cortex of the adrenal gland of the great majority of mammalian species, namely the zona glomerulosa, zona fasciculata and zona reticularis. According to Deane and Greep (1946), these three zones are morphologically distinct, and produce separate secretions.

It is now generally accepted that the activity the mammalian adrenal cortex is mainly regulated by the adrenocorticotrophic hormone (ACTH) secreted by the adenohypophysis. However, marked histological alterations have been observed in the adrenal cortex under the effects of various ACTH treatments by many investigators. Gresh and Grollman (1941) noticed widening of sinusoides and increase of blood supply in the adrenal cortex of mice and rats stimulated with ACTH. Also, Wexler and Rinfert (1955) described certain changes in the adrenal cortex of rats post-treatment with adrenocorticotrophin. These changes included rather complete, or almost complete depletion of the

sudanophilic elements from the component cells of the zona glomerulosam since cholesterol is the precursor of all steroid hormone. Nevertheless. Such changes were noticed by these authors to be highly dependent on the level of the hormone dose given to the animal.

Alterations in zonation and vascularity of the adrenal cortex of young male rats, following administration of adrenocorticotrophin, were also studied by Mikhail (1961). He described significant widening , accompanied with marked congestion of the sinusoids and reduction in thickness of the glomerulosa, whereas the fasciculata became wider than that of control animals.

A large variety of the structural changes which take place experimentally or pathologically in both zona fasciculata and zona reticularis, under the effect of ACTH were listed by Luse (1967). Accordingly, this author concluded that the stimulating effects of ACTH on the mammalian adrenal cortex is indicated by the morphological picture and the increase in the process of the adrenocortical steroidogenisis as a result of ACTH treatment. This view was later confirmed by Kuo and Tchen (1973) and Sharawy and Penny(1977)who noticed that daily injection of ACTH for 7-14 days led to a significant progressive