EXPERIMENTAL STUDY ON THE IMPACT OF HYDROCORTISONE ON THE TISSUES AND FINE STRUCTURES OF MAMMALIAN ADRENAL CORTEX AND TESTIS BEFORE AND AFTER TREATMENT WITH AN INHIBITOR DRUG OF ADRENOCORTICAL STEROID SYNTHESIS

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ABSTRACT

Key Words: adrenal cortex, glucocorticoids, histology, histopathology, hydrocortisone, ketoconazole, rat, steroidogenesis inhibitor drugs, testis, ultrastructure.

Recently, there is increasing concern about natural and man-made compounds that may interfere with endocrine system and thus may adversely affect the human health. Hydrocortisone and ketoconazole are synthetic drugs that had been chosen to be the core of the present study due to their wide utilization in the medical field.

The present study aimed to investigate the impact of hydrocortisone from the histological and ultrastructural point of view on the normal adrenal cortex and testis of adult male rats as well as its influence on such glands post inhibition with ketoconazole. Sixty adult male rats weighing 150-200g were divided into four even groups: ***hydrocortisone-treated group*** *i.m.* injected with *30mg/100g b.wt.* of hydrocortisone sodium succinate dissolved in *0.6ml* bacteriostatic water daily at 9am for *15* days, ***ketoconazole-treated group*** *i.p.* injected with *10mg/100g b.wt.* of ketoconazole dissolved in *1ml* saline solution daily at 9am for *15* days, ***hydrocortisone-treated group post ketoconazole treatment*** *i.p.* injected with *10mg/100g b.wt.* of ketoconazole for *15* days daily *at 9am* followed by *i.m.* injection with *30mg/100g b.wt.* of hydrocortisone for *15* days daily at 9am and 15 rats were kept as ***control group***, each 5 rats were injected with saline solution or bacteriostatic water in the same manner with the corresponding treated group.

Histologically, **hydrocortisone treatment** displayed shrinkage in the thickness of the three cortical zones resulted from

the compression of their component cells which showed signs of necrosis, the presence of clear fibrotic areas in zona glomerulosa and fasciculata, projection of a mass of cortical cells outside the thickened capsule, beside an occupation of medullary cells in between glomerulosa cells. *Ultrastructurally*, adrenocortical cells elucidated deformed mitochondria with abnormal type of cristae (i.e. lamelliform), lysosomes, accumulation of lipid droplets, collagen fibers, fingerprint-like configuration and pyknotic nuclei. Treated testes illustrated marked *histopathological* changes involved; reduction in spermatogenic cells with clear signs of necrosis, dilated intercellular spaces, loss of elongated spermatids and spermatozoa in some seminiferous tubules, in addition to, congested interstitial tissues involved necrotic Leydig cells and vacuoles. *Ultrastructurally,* treated testes showed thickening in basal laminae, clear necrotic signs in spermatogonia, primary spermatocytes and round spermatids, cytoplasmic vacuolation of atrophied Sertoli cells, beside, necrotic Leydig cells.

Ketoconazole-treated adrenal cortex revealed severe *histopathological* changes represented in hypertrophied signs of glomerulosa, fasciculata and reticularis cells which overloaded with lipid droplets that dislodged the deformed nuclei eccentrically. *Ultrastructurally*, adrenocortical cells displayed the presence of an abnormal hypertrophied mitochondria possessed vacuoles which resulted from a complete loss of cristae, extensive accumulation of lipid droplets, as well as pyknotic and karyolysed nuclei. Treated testes illustrated in their *histological pattern* necrotic spermatogonia, primary spermatocytes and round spermatids, deformed elongated spermatids, destructed spermatozoa, deformed Sertoli cells, beside, congested interstitial tissues having vacuoles and necrotic Leydig cells. *Ultrastructurally*, testes showed thickening in basal laminae, distinct necrotic signs in

spermatogonia, primary spermatocyte and round spermatids, deformed elongated spermatid and malformed spermatozoa, in addition to, necrotic Leydig cells in the destructed interstitial tissues.

Whereas, *histological* and *ultrastructural* examination of the **hydrocortisone-treated** adrenal cortex and testis **post ketoconazole treatment** revealed a relatively improvement towards return their normal architectures.

In conclusion, The severe deleterious destructive findings resulted post hydrocortisone or ketoconazole treatment in the normal adrenal cortex and testis should be taken in consideration. Therefore, this study represents a recommendation, that such drugs must be utilized under restricted precautions during the treatment of different types of diseases in the medical field to avoid their hazardous impacts on these glands in particular and on the human health in general.

On the other hand, worth mention that the present histological and ultrastructural results elucidated that hydrocortisone enhanced the architecture of inhibited adrenal cortex and testis which will be reflected in improving their steroidogenesis performance. Thus, the hydrocortisone may be considered as a useful treatment in these cases of inhibited adrenal cortex and testis.

CONTENTS

THE TESTIS

Contents

ABBREVIATIONS

A : Arteriole
AC : Acrosomal Cap
AG : Proacrosomal Granule
AHC : Acrosomal Head Cap
BC : Blood Cell
BL : Basal Lamina
BS : Blood Sinusoid
BV : Blood Vessel
C : Adrenal Cortex
Ca : Adrenal Capsule
Cap : Capillary
CB : Chromatin Body
CF : Collagenous Fibers
CN : Condensed Nucleus
CP : Chromatin Particles
CV : Central Vein
DN : Double Nucleoli
EC : Endothelial Cell
ES : Elongated Spermatid
Eu : Euchromatin
F : Fibrotic Area
Fb : Fibroblast
FE : Fibrous Element
Fl : Flagellum
FP : Fingerprint-like configuration
GA : Golgi Apparatus
GV : Golgi Vesicles

H : Head of Spermatozoon
Ha : Haemorrhagic masses
Ht : Heterochromatin
IT : Interstitial Tissue
Kh : Karyorrhexis
Kl : Karyolysis
LC : Leydig Cell
Li : Lipid Droplet
LM : Lamelliform Mitochondrial Cristae
Ly : Lysosome
M : Mitochondria
Ma : Manchette
MC : Myoid Cell
Md : Medulla
MO : Monocyte Cell
MP : Mid-piece
MV : Microvilli
N : Nucleus
NC : Necrotic Cell
Ne : Neck
NE : Nuclear Envelope
NEC : Necrotic Endothelial Cell
Nm : Nuclear Membrane
Nu : Nucleolus
P : Pseudopodia
Pk : Pyknosis
PS : Primary Spermatocyte
RC : Residual Cytoplasm
RER : Rough Endoplasmic Reticulum
RS : Round Spermatid
SB : Stagnant Blood
SC : Sertoli Cell

SER : Smooth Endoplasmic Reticulum
SG : Spermatogonia
Sl : Sloughed tissue
SN : Single Nucleolus
ST : Seminiferous Tubule
SZ : Spermatozoon
TA : Tunica Albuginea
Tb : Trabeculae
V : Vacuoles
Ve : Venule
VN : Vesicular Nucleus
ZG : Zona Glomerulosa
ZGC : Zona Glomerulosa Cell
ZF : Zona Fasciculata
ZFC : Zona Fasciculata Cell
ZR : Zona Reticularis
ZRC : Zona Reticularis Cell

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INTRODUCTION

Recently, there is increasing concern about natural and man-made compounds that may interfere with endocrine system and thus may adversely affect the human health.

The endocrine system consists of a highly integrated and widely distributed group of organs whose purpose is to maintain a state of metabolic equilibrium, or homeostasis among the various organs of the body. To accomplish this, the endocrine glands secrete a variety of chemical messengers or hormones that regulate the activity of target organs. Related to the endocrine system are the adrenal glands and the testes. These two organs are responsible for the synthesis and secretion of steroid hormones **(Sanderson, 2006)**.

The adrenal gland is the most important steroidogenic tissue in the human body and essential for survival. All steroidogenic processes take place in the adrenal cortex which plays a tremendous number of vital activities in the human body. This importance is being out from the fact that the adrenocortical zones synthesize and secrete steroid hormones, which involved of three major categories; mineralcorticoids, glucocorticoids and sex hormones. Mineralcorticoids, exemplified by aldosterone which is secreted by zona glomerulosa. Aldosterone is an important regulator of salt homeostasis and fluid balance, it can also

potently influence the blood pressure, and it is a major control unit of acid/base balance **(Bielohuby *et al.,* 2007)**. While, zona fasciculata secretes glucocorticoids, exemplified by cortisol, which is essential for life since it has a major role in responding to environmental stimuli; it decreases protein synthesis, thereby increasing the circulating level of amino acids, it elevates blood glucose by stimulating the enzymes involved in gluconeogenesis in the liver, increasing the activity of the urea cycle and it mobilizes fatty acids and glycerol from adipose cells. Cortisol has also anti-inflammatory effects: it stabilizes lysosomal membranes, reducing release of damaging proteolytic enzymes at sites of inflammation and it decreases capillary permeability lead to minimize local swelling. These attributes make cortisol a valuable medication **(Fawcett and Jensh, 2002; Campbell, 2005)**. By zona reticularis, small amounts of androgens are secreted. The two principal adrenal androgens are; androstenedione (Andro) hormone and dehydroepi-androsterone (DHEA), which is far less potent than testosterone and has little physiological significance. Both hormones can serve as substrates for the conversion into testosterone and estradiol **(Fawcett and Jensh, 2002; Keegan and Hammer, 2002)**.

The testis is also an endocrine organ, considered to be the most important organ in the male reproductive system. It is characterized by two main functions, synthesis of steroid