BIOCHEMICAL STUDIES ON SERUM ESTROGEN LEVELS OF FEMALE BILHARZIAL PATIENTS WITH HEPATOSPLENOMEGALY

THESIS

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Master of Biochemistry

BY

Ibrahim Mohamed Abd-Elsalam
B.Sc. Biochemistry (Ain Shams)

Supervised By

Prof. Easa Ahmed Easa
Biochemistry Department
Science Faculty
Ain Shams University

Prof. Mohamed Ibrahim Abdalla
OBS. & GYN. Department
Medicine Faculty
Cairo -- University

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C O N T E N T S

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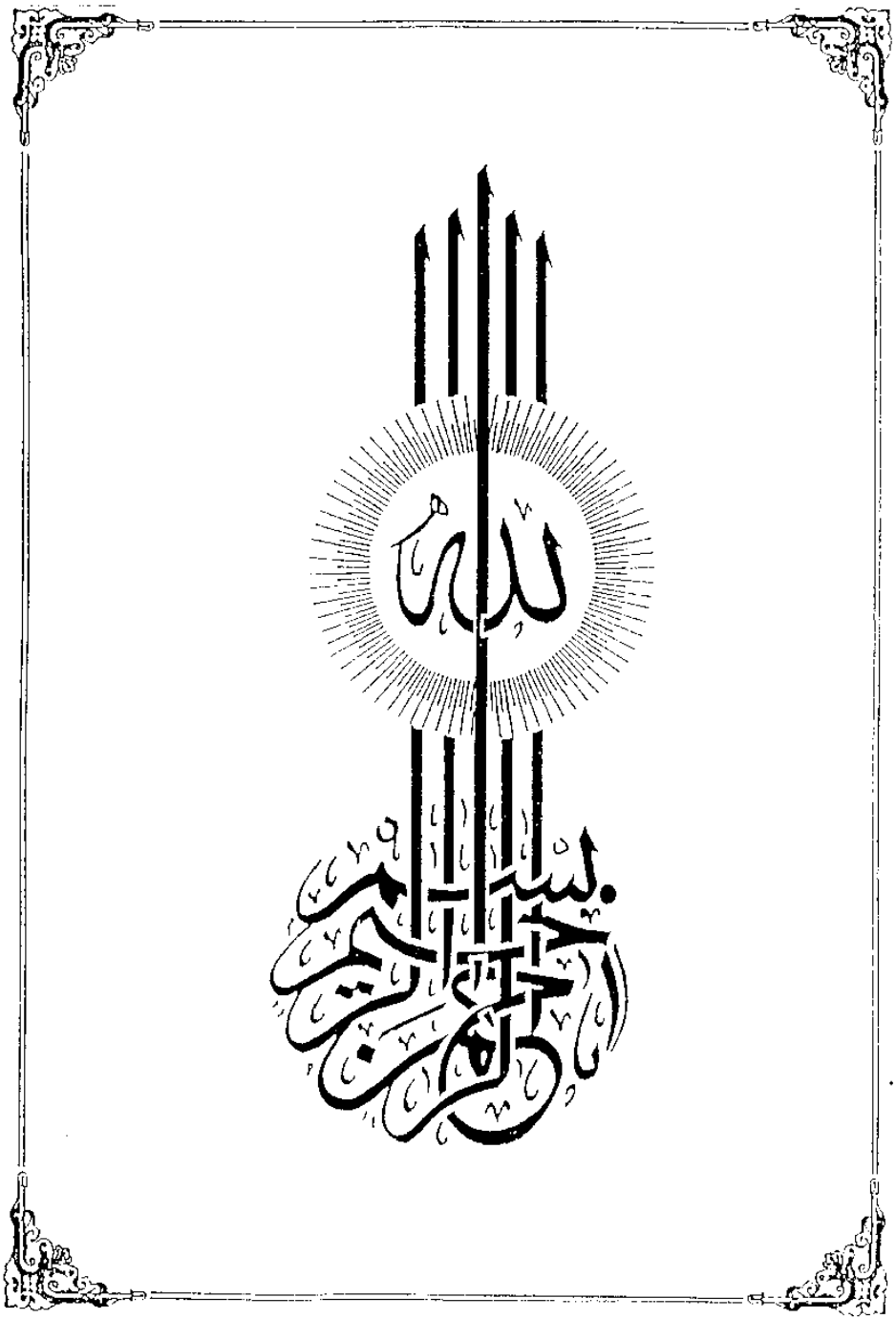

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CHAPTER I

INTRODUCTION AND AIM OF WORK

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Bilharziasis affects 60-70% of population in rural areas in Egypt and this is estimated to be 22 millions. Affection of the liver is the main manifestation of the disease and it has been reported that bilharzia ova reach the liver nearly in every case.

The ova deposition will result in bilharzial granulomata which ultimately end in hepatic periportal fibrosis with various clinical stages:

Stage I, hepatosplenomegaly.

Stage II, Splenomegaly + shrunken liver.

Stage III, Splenomegaly + shrunken liver + Ascites.

Disturbance in the endocrine function of various degrees subsequent to bilharzial liver affection has been reported in both sexes. Signs of feminization such as gynecomastia, testicular atrophy and signs of disturbed liver affection as palmer erythema and spider navei are frequently met with in males with various stages of bilharzial liver disease(Ghareeb, 1962).

Disturbance in the endocrine function of pancreas Ghanem et al. (1973), thyroid (Ghareeb et al., 1969) adrenal gland and gonads (Ghareeb, 1962). (Abdalla et al.,1978 a,b) had been reported in bilharzial liver disease in both males

and females.

The degree of endocrine derangement depends on age at infestation, duration of affection and the institution antibilharzial treatment.

With the evidence of hormonal disturbance in bilharzial patients the need for more determination of hormonal profile with various stages of the endemic disease is apparent. In females the knowledge of the hormonal profile will guide and point to the most appropriate contraceptive steroid to be used for them.

The recent use of competitive protein binding assays and radio-immunoassay had made it possible to determine very low levels of hormones in sera, with high degree of accuracy.

AIM OF WORK

This study is designed to:

(1) To determine the day to day changes in serum estrone, estradiol and estriol, throughout a complete menstrual cycle in normal Egyptian females and in patients with bilharzial hepatosplenomegaly.

(2) To determine the serum level of Alkaline phosphatase, SGOT, SGPT, total protein and albumin in bilharzial patients and to assess whether it is different from normal controls.

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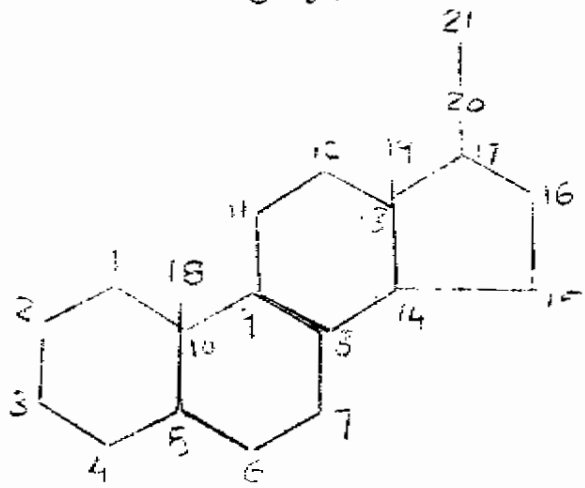
CHAPTER II

REVIEW OF LITERATURE

BIOCHEMISTRY AND METABOLISM OF ESTROGENS

(a) Basic structure and nomenclature of steroid hormones:

All the steroid hormones have the perhydrocyclopentanephenantherene ring system as the basic structure (Fig.1)



(Fig. 1)

The rings being identified by the letters A,B,C & D. The steroids considered here contain up to 21 carbon atoms (C_21) steroids) numbered as shown. Each carbon atom bears two hydrogen atoms except when it is common to two rings (i.e. at C_5, C_{10}, C_8, C_9, C_{13} & C_{14}).

Positions 18 and 19 are usually occupied by angular methyl groups unless otherwise indicated.

NOMENCLATURE

It should be noted that where as androstane and oestrane refer to the 5 α isomers, pregnone refers to the 5 β isomer.

Allo compounds refer to those which differ from the natural or typical steroids with reference to configuration at C_5 e.g. Pregnanediol (5 B) and allopregnanediol (5 α). When the configuration differs at any other carbon atom, the prefix epi is used, e.g. testosterone (17-B hydroxyl group) and epitestosterone (17 α hydroxyl group).

The suffix, ane, indicates a fully saturated nucleus, ene: the presence of one double bond, and -diene, two double bonds etc. The position of the double bond is indicated by the number of the carbon atom from which it originates and it is understood to terminate at the next higher carbon atom unless an alternative is possible, in which case it is indicated. Thus a bond originating at C_5 can terminate at C_6 or C_{10} and is designated 5 -ene or 5-(10) -ene respectively. For convenience a double bond is often indicated by A, thus Δ^4 indicates a double bond originating from C_4.

Alcohols are indicated by the suffixes -ol, -diol, triol, etc. or by the prefixes hydroxy, dihydroxy, trihydroxy, etc. and ketones by the suffixes -one, -dione, or prefix oxo.

The prefix dehydro is used to indicate the elimination of two hydrogen atoms, e.g. in dehydroepiandrosterone,

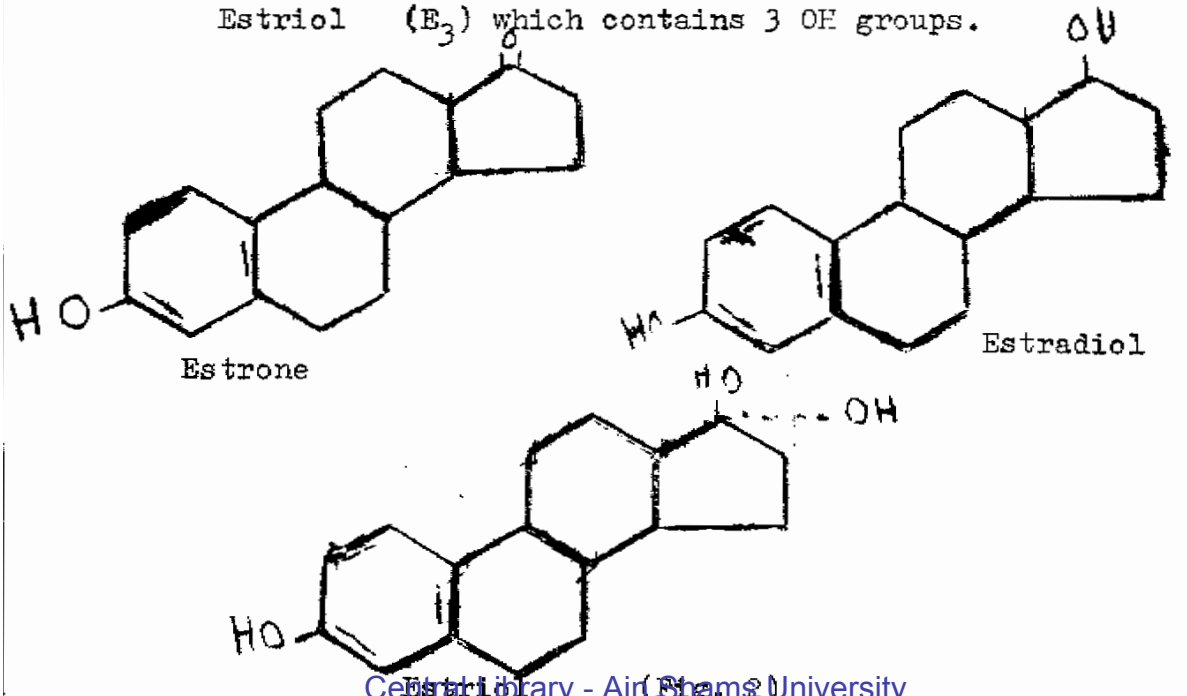
and di (tetra) hydro for the addition of two (four) hydrogen atoms. Deoxy indicates the elimination of an oxygen atom.

Estrogens are very active growth promating substances and produce widespread and complex effects upon the mammalian body. Some of their generalized effects may well be produced by the induction of metabolic changes, but in certain tissues, usually termed target tissues. These target tissues are in the breast, the genital tract and certain hormone sensitive centers of the brain such as the hypothalamus and pituitary gland.

The main site of formation of the estrogens in the female is the ovary. The three main estrogens in the human are estrone (E_1) which contains one O H group.

Estradiol (E_2) which contains 2 OH groups.

Estriol (E_3) which contains 3 OH groups.



Estriol (Fig. 2)

Chemistry:

The three main estrogens in the human are 17-B estradiol, estrone and estriol, which, for purposes of terminology, may be regarded as derivatives of the saturated hydrocarbon, "estrane".

The most potent of these as judged by bioassays being oestradiol-17B.

One of the essential features of these estrogens is the aromatic character of ring A (three double bonds) with absence of a methyl group at C-10. Because of this characteristic, the OH group at C-3 possesses the properties of a phenolic hydroxyl group (weaklyacid). All contain 18 carbon atoms.

Estrone has three double bonds, a hydroxyl (at C-3) and ketone (at C-17) group. Its systematic name is 1,3,5: 10 estratriene -3-01-17-One.

The designation 5:10 indicates the position of the ends of the double bond.

Applying the same system of nomenclature, 17B -Oestradiol is "1,3,5:10 -estratriene -3, 17 (B) -diol". and oestriol is "1,3,5 : 10 -estratriene - 3, 16, 17 (B)- triol.

Estrogen Biosynthesis

Metabolic interrelations and estrogen biosynthesis is

mainly a problem of conversion of androgens to estrogens. Engel (1958) reported that testosterone is converted to estradiol-17B by the human ovary. Baggett et al. (1959) showed conversion of testosterone to estrone and estradiol-17B by the stallion testis, adrenal cortical carcinoma and placental slices. Baggett et al. (1956) demonstrated the conversion of testosterone-3-C^{14} to C^{14} estradiol-17B by human ovarian tissue. Conversion of 19-hydroxy-, 1-4 androstene-, 17 dione to estrone by endocrine tissue was studied by Meyer (1955). The fact that ovarian, adrenal and placental tissues can oxidize androstenedione to 19-hydroxy-androstenedione and convert it to estrone was also recorded by Meyer (1955). Heard et al. (1956) reported the ability of pregnant mares to synthesize testosterone from acetate and to convert the latter to estrone. Perfusion of the dog's ovary with sodium acetate-1-C^{14} and testosterone results in the appearance of radioactive estradiol-17B (Nyman et al., 1959).

The pathways postulated for estrogen biosynthesis from androgens are:

1. Testosterone is first hydroxylated at C^{19} to yield hydroxytestosterone by a C^{19} hydroxylase. This compound can be dehydrogenated to 17B, 19-dihydroxy-1,4 androstene-3-dione. Reactions of this type have been reported by Levy and Talalay (1959 a.b.).

2. Ryan (1959 a,b) has shown that in placental tissue, effective conversion of testosterone to estradiol and conversion of androstenedione to estrone can be achieved by the microsomal fraction fortified with reduced triphosphopyridine nucleotide. He also found that 5-androstene-3B 16$^{\Delta}$, 17B-triol was converted to estriol and that 16 α hydroxytestosterone was converted to 16 α hydroxyestrone which was readily reduced to estriol. This second mechanism is not only for the formation of estriol but also for the ring D ketols formation. The relative importance of these mechanisms remains to be determined.

It is important to note that the formation of estrogens from testosterone does not require the presence of either gonads or adrenals. West et al. (1956) demonstrated this conversion in castrate adrenalectomized patients. However, the extent of such transormation is small and its physiologic importance is unknown.

The ready interconversion of estrogens is of great biological importance and was demonstrated by several investigators. Ryan and Engel (1953) showed that estrone and estradiol-17B are readily interconvertable by the human liver and several other tissues. Langer and Engel (1958) isolated from the human placenta soluble enzymes requiring tri- and di- phosphopyridine nucleotides, which effect estrone and estradiol interconversion.