CLINICAL AND ULTRASTRUCTURAL STUDIES IN MENTAL RETARDATION OF UNKNOWN ETIOLOGY

Thesis

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Ph. D. in Childhood Studies

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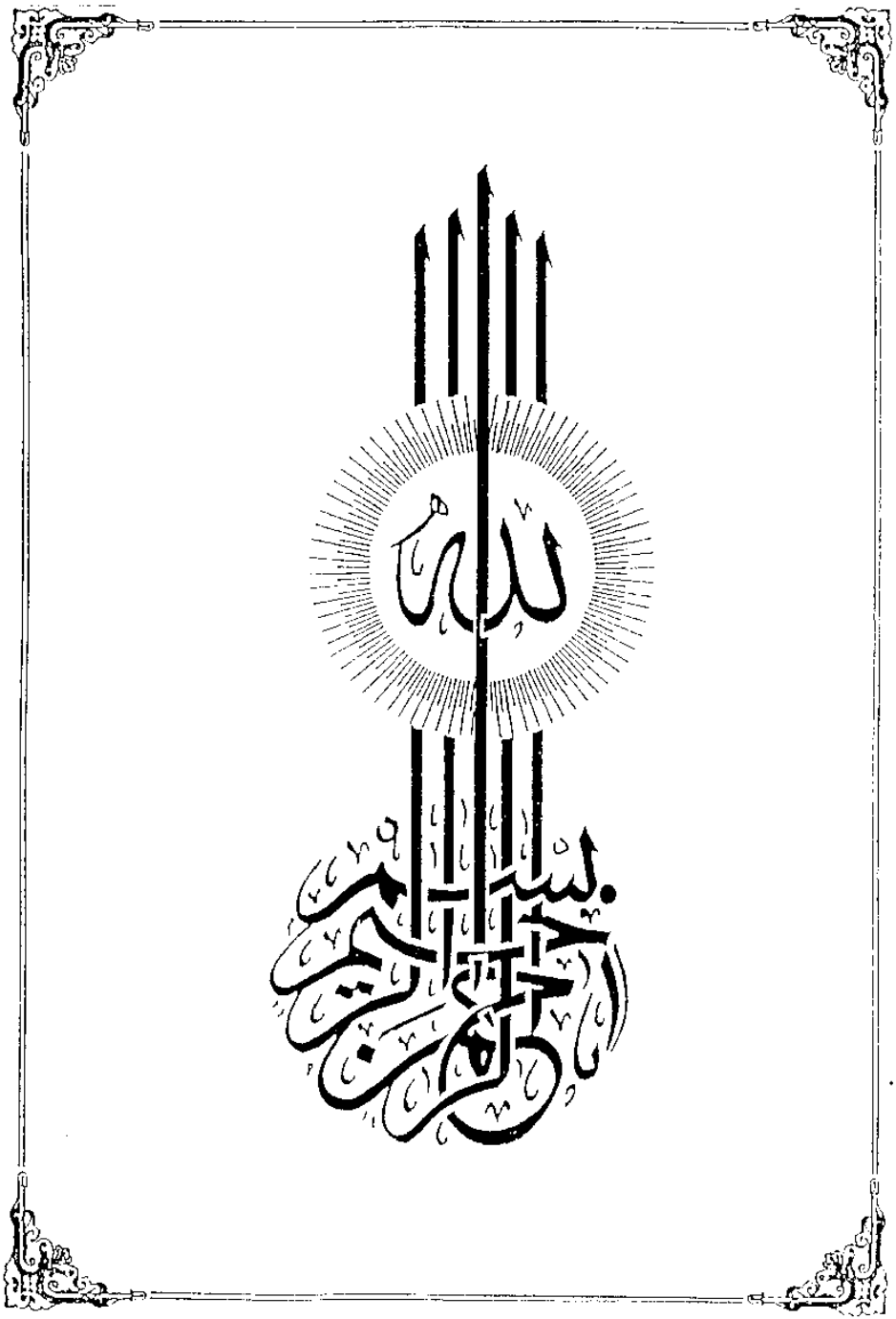

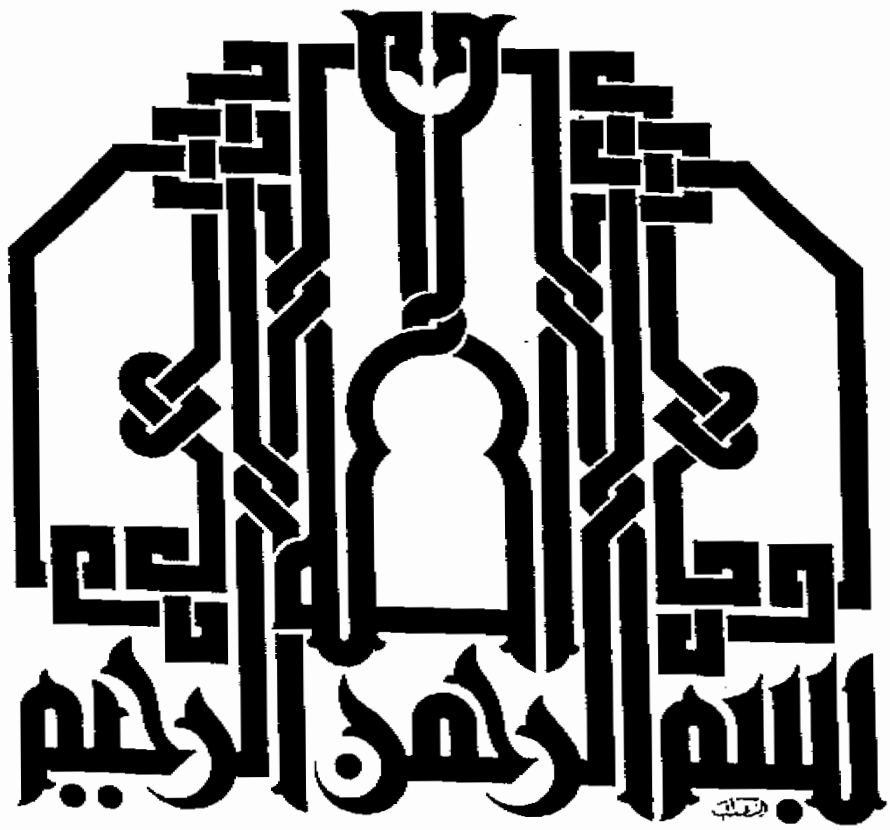

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List of Abbreviations

1 - X - linked ALD ..... X - Linked Adrenoleukodystrophy

2 - Pseudo - NALD ..... Pseudo Neonatal Adrenoleukodystrophy

3 - H_2O_2 ..... Hydrogen Peroxide

4 - μm ..... Micrometer

5 - nm ..... Nano Meter

6 - DNA ..... Deoxyribonucleic Acid

7 - NAD ..... Nicotinamide Adenine Dinucleotide

8 - RCDP ..... Rhizomelic Chondrodysplasia Punctate

9 - AR ..... Autosomal Recessive.

10 - CHRS ..... Cerebrohepatorenal Syndrome

11 - ZS ..... Zellweger Syndrome

12 - SGOT ..... Serum Glutamic Oxalacetic Transaminase

13 - SGPT ..... Serum Glutamic Pyruvic Transaminase

14 - CSF ..... Cerberospinal Fluid

15 - IRD ..... Infantile Refsum Disease

16 - HA ..... Hyperpipecolic Acidemia

17 - VLCFA ..... Very Long Chain Fatty Acid

18 - AMN ..... Adrenomyeloneuropathy.

19 - ERG ..... Electroretinogram

20 - C.T ..... Computerized Tomography

21 - AGT ..... Alanine Glyoxylate Aminotransferase

22 - EEG ..... Electroencephalogram

23 - IQ ..... Intelligence Quotient

Table of Contents

INTRODUCTION

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Genetic studies have always been essential in the diagnosis and classification of diseases of heterogeneous etiology. In addition, they are a prerequisite to prevention, particularly in the field of mental retardation. This can be accomplished by early detection and treatment, proper genetic counselling and prenatal diagnosis (Leroy and Wiesmann, 1993).

Some cases of progressive mental retardation which cannot be at present diagnosed in Egypt by cytogenetic or presently available biochemical techniques may be either peroxisomal, sialic acid or lysosomal abnormalities.

The peroxisomal diseases represent a group of genetically determined disorders in which the major cause of pathology is either the failure to form or maintain the peroxisome or a defect in the function of a single enzyme that normally is located in this organelle. There are several recent reviews with general agreement that peroxisomal disorders can be subdivided into three major categories (Theil et al, 1992).

The first group comprises the disorders of peroxisomal biogenesis in which the organelle fails to form or is unable to be maintained with resultant dysfunction of multiple peroxisomal enzymes eg.are Zellweger syndrome, infantile Refsum disease, neonatal adrenoleucodystrophy and hyperpipecolic acidemia.

The second group is characterised by loss of two or more peroxisomal functions eg: are Rhizomelic chondrodysplasis punctata and Zellweger-like syndrome.

The third group is characterised by loss of only a single peroxisomal function eg: are X-linked adrenoleucodystrophy, acyl CoA oxidase deficiency,

bifunctional protein deficiency, thiolase deficiency, hyperoxaluria type I, acatalasemia and adult Refsum disease.

Sialic acid storage disorders are two types: sialidosis and sialuria. Sialidosis is the term used to designate a group of disorders in which patients have a deficiency of sialidase (neuraminidase) activity. Sialuria differs biochemically from sialidosis by the accumulation and excretion of free sialic acid and normal levels of neuraminidase activity. Therefore, sialuria is a hallmark feature in disorders of free sialic acid metabolism (Mc Kusick,1990).

Patients with mucolipidoses exhibit clinical features of both lipidoses and mucopolysaccharidoses. Despite their name there is little evidence of true storage of lipids or mucopolysaccharides in the organs of affected patients. All the mucolipidoses are inherited as autosomal recessive traits. Unfortunately, there is no specific treatment for these metabolic disorders (Matalon, 1992).

THE AIM OF THE WORK

- To apply specific biochemical screening test to reach a diagnosis in cases suspected by EM to be peroxisomal or sialic acid storage disorders.
- To compare the clinical, genetic features, the ultrastructural results and the biochemical findings.

REVIEW OF LITERATURE

Peroxisomal Disorders

The peroxisomal diseases represent a heterogeneous group of genetically determined disorders in which the major cause of pathology is either the failure to form or maintain the peroxisome as a functional intracellular arganelle or a defect in the function of one or more enzyme that normally is located and active within this organelle. These disorders cause serious disability in childhood. They occur more frequently and present a wider range of phenotypes than has been recognized in the past.

Peroxisomes are subcellular organelles bound by a single membrane and filled with a fine granular matrix. These organelles were first described in 1954 in the cytoplasm of proximal tubular cells in the kidney of the mouse. They were referred to as microbodies. Biochemically, peroxisomes were characterised for the first time in 1960 by De Duve et al. Although peroxisomes are subcellular organelles that are universally found in animal cells (except mature erythrocytes), their functional significance in mammalian metabolism remained unclear until recently (Shutgens et al,1986).

The peroxisomes are organelles specializing in the oxidation of a number of substances, such as amino acids, uric acid, ethanol, phenols and fatty acids (to acetyl CoA), the key enzyme being catalase which removes the H_2O_2 arising through the action of the flavin-containing oxidases. The association of H_2O_2 generating enzymes and catalase in one single organlle led De Duve to introduce the name peroxisome (Wanders et al, 1993).

The peroxisomes range in size from 0.2 to 1.5 µm. The membrane is 6.5 to 7 nm thick and has a trilaminar appearance and a unique polypeptide composition.

Peroxisomes have an unusually high equilibrium density because the membrane contains aqueous protein pores, which permit free passage of metabolites as well. The organelles do not contain DNA and probably also lack glycoproteins.

Although it was thought for a long time that peroxisomes arise by budding from the endoplasmic reticulum, it is now clear that peroxisomes originate by division, or fission, of pre-existing ones (Borst, 1989).

Peroxisomes are frequently found under electron microscopy in close proximity to the endoplasmic reticulum (Soheyla et al,1992).
Another, now generally accepted, principle of microbody biogenesis is that peroxisomal matrix proteins and membrane proteins are encoded by nuclear genes and translated on free polyribosomes in the cytosol , through which they diffuse with half times of 1 to 15 minutes before they are imported into pre-existing peroxisomes (Roermund, 1991).

There are at least eight different genes involved in the formation of normal peroxisomes and in the transport of peroxisomal enzymes (Yajima et al, 1992). Peroxisomes are degraded completely in a random fashion by autophagy.
In recent years, it has become clear that peroxisomes fulfil a number of essential cellular functions. Their main functions in mammalian cells are in lipid metabolism and respiration (Naidu and Moser, 1990).
Peroxisomes are differentiated from mitochondria by their single membrane, electron dense homogeneous matrix, the absence of cristae and lack of DNA (Lazarow, 1987).

The visualization of peroxisomes by staining for their catalase activity with diaminobenzidine (DAB), is a well established and widely used cytochemical method. It was designed by Novikoff and Goldfisher (1969) and modified for human liver and kidney by Roels and Goldfischer (1979). It is an invaluable approach for the reliable examination of peroxisomol features in liver samples from peroxisomal diseases (Espeel et al, 1991). By this means it has been shown that peroxisomes vary in size, shape and number in various organs. They are particularly abundant in liver and kidney, where they have a diameter of 0.5μm. Those in the nervous system in fibroblasts and amniocytes are smaller, with a diameter of 0.1 -0.25μm.

Peroxisomes are numerous in cells that specialize in lipid metabolism, including sebaceous glands, brown fat and the nervous system. The organelles are more abundant in the nervous system in the early postnatal period than they are in the adult brain. Cerebral and cerebellar neurones contain peroxisomes in the first two postnatal weeks, but they are rarely found after the third week of life (Naidu and Moser, 1990).

During active myelination, oligodendroglia contain peroxisomes that can be demonstrated in processes forming myelin sheaths (Adamo et al, 1986). This abundance of peroxisomes in the developing nervous system and glia suggests a major role of peroxisomes in myelinogenesis and may be important in understanding neuronal migration defects noted in the peroxisomal deficiency disorders.

Peroxisomal Enzymes:

In a review in 1981 Tolbert listed 40 peroxisomal enzymes. Since then additional enzymes have been assigned to the peroxisome. The peroxisomal