DETERMINATION OF SALIVARY ANTIMICROBIAL PEPTIDE LEVEL AND ITS RELATION TO ORAL HEALTH PARAMETERS IN A GROUP OF MONGOLIAN CHILDREN.

Thesis

Submitted to the Faculty of Oral and Dental Medicine, Cairo University in partial fulfillment of requirements for Master Degree in Pediatric Dentistry and Dental Public Health.

BY

Nada Mohamed Mostafa Hassan Wassef

(B.D.S.)

Faculty Of Oral And Dental Medicine

Cairo University

٢٠٠٨

SUPERVISORS

Prof. Dr. Eman Sayed El-Masry

Professor of Pediatric Dentistry
and Dental public Health,
Faculty Of Oral And Dental Medicine
Cairo University

Dr. Osama El-Shahawy

Lecturer of Pediatric Dentistry
and Dental public Health,
Faculty Of Oral And Dental Medicine
Cairo University

بسم الله الرحمن الرحيم

(و قل رب زدنى علما)

صدق الله العظيم

(سورة طه–الأية ١١٤)

ACKNOWLEDGMENT

First of all I want to thank GOD for all the mercies and blessings that we are blessed with, and for everything in our lives.

Words can not express how grateful, thankful, and obliged I am to my parents for their endless love, support, trust, and help throughout my whole life.

I would like to express my deepest thanks and appreciation to ***Prof. Dr. Eman El-Masry***, Professor of Pediatric Dentistry and Dental public Health, Faculty Of Oral And Dental Medicine, Cairo University for her great help, guidance, encouragement, and continuous support.

My deepest gratitude and thanks to ***Dr. Osama El-Shahawy,*** Lecturer of Pediatric Dentistry and Dental public Health, Faculty Of Oral And Dental Medicine, Cairo University for his great patience, guidance, effort, and endless encouragement in helping me through this work.

I can not express how grateful and indebted I am to ***Prof. Dr. Mohamed Ali Saber***, Professor of Genetics, Tudor Bilharz research institution, for his great guidance, help, support, effort, and meticulous work with us.

Also I would like to thank my friends and colleagues for their love, help, and support.

And never can we thank enough the children that always help in allowing any of our research work to be done.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF ABBREVIATIONS

AMPs	Antimicrobial peptides
DS	Down's syndrome
ELISA	Enzyme-linked immuno-sorbent assay
GI	Gingival index
hBD	Human β- defensin
HNP	Human neutrophil peptides
LL-٣٧	Cathelicidin
ROB	Robertsonian translocation
S.mutans	Streptococcus mutans
TP	Total protein

INTRODUCTION

Down's syndrome (DS) is an easily recognized congenital, autosomal (non-sex chromosomes) anomaly characterized by generalized growth deficiency and mental deficiency affecting ١ in ٦٠٠ to ١ in ١٠٠٠ live births **(Regezi and Sciubba, ١٩٨٩).**

In past decades, most individuals with Down's syndrome were usually not afforded adequate medical care. Many children with Down's syndrome were institutionalized and they were often deprived of all except the most elementary medical services. Fortunately, there have been major improvements in the health care for them **(Pueschel, ١٩٩٠).**

Dental caries susceptibility is usually low in patients with Down's syndrome, although the prevalence and severity of periodontal diseases are much higher than normal **(Shapira and Stabholz, ١٩٩٦).**

Gingival inflammation in Down's syndrome children develops earlier and is more rapid and extensive than in non-DS children. Abnormalities in host response to the oral flora have been proposed as etiological factors of this gingival inflammation **(Morinushi et al., ١٩٩٧).**

Saliva contains several types of antimicrobial peptides that play a role in innate immunity. Peptides that were recently added to this list are the defensins **(Abiko et al., ٢٠٠٣).**

Antimicrobial peptides (AMPs) are important components of the natural defences of most living organisms against invading pathogens **(Reddy et al., ٢٠٠٤).**

The α-defensins, HNP١–٣, have also been detected in saliva and are elevated in patients with oral inflammation **(Dale and Fredericks, ٢٠٠٥).**

With antimicrobial peptides now strongly implicated in the host innate immune response, in particular in the oral cavity , their availability in unstimulated saliva implies their potential role in protecting tooth structure from bacterially-induced caries, either by direct killing or by prevention of biofilm formation on the tooth surface **(Dale et al., ٢٠٠٦).**

The expression of AMPs in saliva and throughout the oral cavity suggests that they may have a role in protecting tooth structure from caries as well as protecting oral mucosa **(Dale et al., ٢٠٠٦).** Therefore, investigating the α-defensins (antimicrobial peptides) level and its relation to oral health parameters in groups of Down's syndrome and normal children may be beneficial in the prevention and treatment of dental caries and gingival diseases in Down's syndrome children.

Review of Literature

Incidence of Down's syndrome

Approximately one out of every ٨٠٠-١,١٠٠ births results in an extra chromosome of the twenty first group called Trisomy ٢١, or Down's syndrome **(Pilcher, ١٩٩٨).**

In Egypt, the incidence of Down's syndrome has been reported to be ١ per ١٠٠٠ births **(Mokhtar et al., ٢٠٠٣).**

Etiology of Down's syndrome

Approximately (٩٥%) of Down's syndrome cases have extra chromosome ٢١, making the chromosome count ٤٧ instead of the normal ٤٦. The other (٥%) are represented by other chromosomal abnormalities including translocation (٤%) and mosaicism (١%) **(Mutton et al.,١٩٩٦)**

The origin of the extra chromosome is maternal in (٩٥%) of cases and is due to failure of normal chromosomal segregation during meiosis **(James et al., ١٩٩٩).**

Despite extensive studies, it is not possible to clinically differentiate patients with mosaicism or translocation from those with regular trisomy ٢١ **(Mokhtar et al., ٢٠٠٣).**

Robertsonian translocations (ROBs) in humans are whole-arm rearrangements between the acrocentric chromosomes ١٣, ١٤, ١٥, ٢١, and ٢٢. **(Berend et al., ٢٠٠٣).**

In about (١%) of all cases of Down's syndrome, the mistake in the distribution of chromosomes in cell division occurs shortly after fertilization of the ovum by the sperm, so that there is a mixture of cells with different chromosome patterns. This situation is called mosaicism. This means that some individuals who have Down's syndrome have some of their body cells containing ٤٧ chromosomes because of an extra copy of chromosome ٢١, while other cells have the usual ٤٦ chromosomes **(Stewart, ٢٠٠٧).**

Risk factors

The main risk factor for Down's syndrome is maternal age with many studies reporting an increased incidence of Down's syndrome with increased maternal age. There has also been some suggestions of an association with paternal age however this has not been confirmed **(De Michelena, ١٩٩٣).** Other suggested risk factors include race, with an increased rate among Hispanic moters **(Bishop et al., ١٩٩٧),** ionising radiation **(Verger, ١٩٩٧),** increased parity **(Kallen, ١٩٩٧ and Schimmel et al., ١٩٩٧),** although this has not been confirmed by all studies **(Chan et al., ١٩٩٨),** and season, with a peak in births in summer **(Whiting et al., ٢٠٠١).**

Diagnosis of Down's syndrome

The diagnosis of Down's syndrome is made by chromosomal analysis, which can be initiated prenatally (in the first or second trimester of pregnancy) due to given risk factors for pregnancy, or postnatally due to the

characteristic appearance of the newborn child **(Dzurova and Pikhart, ٢٠٠٥).**

Down's syndrome can be diagnosed relatively easily prior to birth by measuring alphafetoprotein, human chorionic gonadotropin and unconjugated estriol in fetal serum, detecting a thickened nuchal fold on fetalultrasound, and by cytogenetic analysis **(Tagliabue et al., ٢٠٠٧).**

Clinical features of Down's syndrome patients

Down's syndrome involves a set of signs and symptoms that characterize a delay in the development of motor and mental functions of its carriers, entailing mental and general alterations **(Coelho and Loevy, ١٩٨٢ & Mustacchi and Rozone, ١٩٩٠).**

Congenital heart disease is diagnosed in approximately (٤٠%) of children with Down's syndrome **(Pueschel, ١٩٩٠).**

It has been estimated that between (١٠%) and (٣٠%) of individuals with Down's syndrome have either atlantoaxial instability, atlantooccipital instability, therefore careful positioning in the dental chair is required to avoid any potential harm to the spinal cord **(Pueschel, ١٩٩٠).**

The primary skeletal abnormality affecting the orofacial structures in Down's syndrome is an underdevelopment or hypoplasia of the midfacial region. The bridge of the nose,

bones of the midface and maxilla are relatively smaller in size. In many instances this causes a prognathic Class III occlusal relationship which contributes to an open bite. Absence or reduction in size of the frontal and maxillary sinuses is common **(Vittek et al., ١٩٩٤).**

Children with Down's syndrome are at greater risk of developing leukemia, usually the acute lymphocytic type **(Wilson, ١٩٩٤ and Desai, ١٩٩٧)**.

There is an increased susceptibility of Down's syndrome individuals to many infections which could be explained by the fact that the neutrophil leukocytes are defective. There are reports of lymphopenia, and eosinopenia, but in an addition, cell mediated immunity is impaired and serum immunoglobin patterns are disturbed. The commonly seen infections in Down syndrome are dermal, mucosal, gastrointestinal and respiratory **(Desai, ١٩٩٧).**

It has been reported that the incidence of upper airway obstruction may be as high as (٣١%) in children with Down's syndrome. The decreased airway size combined with lowered muscle tone predisposes these patients to obstructive sleep apnea **(Pilcher, ١٩٩٨).**

Persons with Down's syndrome vary widely as to their degree of intellectual impairment. Most have IQ's in the mild to moderate range and are able to be treated in a normal

setting. There is often a relatively severe delay in language development. The patient with Down's syndrome will probably understand more than their apparent level of verbal skills **(Pilcher, ١٩٩٨).**

The following general alterations are involved:
-Eyes: slanting, almond-shaped, strabismus, and myopa.
-Nose: flattening of the nose bridge, small, short nose with broad nasal bridge, and pug nose.
-Ears: lop ear with flat or absent helix, auricles with a low implantation.
-wide short neck with abundant skin, congenital cardiopathy, wide hands, short fingers, clinodactyly, brachydactyly, and muscular hypotony **(De Moraes et al., ٢٠٠٧)**

Oral features in Down's syndrome

Common oral features in Down's syndrome children include reduced size of the teeth, agenesis and high frequency of crown irregularities, these abnormalities are all interrelated and result from a decrease in mitotic activity of dental progenitor cells during embryogenesis **(Townsend, ١٩٨٦).**

Oral findings include mouth breathing, open bite, appearance of macroglossia, fissured lips and tongue, angular cheilitis, delayed eruption times, missing and malformed teeth, oligodontia, small roots, microdontia, crowding, and low level of caries **(McDonald and Avery,**