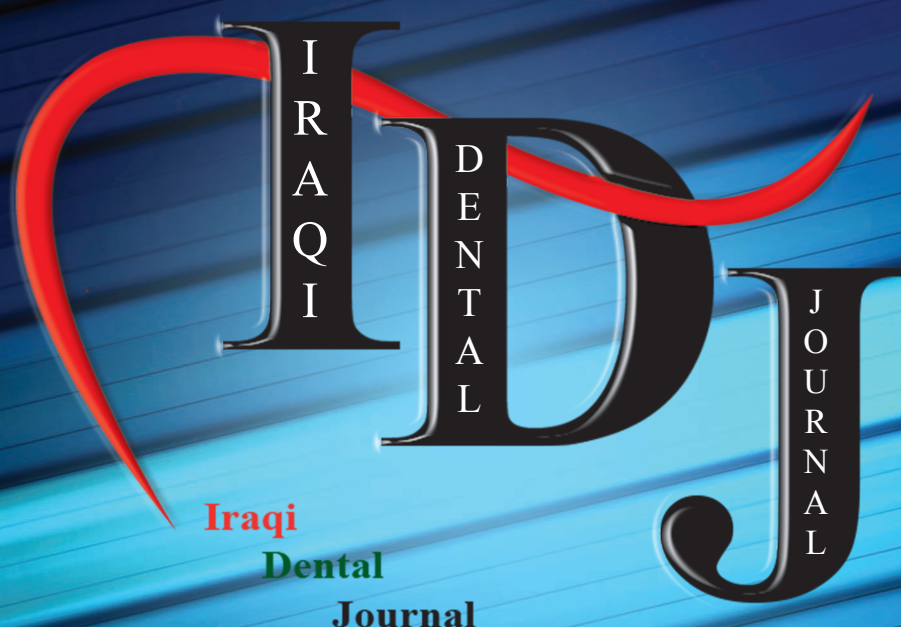




ISSN 2307-4779

# Iraqi Dental Journal



A Triannual Peer-Reviewed Journal , Published by  
**the Iraqi Dental Association**

Volume:38 / Issue:2 / August 2016

# Guidelines For Authors

The “Iraqi Dental Journal” is a Triannual Peer-Reviewed Journal  
Published by the Iraqi Dental Association

## MANUSCRIPT SUBMISSIONS

A covering letter of submission of each manuscript should be sent to the Editor-in-Chief providing complete contact information for the corresponding author.

Manuscripts should be submitted by email to ([submit@iraqidentaljournal.com](mailto:submit@iraqidentaljournal.com)) as an attached Microsoft Word file. Illustrations should be jpeg high resolution files. Tables and figures should be clear and labelled in numerical order as they appear in the text and should be embedded within the text , Illustrated charts should be placed as a picture (not as an Excel chart).

Posted submissions should contain three copies of manuscripts plus CD-Rom containing a soft copy of the manuscripts in the same format mentioned above.

Articles, or any part of them, should not have been previously submitted or published in any other journal.

Manuscript Preparation: All Manuscripts should have: Font size of 12 point and preferred style is Times new Roman. Posted Manuscript should be printed on A4 papers. Do not use the automatic formatting features of Microsoft Word Office such as footnotes and endnotes. All pages should be numbered numerically.

## MANUSCRIPT ARRANGEMENT

- Title: All articles must have a title with the following information and in this particular order: title of the article (ten words maximum), surname, initials, qualifications and affiliation of each author, name, postal address, e-mail address and telephone contact details of the corresponding author.
- Abstract: The abstract should be 200-250 words and should outline the purpose of article including background, material and methods, results and conclusions.
- The abstract should be translated into Arabic. Instant electronic translation is not accepted; therefore, the abstract should be translated by a well-qualified person.
- Keywords: All articles should include “keywords”, up to five words or short phrases should be used. Use terms from the medical subject headings (mesh) of index medicos when available and appropriate. Keywords are used to index the article and may be published with the abstract.
- Articles should be in English and they should be edited by a qualified language advisor.
- Conflict of Interest Information: Authors should declare conflicts of interest clearly.
- Introduction: The introduction should be brief and describe the purpose of the study.

- **Methods:** The methods should include the study design and setting. Statistical analyses have to be included with enough details.
- **Results:** The results should be concisely and reasonably summarize the findings. Graphics or tables should be included. Tables should be self-explanatory, clearly organized, and supplemental to the text of the manuscript. Tables must be inserted in the correct position in the text. Authors should place explanatory matter in footnotes, not in the heading. All figures must be inserted in the appropriate position of the electronic document. Symbols, lettering, and numbering (in Arabic numerals e.g. 1,2, etc... in order the appearance in the text) should be placed below the figure, clear and large enough to remain legible after the figure has been reduced. Figures must have clear descriptive titles.
- **Discussion:** The discussion should deal with new ideas or importance of the article.
- **Conclusion:** The conclusion should be linked with the aims of the study. Put asterisks (\*) to highlight new hypotheses when warranted.
- **References:** Cite references in numerical order in the text, in superscript format. Use brackets in the References section, references must be typed in the order in which they are cited, not alphabetically. Authors are responsible for accuracy of all references. Personal communication and unpublished data should not be referenced. If essential, such material should be incorporated in the appropriate place in the text. List all authors when there are six or fewer, when there are seven or more; list the first three, then “*et al.*”.

#### About IDJ

p-ISSN: 2307-4779

e-ISSN: 2411-9741

KEY TITLE: Iraqi Dental Journal

ABBREVIATED KEY TITLE: Iraqi dent. j.

#### AIMS

Iraqi Dental Journal is a peer-reviewed, open access journal that publishes original research articles, review articles, and clinical studies in all branches of dentistry . It is dedicated to the dissemination of new knowledge and information on all sciences relevant to dentistry and to the oral cavity and associated structures in health and pathological conditions.

The role of the IDJ is to inform its readers of ideas, opinions, developments and key issues in dentistry - clinical, practical and scientific - stimulating interest, debate and discussion amongst dentists of all disciplines. All papers published in the IDJ are subjected to rigorous peer review.

#### Audiance

Postgraduates, undergraduates, members of the dental team, hospital community, academic & general practitioners.

*Managing Editors  
Iraqi Dental Journal*





## EDITOR-IN-CHIEF

### • Consultant Dr. Abdulwahab Al-Nasiri

*B.D.S., F.F.D.R.C.S.I(Ireland), Consultant Maxillofacial Surgery - President Of Iraqi Dental Association - Lecturer in AL-Rafiden College*

## VICE EDITOR-IN-CHIEF

### • Prof. Dr. Hussain F. Al-Huwaizi

*B.D.S., M.Sc., Ph.D.,(Conservative) Conserv. Depart. College of Dentistry - University of Baghdad*

## EDITORIAL BOARD

### • Prof. Dr. Fakhri Alfatlawi

*B.D.S., M.Sc.,(Ortho.) Ortho. Depart. College of Dentistry-University of Baghdad*

### • Prof. Dr. Hussain F. Al-Huwaizi

*B.D.S., M.Sc., Ph.D.,(Conservative) Conserv. Depart. College of Dentistry - University of Baghdad*

### • Prof. Dr. Khulood A. AlSafi

*B.D.S., M.Sc.,(perio.),Ph.D.( Oral Histo. &Bio.),Perio. Depart. College of Dentistry -University of Baghdad*

### • Prof. Dr. Ali F. AlZubaidy

*B.D.S., F.F.D.R.C.S(Ireland),F.D.S.R.C.S.(England),F.D.S.R.C.P.S.(Glasgoww) ,Maxillo facial Depart. Faculty of Dentistry-Hawler medical university*

### • Prof. Dr. Athraa Y. Al-Hujazi

*B.D.S., M.Sc. Ph.D.( Oral Histo. &Bio.),O. Diagnosis Depart. College of Dentistry-University of Baghdad*

### • Assist. Prof. Dr. Adel Al-Khayat

*B.D.S. F.D.S.R.C.S.(England),M.MED.Sci(Sheffield) ,Head of Iraqi Commission for Medical Specialization in Maxillofacial Surgery O.Surgery & Maxillofacial. Depart. - College of Dentistry - University of Baghdad*

### • Assist. Prof. Dr. Lamya H. Al-Nakib

*B.D.S. M.Sc.(O. & Maxillofacial Radiology.),O. Diagnosis Depart.- College of Dentistry-University of Baghdad*

### • Dr. Mudher A. Abduljabbar

*B.D.S., M.Sc. Prosth. Depart. College of Dentistry - University of Baghdad*

## SCIENTIFIC BOARD

### • Prof. Dr. Khalid Merza -B.D.S.,F.R.C.S., Periodontist-Dean of Dentistry College of Al-Rafidain (past)

### • Prof. Dr. Wael Alalousi - B.D.S.,M.Sc.,(Preventive dentistry)

### • Prof. Dr. Ausama A. Almula - B.D.S.,M.Sc.,Ph.D., Orthodontist. , College of Dentistry-University of Baghdad

### • Prof.Dr. Nidhal H. Ghaib - B.D.S., M.Sc. Ortho.. Depart. College of Dentistry - University of Baghdad

### • Prof.Dr. Wasan Hamdi - B.D.S.,M.Sc.,Ph.D., Oral Pathology. ,College of Dentistry - University of Baghdad

### • Assist. Prof. Dr. Abdalbasit A. Fatihallah - B.D.S, M.Sc., Ph.D in Prosthodontics. , College of Dentistry - University of Baghdad

### • Assist. Prof. Dr. Mohammed Nahidh Mohammed Hassan - B.D.S., M.Sc. Ortho. College of Dentistry - University of Baghdad

### • Consult. Dr. Amer Abdullah Jasim Al-Khazraji - B.D.S.(F.I.C.M.S.), High Diploma Laser in Medicine (H. Dip. L.M.) Consultant Oral & Maxillofacial Surgery, Head of Oral & Maxillofacial Department at Al- Nu'man Hospital.

### • Dr. Muhanad M. AL-Janabi - B.D.S.(F.I.C.M.S.),Maxillofacial Surgery- lecturer- Assist. Dean of Dentistry College - University of Tikrit (formerly) .

### • Dr. Hassanien Ahmed Hadi - B.D.S.(F.A.C.M.S.),Maxillosurgery.Lecturer College of Dentistry - University of Baghdad

### • Dr. Ayad M. Mahmoud - B.D.S, M.Sc.Conservative, Assist.Lecturer - College of Dentistry - University of Baghdad

### • Dr. Ahmed Fadhil Faiq Aljard - B.D.S, M.Sc.Ortho, Assist.Lecturer - College of Dentistry - Al Mustansiriya University

## ADVISORY BOARD

### ORAL DIAGNOSIS & BASIC SCIENCES

### • Prof. Dr.Tahani Abdulaziz Jaffer Al-Sandook - Specialist in Pharma.(Ph.D),Dean of Dentistry College-University of Mousel (past)

### • Prof. Dr. Balkees Taha Garib - B.D.S, M.Sc.,Ph.D.,(Oral Pathology),O.Path.depart.- School of Dentistry, University of Sulaimani

### • Prof. Dr. Hajer Ibrahim Abdulla - B.D.S, M.Sc, C.O.M (USA). Oral Medicine Department, College of Dentistry, Al Mustansiriya University

### • Prof. Dr. Raad Muhi AL-Deen Helmi - M.Sc. OMC (USA) O.Medicine Department, College of Dentistry, Al-Iraqiya University

### • Prof. Dr. Sahar H. Alani - B.D.S.,M.Sc.(O.&Maxillofacial Radiology)Ph.D.,(O.patho.), O. Diagnosis Depart. College of Dentistry University of Baghdad

### • Assist. Prof. Dr. Jamal Noori Ahmed - B.D.S., M.Sc.,Ph.D,O. Medicine, O. Diagnosis Depart. College of Dentistry-University of Baghdad

### • Assist. Prof. Dr. Intisar J. Mohamed - B.D.S. ,M.SC. , PH.D. (Oral Histo & Bio.)Assist. Dean of Dentistry College - Tikrit University

- **Assist. Prof. Dr. Dunia W. Sabeaa Alfayad** - B.D.S., M.Sc.( O.Surg.),Ph.D. (O.patho.)Dean of Dentistry College - University of Al-Anbar
- **Assist. Prof. Dr. Abdullah Ibrahim Hamad** - B.D.S.,M.Sc.,(Medical Physiology),Ph.D.,O.Medicine),O.Diag.Depart. Tikrit University
- **Assist. Prof. Dr.Rajaa Souhail Najim** - Ph.D. Medical Physics , Dean of Dentistry College - University of Tikrit
- **Assist. Prof. Dr.Husam Al-Hamadi** - (M.B.Ch.B, M.Sc. Ph.D) -Director of Pharmacology dep. Pharmacy College -Babylon university
- **Assist. Prof. Dr.Adil Hayder Shabeeb Jaber** - B.D.S.,M.Sc.,(O.Patho.),O.Diag.Depart..College of Dentistry,Al-Mustansirya University

## ORAL & MAXILLOFACIAL SURGERY AND PERIODONTICS

- **Prof. Dr. Abbas A.Y. Taher Al-Aboudy** - B.D.S.,LDSRCPS,M.Sc.MBA,FACOMS,FICS,FCIM,(Maxillofacial),Dean of Dentistry College-University of KUFA
- **Prof. Dr. Abdullatif A.H. Al-juboury** - B.D.S., Ph.D, Periodontology& Immunology, Chairman of Dentistry Department , University College of Humanities / AL-Najaf
- **Assist. Prof. Dr. Ali Al-Shawi** - B.D.S,FDSRCS,FFDRCSI,(Maxillo facial)- Dean of Dentistry College-University of Basrah
- **Assist. Prof. Dr. Saif Sehaam Saliem** - B.D.S., M.Sc. (perio.) Assist. Dean of Dentistry College - University of Baghdad
- **Dr. Faez Al-Hamadany** - B.D.S., M.Sc., Ph.D, Lecturer O. & Maxillofacial Surgery Depart. College of Dentistry-University of al-Mustansirya
- **Dr. Faraedon M. Zardaw** - B.D.S., M.Sc., Ph.D,Perio ,lecturer Perio. Depart. College of Dentistry-University of Al-Sulaimaniya
- **Dr. Alaa E. Ali** - B.D.S., M.Sc. Perio. Depart. College of Dentistry - University of Baghdad
- **Dr. Raed Aziz Badea** - B.D.S., M.Sc. (Perio.) College of Dentistry - University of al-Mustansirya

## PROSTHODONTICS

- **Prof. Dr. Abbas F. Al-huwazi** - B.D.S., M.Sc., Ph.D. Prosth, Former Dean of College of Dentistry-University of Kufa
- **Assist. Prof. Dr. Intesar Jamel Ismail** - B.D.S.,M.SC.,Ph.D. Prosth. Depart. College of Dentistry - University of Baghdad
- **Assist. Prof. Dr. Thekra Ismael Hamad** - B.D.S., M.Sc., Ph.D. Prosth. Depart. College of Dentistry - University of Baghdad
- **Dr. Suad Al-Nakash** - B.D.S., , M.MED.Sci(Sheffield) Lecturer- Prosth. Depart. College of Dentistry - University of Al-Mustansirya.

## PEDO,PREVENTION& ORTHO:

- **Prof. Dr. Zainab A. Al-Dahan** - B.D.S., M.Sc., Pedo &Preventive Dentistry Depart. College of Dentistry - University of Baghdad
- **Prof. Dr. Sulafa K. El-Samarrai** - B.D.S., M.Sc., Ph.D., Pedo &Preventive Dentistry Depart. College of Dentistry - University of Baghdad
- **Prof. Dr. Athraa M. Alwahab** - B.D.S., M.Sc., Pedo &Preventive Dentistry Depart. College of Dentistry - University of Baghdad
- **Prof. Dr. Nidhal A. ALI** - B.D.S., M.Sc.Preventive dentistry depart. College of Dentistry - University of Al-Mustansirya.
- **Prof. Dr. Akram Faisal Al-Huwaizi** - B.D.S,M.Sc. (Orthodontics) - Ph.D. (Preventive Dentistry), Orthod. Dep.,College of Dentistry - Baghdad University
- **Assist. Prof. Dr. Hadeel Ali Hussein** - B.D.S.,M.Sc.,(Ortho.), Orthodontics department-College of Dentistry - University of Baghdad
- **Assist. Prof. Dr. Mohammad Rafid Abdulameer Ali** - B.D.S.,M.Sc.,Ortho. Depart. College of Dentistry - University of Al-Mustansirya.
- **Assist. Prof. Dr. Wisam Wahab Al-Hamadi** - B.D.S.,M.Sc. Ortho. Depart. College of Dentistry - University of Babylon
- **Dr. Zaid Saadi Hasan** - B.D.S,M.Sc.,Ph.D.(Preventive dent.)Prevent.Dentist.Depart.College of Dentistry-University of Baghdad
- **Dr. Ammar Salim kadhum** - B.D.S,M.Sc.,(Ortho.)lecturer;Ortho.depart. College of Dentistry-University of Baghdad
- **Dr. Maha Jamal Abbas** - B.D.S.M.Sc.(Preventive dent.) , Lecturer in Collage of Dentistry - Al Mustansyria University

## CONSERVATIVE AND COSMETIC DENTISTRY

- **Prof.Dr. Jamal Aziz Mehdi** - B.D.S., M.Sc. Conserv.. Depart. College of Dentistry - University of Al-Mustansirya
- **Assist.Prof. Dr. Enas Alrawi** - B.D.S., M.Sc., Conserv. Depart. College of Dentistry-University of Baghdad
- **Assist.Prof. Dr.Abdulla Alshamma** - B.D.S., M.Sc.,Ph.D.(Conservative Depart. College of Dentistry - University of Baghdad
- **Assist. Prof.Dr. Manhal Abdul-Rahman Majeed** - B.D.S., M.Sc.,Ph.D.(Conservative depart. College of dentistry - University of Baghdad
- **Assist. Prof, Dr. Hikmet Abdul-Rahim Al-Gharrawi** - B.D.S.,M.Sc.(Cons.),Dean of College of Dentistry- Al-Mustansirya University.
- **Assist. Prof. Dr. Iman M. AL-Zaka** - B.D.S,M.Sc.,(Cons.)Conserv.. Depart. College of Dentistry - University of Al-Mustansirya

## INTRNATIONAL SCIENTIFIC BOARD

- **Prof. Marco Esposito /Italy** B.D.S, Ph.D Italy. -Professor, Gutenberg University, Sweden Editor-in-Chief, European Journal of Implantology (EJOI) - Editor at Cochran Library
- **Prof. Dr. Natheer H. Al-Rawi/UAE** B.D.S,M.Sc, Ph.D Oral Pathology.

- **Prof. Dr. Nagham M. Majeed Al-Shammery/France** B.D.S.,M.Sc. (conservative)
- **Assist. Prof. Elie Azar Maalouf / Lebanon** DDS, DSO Lebanon, President of the Lebanese Dental Association - Chairman of the Department of Periodontology at the Lebanese University
- **Assist. Prof. Dr. Abdulrahman M. Salih/UAE** B.D.S., M.Sc.,Ph.D.(Restorative Depart. College of Dentistry – Ajman University of Science and Technology.
- **Clinical Assist. Prof. Dr.Latifa R Bairam/USA** D.D.S , M.S , Department of Restorative Dentistry - School of Dental Medicine - Buffalo -NY - USA
- **Assist. Prof. Dr. Asmaa T. Uthman/UAE** B.D.S, MSc Oral & Maxillofacial Radiology.UAE
- **Dr. Hassan Maghaireh/ UK** B.D.S, M.Sc, MFDS, RCS Edinburgh .UK. Clinical Lecturer, University of Manchester - Editorial Director, Smile Dental Journal
- **Dr. Walid Nehme/Lebanon** DDS, MS Lebanon - President Lebanese Society of Endodontology - President of Arab Endodontic Society
- **Dr. Mohammed Sultan Al-Darwish/ Qatar** BDS, MSc(Oral Sciences), Ph.D( Public Health), GBOI(Oral Implantology), DICOI(Oral Implantologists), FICD(Oral Implantologists).Consultant Prosthodontics Dental Department. Hamad Medical Corporation. Qatar

## EDITORIAL ASSISTANTS

- **Dr. Alan H. Mawlood AlQassab** - B.D.S, H.D.D (Ortho), M.Sc (O.& Maxillofacial surg.), MOMSRCPS (Glasgow) O.& Maxillofacial Surg.Department. Faculty of Dntistry -Hawler Medical University
- **Assist. Prof. Dr.Mohammed Nahidh M. Hassan** -B.D.S., M.Sc.,Orth. Depart. College of Dentistry - University of Baghdad
- **Dr. Saeed Khalil Al emamy.** - B.D.S. Iraqi Ministry of Health
- **Dr. Ali Ihsan Hadi** - B.D.S

## MEDIA &PUBLIC RELATIONS MANAGERS

- **Dr. Abo Baker Ziad Alrawi** - B.D.S. Iraqi Ministry of Health
- **Dr. Ahmed Salih** - B.D.S. Iraqi Ministry of Health
- **Dr. Bashar H. Hasan** - B.D.S. Iraqi Ministry of Health
- **Dr. Ali Ihsan Hadi** - B.D.S

## GRAPHIC DESIGNERS

- **Dr. Ali Ihsan Hadi** - B.D.S
- **Solange sfeir**
- **Mohammed Aqeel**

# Table of content

» The effect of oral health educational pictures and video on periodontal health and behavior of school children-----	63
<i>Athraa A. Mahmood</i>	
» A comparative Study of Immunohistochemical Expression of Tumor Necrosis Factor-Alpha, Interleukin-6 and Vascular Endothelial Growth Factor in Giant Cell Granuloma of the Jaws and Giant Cell Tumor of Long Bones -----	70
<i>Saba F. Naji , Wasan H. Younis , Bashar H. Abdullah</i>	
» Salivary a-Amylase and Albumin Levels In Patients with Chronic Periodontitis and Poorly or Well Controlled Type 2 Diabetes Mellitus. -----	80
<i>Maha Abdul Aziz Ahmed</i>	
» Effect of Oil Paint Addition on Micro Hardness of Acrylic Ocular Prosthesis -----	87
<i>Firas Abd Kati , Arshad F. Jassem Al-Kaabi</i>	
» Pacifier Sucking Habit and its Relation to Oral Health of Children Aged 1-5 Years (comparative study) -----	90
<i>Aseel Haidar Al-Assadi , Zainab A.A. Al-Dahan , Abdul Khaliq Al- Rammahy</i>	
» Histological and Immunohistochemical Evaluation of Local Exogenous Application of the Green Tea on Bone Healing (Experimental Study On Rats) -----	95
<i>Enas Fadhil</i>	
» Cyclic Fatigue Resistance of New Endodontic Files in Reciprocal vs. Rotational Motion. -----	102
<i>Makdad Chakmakchi , Ashraf Salim Alchalabi</i>	
» Comparing Maximum Stresses and Displacements in A Lower Complete Denture with Different Occlusal Plane Levels and Schemes . A Three Dimensional Finite Element Stress Analysis Study -----	107
<i>Abdalbasit Ahmed Fatiallah , Suza A. Faraj</i>	
» Evaluation of the Involvement of the Temporomandibular Joint in Patients with Psoriasis using Computed Tomography for Detection of Psoriatic Arthritis Changes. -----	113
<i>Hajer Ibrahim , Muntaha fawzi salih</i>	
» Effect of Systemic Administration of Simvastatin on Dental Implant Stability: A Random Clinical Study -----	119
<i>Ali Mohammed Hassan , Adil Al Kayat,</i>	



# *The effect of oral health educational pictures and video on periodontal health and behavior of school children*

*Athraa A. Mahmood*

*BDS, MSc, Assistant Lecturer, Department of Periodontal Dentistry, College of Dentistry, University of Al-Mustansiriyah.*

## **ABSTRACT**

**Background:** Oral health education for students is a fundamental role for maintaining and raising perfect oral health and preventing oral diseases in future. This study was performed to evaluate the effect of oral health educational pictures and video on periodontal health and behavior in dentition of 12-years old school children in Baghdad city.

**Materials and methods:** Oral examination was conducted on (128) children of (Al-Yamama) primary school in Baghdad city. They were divided into four equal groups, each one consisting of (32) children. The first oral examination was done in first visit to children (pre-test). The community periodontal index (CPITN) scores were used to evaluate periodontal health status of children. Then, the first and second groups, experimental groups, were received instruction about oral health enhancement with educational program (pictures and video clip) regularly for one month. The third and fourth groups, control groups, were received instruction about oral health enhancement only regularly for one month. The second oral examination was done after one month to children (post-test). Also, questionnaire were given to assess oral health behaviors of children in pre- and post- tests.

**Results:** Clinical examination display that CPITN of experimental groups (1, 2) was lower with healthier gingiva when compared with control groups (3, 4) with statistical significant differences between pre and post-tests ( $P$ -value  $<0.005$ ). The percentages of using tooth brush and paste, frequency of teeth cleaning at day, time taking for cleaning teeth, preferable time of cleaning, brushing technique, tooth brush replacement, cause of tooth brushing and using of dental floss were improved in post-test in all groups in comparison with pre-test especially in the experimental groups (1, 2).

**Conclusion:** Results of this study clearly reflect the proposed positive effect of the educational program in the lowering CPITN indices scores among the participants in the experimental group. Results of this study also provide another clue that combination of instruction about oral health enhancement with educational program (pictures and video clip) that represent new strategies for community oral health education; which could be targeted in order to enhance the effect of the oral health education campaigns directed to primary school children.

## **KEYWORDS**

Oral pictures, video, gingivitis, CPITN, behavior.

## **INTRODUCTION**

Dental health educational programs were developed particularly for use in schools and clinical settings. These programs utilized a range of educational methods and materials designed to enhance oral health awareness and knowledge for adoption of a non-disease lifestyle and desirable behaviors <sup>(1-3)</sup>. Bad oral health may have a deep influence on general health of the body and many oral diseases are related to persistent diseases <sup>(4, 5)</sup>.

Oral health education affects the individual's oral health learning that is mandatory for optimal oral health. Oral health literacy emphasizes an availability of skills to understand and use information for suitable oral health decisions <sup>(6)</sup>. The School can supply an encouraging circumference and an ideal setting for enhancement oral health <sup>(7, 8)</sup>.

Healthy periodontium plays an important role in the total oral health of the body, especially in childhood, the age when the periodontal disease (PD) may begin. During childhood a variety of biological changes take place, some of them may be considered as predisposing factors to the occurrence of the gingival and periodontal diseases (PD) as tooth eruption <sup>(9,10)</sup>.

Therefore, oral educational program for those students is a fundamental part for elevating, stabilization and maintaining ideal oral health and preventing dental and PD <sup>(11, 12)</sup>.

The most efficient way to prevent oral diseases is to control them in babyhood. It is especially important to take utility of the school health program, in this program it is possible to arrive maximum number of children at an age can profound a valid oral behavior. The most effective education happen in a face to face conditions where there is open exchange of information and where greatest personal sharing in decision making occurs. Actual communication between teacher and student is important in the educational process. However it's not practical to learn each student separately since it becomes expensive and time loss. Therefore, instructions could be performed to educate a large number of students at one time. It could be done by a hygienist, dentist, a teacher or even by student from upper level <sup>(13, 14)</sup>.

The aims of the study was to show the effectiveness of oral health educational program (pictures and video) on behavior and periodontal health status in dentition of 12-years old school children in Baghdad city.

## MATERIALS AND METHODS

Permission was taken from appropriate authorities to conduct this study at (Al-Yamama primary school) in Baghdad city during the period from November 2015 to January 2016 on (128) children. All of them were healthy and of comparable age (11-12 years old). They were chosen on a random basis and divided into 4 equal groups; each group consisting of 32 children; groups (1,2) were called experimental groups while groups (3,4) were called control groups as shown in Table (1).

Two clinical examinations had been carried out to the all children during the present study; the first one carried out before the program conduction (pre-test) and the second one was done one month after (post-test). Experimental groups (1,2) were received instruction about oral health enhancement with oral educational program (pictures and video clip) regularly for one month, while the control groups (3,4) were received instruction about oral health enhancement only regularly for one month.

Clinical oral examination happen through school hours in class rooms under normal light. A dental mirror and a WHO periodontal probe were used. The community periodontal index (CPITN) scores were used (0=healthy gums without bleeding, 1=with gingival bleeding, 2=with calculus). The presence or absence of gingival bleeding and dental plaque was recorded, also the periodontal status was recorded separately for each six index teeth (16, 11, 26, 36, 31, and 46). Missing teeth, retained roots, gingival swelling due to carious lesion were excluded from this study <sup>(15)</sup>.

The questionnaire form was prepared and translate to Arabic language which were filled by children in first visit (pre-test) and after one month (post-test). This questionnaire was used as a mean of data collection to detect the effect of educational programs in student's practices and knowledge about information source of oral heathy which include the following:

- Cleaning teeth means (tooth brush, fingers, I don't use any mean).
- Brushing teeth (tooth paste, salts, others).
- Frequency of teeth brushing at day (one time daily, two times daily, three times daily, infrequent, never).
- Time period of brushing (less than 1 minute, 1-2 minutes, more than 2 minutes).
- Preferable time for tooth brushing (before breakfast, at bed time, after each meals).
- Brushing technique (horizontal, vertical, circular).

- Change of tooth brush (2-3 months, 4-5 months, more than 6 months).
- Reasons of tooth brushing (remove food debris and bacteria, teeth whitening, I don't know).
- Dental floss using (daily, weekly, never).

Then, all groups were given instruction about oral health enhancement. The oral health education program conduction was carried out on experimental groups (1,2) only. This program was based on exposing children in these groups to pictures and video clip (CD) designed for the present study which were based on standing concepts of recommended oral health prevention <sup>(14,16,17)</sup>. Educational contents were the same in both materials (pictures, video) which contain importance of oral health, role of microbial dental plaque, technique and numbers of correct tooth-brushing and tooth flossing, importance of regular dental attention and healthy food. The video clip period lasted 15 minutes which shown in the classroom. Then, video CD was copied and distributed on each child in groups (1,2) and given instructions to show it at least 2 times-weekly for about 1 month in the house.

The statistical analysis of the data was carried out by using (IBM® SPSS® Statistics).

## RESULTS

The sample was consisted of (128) children, the ages of them ranged between (11-12 years old), 50% of patients were males and 50% of patients were females (male to female ratio in this study was 1:1). Furthermore, patients were divided into four equal groups; each group consisting of 32 children; groups (1,2) were called experimental groups while groups (3,4) were called control groups as shown in Table (1).

The mean of CPITN was decreased in all groups (control and experimental) in post-test in comparison to pre-test; where the experimental male group (2) had heathier gingiva than other groups (1,4,3) with means of CPITN scores 0.31 for group (2), 0.37 for group (1), 0.56 for group (4), 0.58 for group (3) as shown in Table (2).

The study showed elevated percentages of score (1) more than score (0) in relation to bleeding on probing in all groups in pre-test, where score 1 were 42.7%, 72.4%, 45.3%, 59.4 % in groups (1,2,3,4) respectively. While, the percentage of scores were changed toward healthy gingiva in post-test in different rates in all groups, where score (0) was elevated in all groups while scores (1,2) were decreased in all groups as shown in Table (3). Also,

the score (0) was higher in experimental groups (1,2) (66.1%, 69.3% subsequently) than control groups (3, 4) (51%, 50% subsequently) in post-test. While scores (1,2) were lesser in experimental groups (1,2) (score 1 was 29.7%, 29.2% and score 2 was 4.2%, 1.6% for experimental groups subsequently) than control groups (3,4) (score 1 was 39.6%, 43.2% and score 2 was 9.4%, 6.8% for control groups subsequently) as shown in Table (3).

For comparisons among groups, ANOVA test was used; the results showed that there were high significant (HS) difference at  $P\text{-value} \leq 0.01$  among and within groups as shown in Table (4).

Least significant difference (LSD) was performed for multiple comparisons among groups in both pre- and post-tests; the results showed that there were HS difference at  $P\text{-value} < 0.01$  between group (1) pre-test and groups (1,2,3,4) post-test, whereas there was non-significant (NS) difference at  $P\text{-value} \geq 0.05$  between group (1) pre-test and groups (2,3,4) pre-test. The group (1) post-test showed HS difference with groups (2,3,4) pre-test while there was NS difference with group (2) and significant (S) difference with groups (3,4) at  $P\text{-value} < 0.05$  post-test. The group (2) pre-test showed HS difference with groups (2,3,4) post-test while there was NS difference with groups (3, 4) pre-test. The group (2) post-test showed HS difference with groups (3,4) pre-test and post-test. The group (3) pre-test showed there was HS difference with groups (3, 4) post-test while there was NS difference with group (4) pre-test. The group (3) post-test showed S difference with group (4) pre-test while there was NS difference with group (4) post-test. The group (4) pre-test showed S difference with group (4) post-test as shown in Table (5).

The using of tooth brush was higher than other means in cleaning teeth of children in post-test) in all groups. The highest rate of tooth brush was 96.9% in group (1) while the lowest rate was 78% in group (4) as shown in Table (6). Also, the using of tooth paste was higher than other materials in cleaning teeth of children in post-test. The highest rate of tooth paste was 96.9% in group (1) while the lowest rate was 78% in groups (3,4) as shown in Table (6).

The frequency of teeth cleaning at day (2-times) was elevated than other in all groups in post-test. The highest rate of 2-times daily was 46.9% in group (1) as shown in Table (6).

The time taken for cleaning teeth was (1-2 minutes) which was higher than other times in all groups in post-test. The highest rate of (1-2 minutes) was 71% in group (1) and lowest rate was 48% in

group (4) as shown in Table (6). Also, the preferable time of cleaning at bed was the best chose by children than other times in all groups in post-test. The highest rate of bed time 56.5% in group (1) while the lowest rate was 51.4% in group (4) as shown in Table (6).

The brushing techniques (vertical and circular) were elevated after educational program (post-test) in all groups, where the highest increase reached to 55% in group (2) for vertical and 30.3% in group (1) for circular, while the least decrease reached to 52% in group (4) for vertical and 8% in group (4) for circular as shown in Table (6).

The tooth brush was replaced every (2-3 months) which was higher than other period of replacement in all groups in post-test. The highest rate was 74.2% in group (1) while the lowest rate was 68% in group (4) as shown in Table (6).

The remove of food debris and bacteria were the most reason for the tooth brushing in children in all groups in post-test. The highest rate was 90.6% in group (1) while the lowest one was 73.5% in group (4) as shown in Table (6).

The using of dental floss (daily and weekly) was elevated after educational program (post-test) in all groups, where the highest increase reached to 25.8% in group (1) for daily using and 53% in group (2) for weekly using, while the least decrease reached to 9.4% in group (3) for daily using and 18.8% in group (4) for weekly using as shown in Table (6).

## DISCUSSION

This study was performed on 128 children aged (11-12) years old which is marked by acceptance of increasing responsibilities by the children including the responsibility for homework and household chores and in addition, the child can begin to assume more responsibility for oral hygiene <sup>(18)</sup>.

The sample was divided into 4 equal groups; each group consisting of 32 children; groups (1,2) were called experimental groups while groups (3,4) were called control groups to determine and compare oral health status and oral hygiene behavior among them by using combination of instruction about oral health enhancement with educational program.

The video has been employed as medium of oral health education to be useful and valuable visual aids. This may be attributed the fact that what's seen is usually better having a lasting impression on target population (children). The video was produced in Arabic language which serves as a culturally appropriate oral health educational tool for children <sup>(19)</sup>.

Improving the oral health status throughout a group directed periodontal health educational programs can be considered as one of the most applied strategies in field of dental and PD prevention, because it can be easily reach a large number of population and can gap the progressively enlarged shortage in the required dental manpower and financial resources especially in the developing countries <sup>(20, 21)</sup>. On the other hand, children have been identified as a special group, which is at great risk of developing dental and PD. These two facts call for directing a special protection and provisions toward this age group <sup>(22)</sup>.

The current study display more healthy gingiva and less mean of CPITN in all groups in post-test in comparison to pre-test, also there was high significant difference between pre- and post-test in all groups, and this study came in agreement with other studies <sup>(23-25)</sup> which supported the role of the educational program in promoting improve oral health in the children over a one month period. The experimental groups (1,2) had healthy gingiva than control groups (3,4) due to the experimental groups received combination of instruction about oral health enhancement with

educational program while control groups received instructions only.

The majority of children in all groups had elevated score (1) (gingival bleeding) than score (0) (healthy gingiva) in pre-test, so that these children were needed motivation only for correction of tooth brushing to decrease the gingival bleeding and return to healthy gingiva. While, the percentage of scores were changed toward healthy gingiva in post-test in different rates in all groups. So that the application of educational program result in effective plaque control and decreases of gingival diseases in future <sup>(26)</sup>.

The changes in the oral hygiene behaviors associated with visual technologies (pictures and video clip) reinforces the impact on learning because what we see and hear has great impact on our behavior <sup>(27,28)</sup>.

The current study established good oral behaviors among children in all groups in post-test in comparison to pre-test especially in experimental groups that will be laid a foundation for further improvements in the children's oral health status in future <sup>(24)</sup>.

**Table (1): distribution of the population sample.**

Groups		Gender	Age	No.
Experimental groups	G1	females	11-12	32
	G2	males	11-12	32
Control groups	G3	females	11-12	32
	G4	males	11-12	32

**Table 2: Descriptive statistical results of CPITN scores for each group.**

Groups		Mean	SE	SD
G1	Pre-test	0.83	0.08	0.47
	Post-test	0.37	0.05	0.31
G2	Pre-test	0.88	0.05	0.32
	Post-test	0.31	0.04	0.24
G3	Pre-test	0.93	0.07	0.39
	Post-test	0.58	0.08	0.45
G4	Pre-test	0.78	0.05	0.29
	Post-test	0.56	0.06	0.35

**Table 3: Percentage of CPITN scores for each groups.**

Groups		Score 0 (%)	Score 1 (%)	Score 2 (%)
G1	Pre-test	37.0	42.7	20.3
	Post-test	66.1	29.7	4.2
G2	Pre-test	19.3	72.4	8.3
	Post-test	69.3	29.2	1.6
G3	Pre-test	29.7	45.3	25.0
	Post-test	51	39.6	9.4
G4	Pre-test	31.3	59.4	9.4
	Post-test	50	43.2	6.8



Table 4: ANOVA test of CPITN.

<i>ANOVA</i>	<i>SS</i>	<i>DF</i>	<i>MS</i>	<i>F-test</i>	<i>P-value</i>	<i>Sig.</i>
Among groups	12.33	7	1.76	13.19	0.000	**
Within groups	33.13	248	0.13			
Total	45.47	255				

$P \geq 0.05$  Non-Significant (NS)       $P < 0.05$  Significant (S)\*       $P < 0.01$  High Significant (HS) \* \*

Table 5: LSD test to compare of CPITN among groups.

<i>Groups</i>		<i>MD</i>	<i>SE</i>	<i>P-value</i>	<i>Sig.</i>
G1 pre-test	G1 post-test	0.45	0.09	0.000	**
	G2 pre-test	-0.05	0.09	0.528	NS
	G2 post-test	0.51	0.09	0.000	**
	G3 pre-test	-0.10	0.09	0.232	NS
	G3 post-test	0.24	0.09	0.007	**
	G4 pre-test	0.04	0.09	0.608	NS
	G4 post-test	0.26	0.09	0.004	**
G1 post-test	G2 pre-test	-0.51	0.09	0.000	**
	G2 post-test	0.05	0.09	0.528	NS
	G3 pre-test	-0.56	0.09	0.000	**
	G3 post-test	-0.20	0.09	0.027	*
	G4 pre-test	-0.40	0.09	0.000	**
	G4 post-test	-0.18	0.09	0.042	*
G2 pre-test	G2 post-test	0.56	0.09	0.000	**
	G3 pre-test	-0.05	0.09	0.573	NS
	G3 post-test	0.30	0.09	0.001	**
	G4 pre-test	0.10	0.09	0.253	NS
	G4 post-test	0.32	0.09	0.000	**
G2 post-test	G3 pre-test	-0.62	0.09	0.000	**
	G3 post-test	-0.26	0.09	0.005	**
	G4 pre-test	-0.46	0.09	0.000	**
	G4 post-test	-0.24	0.09	0.008	**
G3 pre-test	G3 post-test	0.35	0.09	0.000	**
	G4 pre-test	0.15	0.09	0.089	NS
	G4 post-test	0.37	0.09	0.000	**
G3 post-test	G4 pre-test	-0.20	0.09	0.028	*
	G4 post-test	0.01	0.09	0.856	NS
G4 pre-test	G4 post-test	0.21	0.09	0.017	*

$P \geq 0.05$  Non-Significant (NS)       $P < 0.05$  Significant (S)\*       $P < 0.01$  High Significant (HS) \* \*

Table 6: Percentages of child's behavioral in pre- and post-test.

Items		Groups							
		G1		G2		G3		G4	
		Pre-test (%)	Post-test (%)	Pre-test (%)	Post-test (%)	Pre-test (%)	Post-test (%)	Pre-test (%)	Post-test (%)
Cleaning teeth means	Tooth brush	87.5	96.9	43.8	90.6	75	84.4	68.8	78.1
	Fingers	6.3	0	0	0	9.4	6.3	0	0
	I don't use any mean	6.3	3.1	56.3	9.4	15.6	9.4	31.3	21.9
Cleaning teeth material	Tooth paste	65.6	96.9	43.8	87.5	68.8	78.1	68.8	78.1
	Salt	12.5	0	0	0	6.3	6.3	12.5	15.6
	Others	21.9	3.1	56.3	12.5	25	15.6	18.8	6.3
Frequency of cleaning at day	1 time	25	18.8	15.6	25	21.9	28.1	28.1	25
	2 time	18.8	46.9	9.4	31.3	21.9	31.3	18.8	31.3
	3 time	6.3	15.6	6.3	15.6	9.4	15.6	15.6	15.6
	Infrequent	43.8	15.6	12.5	18.8	31.3	15.6	6.3	6.3
	Never	6.3	3.1	56.3	9.4	15.6	9.4	31.3	21.9
Time period of cleaning	< 1 minute	53.3	22.6	35.7	27.9	55.6	34.5	31.8	24
	1-2 minutes	30	71	35.7	51.7	37	48.3	40.9	48
	> 2 minutes	16.7	6.5	28.6	20.7	7.4	17.2	27.3	28
Preferable time of cleaning	Before breakfast	41.7	32.6	47.1	30.8	44.1	33.3	32.1	34.3
	At bed time	52.8	56.5	41.2	56.4	47.1	53.8	50	51.4
	After each meal	5.6	10.9	11.8	12.8	8.8	12.8	17.9	14.3
Brushing techniques	Horizontal	75	24.2	71.4	34.5	74.1	36.7	54.5	20
	Vertical	21.4	45.5	28.6	51.7	25.9	53.3	45.5	72
	Circular	3.6	30.3	0	13.8	0	10	0	8
Change of tooth brush	2-3 months	53.6	74.2	71.4	72.4	62.5	70.4	63.6	68
	4-5 months	10.7	16.1	14.3	20.7	8.3	14.8	27.3	28
	> 6 months	35.7	9.7	14.3	6.9	29.2	14.8	9.1	4
Reason of tooth brushing	Remove food debris and bacteria	62.5	90.6	57.1	89.7	78.1	81.3	58.8	73.5
	Teeth whitening	15.6	6.3	19	5.1	6.3	15.6	17.6	11.8
	I don't know	21.9	3.1	23.8	5.1	15.6	3.1	23.5	14.7
Dental floss using	Daily	3.1	25.8	0	9.4	0	9.4	0	12.5
	Weekly	12.5	29	15.6	53.1	12.5	40.6	18.8	18.8
	Never	84.4	45.2	84.4	37.5	87.5	50	81.3	68.8

## REFERENCES

1. Towner E. The history of dental health education: a case study of Britain. In: Schou L, Blinkhorn A, editors. Oral Health Promotion. Oxford: Oxford University Press, 1993.
2. Murray JJ, Nunn JH, Steele JG. Prevention of oral disease, 4<sup>th</sup> ed. Oxford: Oxford University, 2003; 7-34, 77-95, 123-144, 241-258.
3. Overton DA, Mason J. Community oral health education concept in dental public health. Philadelphia: Lippincott Williams and Wilkins, 2005; 139-157.
4. Bazile A, Bissada NF, Nair R, Siegel B. Periodontal assessment of patients undergoing an-gioplasty for treatment of coronary artery dis-ease. J Periodontol. 2002; 73: 631-636.
5. Ylöstalo PV, Järvelin MR, Laitinen J, Knuuttila ML. Gingivitis, dental caries and tooth loss: risk factors for cardiovascular diseases or indicator of elevated health risks. J Periodontol. 2006; 33: 92-101.
6. Horowitz AM, Kleinman DV. Oral health literacy: The new imperative to better oral health. Dent Clin N Am. 2008; 52: 333-344.
7. Pine CM. Designing school programmes to be effective vehicles for changing oral hygiene behavior. Int Dent J. 2007; 57: 377-381.
8. Jürgensen N, Petersen PE. Promoting oral health of children through schools-Results from a WHO global survey 2012. Community Dental Health. 2013; 30:204-218.
9. Matsson L. Factors influencing the susceptibility to gingivitis during childhood-a review. Int J Paediatr Dent. 1993; 3(3):119-127.
10. Damle SG. Pediatric dentistry, 4th ed. Arya (MEDI) Publishing House-New Delhi, 2012.

11. Rong WS, Bian JY, Wang WJ, Wang JD. Effectiveness of an oral health education and caries prevention program in kindergartens in china. *Community Dent Oral Epidemiol*. 2003; 31(6):412-416.
12. Vonabbergen J, Declerck D, Mwalili S, Martens L. The effectiveness of a 6 year oral health education programme for primary school children. *Community Dent Oral Epidemiol* .2004, Apr; 32(2):143-149.
13. Sarnat H, Arad P, Hanauer D, Shohami E. Communication strategies used during pediatric dental treatment: a pilot study. *Pediatr Dent*. 2001; 23(4):337-342.
14. Petersen PE. The World Oral Health Report: Continuous improvement of oral health in the 21<sup>st</sup> century-the approach of the WHO Global Oral Health Programme. *Community Dent Oral Epidemiol* 2003; 31(Suppl. 1):13-24.
15. WHO. Oral Health Surveys-Basic Methods, 4<sup>th</sup> ed. Geneva: World Health Organization.1997.
16. Daly B, Watt R, Batchelor P, Treasure E. Essential Dental Public Health. Oxford: Oxford University, 2005; 133-152.
17. Chapman A, Copestake SJ, Duncan K. An oral health education programme based on the National Curriculum. *Int J Paediatr Dent*. 2006; 16:40-44.
18. Zimmers, Bizhang M, Seemann R, Barthel CR. Effective of preventive programs on oral hygiene at adults and school children . *Przegl lek*. 2004; 61(8): 880-883.
19. Bankole OO, Ibiyemi O, Oke GA. A dental health education video for Nigerian children in the Yoruba Language. *Afr. J. Biomed. Res*. 2011; 14(1): 77-79.
20. Kay E, locker D. A Systematic review of the effectiveness of health promotion aimed at improving oral health. *Community Dent Health* 1998 Sep; 15(3):132-44.
21. Astrom AN, Okullo I. Validity and reliability of oral impacts on daily performance (OIDP) frequency scale: a cross sectional study of adolescent in Uganda. *BMC Oral Health*. 2003; 3(1):5.
22. Griffiths J, Boyles. Colour guide to holistic oral care: A practical approach. Mosby-yearbook Europe, BPCC, 1993; 42.
23. Biesbrock AR, Walter PA, Bartizek RD. Initial impact of a national dental education program on the oral health and dental knowledge of children. *J Contemp Dent Pract*. 2003; 2(4):1-10.
24. Naaom ER. Effect of preventive periodontal health education on the oral hygiene of primary school children. *MDJ*. 2009; 6(4):402-407.
25. Hussain SR. Effectiveness of dental health education program on periodontal health status of nursery school children in Erbil city. *Zanco J. Med. Sci*. 2012; 16(3):175-179.
26. Liana Bastos, Arthur Belem, Alfredo Carlos. Effectiveness of an Oral Hygiene Program for Brazilian Orphans. *Braz Dent J*. 2002; 13(1): 44-48.
27. Baggio MA, Erdmann AL, Dal Sasso GTM. Cuidado humano e tecnologia na enfermagem contemporânea e complexa. *Texto & contexto enferm*. 2010; 19(2):378-85.
28. Stina APN, Zamarioli CM, Carvalho EC. Effects of an educational video on the oral hygiene of patients with hematologic disorders. *Rev. Eletr. Enf*. .2014; 16(2):304-411.

# *A comparative Study of Immunohistochemical Expression of Tumor Necrosis Factor-Alpha, Interleukin-6 and Vascular Endothelial Growth Factor in Giant Cell Granuloma of the Jaws and Giant Cell Tumor of Long Bones*

**Saba F. Naji**

B.D.S. - Master Student.

**Wasan H. Younis**

Ph.D. - Professor, Department of Oral Diagnosis, College of Dentistry, University of Baghdad

**Bashar H. Abdullah**

Ph.D. - Professor, Department of Oral Diagnosis, College of Dentistry, University of Baghdad

## **ABSTRACT**

**Background:** Central giant cell granuloma (CGCG) and peripheral giant cell granuloma (PGCG) are pathological conditions of the jaws that share the same microscopic features, but differ clinically in terms of their behavior. While the giant cell tumor (GCT) of long bones is a rare benign neoplasm, tend to affect femur and tubular bone, characterized by local aggressiveness, high recurrence rates and metastasis to the lung.

**Objectives:** To evaluate, compare and correlate the expression of TNF- $\alpha$ , IL-6 and VEGF in peripheral and central giant cell granuloma of the jaw and giant cell tumor of long bones.

**Methods:** A total of 60 retrospective formalin- fixed, paraffine-embedded specimens of giant cell lesions of the jaws and long bones, where included in this study. An immunohistochemical staining with TNF- $\alpha$ , IL-6 and VEGF monoclonal antibodies were performed.

**Results:** TNF- $\alpha$ , IL-6 and VEGF were expressed in all lesions. The PGCG compared to the CGCG and GCT showed significantly increased expression of TNF- $\alpha$  and decreased expression of VEGF by the stromal cells. GCT showed increased expression of VEGF by MNGCs and stromal cells. There is a non significant difference between CGCG and GCT regarding the expression of all three cytokines.

**Conclusions:** The present study confirmed the usefulness TNF- $\alpha$ , IL-6 and VEGF in evaluating osteoclastogenesis. The results of this study proved that the biological activity of TNF- $\alpha$ , IL-6 and VEGF was comparable between the central giant cell granuloma and giant cell tumors, supporting the observations that these two lesions are the same entity and have the same biological behavior.

## **KEY WORD**

central giant cell granuloma, Peripheral giant cell granuloma, Giant cell tumor

## **الخلفية**

ورم الخلايا الكبيرة الحبيبي المركزي (CGCG) والمحيطي (PGCG) هما حالتان مرضيتان لهما نفس الخصائص النسيجية ولكنهما يختلفان من ناحية السلوك السريري ويعتبران من امراض الفكين السفلي والعلوي. لكن ورم الخلايا العملاقة للعظام الطويلة (GCT) من الاورام الحميدة النادرة الذي يصيب عظم الفخذ وعظم القصبة. حيث يتميز بالوحشية الموضعية التكرار والانتشار الى الرئة.

**اهداف الدراسة:** ان الغرض من الدراسة هو تقييم ومقارنة السلوك النسيجي المرضي بواسطة اظهار نتائج الدراسة المناعية للانسجة المرضية الكيميائية قد تمت بواسطة (IL-6, TNF- $\alpha$ ) و (VEGF) لكل من الاورام الحبيبية في عظم الفك كانت هدفا لهذه الدراسة.

**المواد وطرائق العمل:** تضمنت العينة الكلية 60 مريضا من اورام الفكين الحبيبية واورام الخلايا الكبيرة للعظام الطويلة وقد ثبتت الشرائح المرضية بالفورمالين وغمرت بشمع البرافين شملت الدراسة استخدام تقنية التصبغ الكيميائي المناعي بواسطة الاجسام المضادة احادية (TNF- $\alpha$ ) و (IL-6) و (VEGF).

**النتائج:** اظهرت الدراسة ان للعوامل السيتوكينية TNF- $\alpha$  و IL-6 و VEGF لها دور مهم في تكوين الخلايا العملاقة المهمة للعظم واثرا في هدم العظم وانهلاله. ان تقييم التعبير المناعي للعامل السيتوكيني TNF- $\alpha$  اظهر فروقا معنوية بين الاورام كما اظهر ورم الخلايا العملاقة الحبيبي المحيطي التعبير المناعي الاكبر لهذا العامل. اما بالنسبة للعامل السيتوكيني IL-6 فلم تكن هناك فروقا معنوية بين اورام العظام الطويلة والاورام الحبيبية في عظم الفك على الرغم من ارتفاع نسبة هذا العامل في ورم الخلايا العملاقة للعظام الطويلة من ناحية اخرى فان تقييم التعبير المناعي للعامل السيتوكيني VEGF فقد اظهر هذا العامل فروقا معنوية بين ورم الخلايا العملاقة للعظام الطويلة حيث سجل الاظهار النسيجي المناعي اعلى معدل له وبين الورم الحبيبي المحيطي والذي سجل اوطأ معدل لهذا العامل كما ان هناك فروقا معنوية بينه وبين الورم الحبيبي المركزي.

**الاستنتاج:** ان نتائج هذه الدراسة اثبتت بان السلوك الحيوي كما تبين من خلال اظهار نتائج العوامل ال سيتوكينية IL-6, TNF- $\alpha$ , VEGF كان متوافقا بين افة الخلايا الكبيرة و ورم الخلايا الكبيرة. وبذلك نقترح بان هاتين الحالتين هما حالة مرضية واحدة مع طيف من السلوك السريري.

## **الكلمات الرئيسية**

الورم الحبيبي المركزي-الورم الحبيبي المحيطي-ورم العظام الطويلة



## INTRODUCTION

Many lesions of the jaws contain giant cells, they include peripheral giant cell granuloma, central giant cell granuloma, aneurysmal bone cyst, brown tumor of hyperparathyroidism and early stage of cherubism<sup>(1)</sup>

Giant cell granuloma which is a benign bone lesion that occurs mainly in the jaws, not tumor but like condition. Giant cell granuloma presented either as central or peripheral giant cell granuloma. They are of unknown origin located more frequently in the mandible than maxilla, occurring in the 2nd and 3rd decades of life. Females are more frequently affected than males<sup>(2,3)</sup>. PGCG is a reactive exophytic lesion occurring on the gingival and the alveolar ridge originating from the periosteum or periodontal membrane<sup>(4)</sup>.

On the other hand giant cell tumor is a low grade locally aggressive neoplasm that develops within the long bone of young adults of 20-40 years of age. It constitutes about 4-5% of all bone tumors and about 18% of all benign bone tumors. It is generally considered a true neoplastic condition with well defined clinical, radiological and histopathological features<sup>(5)</sup>. It apparently arises from the mesenchymal cells of the connective tissue frame work. These cells differentiate into fibroblast-like stromal components and multinucleated cells of osteoclastic type<sup>(6, 7)</sup>.

The histological features of each of these lesions are markedly similar although they vary substantially in their clinical behavior. However, sometimes they switch from relatively indolent growth pattern to become rapidly enlarging and destructive one with recurrence tendency. Controversy still exists whether the CGCG that occurs in the jaws is a true neoplasm and identical to those occurs in the long bones<sup>(3, 8)</sup>.

A diverse array of inflammatory cytokines and chemokines promote the formation of multinucleated osteoclast cells. Osteoclasts are typically present in large numbers in GCT of bone, suggesting that these tumors may contain cell expressing factors that stimulate osteoclast precursor recruitment and differentiation<sup>(9)</sup>. Osteolysis is a common complication of tumors that arise in, or metastasize to, bone. Considerable progress has been made toward an understanding of the mechanisms responsible for physiological osteoclastogenesis.

A large number of growth factors, hormones and cytokines have been identified that can exert direct and indirect stimulatory and antagonistic effects on the development of osteoclasts from hematopoietic precursors.

## MATERIAL AND METHODS

In this study, files of patients with a definite diagnosis of PGCG and CGCG from the Department of Oral and Maxillofacial Pathology, College of Dentistry, University of Baghdad were revised. The cases of GCT from Ghazi – Al-Harriri hospital, the diagnosis in each case having been made on the basis of clinical, radiologic and histologic findings. Formalin- fixed and paraffin-embedded- tissue samples of all the cases were retrieved. All specimens were obtained from surgical excision of the lesions and had been fixed in 10% buffered formalin. The (H&E) stained slides for all cases were reviewed by two histopathologists. The positive control slides were prepared from blocks of patients having tissue known to contain the target antigen against which the primary antibodies used in this study respectively. Positive tissue controls included radicular cyst for TNF- $\alpha$ , cervical squamous cell carcinoma for IL-6 and human kidney for VEGF. For negative controls slides the antibody was omitted. In each section, eight high-power fields were randomly selected, with a 40X magnification restricted to relatively cellular areas containing MNGC, were randomly chosen in each section to obtain the maximum number of high-power fields common to all samples and to allow direct comparisons among them. The MNGC and stromal cells with a clearly defined immunostaining compared with the positive control cells were counted, and the percentage of positively stained cells (PP) of the MNGCs and stromal spindle-shaped cells was assessed in each field by two observers as: 0 (<10% stained cells), 1 ( $\geq 10\%$ ), 2 ( $\geq 25\%$ ), 3 ( $\geq 50\%$ ), and 4 ( $\geq 75\%$ ) this is for TNF- $\alpha$  and IL-6<sup>(10,11)</sup> while for VEGF; 0, no stained cells; 1,  $\geq 25\%$  stained cells; 2, >25% and <50% stained cells; 3, >50% and <75% stained cells; and 4, >75% stained cells<sup>(12)</sup>. Statistical analysis was performed by SPSS version 19 statistical package. Because the data were conformed to abnormal distributions, the non-parametric Mann-hitney was used. The differences were considered as statistically significant at level  $p=0.05$

## RESULTS

All tumors showed similar histological features exhibiting a great number of MNGCs surrounded by cell populations with oval to spindle cell morphology in a loose fibrillar connective tissue stroma with many small blood vessels. TNF- $\alpha$  Fig(1) (A,B,C), Fig (2) (A,B,C). IL-6 Fig (3) (A,B,C) and VEGF Fig(4)(A,B,C) were detected in all cases of PGCGs, CGCGs and GCT. The MNGCs expressed IL-6 and

VEGF in all cases of PGCGs , CGCGs and GCT as cytoplasmic immunostaining while for TNF- $\alpha$  as nuclear and cytoplasmic immunistaining Fig(1) (A,B,C) and cytoplasmic immunostaining as in Fig(2) (A,B,C). There was a statistically highly significant difference between PGCG ,CGCG&GCT considering the expression ofTNF- $\alpha$  by MNGCs & stromal cells as illustrated in Table (1, 2), in contrast to IL-6 which show anon significant difference between CGCG, PGCG & GCT as represented in Table (3). VEGF shows a highly significant difference between CGCG & PGCG &on the other hand between PGCG&GCT by stromal cells as illustrated in Table(4,5). Considering the statistical correlation between the expression of the three markers in CGCG, elucidated that there is a direct moderate relation between the expression

of VEGF in stromal cells( $r=0.50$ ), with significant relation ( $p=0.03$ ) in relation to TNF- $\alpha$  expression as shown in Table(6). Regarding spearman's test correlation in PGCG demonsetrates that the VEGF in stromal cells has an indirect moderate relation with TNF- $\alpha$  ( $r= -0.47$ ), significant relation ( $p=0.04$ ). Similarly, VEGF expression in stromal cells has an inverse moderate relation to IL-6 expression ( $r=-0.59$ ) & ( $p=0.01$ ) as illustrated in Table (7).While in GCT, statistical analysis revealed that, there is a direct significant correlation in expression of TNF- $\alpha$  by stromal cells with MNGCs ( $r=0.52$ ), and ( $p=0.02$ ). On the other hand IL-6 and TNF- $\alpha$  expression by the MNGCs demonstrated a direct significant correlation to ( $r=0.48$ ), and ( $p=0.03$ ) as shown in Table (8).

**Table 1: Comparison for TNF- $\alpha$  expression among studied giant cells lesion types according to cell types**

Mark- er	Cell Type	Lesion Type	Descriptive statistics						Lesion types' comparison		
			N	Mean %	S.D.	S.E.	Min.	Max.	Kruskal Wallis test	P-value	Sig.
TNF $\alpha$	MNGC	C.G.C.G	20	72.2	26.83	6.00	22	100	6.67	0.036	S
		P.G.C.G	20	80.3	17.40	3.89	38	100			
		G.C.T	20	59.5	27.46	6.14	15	99			
	S-cell	C.G.C.G	20	63.05	25.42	5.68	18	98	13.87	0.001	HS
		P.G.C.G	20	84.3	16.86	3.77	45	99			
		G.C.T	20	60.85	22.30	4.99	17	92			

**Table 2: Mann-Whitney U test**

Marker	Cell Type	Lesion type		Mann-Whitney U test	P-value	Sig.
TNF $\alpha$	MNGC	C.G.C.G	P.G.C.G	177.5	0.54	NS
			G.C.T	140	0.104	NS
		P.G.C.G	G.C.T	102.5	0.008	HS
	S-cell	C.G.C.G	P.G.C.G	98.5	0.006	HS
			G.C.T	188.5	0.76	NS
		P.G.C.G	G.C.T	66.5	0.000	HS

Table 3: Descriptive statistics for IL-6 expression among giant cell lesions

Marker	Cell Type	Lesion Type	Descriptive statistics						Lesion types' comparison		
			N	Mean%	S.D.	S.E.	Min.	Max.	Kruskal Wallis test	P-value	Sig.
IL-6	MNGC	C.G.C.G	20	71.8	15.60	3.49	50	100	2.78	0.25	NS
		P.G.C.G	20	68.7	24.57	5.49	10	98			
		G.C.T	20	79.3	14.70	3.29	44	100			
	S-cell	C.G.C.G	20	59.25	17.90	4.00	20	90	1.39	0.49	NS
		P.G.C.G	20	60.55	15.87	3.55	20	90			
		G.C.T	20	65.15	25.68	5.74	15	99			

Table 4: Descriptive statistics for VEGF expression among giant cell lesions

Marker	Cell Type	Lesion Type	Descriptive statistics						Lesion types' comparison		
			N	Mean%	S.D.	S.E.	Min.	Max.	Kruskal Wallis test	P-value	Sig.
VEGF	MNGC	C.G.C.G	20	72.8	23.02	5.15	18	99	0.79	0.68	NS
		P.G.C.G	20	76	20.15	4.51	27	94			
		G.C.T	20	80.1	16.55	3.70	22	94			
	S-cell	C.G.C.G	20	72.05	17.50	3.91	43	99	15.19	0.001	HS
		P.G.C.G	20	49.1	16.11	3.60	27	78			
		G.C.T	20	70.85	18.92	4.23	24	93			

Table 5: Mann-Whitney U test

Marker	Cell Type	Lesion type		Mann-Whitney U test	P-value	Sig.
VEGF	S-cell	C.G.C.G	P.G.C.G	77.5	0.001	HS
			G.C.T	191	0.81	NS
		P.G.C.G	G.C.T	74.5	0.001	HS

**Table 6: Correlation among the markers in central giant cell granuloma.**

Markers	Cells		TNf	IL-6		VEGF	
			S	M	S	M	S
TNF $\alpha$	MNGC	r	0.42	0.16	-0.01	0.19	-0.37
		p	0.07	0.49	0.97	0.43	0.11
	S	r		0.25	0.22	0.50	-0.36
		p		0.28	0.34	0.03 (S)	0.12
IL-6	MNGC	r			0.31	0.07	-0.16
		p			0.19	0.77	0.49
	S	r				0.05	-0.15
		p				0.84	0.53
VEGF	MNGC	r					0.31
		p					0.18

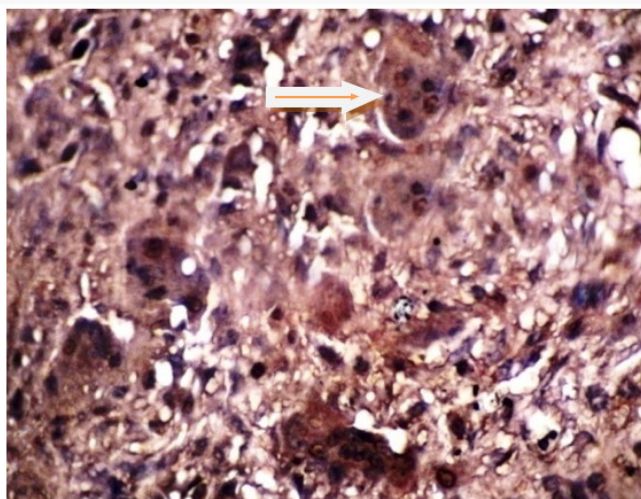
**Table 7: Correlation among the studied markers in peripheral giant cell granuloma**

Markers	Cells		TNS	IL-6		VEGF	
			S	M	S	M	S
TNF $\alpha$	MNGC	R	0.27	0.40	-0.05	0.22	0.10
		P	0.25	0.08	0.84	0.34	0.68
	S	R		0.18	0.29	0.02	-0.47
		p		0.45	0.21	0.94	0.04 (S)
IL-6	MNGC	r			0.18	0.21	-0.10
		p			0.46	0.37	0.67
	S	r				-0.11	-0.59
		p				0.65	0.01 (HS)
VEGF	MNGC	r					0.27
		p					0.26

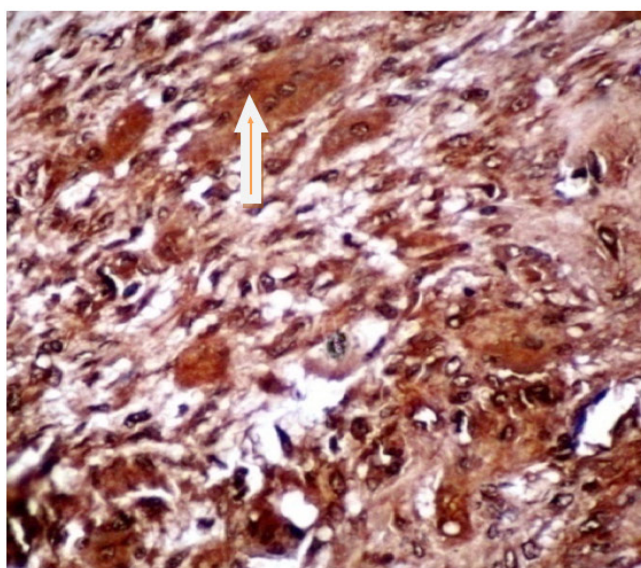
**Table 8: Correlation among the markers in giant cell tumor.**

Markers	Cells		TNf	IL-6		VEGF	
			S	M	S	M	S
TNF $\alpha$	MNGC	R	0.52	0.48	0.20	-0.02	0.00
		P	0.02 (S)	0.03 (S)	0.41	0.93	0.98
	S	R		0.38	0.30	-0.04	-0.13
		P		0.10	0.20	0.85	0.59
IL-6	MNGC	R			0.43	0.13	-0.22
		P			0.06	0.58	0.36
	S	R				-0.34	-0.03
		P				0.14	0.89
VEGF	MNGC	R					0.39
		P					0.09

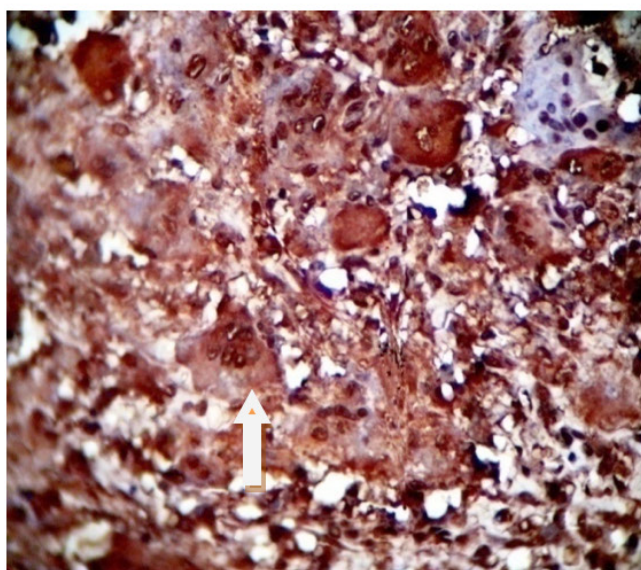




(A)

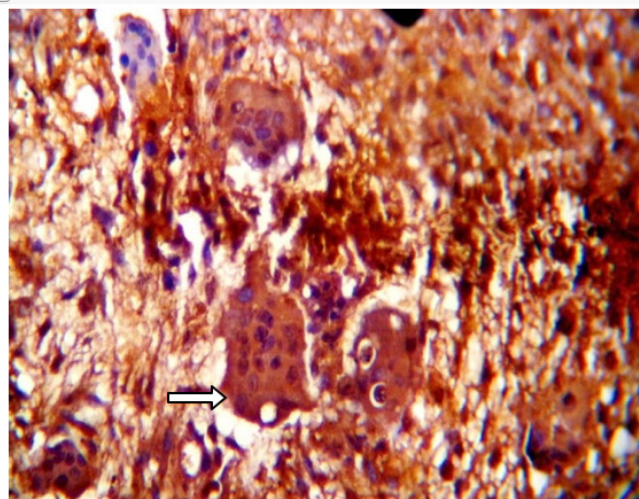


(B)

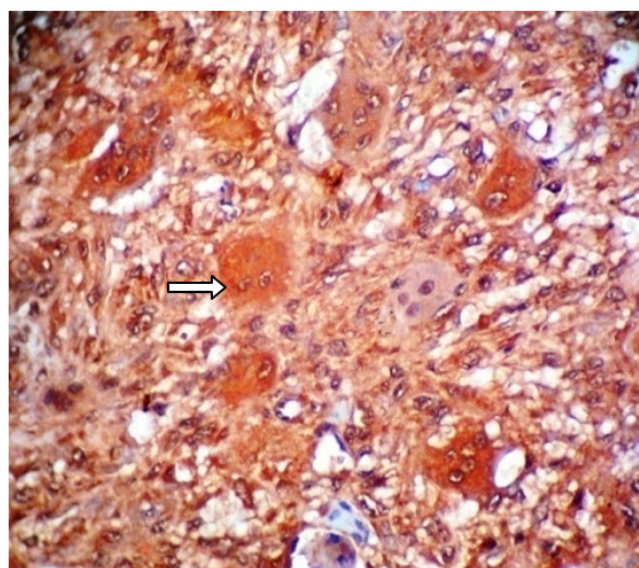


(C)

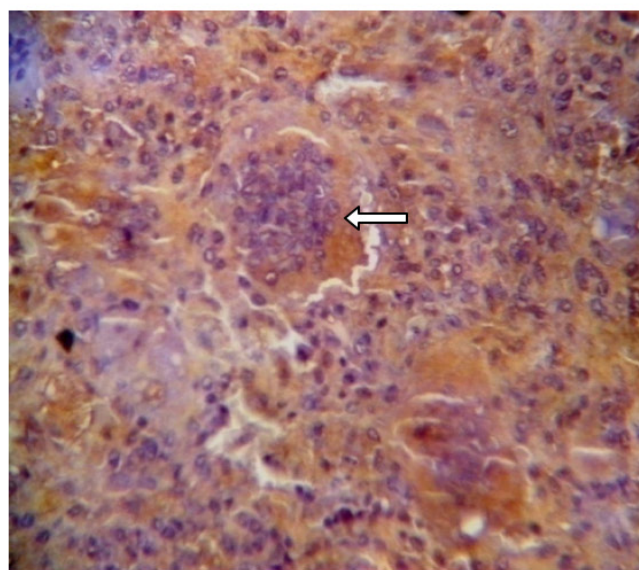
Figure 1: positive nuclear and cytoplasmic expression of TNF- $\alpha$  (A) Central giant cell granuloma 400X, (B) Peripheral giant cell granuloma 400X, (C) Giant cell tumor 400X



(A)



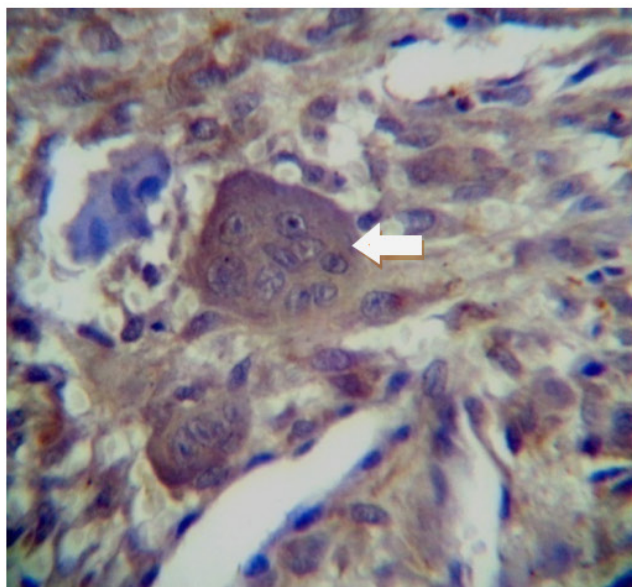
(B)



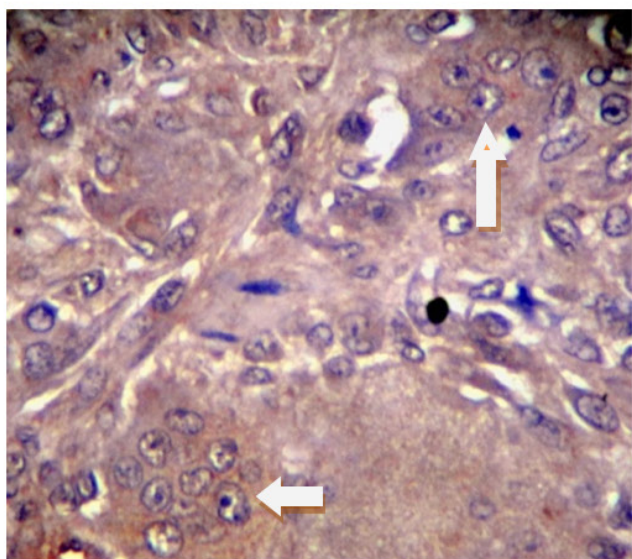
(C)

Figure 2: cytoplasmic expression of TNF- $\alpha$  positive (A) Central giant cell granuloma 400X, (B) Peripheral giant cell granuloma 400X, (C) Giant cell tumor 400X

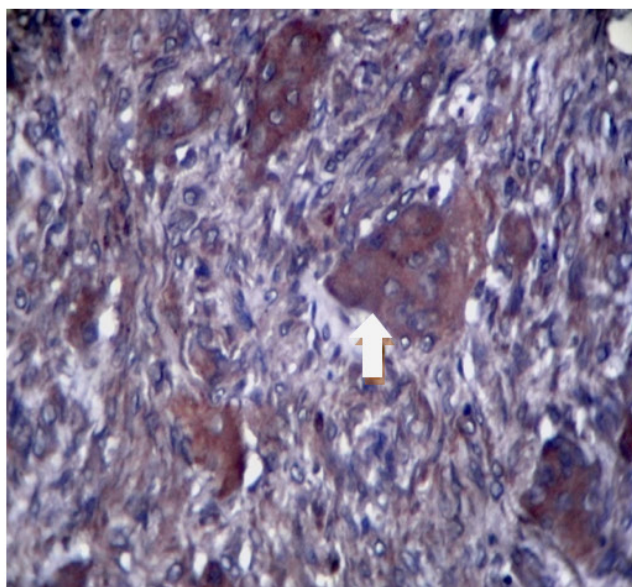




(A)

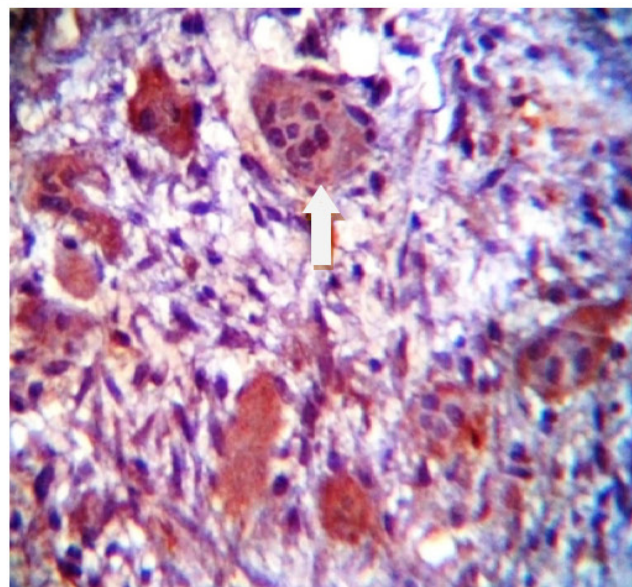


(B)

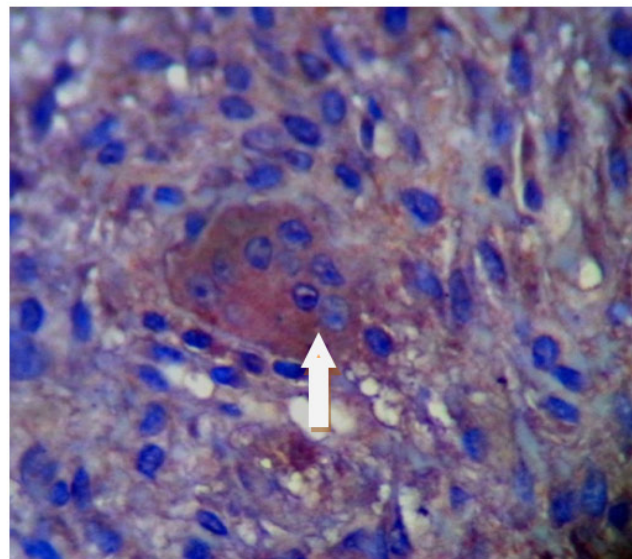


(C)

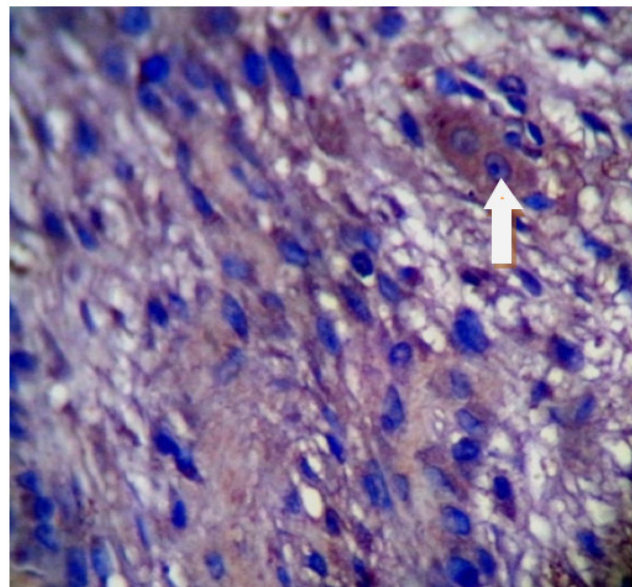
**Figure 3: Positive cytoplasmic expression of IL-6 (A) Central giant cell granuloma 400X, (B) Peripheral giant cell granuloma 400X, (C) Giant cell tumor 400X**



(A)



(B)



(C)

**Figure 4: Positive cytoplasmic expression of VEGF (A) Central giant cell granuloma 400X, (B) Peripheral giant cell granuloma 400X, (C) Giant cell tumor 400X**



## DISCUSSION

Some argument is still found whether the GCGs of the jaws and the GCTs of long bones are really a single pathologic process is an unanswered question.. TNF- $\alpha$  mostly expressed as cytoplasmic, this in agreement with <sup>(11)</sup>. Nuclear and cytoplasmic expression of TNF- $\alpha$  in the present study coincide with <sup>(13)</sup>, who showed increased transcription of nuclear factor of activated T cells (NAFT), "a master of transcription in terminal differentiation of osteoclasts" in the nucleus of MNGCs under the influence of stimuli triggered by TNF- $\alpha$ . In order to compare between the CGCG, PGCG and GCT considering the expression of TNF- $\alpha$  cytokine, the present study results found that, there is a highly significant difference between PGCG and GCT, also between PGCG and CGCG. This finding is confirmed by <sup>(14)</sup>, who demonstrate that the PGCG is the most lesion express TNF- $\alpha$ . This high expression of TNF- $\alpha$  is coherent with the reactive nature of these lesions, in which local irritating factors that trigger an inflammatory response promote a greater release of cytokines such as TNF- $\alpha$ , which may contribute more to angiogenesis rather than bone resorption <sup>(15)</sup>.

On the other hand, there is a non significant difference between CGCG and GCT regarding the expression of TNF- $\alpha$ , this is supported by previous Iraqi study<sup>(16)</sup> who showed that there is a non significant difference between CGCG and GCT. The comparison among these three lesions (CGCG, PGCG & GCT), revealed a none significant differences regarding the expression of IL-6 by MNGCs, this is supported by <sup>(11)</sup>. This result indicate that this cytokine play a critical role in growth process of these lesions <sup>(11,17)</sup> and IL-6 plays a critical role in MNGCs formation and regulation of bone resorption by the same mechanism <sup>(18)</sup>. In order to compare between these lesions, the present results showed that there is a highly significant difference between CGCG and PGCG in VEGF expression by stromal cells, this coincide with <sup>(19,14)</sup>, suggesting that high levels of VEGF-producing cells in a CGCG would be related to a more aggressive biological behavior. . On the other hand, PGCG may provoke the resorption of underlying cortical bone <sup>(20)</sup>, the intensity of this process is significantly lower when compared to that observed in CGCG. Several studies investigating the phenotype of MNGC in CGCL and PGCL have reported consistent immunoreactivity to the anti-CD68 antibody, suggesting that these cells belong to the macrophage lineage <sup>(21)</sup>. Also there is a highly significant difference between PGCG and GCT, this is supported by <sup>(19)</sup>. The elevated level of

VEGF may therefore co-relate with the extent of osteolytic destruction of the lesion, regardless of its primary pathology <sup>(22)</sup>.

On the other hand the non significant relation between CGCG and GCT revealed immunohistochemical similarities between CGCG of the jaws and GCT of long bones, supporting the observation that sometimes these lesions are indistinguishable <sup>(21)</sup>, as well as VEGF expression may provide some prognostic indication of biologically aggressive behavior and local disease recurrence in any osteolytic lesion affecting bone. There was a direct correlation between VEGF & TNF- $\alpha$ , in this study demonstrated in the stromal cells. TNF- $\alpha$  signaling appears to coordinate the expression of specific regulators of endothelial cell survival and metalloproteolytic enzymes <sup>(23)</sup>. Furthermore, MMP-9 mediates the release of extracellular matrix-bound VEGF, increasing vascularization <sup>(24)</sup>. A negative correlation between the expression of VEGF & TNF- $\alpha$  was recorded in this study. Within this context, the lower expression of VEGF in PGCL observed in the present study agrees with <sup>(15)</sup> who found lower expression of VEGF in PGCL compared to CGCL. VEGF may therefore co-relate with the extent of osteolytic destruction of the lesion <sup>(25)</sup>. This finding, together with the discrete trend towards higher expression of TNF- $\alpha$  <sup>(26)</sup>. The present study results suggested that there is a direct correlation between the stromal cells & MNGCs in relation to the expression of TNF- $\alpha$ . This is due to the fact that, an established mechanism by which TNF promotes inflammatory bone resorption is activation of osteoblasts and tissue stromal cells to express receptor activator of NF- $\kappa$ B (RANK) ligand (RANKL) <sup>(27,28)</sup>. On the other hand there is a direct relation between the expression of IL-6 and TNF- $\alpha$  in MNGCs. Proinflammatory cytokines, such as TNF- $\alpha$ , induce IL-6 production through the activation of the p38 MAPK which, in turn, enhances the activity of NF- $\kappa$ B <sup>(29)</sup>.

## CONCLUSION

TNF- $\alpha$  expression in multinucleated giant cells and stromal cells of all cases of CGCG, and GCT confirmed its role in osteoclastogenesis. Its highest expression in PGCG, reflected the reactive nature of these lesions to local irritating factors. IL-6 plays a positive regulatory role in osteoclast with the highest expression was in the GCT. This cytokine might be involved in the growth process and osteoclastogenesis of central giant cell granuloma of the jaw bones and GCT of bone, whereas in PGCG, may contribute

mainly in the mechanism of tumor growth, and occasionally of osteolysis. VEGF showed a significantly higher expression in GCT in comparison to giant cell granuloma of jaw bones emphasized the importance of VEGF in osteoclastogenesis in addition angiogenesis. While in PGCG significantly lower VEGF expression leading to less bone destruction and acting as angiogenic stimulator rather than osteoclastogenic

The similarities in immunohistochemical expression between CGCG and GCT of long bones with a non significant difference between them regarding the expression of TNF, IL-6 and VEGF, supporting the observations that these two lesions are the same entity and indistinguishable and have the same biological behavior.

## REFERENCES

1. Neville, B.W ; Damm, D.D : Bouquot, J.E. Oral maxillo facial pathology. WB. Saunders Company. 2005:p550 .
2. Corso E De , M Politi, [...], and G Paludetti Advanced giant cell reparative granuloma of the mandible: radiological features and surgical treatment. *Acta Otorhinolaryngol* 2006(3):168-172.
3. Neville, B.W; Damm,D.D; Allen,C.M;Bouquot,J.E. Bone Pathology. n: Oral and Maxillofaci l Pathology 3rd ed.. WB. Sanders, Philadelphia. 2009 ;14: 507-629
4. Chaparro-Avendaño AV, Berini-Aytés L, Gay-Escoda C. Peripheral giant cell granuloma. A report of five cases and review of the literature. *Med Oral Patol Oral Cir Bucal*. 2005;10:53-7; 48-52
5. McGough RL, Rutledge J, Lewis VO, Lin PP and Yasko AW. Impact severity of local recurrence in giant cell tumor of bone. *Clin Orthop Rel Res* 2005;438:116-122
6. Werner M. Giant cell tumor of bone : morphological, biological and histogenetical aspects. In *Orthop* 2006;30:489
7. Lanza A., Luigi Laino, [...], and Nicola Cirillo Clinical Practice: Giant Cell Tumour of the Jaw Mimicking Bone Malignancy on Three-Dimensional Computed Tomography (3D CT) Reconstruction. *Open Dent J*. 2008;2:174-178.
8. Bilodeau E, Khalid Chowdhury and Bobby Collins A Case of Recurrent Multifocal Central Giant Cell Granulomas. *Head&neck pathol*. 2009;3(2):174-178
9. Sophie R, Larbi A, Geri M, Anne, G-M, Edwin, Xavie M.. RANK (receptor activator nuclear factor kappa B) and RANK ligand are expressed in giant cell tumor of bone. *Am J Clin Pathol*. 2002;117(2): 210-17
10. Tobón SI, Franco-González LM, Isaza-Guzmán DM, Floréz-Moreno GA, Bravo-Vásquez T, Castañeda-Peláez DA, et al. Immunohistochemical expression of RANK, GR alpha and CTR in central giant cell granuloma of the jaws. *Oral Oncol*. 2005;41:480-8.
11. Papanicolaou P, Chrysomali E, Stylogianni E, Donta C, Vlachodimitropoulos D Increased TNF- $\alpha$ , IL-6 and decreased IL-1 $\beta$  immunohistochemical expression by the stromal spindle-shaped cells in the central giant cell granuloma of the jaws *Med Oral Patol Oral Cir Bucal*. 2012;17 (1): 56-62
12. Floréz-Moreno GA, Henao-Ruiz M, Santa-Saenz DM, Castañeda-Peláez DA, Tobón SI. Cytomorphometric and immunohistochemical comparison between central and peripheral giant cell lesions of the jaws. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2008; 105: 625-32.
13. Amaral FR, Brito JA, Perdigão PF, et al. NFATc1 and TNF alpha expression in giant cell lesions of the jaws. *J Oral Pathol Med* 2010; 39: 269-74.
14. De Matos FR, De Moraes M, Nonaka CF, De Souza LB, Freitas RA.. Immunoreexpression of TNF- $\alpha$  and TGF- $\beta$  in central and peripheral giant cell lesions of the jaws *J Oral Pathol Med* 2012; 41:194-199
15. Matos FR, Nonaka CF, Miguel MC, Galvão HC, Souza LB, Freitas RA. Immunoreexpression of MMP-9, VEGF, and vWF in central and peripheral giant cell lesions of the jaws. *J Oral Pathol Med* 2011; 40: 338-44.
16. Abdul Khafoor S. Histopathological and Immunohistochemical expression of (RANK), (TNF) & (ALP) markers in giant cell tumor of bone and central giant cell granuloma of the jaw. A master thesis, Oral Medicine, Department of Oral Diagnosis, University of Baghdad 2008.
17. Gamberi G, Benassi MS, Ragazzini P, Pazzaglia L, Ponticelli F, Ferrari C, et al. Proteases and interleukin-6 gene analysis in 92 giant cell tumors of bone. *Ann Oncol* 2004;15:498-503.
18. Dai JC, He P, Chen X, Greenfield EM. TNF alpha and PTH utilize distinct mechanisms to induce IL-6 and RANKL expression with markedly different kinetics. *Bone* 2006;38:509-20.
19. Vered M, Buchner A, Dayan D. Giant cell granuloma of the jaw bones: a proliferative vascular lesion? Immunohistochemical study with vascular endothelial growth factor and basic fibroblastic growth factor. *J Oral Pathol Med*. 2006;35(10):613-9.
20. Souza PE, Mesquita RA, Gomez RS. Evaluation of p53, PCNA, Ki-67, MDM2 and AgNOR in oral peripheral and central giant cell lesions. *Oral Dis*. 2000; 6: 35-9
21. Aragão M do S, Piva MR, Nonaka CF, Freitas R de A, de Souza LB, Pinto LP. Central giant cell granuloma of the jaws and giant cell tumor of long bones: an immunohistochemical comparative study. *J Appl Oral Sci* 2007; 15: 310-6.
22. Kumta SM, Huang L, Cheng YY, Chow LT, Lee KM, Zheng MH. Expression of VEGF and MMP-9 in giant cell tumor of bone and other osteolytic lesions. *Life Sci* 2003; 73: 1427-1436.
23. Lehmann W., C.M. Edgar, K. Wang, T-J. Cho, G.L. Barnes<sup>a</sup>, S. Kakar, D.T. Graves<sup>d</sup>, J.M. Rueger, L.C. Gerstenfeld<sup>a</sup>, T.A. Einhorn<sup>a</sup>. Tumor necrosis factor alpha (TNF- $\alpha$ ) coordinately regulates the expression of specific matrix metalloproteinases (MMPs) and angiogenic factors during fracture healing *Bone* 2005; 36, (2): 300-310
24. Rundhaug JE. Matrix metalloproteinases and angiogenesis. *J Cell Mol Med* 2005; 9: 267-85
25. Engsig MT, Chen QJ, Vu TH, Pedersen AC, Therkidsen B, Lund LR, Henriksen K, Lenhard T, Foged NT, Werb Z & Delaissé JM. Matrix metalloproteinase 9 and vascular endothelial growth factor are essential for osteoclast recruitment into developing long bones. *J Cell Biol* 2000; 151: 879-889
26. Parameswaran N, Patial S. Tumor necrosis factor-alpha signaling in macrophages. *Crit Rev Eukaryot Gene Expr* 2010; 20: 87-103



27. Kobayashi, K, Takahashi, N, JIMI, E, UDAGAWA, N, TAKAMI, M, KOTAKE, S, NAKAGAWA, N et al. Tumor necrosis factor alpha stimulates osteoclast differentiation by a mechanism independent of the ODF/RANKL-RANK interaction. *The Journal of Experimental Medicine* 2000; 191; p. 275-286.
28. Lam, J., S. Takeshita, J.E. Barker, O. Kanagawa, F.P. Ross, and S.L. Teitelbaum. TNF-alpha induces osteoclastogenesis by direct stimulation of macrophages exposed to permissive levels of RANK ligand. *J. Clin. Invest.* 2000; 106:1481–1488
29. Kurokouchi, K, Kambe, F, Yasukawa K, Izumi R, IshiguroN, Iwata, H and Seo, H. TNF-alpha increases expression of IL-6 and ICAM-1 genes through activation of NFkappab in osteoblast-like ROS17/2.8 cells. *Journal of Bone and Mineral Research* 1998; 13: 1290-1299.

# Salivary $\alpha$ -Amylase and Albumin Levels In Patients with Chronic Periodontitis and Poorly or Well Controlled Type 2 Diabetes Mellitus.

Maha Abdul Aziz Ahmed

B.D.S.,M.Sc. (periodontics) - Assistant Professor, Department of Periodontics, - College of Dentistry, University of Baghdad.

## ABSTRACT

**Background:** Recent studies suggest that chronic periodontitis (CP) and type2 diabetes mellitus (T2DM) are bidirectionally associated. Analysis of saliva as a mirror of oral and systemic health could allow identification of a amylase ( $\alpha$ -Am) and albumin (A1) antioxidant system markers to assist in the diagnosis and monitoring of both diseases. The aims of study, compare the clinical periodontal parameters in chronic periodontitis patients with poorly or well controlled T2DM, salivary  $\alpha$ -Am, A1, flow rate (FR) and pH then correlate between biochemical, physical and clinical periodontal parameters of each study and control groups.

**Materials and Methods:** 80 males, with an age range of (35-50) years were divided into four groups, (20 subjects at each): two groups had well or poorly controlled T2DM both of them with chronic periodontitis, group of patients with only chronic periodontitis and control group with healthy periodontium and systemically healthy. From all subjects unstimulated whole salivary samples were collected to measure FR, pH, AI and  $\alpha$ -Am, then clinical periodontal parameters (plaque index, gingival index, bleeding on probing, probing pocket depth and clinical attachment level) were recorded.

**Results:** patients had chronic periodontitis with poorly controlled T2DM demonstrated the highest median values of all clinical periodontal parameters and highest increase in levels of salivary  $\alpha$ -Am and AI with lowest median values of FR and pH, in addition to the highly significant differences among the study and control groups regarding biochemical and physical parameters. Positive correlation were revealed between  $\alpha$ -Am with AI and both of them with all clinical periodontal parameters but, they were negative with FR and pH.

**Conclusion:** patients with poor glycemic control had more severe periodontal tissue break down with decrease in FR and pH also obvious increase in levels of AI and  $\alpha$ -Am so, these biochemical markers will provide an objective phenotype to allow practitioners for early diagnosis, which is essential for improved prognosis and effective delay of clinical complications associated with chronic periodontitis and DM and an important strategy to lower the incidence of both diseases world wide.

**Keywords:** periodontitis, T2DM, salivary albumin and  $\alpha$ -amylase.

## المستخلص

**الخلفية:** الدراسات اثبتت ان التهاب اللثة المزمن وداء السكري من النوع الثاني مرتبطان. تحليل اللعاب وتحديد مستوى اميليز و البومين للمساعدة في تشخيص الامراض. اهداف الدراسة قياس مؤشرات اللثة السريري في مرضى التهاب اللثة المزمن مع او بدون السكري من النوع الثاني المسيطر عليه او الغير مسيطر عليه و الفا اميليز و البومين اللعابي و حامضيه و معدل سريان اللعاب ثم تحديد العلاقة بينهم

**المواد والطرق:** ٨٠ ذكر بعمر (٣٥-٥٠) سنة تم تقسيمهم لاربعة مجاميع (٢٠ في كل مجموعة) اثنان لديهم التهاب اللثة المزمن مع السكري من النوع الثاني المسيطر او غير مسيطر عليه و مجموعته التهاب اللثة المزمن و المجموعه الضابطة ثم جمع اللعاب منهم لقياس حامضيه و معدل سريان اللعاب و اميليز و البومين اللعابي و مؤشرات اللثة السريري.

**النتائج:** مرضى التهاب اللثة المزمن مع السكري من النوع الثاني الغير مسيطر عليه اظهروا اعلى قيم لمؤشرات اللثة السريري و اعلى زياده في الاميليز و البومين و اقل قيم لحامضيه و معدل سريان اللعاب مع فروقات معنويه عاليه بين المجاميع. هناك علاقات موجبه بين اميليز و الالبومين و كلاهما مع مؤشرات انسجه ماحول الاسنان السريري لكنها سالبه مع حامضيه و معدل سريان اللعاب

**الاستنتاج:** مرضى السكري النوع الثاني يعانون من تدمير اكثر لانسجه اللثة وانخفاض في حامضيه وسريان اللعاب وزياده في اميليز و البومين وهذين الأخيرين يساعدان في التشخيص المبكر لهذين المرضين لتقليل الاصابة بهما.

## INTRODUCTION

Periodontitis is irreversible inflammatory disorder of the supporting structures of the tooth leading to progressive attachment loss and destruction of alveolar bone. Chronic periodontitis (CP) is the most prevalent form of periodontitis, hence affects about 10%-15% of adult population world wide. Furthermore in the presence of systemic disease (e.g.DM), which modify the host response to plaque accumulation, the disease progression may become more aggressive<sup>(1)</sup>. The DM, is a metabolic disorder characterized by hyperglycemia and T2DM which is the most common type is linked to insulin resistance and patients with DM are prone to oral complications such as periodontal disease (PD), dry mouth and abscesses<sup>(2)</sup>. Hence, today various researches are

being conducted to evaluate possible compound in the oral fluids through which it may possible to assess the presence and severity as well as, to identify the patients at risk for these diseases thus, analysis of saliva which is a complex secretory fluid that can be easily collected through non-invasive means for the screening of large samples in addition, saliva contains locally produced microbial and host response mediators, as well as, systemic (serum) markers<sup>(3)</sup>. Thus the investigation of salivary proteins such as AI and  $\alpha$ -Am in patients with CP and DM may be useful to enhance the knowledge of their roles in these diseases. So, this study was designed to determine the effect of glycemic control in T2DM on periodontal health status as well as, on the levels of salivary AI,  $\alpha$ -Am, FR and pH.

## MATERIAL & METHODS

The participants in this study was 80 males with age range (35-50) years, recruited from specialized center for endocrinology and Diabetes in Baghdad and from periodontics Department, at the teaching hospital in the College of Dentistry, University of Baghdad. They were divided into four groups.

1. Study group of 20 males suffer from CP with well controlled T2DM  $HbA1c < 7\%$ <sup>(4)</sup> (CP+wT2DM).
2. Study group of 20 males suffer from CP with poorly controlled T2DM,  $HbA1c > 9\%$ <sup>(4)</sup> (CP+pT2DM).
3. Study group of 20 males suffer from CP but systemically healthy (CP).
4. Control group of 20 males with clinically healthy periodontium and apparently systemically healthy. Healthy periodontium defined by the absence of any signs and symptoms of gingival inflammation, without periodontal pockets or clinical attachment loss. This group represented a base line data for the salivary A1 and  $\alpha$ -Am levels.

Patients with CP demonstrated the presence of at least four sites with PPD ( $\geq 4$ mm) and clinical attachment loss of (1-2) mm or greater<sup>(5)</sup>.

The inclusion criteria were only males with at least 20 teeth present, T2DM  $\geq 5$  years on oral hypoglycemic therapy only and body mass index within the normal range ( $18.5$ - $24.9$  kg/m<sup>2</sup>)<sup>(6)</sup>. The exclusion criteria were females, presence of systemic diseases other than T2DM, patients administered medications (anti-inflammatory and anti-microbial) or undergone periodontal treatment in the 3 months prior to the study, smoking, alcohol consumption, T1DM and T2DM administering insulin, presence of nephropathy, retinopathy and diabetic foot. Unstimulated whole salivary samples were collected from all participants<sup>(7)</sup>. During that salivary (FR) was measured through dividing the volume of the collected sample by the collection time. After this by using DP universal test paper, the salivary pH was measured, then samples were centrifuged for 15min. at 4000 rpm and frozen, at  $-20^{\circ}\text{C}$ . By using Michigan O periodontal probe, the examination of clinical periodontal parameters was performed on four surfaces (mesial, buccal /labial, distal and lingual / palatal) of all teeth except the 3<sup>rd</sup> molar, which included:

1. Plaque index system (PLI)<sup>(8)</sup>.
2. Gingival Index system (GI)<sup>(9)</sup>.
3. Bleeding on probing (BOP)<sup>(11)</sup>.
4. Probing pocket Depth (PPD).
5. Clinical Attachment level (CAL).

For biochemical analysis of salivary A1, Protein U.S / Syrbio kit was used. While for salivary  $\alpha$ -

Am, (single Reagent GALG2-CNP) /SPECTRUM kit was used, hence the activities were determined by measuring the absorbance at 598 nm and 405 nm respectively both by the spectrophotometer. Descriptive statistics that include mean and median values and inferential statistics which include kruskal – Wallis H test, Mann- Whitney U test and pearson correlation (r) were used. The level of significance (S) was accepted at  $P \leq 0.05$ , highly significance (HS) at  $P < 0.01$  and non-significant (NS) at  $P > 0.05$ . We certify that this study involving human subjects is in accordance with the Helsinki declaration of 1975 as revised in 2000 and that it has been approved by the relevant institutional ethical committee.

## RESULTS

The highest mean of age was found in CP + pT2DM group (45.85), followed by CP + wT2DM (44.95), then CP group (41.7) while, the least mean of age was detected in control group (38). Patients with CP + pT2DM demonstrated the highest median values of the clinical periodontal parameters, then patients suffer from CP + wT2DM, after that CP patients. Inter study groups comparisons regarding all clinical periodontal parameters revealed, HS differences between CP + pT2DM with both CP + wT2DM and CP groups while, they were NS differences between CP + wT2DM with CP groups (Table -1).

From (Table -2), the biochemical analysis of both A1 and  $\alpha$  – Am presented that highest increase in median values were revealed in CP + pT2DM group after that patients with CP + wT2DM, then CP group as compared to the control group hence, HS differences were demonstrated among the four groups. On the other hand, the physical parameters analysis showed decrease in median values of both FR and pH in study groups when compared to control group and the lowest median values demonstrated in CP + pT2 DM group. Again, HS differences among the study and control groups were found.

The comparisons between all pairs of the study and control groups regarding  $\alpha$ -Am, A1, FR and pH demonstrated HS and S differences except the NS differences between CP+wT2DM with CP groups concerning  $\alpha$ -Am, A1 and pH (Table -3).

The results of correlations (Table -4&5) between  $\alpha$ -Am and A1 with clinical periodontal parameters were positive but they were negative with FR and pH at all groups, although  $\alpha$ -Am revealed moderate positive correlations with PLI and GI at CP + pT2 DM and CP + wT2DM groups respectively.

The correlations between  $\alpha$ -Am with A1 were

positive at all groups (table -6).

## DISCUSSION

The CP + T2DM patients revealed higher mean of age, this can be explained by the greater incidence of both diseases in adults<sup>(10)</sup>.

In diabetic patients, the vascular changes, neutrophil dysfunction, altered collagen synthesis, accumulation of advanced glycation end products leading to impaired tissue repair capacity<sup>(1)</sup>, as well as increased glucose level in gingival crevicular fluid (GCF) and saliva<sup>(11)</sup>, decrease FR that disrupt the cleaning and buffering capacities and clearance of bacterial substrate which then increase accumulation of plaque and calculus<sup>(12)</sup>, in addition increased levels of  $\alpha$  - Am and AI, in which the former favored proliferation of both aerobic and anerobic bacteria in plaque, while the latter considered potential energy sources and enable the attachment of pathogenic bacteria thus alter the composition of plaque<sup>(13)</sup>. So, diabetics had 3fold increase in risk of having periodontitis compared to non-diabetics, hence adults with an HbA1c level of 9% had significantly higher prevalence of severe periodontitis thus, the gingival inflammation and bleeding are intensified, greater prevalence and extent of pockets with twice as likely a non- diabetics to have attachment loss<sup>(2,10,12)</sup>.

Saliva contains numerous defense antioxidant proteins e.g. AI and  $\alpha$ - Am which able to inhibit the generation of free radicals<sup>(14)</sup>. The highly significant increase in  $\alpha$ -Am level in CP patients as compared to control group revealed by this study are in accordance with other studies<sup>(15-19)</sup>, the same result was found when comparing CP + T2 DM groups with control group, hence different researchers had reported that salivary  $\alpha$ -Am concentrations from T2DM patients were higher<sup>(11,20-22)</sup> or lower<sup>(23-26)</sup> than its levels in non-diabetics. The response of salivary gland to inflammatory diseases, resulting in enhanced synthesis and secretion of defense proteins<sup>(15)</sup>. The increased basement membrane permeability of salivary glands in diabetics leads to increased passage of proteins into the saliva, moreover the sialosis in the parotid gland in T2 diabetics, hence most of  $\alpha$  -Am being synthesized in this gland, could result in variations in the salivary composition<sup>(22)</sup>. Studies showed that  $\alpha$  - Am is a major lipopolysaccharide binding protein of *Agri*, *gatibacter actinomycetem comitans* and *Porphyromonas gingivalis* (*P.gingivalis*) and interfere with bacterial adherence and biofilm formation also performs a direct inhibitory effect on the growth of *Neisseria Gonorrhoea* and *P.gingivalis*

<sup>(13)</sup>. The notable increase in AI level in CP patients in comparison to control subjects in this study was in consistent with findings of previous studies<sup>(27-29)</sup>, while others<sup>(14,30)</sup> demonstrated decrease in AI levels with deterioration of periodontal tissue condition. Although the significant increase of AI in T2 diabetics found by researchers<sup>(31,32)</sup> were in agreement with this study, but disagree with other results<sup>(33,34)</sup>. AI accounting for more than 50% of all plasma proteins, thus is regarded as markers for plasma protein leakage occurring as a consequence of inflammatory process, so the high salivary AI level in CP patients due to ulceration in sulcular epithelium confirming the sulcular origin of AI from GCF, thus 4-5 times rise in AI level was noted during periodontal tissue destruction when compared with that of the control<sup>(28)</sup>, moreover the presence of *Treponema Denticola* seemed to increase AI in periodontitis patients<sup>(35)</sup>. On the other hand, disregulation in the factors that regulate AI synthesis during DM occur which include nutrition, hormonal balance and osmotic pressure and the inflammation of salivary gland causing increased leakage of serum proteins into the saliva<sup>(33)</sup>. Finally, studies measured AI and  $\alpha$  - Am levels in T2 diabetics, they ignore their periodontal health status.

The more acidic pH in CP patients was in line with some studies<sup>(29,36)</sup>, hence significant correlation did exist between pH and PPD on the other hand increase in pH was found<sup>(28,37)</sup> in CP patients. From the present study the decrease in pH of diabetics was coincide with other reports<sup>(12,25,38)</sup>, hence significant decrease in pH was demonstrated when comparing uncontrolled T2DM with healthy and controlled T2DM as well as, healthy with controlled T2DM<sup>(39)</sup>. The decrease in salivary FR and bicarbonate content consequently contributed to the more acidic saliva<sup>(38)</sup>. The higher concentrations of hydrogen ions (from salivary glands or oral microbiota), the lowest the pH, since pH level negatively correlated with proportion of periodontal pathogens, that grow in mildly acidic pH, either utilize or create products that are mild to moderately acidic in nature<sup>(29)</sup>.

The decrease in salivary FR in this study coincide with others concerning CP<sup>(15,19,29)</sup>, and DM<sup>(12, 24,25,40)</sup> but diverge with previous studies about CP<sup>(37)</sup> and DM<sup>(41)</sup> who reported increased of FR, on the other hand some researchers found that FR levels not affected by periodontal health status<sup>(28)</sup> or presence of DM<sup>(42)</sup>. There are multiple causes of salivary hypofunction including inflammation e.g. periodontal disease<sup>(19)</sup>, hydrogen concentration, aging<sup>(40)</sup> or systemic disease e.g. DM<sup>(24)</sup>, so in this case the



decrease in pH, medication given for diabetics, poly urea and dehydration, neuropathies, microvascular changes, metabolic disturbances also, hypertrophy of salivary glands can be attributed to decrease in FR <sup>(12, 25)</sup>.

Positive correlations of  $\alpha$  -Am and A1 with each other and with clinical periodontal parameters, but they were negative with FR and pH, this can be explained by the presence and increased inflammation with periodontal tissue destruction due to CP and DM which lead to increased levels of  $\alpha$ -Am and A1 but decrease in FR and pH. These results were in concurrent with other results <sup>(16,17,19)</sup> who found

significant positive correlations between  $\alpha$ -AM with PPD and CAL, while significant negative correlation with FR<sup>(19)</sup> in CP patients. In general, there were correlations between  $\alpha$  - Am with glycemic control <sup>(23,24,26)</sup>, but non significant with FR at controlled and uncontrolled T2DM<sup>(23)</sup>. Significant positive correlation was detected between A1 levels with GI in T2 diabetes <sup>(43)</sup>.

Finally, the results may differ from one study to another these may be due to e.g. the diversity in selection criteria of samples, metabolic control, wide range of age, different types of saliva, that can limit direct comparison.

**Table (1): Median values of the clinical periodontal parameters and the inter groups comparisons between all pairs of the study groups**

Clinical periodontal parameters	Groups	Median	CP+ pT2DM & CP+ wT2DM		CP+ pT2DM & CP		CP & CP+ wT2DM	
			Mann Whitney U test	P-value Sig.	Mann Whitney U test	P-value Sig.	Mann Whitney U test	P-value Sig.
PLI	CP+ pT2DM	2.682	4.735	0.00 HS	5.411	0.00 HS	1.948	0.51 NS
	CP+ wT2DM	1.815						
	CP	1.341						
	Control	0.232						
GI	CP+ pT2DM	2.553	5.410	0.00 HS	5.42	0.00 HS	0.677	0.499 NS
	CP+ wT2DM	1.556						
	CP	1.5						
	Control	0.108						
BOP score1	CP+ pT2DM	60.5	4.390	0.00 HS	3.993	0.00 HS	1.233	0.217 NS
	CP+ wT2DM	46						
	CP	42						
PPD	CP+ pT2DM	6.67	4.363	0.00 HS	4.255	0.00 HS	0.825	0.409 NS
	CP+ wT2DM	6.13						
	CP	5.945						
CAL	CP+ pT2DM	4.4	4.372	0.00 HS	4.749	0.00 HS	0.989	0.323 NS
	CP+ wT2DM	3.08						
	CP	2.435						

\*P<0.01 High significant

**Table (2): Median values of salivary  $\alpha$ -Amylase , Albumin ,FR and pH and the significance of differences among the study and control groups.**

parameters	CP+ pT2DM	CP+ wT2DM	CP	Control	Kruskal-Wallis H test	
	Median	Median	Median	Median	Chi square	P-value Sig.
$\alpha$ -Amylase U/L	162.14	99.25	90.86	65.47	43.62	0.00 HS
Albumin mg/dl	104.8	79.18	75.72	56.51	30.568	0.00 HS
FR ml/min	0.23	0.725	0.75	1.2	65.6	0.00 HS
pH	5	6	6	7	24.96	0.00 HS



**Table(3): Inter groups comparisons of the median values of salivary  $\alpha$ -Amylase ,Albumin ,FR and pH between all pairs of the study and control groups**

parameters	CP+ pT2DM&CP+ wT2DM		CP+ pT2DM&CP		CP+ pT2DM&- Control		CP+ wT2DM&CP		CP+ wT2DM&- Control		CP& Control	
	Mann Whitney U test	P-value	Mann Whitney U test	P-value	Mann Whitney U test	P-value	Mann Whitney U test	P-value	Mann Whitney U test	P-value	Mann Whitney U test	P-value
$\alpha$ -Amylase U/L	3.354	0.001 S	3.517	0.00 HS	5.410	0.00 HS	0.352	0.725 NS	4.436	0.00 HS	4.003	0.00 HS
Albumin mg/dl	2.998	0.021 S	2.976	0.03 S	4.816	0.00 HS	0.864	0.322 NS	2.332	0.044 S	4.275	0.00 HS
FR ml/min	4.998	0.00 HS	5.437	0.00 HS	5.444	0.00 HS	4.870	0.00 HS	5.278	0.00 HS	4.809	0.00 HS
pH	4.275	0.00 HS	4.925	0.00 HS	5.231	0.00 HS	1.274	0.203 NS	3.213	0.01 S	2.453	0.014 S

**Table (4): Correlations between the levels of  $\alpha$ -Amylase with the clinical parameters of each study and control groups.**

parameters	Statistical analysis	CP+ pT2DM	CP+ wT2DM	CP	Control
PLI	r	0.56	0.188	0.254	0.248
	p	0.816 NS	0.427 NS	0.281 NS	0.292 NS
GI	r	0.164	0.57	0.222	0.290
	p	0.489 NS	0.012 S	0.348 NS	0.214 NS
BOP score1	r	0.207	0.227	0.003	-
	p	0.381 NS	0.330 NS	0.990 NS	-
PPD	r	0.173	0.039	0.199	-
	p	0.466 NS	0.871 NS	0.400 NS	-
CAL	r	0.154	0.201	0.068	-
	p	0.516 NS	0.395 NS	0.775 NS	-
FR	r	-0.268	-0.442	-0.009	-0.156
	p	0.254 NS	0.049 S	0.969 NS	0.511 NS
pH	r	-0.131	-0.035	-0.144	-0.096
	p	0.582 NS	0.884 NS	0.543 NS	0.687 NS

**Table(5): Correlations between the levels of Albumin with the clinical parameters of each study and control groups.**

parameters	Statistical analysis	CP+ pT2DM	CP+ wT2DM	CP	Control
PLI	r	0.148	0.134	0.085	0.131
	p	0.533 NS	0.573 NS	0.721 NS	0.581 NS
GI	r	0.327	0.186	0.070	0.224
	p	0.159 NS	0.434 NS	0.771 NS	0.343 NS
BOP score1	r	0.378	0.157	0.186	-
	p	0.100 NS	0.508 NS	0.434 NS	-
PPD	r	0.121	0.268	0.255	-
	p	0.611 NS	0.253 NS	0.277 NS	-
CAL	r	0.482	0.189	0.107	-
	p	0.032 S	0.424 NS	0.653 NS	-
FR	r	-0.321	-0.214	-0.151	-0.046
	p	0.167 NS	0.365 NS	0.526 NS	0.847 NS
pH	r	-0.197	-0.273	-0.045	-0.235
	p	0.406 NS	0.245 NS	0.849 NS	0.318 NS

**Table (6): Correlations between salivary levels of ( $\alpha$ -Amylase and Albumin of each study and control groups.**

Parameters	Statistical analysis	CP+ pT2DM	CP+ wT2DM	CP	Control
$\alpha$ -amylase and albumin	r	0.291	0.103	0.195	0.511
	p	0.214 NS	0.665 NS	0.411 NS	0.831 NS

## REFERENCES

- Michael G Newman, Henry H Takei, Perry R Klokkevold, Fermin A Carranza. Carranza's clinical periodontology. 12<sup>th</sup> ed. St. Louis MO: Souaders Elsevier; 2015.
- Mealey BL, Oates TW. Diabetes Mellitus and Periodontal disease. J Periodontal. 2006; 77:1289-1303.
- Anil KN, Neh B. Saliva as a detective biofluid. International J. of Med. and App Sciences. 2015, Vol.4, issue1.
- Diabetes Care. Diagnosis and Classification of Diabetes Mellitus .American Diabetes Association. 2014; 37(1):14-80.
- Lang NP, Bartold PM, Cullinam M, et al. International classification work shop. Consensus report: chronic periodontitis. Annals of periodontology. 1999;4-53.
- World Health Organization. WHO expert consultation. Appropriate body mass index for Asian populations and its implications for policy and intervention strategies. The lancet. 2004; 363: 157.163.
- Tenovuod, Saliva. In text book of clinical cardiology by thy lstrup A and Fejers kov O. 2<sup>nd</sup> ed. Munks gaard, Copenhagen. 1994; 17.43.
- Silness P and Loe H: Periodontal disease in pregnancy. Acta Odontol Sand.1964; 22:121.
- Löe H. The gingival index, the plaque index and the retention index system. J Periodontal. 1967;38(6): 610-616.
- Ghasaq A Abdul – Wahab, Maha A Ahmed. Assessment of some salivary enzymes levels in type 2 diabetic patients with chronic periodontitis (clinical and biochemical study). J Baghdad College Dentistry .2015, 27(1):138-143.
- Pal Prabal, Desai NT, Kannan N, et al. Estimation of salivary amylase, salivary total protein and periodontal microflora in diabetes mellitus. JIDA. 2003;74:143-49.
- Deelan Amanj Sabir and Maha Abdul Aziz Ahmed. Assessment of salivary leptin and resistin levels in type 2 diabetic patients with chronic periodontitis (A comparative study). J Baghdad College Dentistry. Accepted for publication, 2015.
- John J Taylor. Protein biomarkers of periodontitis in saliva .ISRN. 2014, P.18.
- Miricescu D, Maria Greabu, Alexandra Totan, et al. The antioxidant potential of saliva: Clinical significance in oral diseases. Ther. Pharmacol. Clin. Toxicol. 2011; 15(2):139-143.
- Sanchez GA, V Miozza, A Delgado, et al. Determination of salivary levels of mucin and amylase in chronic periodontitis patients. J of Periodontal Res. 2011, 46(2):221-7.
- Hady H, Bertl K, Laky M, et al. Salivary and serum chromogranin A and amylase in periodontal health and disease. J Periodontal. 2012, 83(10): 1314-21.
- Sanchez GA, VA Miozza, A Delgado, et al. Relationship between salivary mucin or amylase and the periodontal status. Oral Dis. 2013, V(19) Issue (6), page :585-591.
- Swati K, Rahul B, Biju T, et al. Estimation of levels of salivary mucin, amylase and total protein in gingivitis and chronic periodontitis patients. J. Clin. Diag. Res. 2014,8(10): ZC 56-ZC 60.
- Andrea BA, Aljandra KD, et al. Comparison of salivary levels of mucin and amylase and their relation with clinical parameters obtained from patients with aggressive and chronic periodontal disease. J.Appl. Oral Sci. 2015, V(23), No(3).
- Syleman Aydin. A comparison of Ghrelin, Glucose, alpha-amylase and protein levels in saliva from diabetics. J Biochemistry and Nuclear biology. 2007, V(40), No. (1): pp.29-35.
- Sathy apriyas S, Bharani GO, Nagalingam M, et al. Potential of salivary proteins as a biomarkers in prognosis of diabetes mellitus. J of Pharmacy Res. 2011;4(7): 2228-29.
- L Malathi, KMK Masthan, N Balachander, et al. Estimation of salivary amylase in Diabetic patients and saliva as a diagnostic tool in early diabetic patients. J. Clin Diagn Res. 2013, Nov,7 (11): 2634 – 2636.
- Artis SP, Deg wekar SS, Bhwtte RR. Estimation of salivary glucose, salivary amylase, salivary total protein and salivary flow rate in diabetics in India. J of Oral Science. 2010; 52:359-68.
- Shukria M AL.Zahawi, Hassan A Mahmood, Zewar A Al-Qassab. Effects of diabetes mellitus type II on salivary flow rate and some salivary parameters (total protein, glucose and amylase)in Erbil city .J Bagh College Dent. 2012, V. (24). Issue (2) page 123.
- Prathibha KM, Priscilla Johnson, Mathangi Ganesh, et al. Evaluation of salivary profile among a adult type 2 Diabetes mellitus in south India. J Clin Diagn Res.2013, 7(8): 1592 – 1595.
- M Indria, P Chandra shekar, et al. Evaluation of salivary glucose, amylase and total protein in type 2 diabetes mellitus patients. Indian J. of Dental Res. 2015, V(26) issue (3) page: 271.
- L da.R Conclaves, Soares MR, Noqueira FC, et al. Comparative proteomic analysis of whole saliva from chronic periodontitis patients. J of Proteomics. 2010; 73(7): 1334-41.
- Mulki Shaila, G Prakash Pai and Push para j Shetty. Salivary protein concentration, flow rate, buffer capacity and pH estimation: A comparative study among young and elderly subjects both normal and with gingivitis and periodontitis. J. Indian Soc. Periodontology. 2013. 17(1): 42-46.
- Yadgar Gazy, Bakhtiar Mohiadeen, Ziwar AL-Kasab. Assessment of some salivary biochemical parameters in cigarette smokers with chronic periodontitis. J Baghdad College Dentistry. 2014. V. 26(1), Page: 144-149.
- Scully DV, Langley-Evans SC. Periodontal disease is associated with lower antioxidant capacity in whole saliva and evidence of increased protein oxidation. Clin Sc. 2003;105(2):167-72.
- Doods MWJ, Chih –Koyeh, Dorthea A Johnson. Salivary alterations in type 2 (non-insulin dependent) diabetes mellitus and hypertension. Community Dental Oral Epi. 2000; V(28), Issue (5),Page:373-381.
- Vaziri PB, M Vahedi, SH Adollahzadeh, et al. Evaluation of salivary albumin in diabetic patients. Iranian J Publ. Health.

- 2009;38(3): 54-59.
33. Carmen carda, Nezly Mosquera – Lioreda, Lucas Salom, et al. Structural and functional salivary disorders in type 2 diabetic patients. *Med. Oral Patol Oral Cir Buccal*. 2006; 11: E 309-14.
  34. Hassan HR, Abdul Sattar A. Influence of diabetes disease on concentration of total protein, albumin and globulins in saliva and serum: A comparative study. *Iraqi National of Chemistry*. 2015; 15(1).
  35. Yakob Mahald, Karik, Tervahartiala T, et al. Association of Periodontal microorganisms with salivary proteins and MMP-8 in gingival crevicular fluid. *J.Clin. Periodontol*. 2012; 39(3): 256-63.
  36. Sharmila B, Sangeeta M, Rahul K. Salivary pH: A diagnostic biomarker. *J of Indian Society of Periodontology*. 2013;17(4):461-465.
  37. Basima Gh Ali and Omar Husham Ali. Detection of salivary flow rate and minerals in smokers and non smokers with chronic periodontitis (clinical and biochemical study). *J Baghdad College Dentistry*. 2012; Vol.24(1):68-71.
  38. Eslami H, Fakhrzadeh V, Pakdel F, et al. Comparative evaluation of salivary pH level in type II diabetic patients and Healthy subjects. *VISI J Acemdic*. 2015(4):144.148.
  39. Arul Asrikemath J, R Sanjay and Palanivelu peramachi. Evaluation of correlation between salivary pH and prevalence of dental caries in subjects with and without diabetes mellitus. *Research J of Recent Sciences*. 2014, V.3, 224-226.
  40. Abdulla I Hamad, Riyadh O Alkiai, Intesar JAl kaisi. Flow rates of resting whole saliva of diabetic patients in relation to age and gender. *Tikrit J of Dental Sciences*. 2012; 1: 1-5.
  41. Jose Roberto C, Regina Marcia SP, Fernando de OC, et al. Salivary and microbiological parameters of chronic periodontitis subjects with and without type 2 diabetes mellitus: a case – control study. *Rev. Odontol. UNESP*. 2014; Vol. 43, No. 3.
  42. Collin HL, Niskanen L, Uusitupa M, et al. Oral symptoms and signs in elderly patients with type 2 diabetes mellitus. A focus on diabetic neuropathy. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 2000;90 (3): 299-305.
  43. Ben – Aryeh H, Serouya R, Kanter Y, et al. Oral health and salivary composition in diabetic patients. *J diabetes Complications*. 1993; 7(1): 57-62.

# Effect of Oil Paint Addition on Micro Hardness of Acrylic Ocular Prosthesis

**Firas Abd Kati**

(Assistant Lecturer) - College of Health & Medical Technology / Middle Technical University

**Arshad F. Jassem Al-Kaabi**

(Assistant Lecturer) - College of Health & Medical Technology / Middle Technical University

## ABSTRACT

**Introduction:** Medical grade acrylic resin is the material of choice for many dental and facial restorations. When making artificial eye from this polymer, it is highly required to apply the perfect shade matching the original one. However, this may have some significant effect on the mechanical properties as well as surface integrity since most types of paints used with acrylic resin are oil paints. The aim of this study was to evaluate the effect of oil paint addition on acrylic resin surface hardness.

**Methods:** The study consisted of 3 main groups (control, white 1, and white 2). Each group consisted of 10 samples. The paint concentration was increased from white 1 to white 2 respectively. Vickers hardness test was applied on all samples.

**Results:** The statistical results showed that there was a significant increase in the acrylic surface hardness as the oil paint concentration increases (p-value < 0.05).

**Conclusion:** A conclusion drawn from this study that the oil paint would have significantly increase the acrylic eye prosthesis hardness but it might be to a certain level.

## KEYWORDS

ocular prosthesis; acrylic resin coloring; acrylic resin surface hardness.

## المستخلص

الأكريليك هو المادة المستخدمة بشكل واسع في مجالات طب الأسنان وتعيضات الوجه (على سبيل المثال العين الاصطناعية). يتطلب تصنيع العين الاصطناعية استخدام الألوان الزيتية لغرض الحصول على لون مطابق للعين الطبيعية وهذا قد يكون له بعض التأثير على الخواص الميكانيكية لمادة الأكريليك. إن الهدف من هذه الدراسة هو تقييم تأثير إضافة الطلاء الأبيض بتركيز معينة على صلابة سطح الأكريليك. شملت الدراسة على ثلاث مجموعات رئيسية. تتألف كل مجموعة من 10 عينات. وقد زاد تركيز نسبة الطلاء الأبيض من 1 مل إلى 2 مل على التوالي. تم تطبيق اختبار صلابة في جميع العينات.

النتائج: أظهرت النتائج أن هناك زيادة كبيرة في صلابة سطح الأكريليك مع زيادة تركيز الطلاء.

الخلاصة: الاستنتاج من هذه الدراسة أن استخدام الأصباغ الزيتية كان له تأثير كبير على صلابة مادة الأكريليك ولكن قد يكون على مستوى معين.

## INTRODUCTION

Ocular prosthesis refers to the object that is manufactured from glass or plastic materials to replace the missing eye. Since one of the first noticeable features of the human face is the eye<sup>(1)</sup>, any deformity or defect that has a congenital or accidental origin would significantly impact both the physiological and the psychological status of the individual<sup>(2,3)</sup>. It is suggested to replace the lost eye as soon as possible in order to avoid the patient stress due to the societal reaction to the patient's deformity<sup>(4)</sup>. The biggest challenges associated with ocular prosthesis treatment is to make the patient capable of coping with the rehabilitation process which would enhance its esthetic acceptability<sup>(5)</sup>. Regarding esthetics, ocular prosthesis are mainly made from medical grade acrylic resin (polymethylmethacrylate) in a process involving proper shaping and adding white stain as well as blood vessel simulation to match the appearance of the natural eye<sup>(6)</sup>. When it comes to coloring, different materials are used to for this purpose such as hydrosoluble gouache and nitrocellulose that are chemically compatible with acrylic resin. However, the main type of stain used is the oil paint<sup>(7, 8)</sup>. It was reported in the literature

that color combination with the acrylic resin would have some effects on the mechanical properties of the resin itself<sup>(9)</sup>. Oil paint addition would have an optimum esthetic results for the restoration due to their compatibility with the acrylic resin. However, oil acts as a plasticizer that could change the mechanical properties of the final restoration which might later affect its color stability<sup>(10, 11)</sup>. Therefore, it is necessary to understand what kind of change the oil paint addition would happen on the acrylic resin surface properties as well as the mechanical properties. The aim of this study was to evaluate the effect of oil paint addition on acrylic resin surface hardness.

## MATERIALS AND METHODS

### Materials

In this study, a total of 30 specimens of acrylic resin (Spofadental, Czech Republic) were used. They were divided into 3 groups; the control, white 1, and white 2. Each group had 10 specimens. The experimental groups (white 1 and white 2) differ from each other in the amount of white oil paint added. The oil paint used was Grumbacher Titanium white (USA) (Picture 1). Dental stone (Spofadental, Czech Republic) was used for molds making, and petroleum



jelly (China) was used as a separating medium.



Picture (1): Oil paint (acrylic compatible) Grumbacher Titanium white

### Sample preparation

Metal models (65 mm x 10 mm x 3 mm) were used to make acrylic samples. The sample preparation started by applying separating medium to the upper and lower halves of metal flask to facilitate removal of stone mold following deflasking. Then, the mixed dental stone at a creamy state was placed into lower half of flask and patterns gently in the middle part while taking into consideration half of patterns must be exposed so that they can be easily removed. After complete set of the dental stone layers, the separating medium was added and upper half was placed into its position. Another mix of dental stone was prepared and placed over both patterns and stone surface. The mold was left for one hour. It was then opened to remove the metal sample (Picture 2). After that, the mold was lubricated with separating medium and left to dry. The second layer of the medium was applied. The acrylic powder and liquid were mixed for the control group by adding 22g of the powder to 10 ml of the monomer at each mix. The dough acrylic was packed into the mold, the flask was compressed under hydraulic press and then cured. Once curing completed, the flask was left to cool. The samples were removed from the flask, finished and polished.

They were then stored in water to avoid acrylic shrinkage until conducting the hardness test. The White acrylic samples (groups 2 and 3) were fabricated using the above process with adding 1ml white oil paint to 9ml liquid monomer for the White 1 group, and adding 2ml white oil paint to 8ml of monomer for the White 2 group.



Picture (2): The dental stone mold and the metal model

### Hardness test

Hardness tester (Matsuzawa Japan for Vickers hardness test) was used for measuring acrylic samples' hardness and the Vickers micro-hardness test was selected according to the samples' material (Picture 3). The test protocol consist of applying a load of 50 grams on the sample surface for 10 seconds. The samples were tested three times (left, middle, right) and average readings were calculated for each specimen.



Picture (3): Matsuzawa hardness testing machine

### RESULTS

Each sample was subjected to Vickers hardness test for three times at three different spots. An average value for the three different measurements was calculated to represent the sample hardness strength. The IBM SPSS V.20 software was used for the statistical analysis. As presented in Tables (1 and 2), there was a noticeable increase in the mean values of hardness strength as the concentration of the oil paint increases. That was associated with the slight but insignificant increase in the standard deviation values for the white 1 and the white 2 groups. The One-way ANOVA test confirmed the tendency of the oil paint to increase the hardness strength of the acrylic resin material  $P\text{-value} < 0.05$  as both white 1 and 2 were significantly different in the amount of hardness strength than the control. They were also significantly different from each other in the amount of hardness strength as illustrated in the Tukey HSD multiple comparison test (Table 3).



Table (1): Hardness values for the groups' samples (g/mm<sup>2</sup>)

Control	White 1	White 2
129.60	226.00	245.40
128.40	230.40	243.80
130.70	225.30	246.30
126.50	227.20	249.60
128.30	229.80	250.40
127.60	231.50	241.50
131.20	219.20	242.20
125.90	228.40	243.30
124.20	231.20	248.20
123.80	230.90	247.80

Table (2): Means and standard deviation values for the groups

	Groups	N	Mean	Std. Deviation
Hardness (g/mm <sup>2</sup> )	Control	10	127.62	2.53412
	White 1	10	227.99	3.79223
	White 2	10	245.85	3.11707

Table (3): Tukey HSD multiple comparison test (ANOVA)

Groups	N	Subset for alpha = 0.05		
		1	2	3
Control	10	127.6200		
White 1	10		227.9900	
White 2	10			245.8500
Sig.		1.000	1.000	1.000

## DISCUSSION

Acrylic resin material (polymethylmethacrylate) has been used extensively for a variety of medical applications due to its chemical and mechanical properties as well as being biologically inert when it is used for making a dental or facial prosthesis. When it comes to ocular prosthesis which is mainly constructed from acrylic resin, it is necessary to evaluate the functionality and the suitability of this material as well as its longevity. Acrylic eyes need to match the real ones as much as possible. Therefore, several paints and stains are used for prosthetic coloring. Since oil paints are the material of choice for most technicians, it is believed that it might have an effect on the mechanical properties of the acrylic restoration since oil paint acts as a plasticizer. The aim of this study was to examine and evaluate the effect of oil paint addition to the acrylic resin on its surface hardness. Three study groups were used; the control and 2 different paint-loaded groups. The Vickers hardness test results showed a significant variation

among the groups with the increase in surface hardness as the oil paint concentration increases. The statistical results demonstrate the surface hardness increase in relation to oil paint concentration. The study results disagrees with a study conducted by Parker S. and associates [12] who stated that surface hardness of soft acrylic resin decreases with the increase of plasticizer content which is oil soluble compound.

## CONCLUSION

This study was an attempt to evaluate the effect of oil paint addition on the acrylic resin (polymethylmethacrylate) material in terms of hardness strength. The study results clearly stated that the oil paint would significantly increase the hardness strength of the acrylic resin. However, it is still ambiguous that at what lever of oil paint concentration this hypothesis would still valid. Therefore, further considerations are highly recommended.

## REFERENCES

1. Doshi PJ, Aruna B. Prosthetic Management of patient with ocular defect. J Ind Prosthodont Soc 2005; 5: 37–38.
2. Lubkin V, Sloan S. Enucleation and psychic trauma. Adv Ophthalmic Plast Reconstr Surg 1990; 8: 259–262.
3. Raflo GT. Enucleation and evisceration. In: Tasman W, Jaeger E eds. Duane's Clinical Ophthalmology, Revised edn, Vol. 5. Philadelphia: Lippincott-Raven, 1995: 1–25.
4. Artopoulou II, Montgomery PC, Wesley PJ, Lemon JC. Digital imaging in the fabrication of ocular prostheses. J Prosthet Dent 2006; 95: 327–330.
5. Ow RKK, Amrith S. Ocular prosthetics: use of a tissue conditioner material to modify a stock ocular prosthesis. J Prosthet Dent 1997; 78: 218–222.
6. Benson P. The fitting and fabrication of a custom resin artificial eye. J Prosthet Dent 1977; 38: 532–538.
7. Fernandes AU, Goiato MC, Batista MA, Santos DM. Color alteration of the paint used for iris painting in ocular prostheses. Braz Oral Res. 2009 Oct-Dec;23(4):386-92.
8. Rutkunas V, Sabaliauskas V, Mizutani H. Effects of different food colorants and polishing techniques on color stability of provisional prosthetic materials. Dent Mater J. 2010 Mar;29(2):167-76.
9. Heath JR, Wilson HJ. Surface roughness of restorations. Br Dent J 1976; 140: 131-137.
10. Wright PS. Composition and properties of soft lining materials for acrylic dentures. J Dent 1981;9:210–23.
11. Braden M, Wright PS. Water absorption and water solubility of soft lining materials for acrylic dentures. J Dent Res 1983; 62:764–8.
12. Parker S, Martin D, Braden M. Soft acrylic resin materials containing a polymerisable plasticiser I: mechanical properties. Biomaterials. 1998 Sep;19(18):1695-701.

# *Pacifier Sucking Habit and its Relation to Oral Health of Children Aged 1-5 Years (comparative study)*

*Aseel Haidar Al-Assadi,*

B.D.S, M.Sc. Assist. Prof.

*Zainab A.A. Al-Dahan,*

B.D.S, M.Sc. - Prof.

*Abdul Khaliq Al- Rammahy*

Assist. Prof.

## **ABSTRACT**

**Background:** For many infants and children non-nutritive sucking habits are very common and one of those habits is pacifier sucking, however, if this habit persist beyond the age of 3 years it may cause esthetic, occlusal and psychological changes. This study was conducted to determine the effect of pacifier sucking habit on the oral health of children aged 1-5 years old in Baghdad city and to assess its role in the modification of the oral microflora.

**Materials and methods:** The study was carried out among children aged 1-5 years old with no history of any systemic diseases nor taking any medical treatment for the past two weeks prior to the examination, 50 children with continuous pacifier sucking habit were chosen to be the study group, compared to 50 children without any sucking habit (control group) matching the study group in age and gender. Information sheet filled by the parents concerning general health and frequency of oral infections of their children was taken. Oral microorganisms samples were obtained from children and cultured aerobically using blood agar, MacConkey agar, chocolate agar and sabouraud, s dextrose agar.

**Results:** Children with pacifier sucking habit showed higher frequency of having continuous oral infections (44%) whereas in non pacifier sucking group it was zero. Concerning the oral infections, a statistical difference was found by which the pacifier sucking children oral infections exceed significantly that of the non pacifier sucking children ( $P<0.01$ ). Regarding the oral microorganisms, more types were found among pacifier sucking children. *Candida*, *Strep.pyogenes*, *strep.faecalis*, *E.coli*, *Acinetobacter* and *strep. pneumonia* were more common among pacifier sucking children.

**Conclusion:** Pacifier use affect types and frequency of microorganisms found in the oral cavity which may affect the frequency of oral infections. Health programs should be constructed to improve parents' knowledge concerning the effect of pacifier use in the oral health of their children and how to clean it if it used to reduce its contamination.

## **KEY WORDS**

Pacifier sucking habit, candidal infection, oral microflora.

## **INTRODUCTION**

Two types of sucking habit are found, nutritive sucking habit by which the child sucks for nutrition and non nutritive sucking habit and that is when the child sucks to satisfy psychological need<sup>(1,2)</sup>. Both are considered sucking habits as they involve frequent conscious and unconscious neuromuscular activities<sup>(3,4)</sup>.

Pacifier sucking habit is considered as a nonnutritive sucking habit<sup>(5)</sup>. It is a nursing device shaped for a baby's mouth with imperforated teat that is attached to a shield designed in a way to prevent the child from putting the entire pacifier in his mouth. It also has rings attached to the flange for easy removal if aspirated<sup>(6)</sup>.

The use of pacifier may be initiated by the parents or the caregiver of the child for many reasons and the most common one is that, as its name indicate, soothing and comforting a baby<sup>(7,8)</sup>, however, adverse health effects have been associated with its use including inverse relation to breast feeding duration which increas the risk of early weaning<sup>(9,10)</sup>, otitis

media<sup>(11)</sup>, thrush and candidal infection<sup>(12)</sup>. It could also associated with wheezing, respiratory illness, vomiting, fever, diarrhea and colic<sup>(13)</sup>.

In Iraq, there is no available data or study concerning the effect of the pacifier in the oral health of children, so this study was conducted to determine its role in the oral infections and to assess its effect on the oral microflora among Iraqi children 1-5 years.

## **MATERIALS AND METHOD**

Approval from the ministries of work and social affairs, education and health to carry out this study was taken. A contact with school authorities was made to explain the purpose of the study. For including children in this study permission was obtained from the parents and questionnaires were designed to obtain information from them including general health and the sucking habits of their children. The study group was consisted of fifty healthy children with continuous pacifier sucking habit aged 1-5 years, they were selected from thirty kindergarten and nursery schools in both sides of Baghdad city. While the control group were 50 children matching the age and gender selected from the same school of the study

group. They were without any sucking habit. All children in both groups had not taken antibiotic and/or antimycotic treatment during at least the previous three months and without any chronic disease.

Microbiological samples were collected from children by swabbing their oral mucosal surface (cheek, hard palate, dorsum of the tongue and floor of the mouth) by sterile cotton swap (14). Each swab was streaked on blood agar, MacConkey agar, chocolate agar and Sabouraud's dextrose agar and then incubated aerobically for 24 hours at 37°C. The morphology of different types of colonies was recorded and smears of these different colonies were done to study the Gram's reaction and microscopical characteristic (15).

Different types of colonies were sub cultured and stored for further biochemical tests to reach complete identification of each isolate. These tests include: Hemolysis on blood and chocolate agar plates, catalase test, oxidase test, slide coagulase test, Imvic, urease test, Kliger iron agar (KIA) test, bacitracin differentiation test, optochin sensitivity test and API 20 E system (16,17).

## RESULTS

Concerning oral infections, continuous oral infections occurred only among pacifier sucking group (44%), however, children affected by oral infections at few and distant times among non pacifier sucking group were 46 (92%) whereas among pacifier sucking group was 9 (18%). A statistical difference was found by which the pacifier sucking group exceed significantly that of non pacifier sucking group ( $P < 0.01$ ), Table (1).

There are 17 different types of microorganisms in pacifier sucking group, while in non pacifier sucking group there are only 14 types of microorganisms, Table(2). In the pacifier sucking group the highest percentage appear to be found for *Strep. viridans*, *Candida* and *Moraxella* which are 100%, 96% and 94% respectively, however, in non pacifier sucking group the highest percentage appear to be found for *Strep.viridans* 100% followed by *Moraxella* 96% and then *Candida* 60%.

Table (3) and (4) demonstrated the distribution of microorganisms by age group in the pacifier and non pacifier sucking children respectively.

## DISCUSSION

The results of the present study revealed that there was a significant relation between the occurrence of oral infection and the pacifier use as it is shown in

Table (1), this finding is in accordance with that found by other studies (12,14,18).

The association between pacifier sucking habit and the occurrence of oral infection may be related to the presence of a foreign body in the mouth which may be contaminated in a way or another like its contamination when it's dropped down or its use by another child.

In this study the presence of candida in the pacifier sucking children was 36% higher than that of the non pacifier sucking children ( Table 2) which suggests that the use of pacifier may be a local factor that influences and enhances the colonization and proliferation of candida in the oral cavity (14).

The microorganisms identified according to the systematic manner which comprises the colony morphology and the selective media in addition to the biochemical tests, the microbiological study of pacifier and non – pacifier sucking groups demonstrates the differences in the types and frequency of microorganisms. Table (2) shows the number of microbial isolates in pacifier sucking group were 17 different types of microorganisms whereas in non pacifier sucking group they were 14. In general the number and frequency of microbial isolates in pacifier sucking group were higher than in the non pacifier sucking group.

Like any other removable appliance used orally, a pacifier after a period of time will itself become colonized with microorganisms and may modify the oral flora (8, 19), also the horizontal transmission of microorganisms could be occurred by a pacifier as a cross infectional factor, facilitating for example yeast and *strept mutants* infections (14). Also the continuous use of pacifier would favor the growth of aciduric microorganisms like the yeast and lactobacilli as a result of the drop of saliva pH due to the stagnation of saliva that is contributed to the use of pacifier (14, 19).

The predominance bacterial isolates which had been found in this study in both pacifier and non – pacifier sucking group were *Strep.viridans* (100% for both groups) and *Moraxella* (94% and 96% respectively). These two microorganisms are normal flora in the mouth and the presence of *Moraxella* in this high percentage is in agreement with Nolte (20).

The next most predominance microbial isolates in the pacifier sucking group was *Candida albicans* 96% compared to 60% in non pacifier sucking group, which mean that pacifier sucking group exceed by 36% the non pacifier group. This finding is in accordance with the results presented by other studies (14, 19, 22), however, it is near that found in Jordan in

1995 by Darwazeh and AL – Bashir <sup>(22)</sup> which was 30%, and that found in Brazil in 2001 by Mattos-Graner et al.<sup>(14)</sup> which was 32%.

Staph. epidermidis is higher in pacifier sucking group (86%) compared to the non pacifier sucking group(24%). This may be due to the transmission of this microorganisms by the pacifier from another area of the body such as the skin or by fomites as cloths.

*Strep.pyogenes* is a  $\beta$ -haemolytic microorganism, it is one of the commonest bacterial pathogens that cause pharyngotonsillitis all over the world <sup>(23)</sup>. In the present study, *Strep.pyogenes* represent 6% in non pacifier sucking group compared to 78% in pacifier sucking group, this high percentage may be due to seasonal variation as the sample were collected in winter in which tonsillitis and other related infections are known to be common and so pacifier may act as a source of transmission of these microorganisms either directly (from other infected children as an example),

or indirectly (contaminated hands).

## CONCLUSIONS

Oral infections were higher among pacifier sucking children than the non pacifier sucking children which were ensured by that aerobic microbial isolates in pacifier sucking group was (17) isolates compared with (14) isolates in non pacifier sucking group, and the presence of candida and coliform bacteria (*E-coli*, *Klebsiella*, *Enterobacter*) was higher in pacifier sucking group.

If there is no way other than give the child a pacifier certain precautions and strict hygiene rules should be kept in mind, in which the pacifier should be efficiently cleaned or rinsed before and after each use to decrease exposure to germs, also never coating a pacifier with any sweet fluid that may increase the risk of developing dental caries.

**Table (1): Occurrence of oral infection among pacifier and non pacifier sucking children.**

Occurrence of oral infection	Pacifier sucking		Non pacifier sucking		Sig.
Continuously	22	(44%)	0	-	H.S**
Occasionally	19	(38%)	4	(8%)	H.S
Few and distant times	9	(18%)	46	(92%)	H.S
Total	50	100%	50	100%	

\*\* Highly significant,  $P<0.01$

**Table (2): Distribution of M.O. by frequency and percentage in the pacifier and non pacifier sucking children.**

Type of M.O	Pacifier sucking		Non pacifier sucking	
	Frequency	%	Frequency	%
Strep. viridans	50	100	50	100
Strep. faecalis	31	62	15	30
Strep. pneumonia	10	20	1	2
Strep. pyogenes	39	78	3	6
Moraxella	47	94	48	96
Staph.epi.	43	86	12	24
Staph.aureus	7	14	2	4
Sarcinae	8	16	5	10
Lactobacilli	11	22	8	16
corynebacterium	2	4	1	2
Candida	48	96	30	60
E-coli	16	32	1	2
Acinetobacter	15	30	1	2
Enterobacter	3	6	-	-
Pseudomonas	3	6	-	-
Pantoea	3	6	-	-
Klesbsiella	10	20	5	10



**Table (3): Distribution of M.O among pacifier sucking children**

<i>M.O</i>	<i>Age Group</i>							
	<i>1-2 years n=21</i>		<i>2-3 years n=15</i>		<i>3-4 years n=8</i>		<i>4-5 years n=6</i>	
	<i>Frequency</i>	<i>%</i>	<i>Frequency</i>	<i>%</i>	<i>Frequency</i>	<i>%</i>	<i>Frequency</i>	<i>%</i>
Moraxella	20	95.2	14	93.3	8	100	5	83.3
Candida	19	90.5	15	100	8	100	6	100
Staph. epi.	17	80.9	14	93.3	6	75	6	100
Stap. aureus	2	16.7	2	13.3	2	25	1	16.7
Sterp.pyogenes	14	66.7	12	80	7	87.5	6	100
Sterp.faecalis	11	52.4	9	60	6	75	5	83.3
Sterp.pneumonia	5	23.8	4	26.6	1	12.5	0	-
Sterp.viridans	21	100	15	100	8	100	6	100
Lactobacilli	1	4.8	1	6.7	4	50	5	83.3
Corynebacterium	0	-	0	-	1	12.5	1	16.7
Sarcinae	4	19.1	3	20	1	12.5	0	-
E.coli	5	23.8	4	26.6	4	50	3	50
Enterobacter	2	16.7	0	-	2	12.5	0	-
Acinetobacter	8	80.1	3	20	2	25	3	50
Pantoea	2	9.5	1	6.7	0	-	0	-
Pseudomonas	2	9.5	0	-	1	12.5	0	-
Klebsiella	5	23.8	5	33.3	0	-	0	-

**Table (4): Distribution of M.O among non pacifier sucking children.**

<i>M.O</i>	<i>Age Group</i>							
	<i>1-2 years n=21</i>		<i>2-3 years n=15</i>		<i>3-4 years n=8</i>		<i>4-5 years n=6</i>	
	<i>Frequency</i>	<i>%</i>	<i>Frequency</i>	<i>%</i>	<i>Frequency</i>	<i>%</i>	<i>Frequency</i>	<i>%</i>
Moraxella	19	90.5	15	100	8	100	6	100
Staph. epi.	7	33.3	5	33.3	0	-	0	-
Strep. viridans	21	100	15	100	8	100	6	100
Strep. faecalis	6	28.6	7	46.7	1	12.5	1	16.7
Strep. Pneumonia	1	4.8	0	-	0	-	0	-
Strep. Pyogenes	0	-	0	-	1	12.5	2	33.3
Staph. aureus	0	-	0	-	1	12.5	1	16.7
Lactobacilli	0	-	1	6.7	4	50	3	50
Corynebacterium	0	-	0	-	0	-	1	16.7
Sarcinea	0	-	1	6.7	2	25	2	33.3
Candida	21	57.1	9	60	5	62.5	4	66.7
E. coli	-	-	-	-	1	12.5	-	-
Acinetobacter	-	-	-	-	1	12.5	-	-
Klebsiella	0	-	0	-	2	25	3	50

## REFERENCES

- Warren JJ, Levy SM, Nowak AJ, Tang S: Non nutritive sucking behaviors in preschool children: a longitudinal study. *Pediatr Dent.* 2000; 22:187-191.
- Maguire JA. The evaluation and treatment of pediatric oral habits. *Dental Clin. North Am.* 2000; 44(3): 659-669.
- Katz CRT, Souto JRS, Feitosa SVHS, Souza AS, Zisman M, Rosenblatt A: Harmful oral habits: a multidisciplinary approach. *Arq Odontol* 2002; 38:35-42.
- Maia-Nader M, Figueiredo CSA, Figueiredo FP, Silva AAM, Thomaz EBA, Saraiva MCP, Barbieri MA, Bettiol H: Factors associated with prolonged non-nutritive sucking habits in two cohorts of Brazilian children. 2014; 14:743-753.
- Castilho SD, Rocha MA. Pacifier habit: history and multidisciplinary view. *Jornal de Pediatria* 2009; 85(6):480-488.
- Al-Assadi AH. Pacifier effects on oral health of children in Baghdad city. MSc. Thesis College of Dentistry University of Baghdad 2004.



7. Greenberg CS. A sugar-coated pacifier reduces procedural pain in newborns. *Pediatr Nurs.* 2002; 28(3):271-277.
8. Al-Hashemi EH, Al-Shaheed MIA: Isolation and identification of bacteria in dummy sucking children with gastroenteritis (comparative study). *J Bagh College dent* 2010; 22(4); 111-114.
9. Collins CT, Ryan P, Crowther CA, McPhee AJ, Paterson S, Hiller JE. Effect of bottles, cups, and dummies on breast feeding in preterm infants: a randomized controlled trial. *BMJ.* 2004; 329(7459):193-198.
10. Vogel AM, Hutchison BL, Mitchell EA. The impact of pacifier use on breastfeeding: a prospective cohort study. *J Paediatr Child Health.* 2001; 37(1):58-63.
11. Rovers MM, Numans ME, Langenbach E, Grobbee DE, Verheij TJ, Schilder AG. Is pacifier use a risk factor for acute otitis media? A dynamic cohort study. *Fam Pract.* 2008; 25(4):233-236.
12. Niemela M, Pihakar O, Pokka T, Uhari M. Pacifier as a risk factor for acute otitis media. A randomized controlled trial of parental counseling. *Pediatrics.* 2000; 106: 483 – 488.
13. North Stone K, Fleming P, Golding J. Socio- demographic association with digit and pacifier sucking at 15 months of age and possible associations with infant infection. *Early Hum Dev* 2000; 60(2): 137 – 48.
14. Mattos-Graner RA, A; DeMoraes; Regina M. Rontanti; Esther G. Briman. Relation of oral yeast infection in Brazilian infants and use of a pacifier. *J Dent Children* .2001; 68(1): 33 – 36.
15. Davis JR, Stager CE, Wnde RD, Qadri SMH. Clinical laboratory evaluation of the automatic system *Enterobacteriaceae* *Biochemical Card. J Clin Microbiol*1981; 4:370-5.
16. Fingold S.M. and Baron E.J. *Diagnosis microbiology*.7th ed. CV. Mosby Co., USA, 1986:250-258.
17. Mellevilli T.H. and Russel C.L. *Microbiology for dental students*. 3rd ed. CV Mosby Co., 1981:299-373.
18. Kramer MS; Bair RG; Dagenais S; Yang H; Jones P; Ciofari L; Jane F.: Pacifier use, early weaning and cry/fuss behavior: A randomized controlled trial. *JAMA* .2001; 286 (3): 322 – 6.
19. Yonezu T, Yakushiji M. Longitudinal study on influence of prolonged non-nutritive sucking habits on dental caries in Japanese children from 1.5 to 3 years of age. *Bull Tokyo Dent Coll.* 2008; 49:59-63.
20. Nolte W.A. *Oral microbiology with basic microbiology and immunology*. Fourth edition. CV Mosby Co. 1982: 424, 362, 413.
21. Elodie C, Karine M, François N R. Renaud, Jeanne D, Emmanuelle B, Jean F. Pacifiers: A microbial reservoir. *Nursing & Health Sciences.* 2006; 8(4):216-223.
22. Darwazeh, A.M. G, and AL- Bashir; A. Oral candidal flora in healthy infants. *Oral. Pathol.* 1995; 24:361 – 364.
23. Abdullah M.A. Group A Streptococcal Tonsillar infection among primary school children in Samara. MSc. Thesis College of Medicine University of Tikrit.2003.

# *Histological and Immunohistochemical Evaluation of Local Exogenous Application of the Green Tea on Bone Healing (Experimental Study On Rats)*

**Enas Fadhil**

B.D.S., MSc., Ph.D., in oral histology  
College of Dentistry University of Baghdad, Iraq

## **ABSTRACT**

**Background:** The study was designed to identify the role of local exogenous green tea in bone healing .

**Material and method:** Thirty male albino rats, were subjected for bone defects in medial sides of both tibia bone (right tibia was considered as experimental site with 1μl of extract green tea, while the left be the control one, (treated with 1μl of normal saline). The rats were scarified at 3,7,10 days post surgery. Bone healing was histologically examined with immunohistochemical for localization of bone morphogenic protein 2.

**Results:** show bone healing treated with extracted green tea illustrated to be faster in apposition of osteoid and bone trabeculae that is detected to fill the most of bone defect in comparison with control group.

**Conclusion:** Our results indicate that low application of exogenous green tea could be an effective therapeutic for bone injuries, these data are promising for a possible future clinical usage especially for enhancement of bone healing.

## **INTRODUCTION**

Bone is a composition tissue that consists of organic components, type I collagen, non collagenous proteins and inorganic components, mainly hydroxyapatite crystals (Nanci, et al., 2008). The two main categories of bone cells are osteoblasts that form the bone and osteoclasts that resorb (dissolve) the bone. The combined and cooperative activities of osteoblasts and osteoclasts result in a bone architecture that provides mechanical support and protection for the body. In addition, bone serves as a vital reservoir of minerals, principally calcium and phosphorus, necessary for maintaining normal cellular, neurologic, and vascular activities of the body (Nijweide et al., 2010).

Both osteoblastic and osteoclastic cells regulate bone metabolism, and both cell types are involved in the development of osteoporosis (Nijweide et al., 2010). Osteoblasts, the bone-forming cells, locate near the surface of the bone and produce cytokines that affect osteoclasts. viable osteoblasts acting in concert with multiple growth factors (Matsuo et al., 2008), which are essential for the repair and regeneration of bone. Specific growth factors important to bone formation include fibroblast growth factor 2, platelet-derived growth factor, vascular endothelial growth factor, Insulin like growth factor, bone morphogenic protein 2, 4 and 7, and transforming growth factor-beta (Singer et al., 2011).

Tea, the dried leaves of the *Camellia sinensis* species of the Theaceae family, is a popular beverage with an annual production of three billion kilograms worldwide (Yang et al., 2000). In the past decade, epidemiological evidence has shown an association between tea consumption and the

prevention of age-related bone loss in the elderly population. The objective of this research is to study the benefits of local application of green tea as exogenous biomaterial on bone healing. As the use of green tea is a first trial to study in healing process.

Green tea polyphenols (GTP) have positive effects on bone mass and microarchitecture in both models of bone loss (She et al., 2008; Shen et al., 2009). Green tea prevented the aging-induced as well as aging plus estrogen deficiency-induced reduction in femoral bone mass (i.e., bone mineral content (BMC) and density (BMD), respectively (Chwan-Li et al., 2011). Serum osteocalcin (a bone formation biomarker) was elevated by the GTP treatment, while serum tartrate-resistant acid phosphatase (TRAP, a bone resorption biomarker) was suppressed by the GTP treatment (Wang et al., 2008).

Green tea poly phenols supplementation was shown to suppress oxidative stress (as shown by decreased urinary 8-hydroxy-2'-deoxyguanosine levels) and inflammation (as shown by suppressed mRNA expression of tumor necrosis factor- $\alpha$  and cyclooxygenase-2 in spleen). Authors concluded that GTP mitigates bone loss in a chronic-inflammation-induced bone loss model by reducing oxidative stress-induced damage and inflammation (Shen et al., 2009; Shen et al., 2010).

One particular green tea compound called Epigallocatechin (EGC), led to an increase in this enzyme's activity by 79%. It also increased levels of bone mineralization in these cells: a vital part of strengthening bones (Mount et al., 2006).

Epigallocatechin also suppressed the activity of cells called osteoclast cells that weaken and break down old bone, part of the natural process of bone

remodelling. (Choi et al.,2003).

## MATERIALS AND METHODS

The study was designed to illustrate the effectiveness of application of the local exogenous extracted green tea gel by studying bone specimens histologically and identification of bone morphogenic protein 2 in bone tissue.

All experimental procedures were carried out in accordance with ethical principles of animal experimentation.

### Experimental animals

In this research, thirty male albino rats, weighing (320-500) gram, aged (6.5-8) months were used and maintained under control conditions of temperature, drinking and food consumption.

The animals were subjected for bone defects in medial sides of both tibial bone ( right tibia was considered as experimental site, while left be the control one), the animals were divided into two groups

1. Control group the bone defect treated with 1µl normal saline.

2. Experimental group includes 10 rats, the bone defect treated with 1µl extracted green tea gel .

The rats were sacrificed at 3, 7,10 days post surgery.

### Materials

Extracted green tea gel

### Methods

The animals were subjected for a surgical operation. The surgery was performed under a well sterilized condition and gentle technique. Every animal was weighted to calculate the dose of general anesthesia that was given to it. The general anesthesia was induced by Intra muscular injection of xylazine 2% (0.4 mg/kg B.W.), plus ketamine HCL 50mg (40 mg/kg- B.W.also an antibiotic cover with oxy-tetracycline 20% ( 0.7 ml/kg) intramuscular injection was given. Then the animal was placed on the surgical table and the surgical towel was placed under the site of the operation . Both tibiae were shaved using cream from the inner side and the skin was cleaned with a mixture of ethanol and iodine then a piece of cotton damped with alcohol and left covering the shaved skin for one minute . Surgery was performed under sterile condition and gentle surgical technique. The cotton was removed. Incision was made on the lateral side to expose the medial side of the tibia , the skin and fascia flap was reflected. By intermittent drilling, and continuous cooling with irrigated saline, a hole of 1.8mm was made with small round bur. Bone penetration was performed at a rotary speed of 1500

rpm. Following the hole preparation, the operation site was washed with saline solution to remove debris from the drilling site. Bone defect was made on both medial sides of right and left tibia bone, experimental and control sites.

After operation drying the area by air, then applied extracted green tea gel 1µl .While normal saline 1 µml was used for control. Suturing of the muscles was done with absorbable catgut followed by skin suture . The operation site was sprayed with local antibiotic (tetracycline spray), also long acting systemic antibiotic oxy-tetracycline 20% (0.7 ml/1kg) was given to the animals after the surgery. Post operative care was performed by giving an antibiotic ( local and systemic). The animals were scarified at intervals 3,7,10 day , using over dose anesthesia . Five animals for each period, for each group. The tibia bone was dissected and fixed in 10% buffered formalin. Histological examination was done for all samples under light microscope.

### Assessment for immunohistochemistry results

Positive reading was indicated when the cells display a brown cytoplasmic stain, while negative reading was indicated for absence of immune-reactions depends on positive and negative control.

### Immunohistochemical scoring of BMP2

Quantification method of Immunoreactivity was semiquantitatively estimated the immune-staining score that was calculated as the sum of a proportion score . The proportion score reflects the estimated fraction of positively stained infiltrating cells: score 0: none. score 1:<10%.score 2:10-50%. score 3:51-80%.score 4: >80%. The intensity score represents the estimated staining intensity (score0, no staining, score1weak,score2 moderate, score3 strong), Hillmann et al.,1999.

## RESULTS

### Histological findings

Control group at 3 Days duration shows bone marrow infiltrated by inflammatory cells with newly formed blood vessels; (Figure 1). Bone healing site for Control 7 Days duration shows fibrous tissue, with progenitor cell and fibroblast cell; (Figures 2). Trabeculated bone coalesce with cutting bone. Osteoblast, osteocyte and reticular cells aew detected at 10 day post operative period ( Figures 3,4).

**Figure 3.1** Microphotograph view for control group (3 days) shows progenitor cell (arrow head), inflammatory cell (arrow).H&EX20



(CB) in control group 10 days. H&EX20

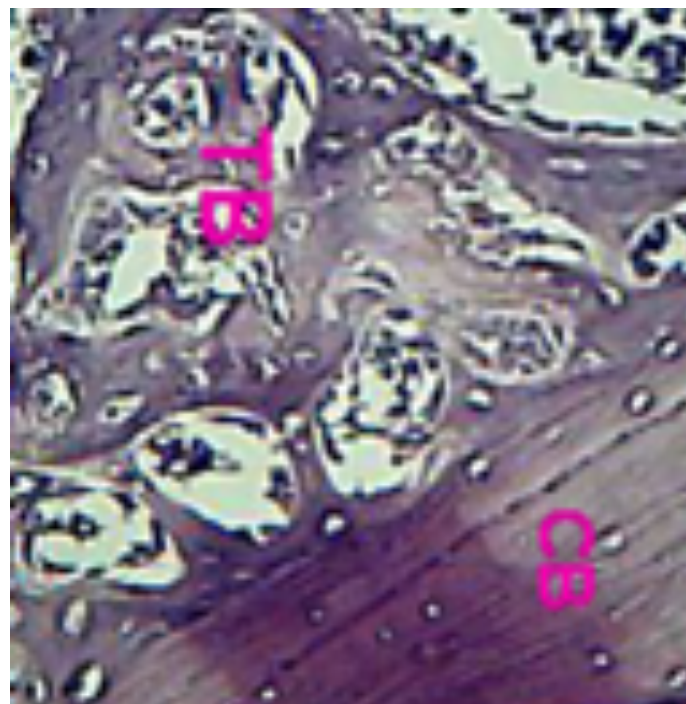


Figure 4 View for osteoid tissue (OT) filled the bone defect in green tea gel group at 3 days. H&EX20

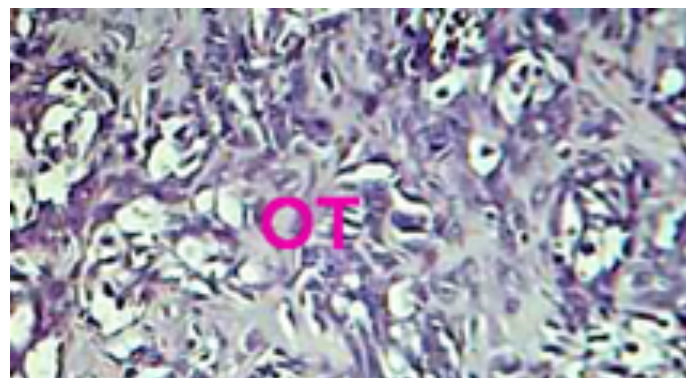


Figure5: New blood vessels (arrow heads) in developing osteoid tissue. H&EX40

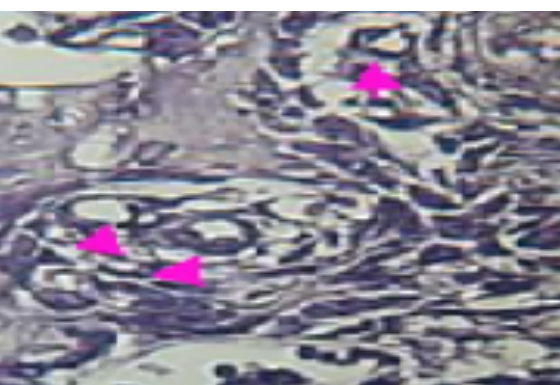


Figure2:View for bone healing site (control 7 days) shows fibrous tissue (FT) with progenitor cell (arrow head), fibroblast (arrow).H&EX20

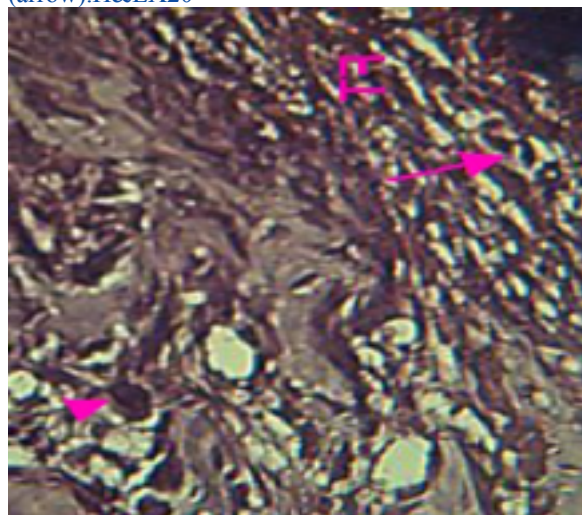


Figure3:View for trabeculated bone (TB) coalesce with cutting bone

Figure 6:Trabeculated bone (TB), Coalesce with cutting bone (CB) in green tea gel group 7 days. H&EX20



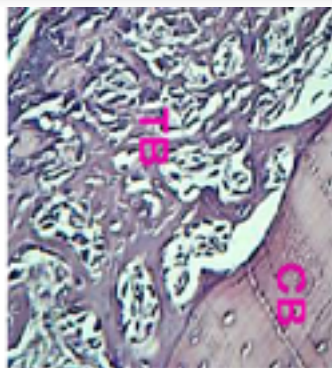


Figure7:View for highly cellular osteoid tissue.H&EX40

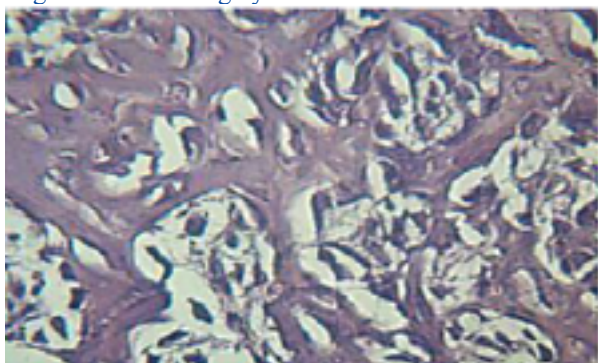
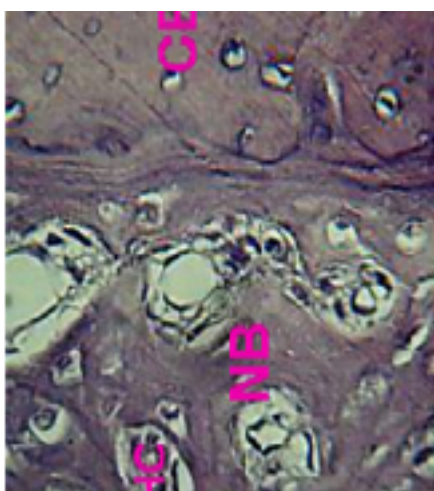


Figure 8: New bone (NB) with multiple Haversian canal (HC) , in nearly cutting bone (CB). H&EX20



**Experimental group:** Bone treated with extracted green tea gel illustrated osteoid tissue filled the bone defect with newly formed blood vessels is detected at 3 day Figures (4,5). Trabeculated bone, coalesce with cutting bone in extracted green tea gel group 7 days with highly vascular osteoid tissue; Figures (6,7) and the period 10 days illustrated new bone with multiple Haversian canal in nearby cutting bone; Figure (8).

### 3.2 Immunohistochemical evaluation for expression of bone morphogenic protein 2

Figure(9) Immunohistochemical view for control group 3 days shows positive bone morphogenic protein 2 expression by progenitor (arrow heads), inflammatory cell (green arrow), endothelial cell (pink arrow). DAB stainX20.

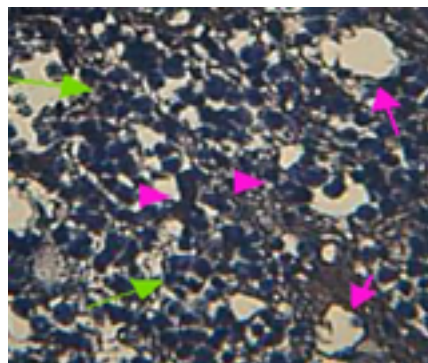


Figure (10) Osteoprogenitor cell (arrow heads) and osteoblast (arrow) show positive BMP2 expression. DAB stainX40

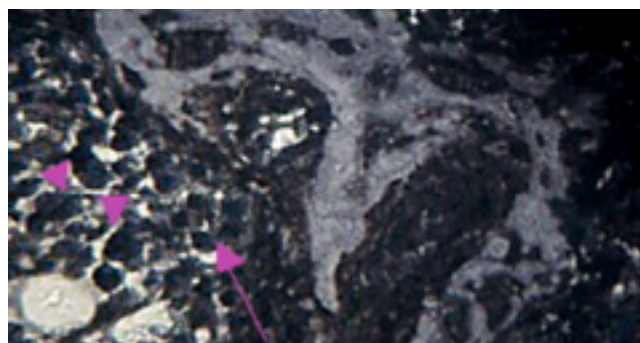
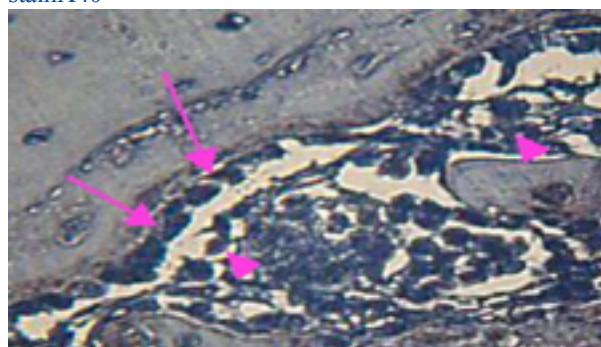


Figure (11) Osteoblast (arrows), reticular cell (arrows heads) show positive BMP2 expression in control group 10 days. DAB stainX40



Figure(12) Immunohistochemical view for positive BMP2 expressed by osteoprogenitor cell (arrows) in green tea gel group 3 days. DAB stainX20

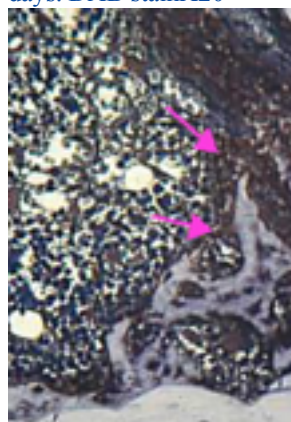
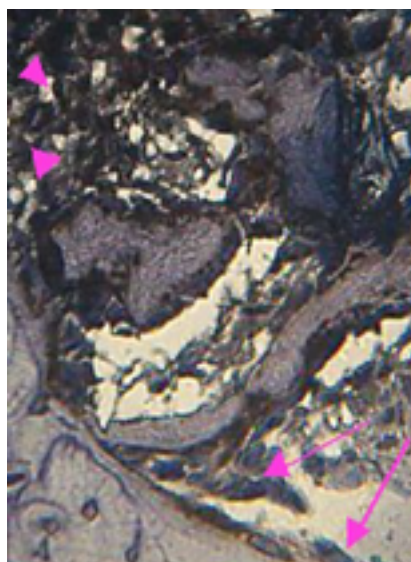


Figure (13) Osteoblast (arrows) and progenitor cell (arrow heads) expressed positive BMP2. DAB stainX40



**Control group** shows an immunohistochemical view shows positive BMP2 expression by progenitor cell , inflammatory cell and endothelial cell in control group 3 days (Figure 9). Osteoprogenitor cell and osteoblast show positive BMP2 expression at 7 day (Figure 10). Osteoblast and reticular cell show positive BMP2 expression at 10 day (Figure 11).

**Experimental group of extracted green tea gel** illustrates an immunohistochemical view for positive BMP2 expressed by progenitor cell and endothelial cell in green tea gel group 3 days (Figure 12). Osteoblast and reticular cell show positive BMP2 expression at 7 day (Figure 13). Haversian canal (HC) content and active osteocyte in green tea gel 10 days show positive BMP2 expression (Figure 14).

### Statistical analysis of results

Results for the mean of bone cells count at 7 period for all study groups illustrates a high significant difference value for green tea gel group in comparism to control group

Figure(14) Osteoblast (arrow), osteocyte (arrow heads) expressed positive BMP2 in green tea gel group 10 days. DAB stainX40

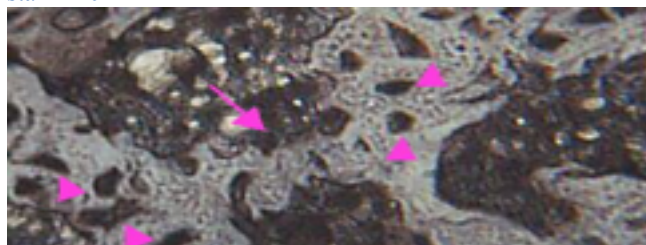


Table 1: Descriptive statistics of the bone cells count (H&E) and groups' difference in each duration

Bone cells	Duration	Groups	Descriptive Statistics				Groups' difference	
			Mean	S.D.	Min.	Max.	F-test	p-value
Osteoblast	7 days	Control	9.75	0.14	9.6	8.9	314.64	0.000 (HS)
		Green tea gel	18.23	0.19	19	19.4		
	10 days	Control	7.83	0.12	7.7	7.9	25.26	0.000 (HS)
		Green tea gel	7.85	0.08	7.8	7.9		
Osteocyte	7 days	Control	13.80	0.10	12.7	12.9	310.89	0.000 (HS)
		Green tea gel	15.25	0.18	16.1	16.4		
	10 days	Control	9.68	0.11	9.6	9.8	676.42	0.000 (HS)
		Green tea gel	15.13	0.12	15	15.2		
Osteoclast	7 days	Control	1.75	0.96	1	3	9.03	0.002 (HS)
		Green tea gel	1.88	0.11	2.8	3		
	10 days	Control	0.80	0.46	0.4	2	8.45	0.003 (HS)
		Green tea gel	0.20	0.09	0.2	0.4		

It has been shown that at 7 days duration the mean of the no. of osteoblast cells was significantly higher (18.23)  $P<0.001$  in the study group treated with green tea gel.

The mean of the no. of osteocyte cells was

significantly higher ( 15.25)  $P<0.001$  in the study group treated with green tea gel at 7 days duration.

The mean of no. of osteoclast cells was significantly higher ( 1.88)  $P<0.001$  in the study group treated with green tea gel at 7 days duration.



**Table 2: Descriptive statistics and duration difference of the positive stromal cells expressed by bone morphogenic protein 2**

Groups	Duration	Descriptive Statistics				Duration difference	
		Mean	S.D.	Min.	Max.	F-test	p-value
Control	3 days	33.53	2.47	30.2	35.5	252.24	0.000 (HS)
	7 days	30.23	0.19	30.1	30.5		
	10 days	12.35	0.21	12.1	12.6		
Green tea gel	3 days	60.33	0.17	60.1	60.5	4914.83	0.000 (HS)
	7 days	49.55	0.41	49.1	50.1		
	10 days	35.90	0.41	35.3	36.2		

**Table 3: LSD test after ANOVA test**

Groups	Duration		Mean Difference	p-value
Control	3 days	7 days	3.30	0.010 (HS)
		10 days	21.18	0.000 (HS)
	7 days	10 days	17.88	0.000 (HS)
Green tea gel	3 days	7 days	10.78	0.000 (HS)
		10 days	24.43	0.000 (HS)
	7 days	10 days	13.65	0.000 (HS)
		10 days	48.98	0.000 (HS)
	7 days	10 days	12.15	0.000 (HS)
		10 days		

Table 2,3 show the statistics and duration difference of the positive stromal cells expressed bone morphogenic protein 2 and it represents a highly significant difference values for green tea gel group in comparism to control group.

## DISSCUSION

Bone formation is a continuous process that begins during fetal development and persis throughout life as a remodeling process. In the event of injury, bones heal by generating new bone rather scar tissue (Carvalhol et al., 2012), to elucidate the extent to which skeletal regeneration affected by exogenous extracted green tea gel , we used detailed expression stages of bone apposition and ossification.

The present study shows an early ostoid deposition in extracted green tea gel group at 3 day period, as ostoid tissue formation, accompanied with significant immunopositive expression for bone morphgenic protein 2 in bone defect site. These results

could be attributed to, that the traumatic site includes, stem cells differentiate into osteoblasts (Catharino et al., 2014) that enhanced by bone morphogenic protein 2 which has been implicated in angiogenesis and the proliferative osteoblast and active osteocyte were included in deposition of collagen fiber.

At 7&10 day periods, extracted green tea gel groups show trabeculae filled a proximately the whole defect in comparism to histologic views for control group . This result can be attributed that green tea gel directly promotes the differentiation of primary osteoblast, which is responsible for bone apposition and mineralization. The placebo-treated defects were not able to create a bony bridge across the gap. In contrast, green tea gel treatment caused significant filling with bone.

The present statistics analysis for the mean value of osteoblast, active osteocyte in green tea gel group, and stromal cells that expressed immunopositive reaction for bone morphogenic protein 2 were

recorded to be a highly significant differences in comparison to control group.

By direct exposing the extracted green tea gel to bone defect, bone -forming cells from rats – called osteoblasts – to these green tea compounds, the research found that both the rate of bone growth and bone strengthening was significantly increased within a few days.

Some of this bone boosting affect comes down to the activity of a key enzyme that promotes bone growth. One particular green tea compound called Epi- gallo-catechin (EGC), led to an increase in this enzyme's activity by 79%. It also increased levels of bone mineralization in these cells: a vital part of strengthening bones. Epi- gallo-catechin also suppressed the activity of cells called osteoclast cells that weaken and break down old bone, part of the natural process of bone remodelling. This study was conducted on rat bone cells, so further research is need to see if the same affect will hold true for human bone cells, this is acoincied with Chun et al.,2009 who was founded that catechin compound that present in green tea increase the harnessed of osteoporotic bone.

Our study shown that polyphenols which present in green tea (GTP) have positive effects on bone healing, this coincide with shen et al.,2008 who stated that the GTP prevented the aging-induced as well as aging plus estrogen deficiency-induced reduction in femoral bone mass.

## REFERENCE

1. Ando K, Mori K, Rédini F, Heymann D. RANKL/RANK/OPG: key therapeutic target in bone oncology. *Curr Drug Discov Technol*. 2008;5(3):263–8.
2. Carvalhol, L.R., Breyner, N., Hell, R., Valerio P. Healing Pattern in Calvaries Bone defects following bone regeneration in rats guided by chitosan scaffold and adipose tissue-derived mesenchymal stem cells. *The open Tissue Engineering and Regenerative Medicine Journal*. 2012; 5, 25-34.
3. Catharino P.C., Dominguez GC., Pinto Ddos S. Jr., Morea. Histomorphometric, and Radiographic Monitoring of Bone Healing Around Inoffice-Sterilized Orthodontic Mini-implants With or Without Immediate Load: Study in Rabbit Tibiae. *IntJ. Oral Maxillofac Implants*. 2014;Mar-Apr 29(2), 321-30.
4. Choi EM, Hwang JK. Effects of (+)-catechin on the function of osteoblastic cells. *Biol Pharm Bull*.2003;26(4):523–6.
5. Chun Hay Ko, Kit Man Lau, Wing Yee Choy, Ping Chung Leung. Effects of Tea Catechins, Epigallocatechin, Galliccatechin, and Galliccatechin Gallate, on Bone Metabolism. *Journal of Agriculture and Food Chemistry*. 2009; 57, 7293–7297.
6. Chwan-Li Shen, James K. Yeh, Jay J. Cao, Ming-Chien Chyu, and Jia-Sheng Wang. Green Tea and Bone Health: Evidence from Laboratory Studies *Pharmacol Res*. 2011Aug; 64(2): 155-161.
7. Hillmann, G., Gebert, A., Geurtsen W. Matrix expression and proliferation of primary gingival fibroblasts in three dimensional cell culture mode, *Jounal of Cell Science*. 1999; 117. 2823-2832.
8. Matsuo K, Irie N. Osteoclast-osteoblast communication. *Arch BiochemBiophys*. 2008;473(2):201–9
9. Nijweide PJ, Burger EH, Feyen JH. Cells of bone: proliferation, differentiation, and hormonal regulation. *Physiol Rev*. 1986;66(4):855–86. Review].
10. Shen CL, Wang P, Guerrieri J, Yeh J, Wang JS. Protective effect of green tea polyphenols on bone loss in middle-aged female rats. *Osteoporosis Int*. 2008;19(7):979–90.
11. Shen CL, Wang P, Guerrieri J, Yeh J, Wang JS. Protective effect of green tea polyphenols on bone loss in middle-aged female rats. *Osteoporosis Int*. 2008;19(7):979–90.
12. Shen CL, Yeh JK, Cao JJ, Tatum OL, Dagda RY, Wang J-S. Green tea polyphenols mitigate bone loss of female rats in a chronic inflammation-induced bone loss model. *J Nutr Biochem*. 2009;21:968–74. 2010.
13. Shen CL, Yeh JK, Samathanam C, Cao JJ, Stoecker BJ, Dagda RY, Chyu MC, Dunn DM, Wang JS. Green tea polyphenols attenuate deterioration of bone microarchitecture in female rats with systemic chronic inflammation. *Osteoporosis Int*. 2010 Mar 20; [Epub ahead of print]. PMID: 20306019.
14. Shen CL, Yeh JK, Stoecker BJ, Chyu MC, Wang JS. Green tea polyphenols mitigate deterioration of bone microarchiteture in middle-aged femalerats. *Bone*. 2009;44(4):684–90.
15. Singer F.R, and Eyre D.F. 2011. Using biochemical markers of bone turnover in clinical practice. *Cleveland Clin J Med*. 75 (10): 73-750.
16. Wang JS, Luo H, Wang P, Tang L, Yu J, Huang T, et al. Validation of green tea polyphenol biomarkers in a phase II human intervention trial. *Food Chem Toxicol*. 2008;46(1):232–40.
17. Yang CS, Landau JM. Effects of tea consumption on nutrition and health. *J Nutr*. 2000;130:2409–12.



# Cyclic Fatigue Resistance of New Endodontic Files in Reciprocal vs. Rotational Motion.

**Makdad Chakmakchi**

B.D.S, Ph.D. Assistant Professor. University of Mosul, College of Dentistry

**Ashraf Salim Alchalabi**

B.D.S, M.Sc. Ph.D. Student. Department of Conservative Dentistry. College of Dentistry

## ABSTRACT

**Objectives:** To evaluate the flexural fatigue resistance of WaveOne Primary and Protaper NEXT X2 NiTi files used in reciprocating and rotation motions.

**Materials and methods:** A 20 files, 25mm long for use in reciprocating [Group 1: WaveOne Primary, n=10] and continuous rotation [Group 2: Protaper NEXT X2, n=10] were selected. All instruments were operated in a static model for cyclic fatigue testing which was conducted in a device that allowed for a reproducible simulation of an instrument confined in a curved canal. The instruments were activated by a torque-controlled motor [X-smart plus, Dentspy Maillefer] using the preset program WaveOne ALL for the WaveOne instruments and the preset program for the Ppotaper NEXT X2 instruments. All instruments were rotated until fracture occurred. The number of cycles to failure [NCF] for each instrument was calculated. The length of the fractured file tip was measured. NCF data were statistically analyzed by Student *t*-test.

**Results:** The statistical analysis revealed a statistically significant differences [ $p < 0.001$ ] between two groups. WaveOne primary instruments were associated with a significantly greater NCF.

**Conclusions:** The test results showed significantly increased cyclic fatigue resistance of the file with reciprocal motions compared to continuous rotation. No differences were found between the lengths of the fractured segment.

## KEYWORDS

number of cycles to failure, nickel-titanium file, reciprocation, fatigue resistance, endodontic files.

## مقاومة التعب الدوري للملفات اللبية الجديد في الحركة المتبادلة مقابل الحركة الدورانية.

مقداد جقماقجي

استاذ مساعد, كلية طب الاسنان - جامعة الموصل

أشرف سالم الجليبي

مدرس, كلية طب الاسنان - جامعة الموصل

## المستخلص

اهداف: لتقييم مقاومة التعب للملفات WaveOne Primary و Protaper NEXT X2 المستخدمة في الحركات التبادلية والدورانية. المواد والعمل: تم اختيار ٢٠ ملف ذات طول ٢٥ ملم [المجموعة ١ WaveOne Primary،  $n = 10$ ] والمجموعة ٢ في الحركة الدورانية [المجموعة ٢: Protaper NEXT X2،  $n = 10$ ] وتعمل جميع الملفات في نموذج ثابت لاختبار التعب الدوري. وتم اختبار فعالية هذه الملفات باستخدام جهاز بديل عن قناة عصب السن. حيث تدور الملفات داخل قناة منحنية، حيث يتم السيطرة على عزم الدوران بواسطة جهاز [X-smart plus, Dentspy Maillefer] تعمل ببرامج متخصصة للملفات المستخدمة WaveOne ALL { لفايلات WaveOne, Protaper NEXT X2 } بهذا الفحص يتم تناوب الفايلات لغايه حدوث كسر. ومن ثم حساب عدد دورات الفشل [NCF] لكل أداة وقياس طول حافة الملف الممزق. ومن ثم تحليل هذه البيانات إحصائياً باستخدام اختبار *t*. النتائج: أظهرت نتائج التحليل الإحصائي فروق ذات دلالة إحصائية [ $p > 0.001$ ] بين مجموعتين. ارتبطت الأدوات الأساسية WaveOne مع أكبر بكثير من [NCF]

الاستنتاجات: نتائج الاختبار أظهرت زيادة كبيرة في مقاومة التعب دوري الملف مع حركات متبادلة مقارنة مع دوران مستمر. لم يتم العثور على الاختلافات بين أطوال الجزء الممزق.

## INTRODUCTION

The introduction of rotary nickel-titanium [NiTi] endodontic instruments into clinical practice has improved the efficacy of endodontic treatment in terms of procedural time, accuracy, and risk reduction (1-3). However, despite these advantages, unexpected instrument fracture is not uncommon (1). Sattapan et al identified two modes of fracture for arotary NiTi instruments: torsional failure and cyclic fatigue (4). The torsional failure occurs when the tip or any part of the instrument is locked into the canal, while rotary motion continues. The fracture caused by fatigue failure is caused by flexure. The rotating instrument in a root canal curvature, is subjected to tension/compression cycles at the point of maximum flexure,

until fracture occurs (5,6). During rotation, half of the instrument shaft is in tension (outer curved part), whereas the rest is in compression (concave part). This repeated tension-compression cycle loading, increases cyclic fatigue of the instrument over time and has been identified as an important factor in instrument fracture (5, 6).

Cyclic fatigue resistance of a file system is usually tested by counting the number of rotations of the instrument in an artificial root canal until fracture (7). The clinical concern is the fact that cyclic fatigue occurs with no visible signs of plastic deformation. Therefore, clinicians should be aware of the most influential factors that contribute to this type of failure, to prevent instrument fracture (8).

To improve the fracture resistance of rotary nickel titanium files, manufacturers have introduced new alloys and developed new manufacturing processes<sup>(8)</sup>. M-wire is a new NiTi alloy prepared by a special thermal process incorporating three crystalline phases (deformed and micro-twinned martensite, R-phase and austenite). This structure is believed to offer increased flexibility and resistance to cyclic fatigue in comparison with conventional nickel titanium alloys<sup>(9)</sup>.

In addition, the use of reciprocating motion has been shown to extend the lifespan of a NiTi instrument in comparison with continuous rotation<sup>(10,11)</sup>.

Recently, a new brand of NiTi instruments were introduced to the market based on the above developments: WaveOne Primary (Dentsply Maillefer, Ballaigues, Switzerland), a 4<sup>th</sup> generation reciprocal file, has been claimed to reduce torsional stress by periodically reversing the file and therefore increasing the instrument life span<sup>(10,12)</sup>. Protaper NEXT X2 [Dentsply Maillefer] a 5<sup>th</sup> generation rotary file has been launched with an exceptional shaping capacity of severely curved narrow canals, a procedure that was impossible with most NiTi systems commercially available so far.

The reciprocal motion has been shown to offer important advantages for the preparation of curved canals by using a single NiTi file<sup>(13-16)</sup>. The concept of using a single NiTi instrument to prepare the entire root canal is interesting, because the technique is simple, cost effective with a shorter learning curve<sup>(17)</sup>.

Currently, there is limited information about the comparative fatigue performance of these systems. The aim of the present study was to evaluate the flexural fatigue resistance of WaveOne Primary and Protaper NEXT X2 NiTi files used in reciprocating and continuous rotation motions. The null hypothesis was that there is no statistically significant difference between the two systems in the property tested.

## MATERIALS AND METHODS

A total number of 20 files, 25mm long for use in reciprocating motion [Group 1: WaveOne Primary, n=10] and continuous rotation [Group 2: Protaper NEXT X2, n=10] were selected for this study. The reciprocating instruments had an ISO size 25 at the tip, a taper of .08 at the apical 3 mm with a decreasing and variable taper up to the end of their working part. The continuous rotation instruments had an ISO size 25 at the tip, a taper of .06 at the apical 3 mm with a decreasing and variable taper up to the end of their working part and an off-centered, rectangular cross

section. All files were free of defects or deformities confirmed under a stereomicroscope [SZR-10; Optika, Bergamo, Italy].

A static model for cyclic fatigue testing was conducted in a custom-made device that allowed for a reproducible simulation of an instrument confined in a curved canal, similar to that described by Plotino et al<sup>(17)</sup>. The artificial canal was manufactured by reproducing the instrument's size and taper, thus providing the instrument with a suitable trajectory that conforms to the parameters of the chosen curvature (Figure 1).

Tempered steel was subjected to computer-aided milling to produce an artificial canal of the following dimensions: Total length of 20 mm starting with a 12 mm straight part with entrance of 1.3 mm diameter, 60° angle of curvature measured according to Schneider<sup>(18)</sup>, 5 mm radius of curvature and length of curved segment, 3 mm straight part after curvature and 0.6 mm apical diameter.

The instruments were activated by using a 6:1 reduction handpiece powered by a torque-controlled motor [X-smart plus, Dentsply Maillefer] using the preset program WaveOne ALL for the WaveOne instruments and the preset program for the Protaper NEXT X2 instruments.

To reduce file friction with artificial canal walls, high-flow synthetic lubricating oil (Super Oil, Singer Co Ltd, Elizabethport, NJ, USA) was applied and the free canal surface was covered with glass to prevent the instruments from slipping out<sup>(19)</sup>.

All instruments were rotated until fracture occurred. The time to fracture was recorded and stopped as soon as a fracture was detected visually and/or audibly using a chronometer (accuracy  $\pm 0.01$  s) and registered to the nearest integer second. The length of the fractured tip was also recorded for each instrument.

The number of cycles to failure (NCF) for each instrument was calculated by multiplying the time (in s) to failure by the number of rotations or cycles per second regardless of the rotational direction (350 rpm for WaveOne all and 300 rpm for PTN X2 according to the manufacturer). The length of the fractured file tip was measured by a digital caliper (Whitworth, China), accurate to  $\pm 0.01$  mm.

NCF data were statistically analyzed by Student *t*-test with a significance level set at 95% ( $\alpha=0.05$ ) employing IBM SPSS statistics v 19.0.0 software.

## RESULTS

The results of NCF values of the groups tested are

presented in Table 1. The statistical analysis revealed a statistically significant differences ( $p < 0.001$ ) between two groups. WaveOne primary instruments were associated with a significantly greater NCF.

The mean length of the fractured segment was also recorded in millimeter (mm) to evaluate the correct positioning of the tested instrument inside the canal curvature and whether similar stresses were being induced. No statistically significant difference ( $P < 0.01$ ) in the mean length of the fractured fragments was evident for the instruments (Table 2).

## DISCUSSION

The aim of this study was to investigate the effect of the type of the file movement on the cyclic fatigue of nickel–titanium instruments of the reciprocal WaveOne and latest rotary PTN X2 files. The results showed a statistically significant difference in the number of cycles to failure (NCF) but not in the fragment lengths between the two products. Therefore the testing hypothesis should be partially rejected for NCF.

File fracture is a major concern during endodontic treatment in curved root canals when using NiTi instruments. The structural characteristics and geometric designs have a critical influence on the fracture susceptibility of NiTi files<sup>(20)</sup>. Movement kinematics (continuous or reciprocating motion) and speed are included among the factors determining the resistance of rotary NiTi instruments to cyclic fracture<sup>(10)</sup>. Previous studies have shown that the fatigue life is affected by the radius and angle of the curvature<sup>(21,22)</sup>.

Several different methods have been used to test cyclic fatigue resistance of NiTi files. According to Yao et al<sup>(23)</sup>, the use of standardized artificial canals in cyclic fatigue experiments minimizes the influence of other variables. In this study, the artificial canal used was manufactured reproducing the instrument's size and taper to standardize the conditions.

Selection of stainless steel to simulate canals was made to minimize wear of canal walls during instrumentation<sup>(20,24,25)</sup>. Also a 5-mm radius was chosen to simulate an abruptly curved canal.

Several non-tooth devices<sup>(26)</sup> were used to investigate in vitro cyclic fatigue resistance in both static and dynamic models. Although dynamic models better simulate clinical brushing or pecking motions<sup>(27)</sup>, the instruments tested are not constrained in a precise trajectory. Moreover, regardless standardization of amplitude and frequency of the axial movements these variables are completely subjective on reproducing the motions manually<sup>(28)</sup>.

In the present study, both instruments were made by

the same alloy produced with the same proprietary thermal treatment (M-wire). Thus, even in this case, different results between the instruments should not be related to their metallurgical behavior.

In static models all confounding causes apart from cyclic fatigue are minimized. The results of the present study showed that reciprocal motions (ie, WaveOne Primary) significantly increased the cyclic fatigue resistance of tested file compared with continuous rotation independently from different cross-sections. In agreement with these results, other recent articles reported a higher cyclic fatigue resistance of reciprocating motion than continuous rotation in instruments specifically designed to be used in reciprocal motion as well as in those manufactured for continuous rotation use<sup>(12,29)</sup>. It has been postulated that the increased fatigue resistance occurs because of the release of reaction stresses built up in the material by reversing the rotational direction<sup>(10,11)</sup>.

Although the PTN X2 system had a lower rotations speed (300 rpm) compared to WaveOne Primary system (350 rpm) but the latter showed a higher cyclic fatigue resistance, probably because of the reciprocating movement. The instrument rotates 60° in each cycle, which means that after 6 cycles, the instrument completes one entire rotation (360°)<sup>(30)</sup>. Thus the instrument is subjected to lower tensile and compressive stress, operating for a longer period of time before failure<sup>(28)</sup>. This research reached similar results as in other studies. A reciprocating motion may decrease the impact of cyclic fatigue on NiTi rotary instrument life compared with rotational motion<sup>(10-12)</sup>.

Also a possible explanation of the different results among the instruments tested in this study can be related to the different cross sectional designs. It has been shown that cross-sectional design has an impact on the stress developed by an instrument under either tension or bending<sup>(16,31)</sup>. Protaper NEXT X2 is made of M-Wire and has an off-centered rectangular cross-section, whereas WaveOne Primary files have a modified convex triangular cross-section at the tip and a convex triangular cross-section in the middle and coronal portion of the instrument.

Although comparisons among the different brands is difficult to make because of their differences in movement, design and cross-sectional areas, the results presented here can be useful for clinical practice.

In conclusion, under these experimental conditions, the reciprocal motions showed significantly increased cyclic fatigue resistance of the file tested

compared with continuous rotation independently from their cross section. No differences were found between the lengths of the fractured segment in the two tested files.

#### Acknowledgement

Sincere appreciation to Professor Dr George Eliades, Director, Department of Biomaterials,

University of Athens, School of Dentistry, Greece, for revising the original manuscript.

#### COMPLIANCE WITH ETHICAL STANDARDS

##### Conflict of interest:

The author Makdad Chakmakchi confirm that

this article content has no conflicts of interest.

The author Ashraf Salim Alchalabi confirm that this article content has no conflict of interest.

##### Funding:

The work was supported by the Department of Conservative Dentistry, College of Dentistry, the University of Mosul, Iraq.

##### Ethical approval:

This article does not contain any studies with human participants or animals performed by any of the authors.

##### Informed consent:

For this type of study, formal consent is not required.



**Figure 1.** The static model used for cyclic fatigue testing.

Table 1. Results of NCF.

Groups	N	Minimum	maximum	Mean±SD	SE	t	p-value
1.WaveOne Primary	10	310	580	424±82	26	6.06	p<001S
2. Protaper NEXT X2	10	180	300	240±33	10		

SD= std.deviation

S= significant

Table 2. Results of fragment lengths [in mm].

Groups	N	Minimum mm	maximum mm	Mean±SD	SE	t	p-value
1.WaveOne Primary	10	3.97	5.0	4.66± 0.36	0.12	1.84	P<0.09NS
2. Protaper NEXT X2	10	3.61	4.97	4.34±0.43	0.14		

SD= std.deviation

NS= not significant



## REFERENCES

1. Parashos P, Messer HH. Rotary NiTi instrument fracture and its consequences J Endod 2006; 32: 1031–43.
2. Taschieri S, Necchi S, Rosano G, et al. Advantages and limits of nickel-titanium instruments for root canal preparation: a review of the current literature Schweiz Monatsschr Zahnmed 2005; 115 (11): 1000–5.
3. Walia HM, Brantley WA, Gerstein H. An initial investigation of the bending and torsional properties of Nitinol root canal files J Endod 1988; 14: 346–51.
4. Sattapan B, Palamara JE, Messer HH. Torque during canal instrumentation using rotary nickel-titanium files J Endod 2000; 26: 156–60.
5. Parashos P, Gordon I, Messer HH. Factors influencing defects of rotary nickel titanium endodontic instruments after clinical use J Endod 2004; 30: 722–5.
6. Peters OA. Current challenges and concepts in the preparation of root canal systems: a review J Endod 2004; 30: 559–65.
7. Lee M, Versluis A, Kim B, et al. Correlation between experimental cyclic fatigue resistance and numerical stress analysis for nickel-titanium rotary files J Endod 2011; 37: 1152–7.
8. Saber SE. Factors influencing the fracture of rotary nickel-titanium instruments. ENDO [Lond Engl] 2008; 2: 273–283.
9. Saber SE. The effect of instrument material, taper and degree of root canal curvature on cyclic fatigue of rotary nickel-titanium instruments. ENDO [Lond Engl] 2013; 1: 59–64.
10. De-Deus G, Moreira EJ, Lopes HP, Elias CN. Extended cyclic fatigue life of F2 ProTaper instruments used in reciprocating movement. Int Endod J 2010; 43: 1063–8.
11. You SY, Bae KS, Baek SH, Kum KY, Shon WJ, Lee W. Lifespan of one nickel-titanium rotary file with reciprocating motion in curved root canals J Endod 2010; 36: 1991–4.
12. Varela-Patino P, Ibanez-Parraga A, Rivas-Mundina B, Cantatore G, Otero XL, Martin-Biedma B. Alternating versus continuous rotation: a comparative study of the effect on instrument life J Endod 2010; 36: 157–9.
13. De-Deus G, Brandao MC, Barino B, Di Giorgi K, Fidel RA, Luna AS. Assessment of apically extruded debris produced by the single-file ProTaper F2 technique under reciprocating movement Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2010; 110: 390–4.
14. Franco V, Fabiani C, Taschieri S, Malentacca A, Bortolin M, Del Fabbro M. Investigation on the shaping ability of nickel–titanium files when used with a reciprocating motion J Endod 2011; 37: 1398–1401.
15. Paque F, Zehnder M, De-Deus G. Microtomography based comparison of reciprocating single-file F2 ProTaper technique versus rotary full sequence J Endod 2011; 37: 1394–7.
16. You SY, Kim HC, Bae KS, Baek SH, Kum KY, Lee W. Shaping ability of reciprocating motion in curved root canals: a comparative study with micro-computed tomography J Endod 2011; 37: 1296–300.
17. Plotino G, Grande M, Testarelli L and Gambarini G. Cyclic fatigue of Reciproc and WaveOne reciprocating instruments Int Endod J 2012; 45: 614–8.
18. Schneider SW. A comparison of canal preparations in straight and curved root canals. Oral Surg 1971; 32: 271–5.
19. Pedulla E, Grande NM, Plotino G, Gambarini G, Rapisarda E. Influence of continuous or reciprocating motion on cyclic fatigue resistance of 4 different nickel-titanium rotary instruments J Endod 2013; 39: 258–61.
20. Kim HC, Cheung GS, Lee CJ, Kim BM, Park JK, Kang SI. Comparison of forces generated during root canal shaping and residual stresses of three nickel-titanium rotary files using three-dimensional finite-elements analysis J Endod 2008; 34: 743–7.
21. Grande NM, Plotino G, Pecci R, Bedini R, Malagnino VA, Somma F. Cyclic fatigue resistance and three-dimensional analysis of instruments from two nickel titanium rotary systems Int Endod 2006; J 10: 755–63.
22. Pruett JP, Clement DJ, Carnes DL. Cyclic fatigue testing of nickel-titanium endodontic instruments J Endod 1997; 23: 77–85.
23. Yao JH, Schwartz SA, Beeson TJ. Cyclic fatigue of three types of rotary nickel-titanium files in a dynamic model J Endod 2006; 32: 55–7.
24. Gambarini G. Cyclic fatigue of Profile rotary instruments after prolonged clinical use Int Endod J 2001; 34: 386–9.
25. Oh SR, Chang SW, Lee Y, Gu Y. A comparison of nickel-titanium rotary instruments manufactured using different methods and cross-sectional areas: ability to resist cyclic fatigue Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2010; 109: 622–8.
26. Plotino G, Grande NM, Cordaro M, et al. A review of cyclic fatigue testing of nickel-titanium rotary instruments J Endod 2009; 35: 1469–76.
27. Li UM, Lee BS, Shin CT, et al. Cyclic fatigue of endodontic nickel titanium rotary instruments: static and dynamic tests J Endod 2002; 28: 448–51.
28. Wan J, Rasimick BJ, Musikant BL, Deutsch AS. A comparison of cyclic fatigue resistance in reciprocating and rotary nickel-titanium instruments Aust Endod J 2011; 37: 122–7.
29. Gavini G, Caldeira CL, Akisue E, et al. Resistance to flexural fatigue of Reciproc R25 files under continuous rotation and reciprocating movement J Endod 2012; 38: 684–7.
30. Webber J, Machtou P, Pertot W, Kuttler S, Ruddle C, West J. The WaveOne single-file reciprocating system. Roots 2011; 1: 28–33.
31. Cheung GS, Zhang EW, Zheng YF. A numerical method for predicting the bending fatigue life of NiTi and stainless steel root canal instruments Int Endod J 2011; 44: 357–61.

# Comparing Maximum Stresses and Displacements in A Lower Complete Denture with Different Occlusal Plane Levels and Schemes . A Three Dimensional Finite Element Stress Analysis Study

Abdalbasit Ahmed Fatiallah

B.D.S M.Sc., Ph.D

Assistant Professor college of dentistry University of Baghdad Iraq

Suza A. Faraj

B.D.S M.Med.Sci

Professor college of dentistry University of Baghdad Iraq

## ABSTRACT

**Purpose of study:** A three dimensional stress analysis method was used to assessed the stress distribution and displacement in a lower complete denture

**Materials and Methods:** The sample consists of three groups , First group :three set of lower complete denture constructed using cast poured from ready made teaching mould to study the effects of balanced ,lingualized and monoplane occlusal schemes on the stress distribution when the occlusal plane leveled with upper, middle and lower third of the retro molar pad .the load used in this study was 58.8 Newton directed axially downward applied on specific sites differ with different occlusal schemes.

**Results:** Both balanced and lingualized occlusal schemes exert minimal stresses and rotational movements around ridge crest when leveled with middle third of retro molar pad while occlusal plane leveled with the lower third of retro molar pad shows more incidence for rotational movements

**Conclusion:** Stability improved in monoplane occlusal plane over the three occlusal plane levels the posterior of teeth should be adjusted over the ridge crest so that the lateral stresses reduced and increased denture stability.

## KEYWORD:

Occlusal plane ,lower denture and finite element stress analysis.

محصلة الاجهادات و الازاحات الناتجة في الطقم السفلي الكامل لعدة مفاهيم و مستويات للاتباق دراسة  
بواسطة

طريقة العناصر المحددة ذات الثلاثة أبعاد

## المستخلص

ان واحدة من أصعب المقاييس التجريبية هي محاولة توزيع القوة على النسيج الساند للطقم بشكل كامل. استعملت طريقة العناصر المحددة ذات الثلاثة أبعاد لتحليل توزيع الاجهادات و الأزاحة لطقم الأسنان السفلي الكامل. تتكون العينات من ثلاثة مجموعات: المجموعة الأولى: يتكون من ثلاث مجموعات لأطقم الأسنان الكاملة، كل مجموعة شيدت بنظام أطباق مختلف عن نظام أطباق المجموعتين الأخريين «أطباق متوازن و أطباق حنكي و أطباق أحادي الأسطح» مستوى الأطباق لهذه المجموعات ضبط مع الثلث العلوي لرفادة خلف الأرحاء. المجموعة الثانية: يتكون من ثلاث مجموعات لأطقم الأسنان الكاملة، كل مجموعة شيدت بنظام أطباق مختلف عن نظام أطباق المجموعتين الأخريين «أطباق متوازن و أطباق حنكي و أطباق أحادي الأسطح» مستوى الأطباق لهذه المجموعات ضبط مع الثلث الوسطي لرفادة خلف الأرحاء. المجموعة الثالثة: يتكون من ثلاث مجموعات لأطقم الأسنان الكاملة، كل مجموعة شيدت بنظام أطباق مختلف عن نظام أطباق المجموعتين الأخريين «أطباق متوازن و أطباق حنكي و أطباق أحادي الأسطح» مستوى الأطباق لهذه المجموعات ضبط مع الثلث السفلي لرفادة خلف الأرحاء. الحمل الذي استعمل في هذه الدراسة 58.8N يشكل تحتي محوري مسلط على مواقع محددة تختلف باختلاف مفاهيم الأطباق. النتائج كشفت بأن كلا من الأطباق المتوازن والحنكي سلط اجهاد وحركات تدويرية أقل مايمكن عندما كان مستوى الأطباق مع الثلث الوسطي لرفادة خلف الأرحاء , أيضا بأقتراب السطح الأتباقي للأسنان من الحافة العليا للحرز السنخي الساند للطقم تزداد احتمالية حدوث الحركات التدويرية. بينما أظهر الأطباق أحادي الأسطح استقرارا أكثر و قلة في الحركات التدويرية.

## INTRODUCTION:

Conflict thought concerning the relative merits of occlusal scheme and the debated problems whether the anatomic or non anatomic posterior teeth are more desirable used for constructing complete denture, so to resolve these controversy stresses and denture base deformation reaching the residual ridge determined utilizing a method more concerning with geometrical concept superimposed by physiological requirement to restore and maintain oral health.

Verifying complete denture occlusion intra orally is difficult and unreliable because of the resiliency of the supporting tissue and temporomandibular joint and also because of defective tooth contact so that remounting the denture on an articulator and equilibrating it is an accepted method, then a simulated model transferred to computer in which the analysis carried out. <sup>(1)</sup>

There has been a great deal of interest in the force distribution and base deformation of complete

denture with various tooth forms during function. <sup>(2)</sup>

The distance between occlusal plane and alveolar crest has an influence on the stability of lower complete denture; the precise location of the occlusal plane is rather a controversial issue. <sup>(3)</sup>

## **MATERIALS AND METHOD**

The sample grouped so that three sets of complete denture constructed using cast poured from ready made teaching mould Colombia dentoform corporation U.S.A to study the effect of balanced , lingualized and monoplane occlusal schemes on the stress distribution and displacements when the occlusal plane leveled with upper , middle and lower third “<sup>(4)</sup>, <sup>(5)</sup>”

The imaginary occlusal plane of occlusal rim corresponding to the horizontal plane and the relation of the edentulous arch is class I , then the articulator set so that the sagital and condylar guidance at 30 and 15 degree respectively, and with sagital and condylar inclination at 10 and 0 degree respectively. <sup>(6)</sup>

The lower posterior teeth were first arranged in the three concept of occlusion with the reliable landmarks considered to orient the occlusal plane and position the teeth which is the retro molar pad.

After curing , a section made in each denture at the first molar region mesially and distally to obtain a Bloch section by means of laboratory hand piece and engine ( Q.D. England) with 0.6 mm disk bur. To facilitate the modeling process and the data input to the programm used (ANSYS 5.4 program version “Swenson analysis system, Houston ,Pennsylvania) the following steps were applied;

- 1.The mesial and distal area of the denture base were drawn on a grid paper with its exact dimensions through super imposition of them on the grid paper, x and y coordinate system obtained for a specific key points to both mesial and distal areas of the denture base.
- 2.The distance between the mesial and distal area of the denture base block section obtained by the means of Vernier which represent the Z-value in space that change the model from two dimensional to three dimensional model.
- 3.The mesial and distal areas joined at their key points by lines then the lines converted to areas and the areas converted to volumes which represent the denture base.
- 4.Modeling of the tooth made by the aid of Vernier through obtaining the real dimension with the selection of a specific key points.
- 5.the mucosal thickness under the denture supposed to be 1.5mm <sup>(7)</sup>

6.The model “tooth, denture base and mucosa” glued together in order to act as one unit through ANSYS options when applying the boundary conditions.

7.Materials properties used in the study mentioned in Table 1.

Boundary condition include the following :

Displacements and load application. For displacements the surface of the mucosa that cover the residual ridge its nodes considered to be fixed in all degree of freedom while load application done according to the site of contacts with the opposing artificial upper teeth in centric

## **RESULTS**

A load of 58.8 Newton applied and Von mices stresses obtained at specific selected nodes located at the crest of the ridge, middle of the buccal flange and the middle of the lingual flange. <sup>(10)</sup> Fig. 1

When Comparing maximum stresses among occlusal plane levels in each occlusal scheme a highly significant difference shown among Balanced occlusal scheme leveled with upper third , lower third and middle third of retro molar pad “B1,B2 and B3 Respectively’ with the lowest value in B2 and the highest value in B1.for lingualized occlusal schemes a similar results obtained. as shown in Table 2 . In case of monoplane occlusal scheme there is no significant different among the three occlusal plane levels in the three sites of measurements

Comparing maximum stresses among occlusal scheme within each level revealed that a highly significant difference among occlusal schemes when occlusal plane leveled with the upper third of retro molar pad with the highest stress value in M1 and the lowest stress value in L1 similar results for the occlusal plane leveled with the middle third of retro molar pad obtained. While for occlusal plane leveled with lower third of retro molar pad also highly significant appeared among them with the highest stress value at B3and the lowest one at L3. Table 2

## **DISCUSION**

When comparing the value of stresses in B1,B2 and B3, B2 transmits minimal vertical and lateral stresses to the underlying mucosa and similar results with lingualized occlusal scheme this is may be due to the distance between the occlusal surface and ridge crest, the denture base thickness also reduced result in more stresses generated within it. Fig 2,3

While monoplane occlusal scheme posses no statistical difference among the three levels of occlusal plane this is may be due to the fact that the load distributed over a wide area of occlusal surface results in even distribution of stresses through out the

denture base. Fig 4

When comparing the three occlusal schemes, there were highly significant differences in the stress values at some measurement points on the supporting structure at mucosal-denture base interface in centric occlusion, this is may be due to the buccolingual position of mandibular denture teeth and the point of occlusal load application were differ and this is in agreement with Ohguri et al <sup>(7)</sup> who conducted a study to show the influence of occlusal schemes on the pressure distribution under a complete denture by comparing fully balanced and lingualized occlusion using pressure transducer attached to simulated dentures.

The monoplane occlusion in all levels considered to transmits the highest stress values to the underlying mucosa than the other occlusal schemes this is in agreement with Kelsey et al <sup>(11)</sup> who found that greater pressure values obtained with flat zero degree teeth than other type of occlusion “anatomic teeth”.

Lingualized occlusion on the other hand transmits minimal occlusal load than the other occlusal schemes due to the fact that the number of occlusal contacts are reduced considerably in that only the palatal cusps of the posterior teeth made contacts in centric relation with the central fossa of the lower posteriors, while the buccal cusps are out of contact. Therefore there is only one centric stopper between upper and lower antagonist posterior teeth.

When occlusal plane leveled with middle third of retro molar pad minimal resultant displacements take place in balanced and lingualized occlusion through out the denture base with no rotational movements occurs, this is because the direction of occlusal force more vertical than lateral force causing the denture base to displace vertically. Fig. 5

As the balanced occlusion leveled with the upper third of retro molar pad , it posses minimal rotational movements around the ridge crest in buccal

direction associated with vertical one. On the other hand lingualized occlusal schemes when compared with balanced occlusal scheme performed a more stable movements because it displaced in vertical and slightly lingual direction. Fig 6

In Fig 7 monoplane occlusal scheme results in less trauma to the underlying mucosa because it displaced only in vertical direction.

When occlusal plane leveled with the upper third of RMP the displacement in balanced occlusion was significantly higher than the displacement in both monoplane occlusion and lingualized occlusion, this is in agreement with Stromberg <sup>(12)</sup> in which he found that by eliminating cusps and using non anatomic teeth we are going to reduce the lateral displacements against the supporting tissue and with Kydd <sup>(2)</sup> who found that 33 degree posterior teeth produce greater denture base deformation nearly 50% more than non anatomic flat teeth

While the resultant displacements in monoplane occlusal scheme, when leveled with the middle third of retro molar pad is significantly higher than both balanced and lingualized occlusal scheme with more vertical displacement taking place this is in disagreements with <sup>(2), (13)</sup>

## CONCLUSION

- 1.Lingualized occlusion transmits minimal stresses to the underlying mucosa at all levels when compared with balanced and monoplane occlusion.
- 2.In balanced and lingualized occlusal schemes, when reducing the occlusal plane surface of teeth more incidences of rotational movements will occur.
- 3.In monoplane occlusal scheme stability improved over the three occlusal plane levels and does not exert rotational movements around the ridge crest.
- 4.Posterior teeth should be adjusted over the ridge crest so that the lateral displacements and stresses reduced and increase denture stability.

**Table 1 shows materials properties used in the finite elements study**

<i>Material</i>	<i>Young modulus “GPa”</i>	<i>Poison’s Ratio</i>	<i>Authors</i>
Tooth	2.65	0.35	Craig <sup>(8)</sup>
Acrylic resin denture base	2.65	0.35	Craig <sup>(8)</sup>
Alveolar Mucosa	7.5	0.45	Larabee and Glate <sup>(9)</sup>



**Table 2 Stress values in (MPa) At the mucosa-denture base interface in centric occlusion.**

	<i>Balanced occlusal concept</i>			<i>Lingualized occlusal concept</i>			<i>Monoplane occlusal concept</i>		
	<i>Level of Occlusal plane</i>	<i>Mean stresses (MPa)</i>	<i>St.D Of Mean</i>	<i>Level of Occlusal plane</i>	<i>Mean stresses (MPa)</i>	<i>St.D Of Mean</i>	<i>Level of Occlusal plane</i>	<i>Mean stresses (MPa)</i>	<i>St.D Of Mean</i>
Crest of the Ridge	B1	0.098	0.024	L1	0.050	0.010	M1	0.087	0.0057
	B2	0.092	0.0025	L2	0.056	0.0043	M2	0.088	0.010
	B3	0.311	0.0012	L3	0.198	0.0046	M3	0.090	0.011
Middle of Buccal flange	B1	0.214	0.0041	L1	0.148	0.0042	M1	0.261	0.0028
	B2	0.078	0.011	L2	0.036	0.0087	M2	0.253	0.0046
	B3	0.279	0.0052	L3	0.171	0.0028	M3	0.258	0.0004
Middle Of Lingual flange	B1	0.177	0.0029	L1	0.094	0.0057	M1	0.182	0.0053
	B2	0.043	0.011	L2	0.036	0.0072	M2	0.186	0.011
	B3	0.246	0.0079	L3	0.128	0.0077	M3	0.185	0.0050

**B1** Balanced occlusion leveled with the upper third of Retro molar pad

**B2** Balanced occlusion leveled with the middle third of Retro molar pad

**B3** Balanced occlusion leveled with the lower third of Retro molar pad

**L1** Lingualized occlusion leveled with the upper third of Retro molar pad

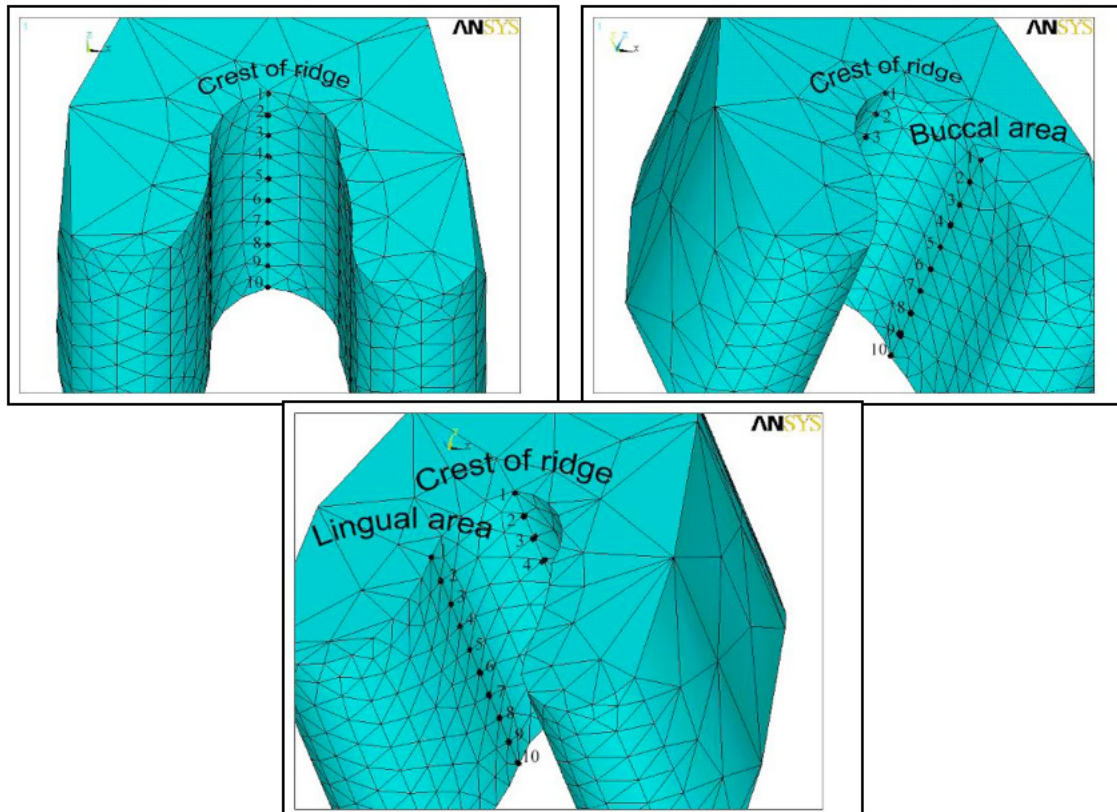
**L2** Lingualized occlusion leveled with the middle third of Retro molar pad

**L3** Lingualized occlusion leveled with the lower third of Retro molar pad

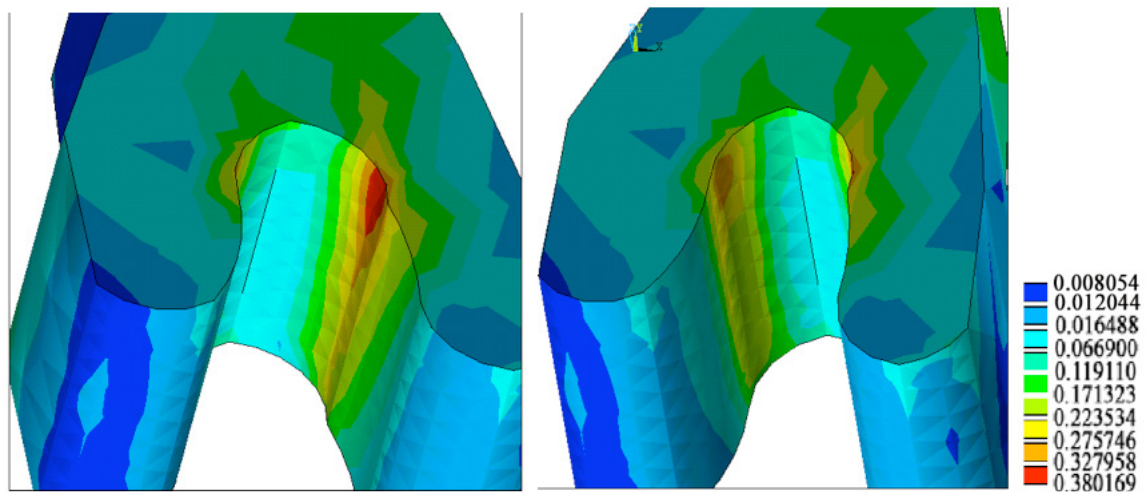
**M1** Monoplane occlusion leveled with the upper third of Retro molar pad

**M2** Monoplane occlusion leveled with the middle third of Retro molar pad

**M3** Monoplane occlusion leveled with the lower third of Retro molar pad



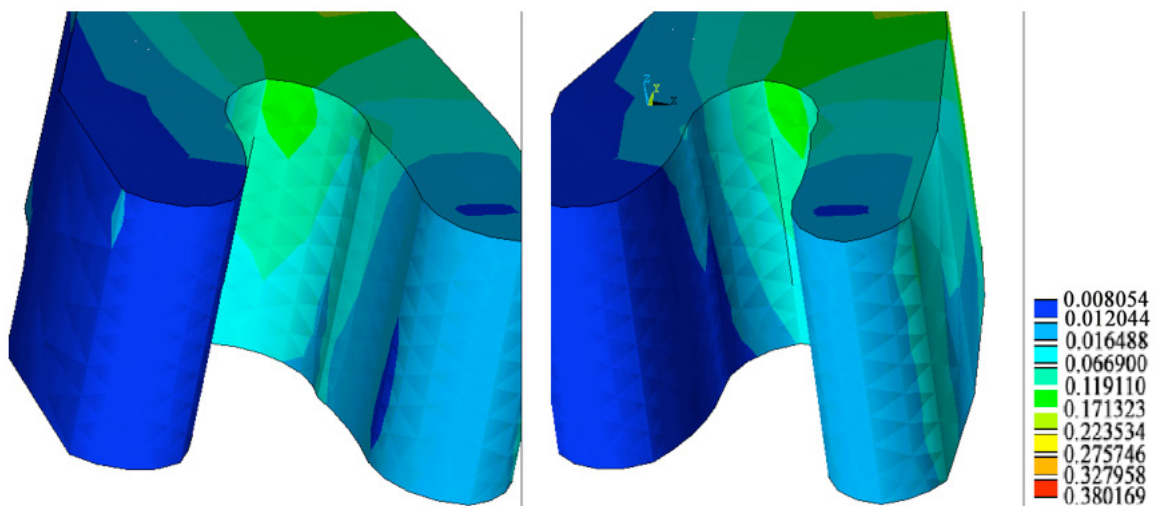
**Fig. 1 Three dimensional model represent the site of nodes selected.**



Crest of the ridge, buccal flange

Crest of the ridge, lingual flange

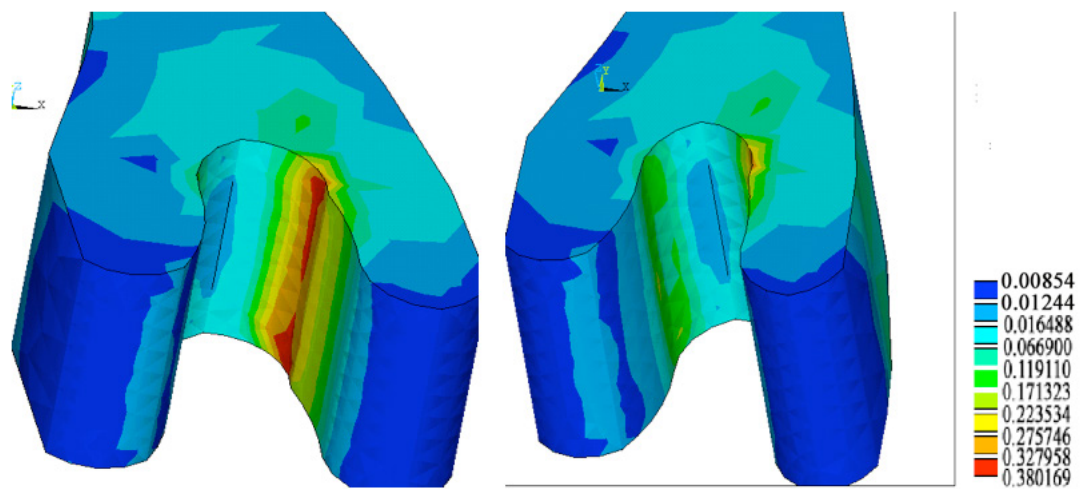
**Fig. 2** Balanced occlusion leveled with the upper third of retro molar pad



Crest of the ridge, buccal flange

Crest of the ridge, lingual flange

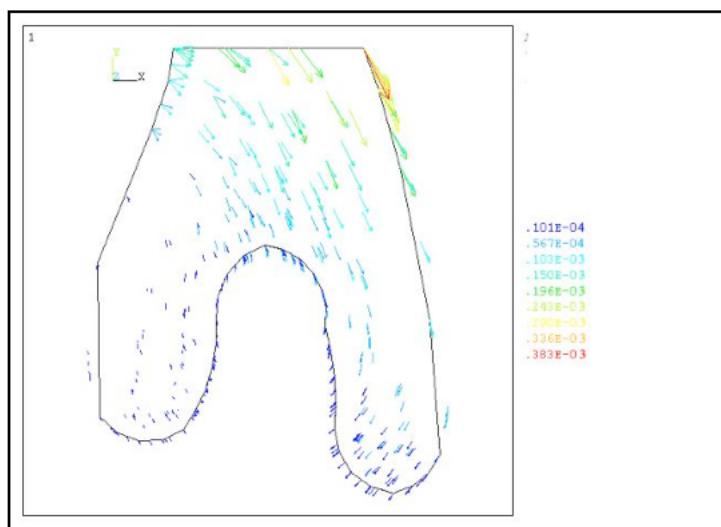
**Fig. 3.** Balanced occlusion leveled with middle third of retro molar pad



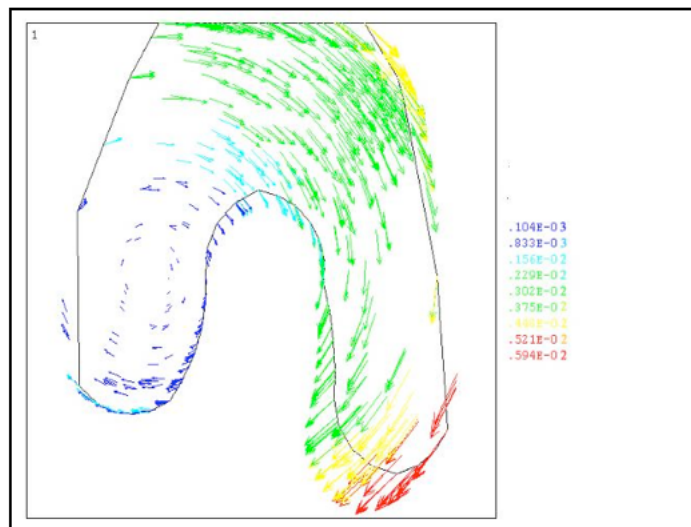
Crest of the ridge, buccal flange

Crest of the ridge, lingual flange

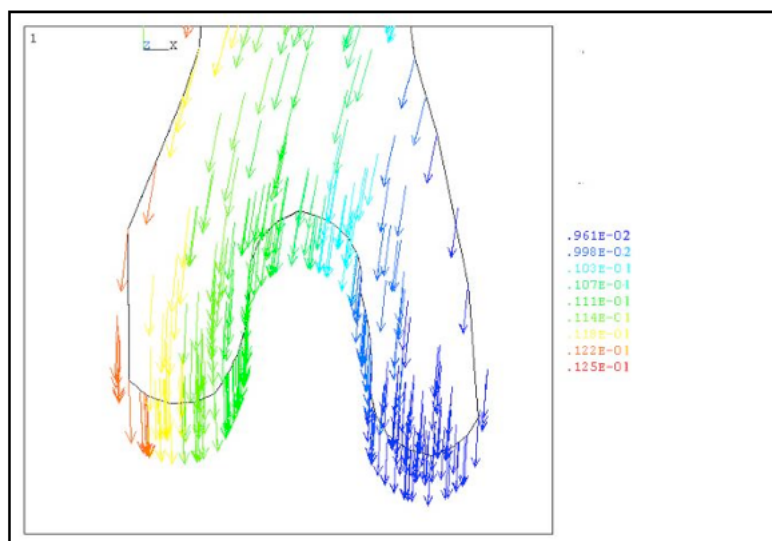
**Fig. 4.** Monoplane occlusion



**Fig. 5** Shows the displacement Vectors in balanced occlusion leveled with the middle third of RMP. (mm)



**Fig.6:** Shows the displacement Vectors in lingualized occlusion leveled with the lower third of RMP. (mm)



**Fig..7** Shows the displacement Vectors in monoplane occlusion (mm)

## REFERENCES

1. Boening K. and Waker M. Computer aided evaluation of occlusal load in complete denture. J. prosth. Dent. 1992, 67(3)339-44.
2. Kydd W. Complete denture base deformation with various occlusal teeth form. J. prosth. Dent 1956, 4(5):714-18.
3. Ismail YH. and Bowman JF. Position of the occlusal plane in natural and artificial teeth. J. prosth. Dent 1968, 20:407-10.
4. Jones PM.: The monoplane occlusion for complete denture JADA 1972, 85: 94-100.
5. Gromas DG. and Stout CJ. Linear occlusion concept for complete denture. J. prosth. Dent 1974, 32(2):122-129.
6. Inowe S.; Kawano F.; Nagao K. and Matsumoto N. Occlusal scheme and pressure distribution of supporting tissue. The international journal of prosth. 1996, 9 (2):179-87.
7. Ohguri T.; Kawano F.; Ichikawa T. and Matsumoto N. Occlusal scheme and pressure under complete denture. The international journal of prosth. 1999, 12(4):353-58.
8. Craig R. Restorative dental material.10th ed. Mosbey St. Louis 1997: 59-103.
9. Larrabee W. and Glat J. A finite element method of skin deformation .Laryngoscope 1986, 96:413-19.
10. Roedema WH. Occlusal table width and pressure under denture. J. prosth. Dent 1976, 36:24-34.
11. Kelsey CR.; Coplostz and Schoonmarker M. The effect of occlusal forms on pressure and bending during mastication with complete denture. J. prosth. Dent 1976, 55 (2):312.
12. Stromberg WR. A method of measuring forces of denture bases against supporting tissue J. prosth. Dent. 1954, 5(2):268-88.
13. Kydd W. Complete denture base deformation with various occlusal teeth form. J. prosth. Dent 1956, 4(5):714-18.



# Evaluation of the Involvement of the Tempromandibular Joint in Patients with Psoriasis using Computed Tomography for Detection of Psoriatic Arthritis Changes.

Hajer Ibrahim

B.D.S, Msc.oral medicine(U.S.A)

Muntaha fawzi salih

B.D.S.

## ABSTRACT

**Background:** Tempromandibular disorders is a collective term, embracing several clinical problems involving the muscles of mastication, temporomandibular joint (TMJ), or both. Tempromandibular disorders is a symptom complex rather than a single condition. TMDs form a cluster of related disorders with common symptoms which include localized pain, limited or asymmetric movement, and clicks or grating on opening. Psoriatic arthritis is a chronic inflammatory arthropathy that affects patients with psoriasis. The clinical findings for Tempromandibular PSA are pain, tenderness, limitation of movement, joint stiffness, clicking as the jaw is opened or closed, difficulty in opening the mouth, locking of the jaw, and crepitations. These findings are essentially the same as for myofascial pain/dysfunction<sup>(3)</sup>. The objective of the study was to determine the extent of tempromandibular joint involvement in patients with psoriatic arthritis and to evaluate the correlation between clinical findings and radiographic finding.

**Materials and method:** In this study 98 patients were selected, 50 male and 48 female, their age ranged between (18-68) years. They were collected at Al-Yarmook teaching hospital/department of. They were diagnosed as having psoriatic arthritis by a dermatologist according to (Moll and Wright diagnostic criteria 1973)<sup>(11)</sup>. These patients were subjected to rheumatoid factor test RF to exclude the presence of rheumatoid arthritic disease, then the patients were exposed to CT scanning for the Tempromandibular joint in Al-Yarmook teaching hospital for screening the involvement of TMJ with psoriatic arthritis by the presence of any radiographic changes such as erosion, flattening, osteophyte and sclerosis in the condylar head.

**Results:** The result of the study showed that the mean age of psoriatic arthritic patients in TMJ was (44.2) years, and female percentage was (48.4%) and male was (51.6%). Psoriatic arthritis in TMJ showed high significance in the unilateral side complaining (100%) and it was significant in the positive family history (54.8%), and non significant in the right/left ratio (41.9%). Oligo type was found in twenty six patients (83.9%) with significant p-value. Clinically: all symptoms including clicking, tenderness and tempromandibular joint pain were highly significant, limitation showed (54.8%), TMJ pain showed (64.5%) and tenderness showed (54.8%). Radiologically: CT scan showed sensitivity for erosion in TMJ (54.8%), for osteophyte (32.3%), for condylar flattening (16.1%) and for sclerosis (9.7%) while specificity for all were (100%). Association between clinical and radiological changes showed non significant correlation.

**Conclusions:** Psoriatic arthritis of TMJ is unilateral (oligo type). Female/ male ratio is about 1:1. About 50% of patients have limitation in mouth opening. Most of the patients have positive family history. Duration of the disease is related to the development of psoriatic arthritis in TMJ. Plaque psoriasis is the most associated type with the psoriatic arthritis in TMJ. Most of patients with psoriatic arthritis show psoriatic nail changes. The most radiographic findings found in patients with psoriatic arthritis was erosion in the condylar head. There is association between clinical and radiologic findings for psoriatic arthritis in TMJ.

(Keywords: Tempromandibular joint, Tempromandibular disorders, psoriatic arthritis, Computed tomography).

## تقييم إصابة المفصل الفكي الصدغي عند مرضى الصدفية باستعمال التصوير المقطعي المحوسب للكشف عن

### التغيرات المفصالية

د. هاجر إبراهيم عبد الله

استاذ في طب الفم اكلية طب الاسنان الجامعة المستنصرية

د. منتهى فوزي صالح

طبيب اسنان اكلية طب الاسنان الجامعة المستنصرية

المستخلص:

### خلفية البحث

اضطرابات المفصل الفكي الصدغي تشير الى مصطلح مركب يحتوي على عدة حالات تتضمن المفصل الصدغي، التركيبات المحيطة او كلاهما. اضطرابات المفصل الفكي الصدغي هي مجموعة اعراض معقدة اكثر من كونها حالة معينة واحدة، ويعتقد انها تنتج من عدة عوامل مثل: العوامل الجينية، التنموية والايضية وانها تكون مجموعة من الحالات المرتبطة مع اعراض شائعة تتضمن ألم موضعي، حركة متحددة او غير متناظرة، قلة او تشابك أثناء الفتح. الروماتيزم الصدفي او التهاب المفصل في الصدفية هو التهاب المفاصل المزمن الذي يؤثر على المرضى المصابين بداء الصدفية. الروماتيزم الصدفي الاكثر شيوعا هو السلبي المصل الاحادي الجانب الموجود عند المرضى المصابين بداء الصدفية لذلك هو يحصل اكثر عند المرضى ذوي النسيج نوع كريات الدم البيضاء المناعية الانسانية ب ٢٧.

النتائج السريرية لمرض الروماتيزم الصدفي الذي يصيب المفصل الفكي الصدغي هي ألم، مضض، تحدد الحركة، تصلب المفصل، قلة عند فتح او غلق الفك، صعوبة عند فتح الفم، ثقل المفصل وفرقة. هذه النتائج السريرية تشبه الى حد ما تلك الموجودة في الألم المرتبط بخلل الاداء المرتبط باللفافة العضلية

### اهداف البحث

لتحديد مدى شمول المفصل الفكي الصدغي لدى المرضى المصابين بالتهاب المفصل في الصدفية.



في هذه الدراسة تم اخذ ثمانية وتسعون مريضاً، خمسون ذكراً وثمانية واربعون امرأة اعمارهم كانت بين (١٨-٦٨) سنة من مستشفى اليرموك التعليمي قسم الامراض الجلدية من الفترة (تشرين الثاني ٢٠١٤- ايار ٢٠١٥). شخضوا من قبل اخصائي الجلدية بانهم مصابون بالتهاب المفصل في الصدفية استناداً الى معايير خاصة.

هؤلاء المرضى خضعوا لتحليل العامل الروماتويدي لاستبعاد وجود مرض التهاب المفصل الروماتويدي، ثم خضعوا للتصوير المقطعي المحوسب للمفصل الفكي الصدغي في مستشفى اليرموك التعليمي لتوضيح شمول المفصل بمرض التهاب المفصل في الصدفية بوجود اي تغيرات في التصوير الشعاعي مثل التآكل، التسطح، نابطة عظمية والتصلب في الرأس اللقيمي.

النتائج

نتائج الدراسة اظهرت ان متوسط اعمار المصابين بالتهاب المفصل في الصدفية في المفصل الفكي الصدغي هو (٤٤,٢ سنة) ونسبة الاناث كانت (٤٨,٤) ونسبة الذكور كانت (٥١,٦). التهاب المفصل في الصدفية في المفصل الفكي الصدغي اظهر توافق احصائي عالي في التأثير وحيد الجانب (١٠٠٪) وتوافق احصائي في التاريخ الطبي العائلي مع المرض (٥٤,٨٪) ولا يوجد توافق في نسبة الجانبين الايمن واليسار (٤١,٩٪).

النوع ذات التأثير الاحادي الجانب وجد عند ستة وعشرون مريضاً (٨٣,٩٪) مع وجود توافق احصائي. جميع الاعراض المتضمنة القلقة، المضض والالم في المفصل الفكي الصدغي كانت عالية التوافق الاحصائي، التحدد ظهر بنسبة (٦٤,٥٪) والمضض بنسبة (٥٤,٨٪). النتائج الشعاعية: التصوير المقطعي المحوسب اظهر حساسية للتآكل في المفصل الفكي الصدغي بنسبة (٥٤,٨٪) وللنابطة العظمية بنسبة (٣٢,٣٪) ولتسطح الراس اللقيمي بنسبة (١٦,١٪) وللتصلب بنسبة (٩,٧٪) فيما كانت نسبة النوعية لجميع النتائج هي (١٠٠٪). العلاقة بين التغيرات السريرية ونتائج التصوير الشعاعي اظهرت عدم وجود توافق احصائي فيما بينهما.

الاستنتاجات

التهاب المفصل في الصدفية في المفصل الفكي الصدغي هو من النوع ذات التأثير الاحادي الجانب. نسبة الذكور الى الاناث هي ١:١، ما يقارب ٥٠٪ من المرضى لديهم تحديد في القدرة على فتح الفم. اغلبية المرضى كان لديهم تاريخ طبي عائلي مع المرض. مدة مرض لها علاقة مع تطور التهاب المفصل في الصدفية في المفصل الفكي الصدغي. الصدفية اللويحية هي اكثر انواع الصدفية ارتباطاً مع التهاب المفصل الصدغي في المفصل الفكي الصدغي. معظم المرضى كان لديهم تغيرات الاظافر الصدفية. معظم نتائج التصوير الشعاعي للمرضى اظهرت وجود تآكل في الراس اللقيمي. هناك علاقة بين النتائج السريرية والشعاعية لالتهاب المفصل في الصدفية في المفصل الفكي الصدغي.

**INTRODUCTION**

Psoriasis is a chronic, autoimmune disease that appears on the skin. It is not contagious, yet is the most common autoimmune disease in the United states. Psoriasis has been linked to other serious health conditions, such as cardiovascular disease and depression. About 30 percent of people with psoriasis develop psoriatic arthritis, which causes pain, stiffness and swelling in and around the joints <sup>(1)</sup>.

Psoriatic arthritis can develop at any age. Genes, the immune system, and environmental factors are all believed to play a role in the onset of the disease. Arthritis is not correlated with the extent of skin disease <sup>(2)</sup>. psoriatic arthritis result in destructive arthritis in which the inflammatory process leads to bony erosion and loss of joint architecture

Psoriatic arthritis (PsA) has historically been considered a milder rheumatic disease not yielding significant clinical damage. However, recent studies have shown that PsA can be deforming and debilitating and that joint damage can be severe <sup>(5)</sup>. Traditionally, joint damage has been recorded using plain radiographs. Characteristic radiographic features of PsA include joint erosions, joint space narrowing, bony proliferation including periarticular and shaft periostitis, osteolysis including "pencil in cup" deformity and acro-osteolysis, ankylosis, spur formation, and spondylitis <sup>(4)</sup>.

In 80% of patients, psoriasis usually precedes the development of arthritis Treatment is by using anti-inflammatory agents, anti-malarials, and adding methotrexate <sup>(15)</sup>. Treating the skin alone seems

to have little impact on joint disease, and the relationship between skin and joints is still unclear. Psoriatic arthritis of the temporomandibular joint (TMJ) was described in 1965, and in the twenty cases reported the TMJ affection has been part of polyarthritis in patients who have suffered from cutaneous psoriasis for years. Clinically, the temporomandibular joint has presented symptoms of chronic arthritis (pain, TMJ tenderness and restricted movement). Radiologic imaging has revealed abnormalities in condyle position, erosion, Osteoporosis <sup>(6)</sup>.

**SUBJECT, MATERIALS AND METHOD**

Patients were diagnosed as having psoriasis by the dermatologist, then Moll and Wright diagnostic criteria were applied to these patients for diagnosis of psoriatic arthritis. If two of the first four criteria and the fifth one were achieved the patient diagnosed as having PsA. an inflammatory arthritis (peripheral arthritis and/or sacroiliitis or spondylitis).

- The presence of psoriasis.
- Nail involvement such as pitting and separation from the nail bed (onycholysis), as well as yellow-pink discoloration (the oil-drop sign).
- Sausage digits (dactylitis).
- The (usual) absence of serological tests for rheumatoid factor <sup>(11)</sup>.

**Clinical examination:** The clinical examination started with examination of psoriatic skin lesions, then examination of nails for psoriatic nail changes.

**TMJ pain on palpation:**

The tip of the index finger was placed over the

lateral aspects of both joint area and slight pressure was applied, pain or tenderness was recorded in a static position or during opening and closing of the mouth. The same procedure was repeated to examine the posterior aspect of the TMJ via external auditory meatus using the index and middle fingers, pain on movement or tenderness was recorded whenever palpation resulted in reflex or the patient reported a subject discomfort <sup>(13)</sup>. Computed tomography scans were carried on Philips Brilliance CT. Bilateral tempromandibular joint CTscans were obtained for all 98 patients. All CT scans were evaluated in details by Radiologist. Each condyle was evaluated for changes like erosion, flattening, osteophyte formation and sclerosis. The clinical data then was correlated with the CT scan findings for each joint and subjected to statistical evaluation.

## RESULT

The result of the study showed that the mean age of psoriatic arthritic patients in TMJ was (44.2) years, and female percentage (48.4%) and male (51.6%). Psoriatic arthritis in TMJ showed highly significant in the unilateral side complaining (100%) and its significant in the positive family history (54.8%) and non significant the right/left ratio (41.9%). Oligo type was founded in twenty six patients (83.9%) with significant p-value, clinically: all symptoms including clicking, tenderness and tempromandibular joint pain were highly significant, limitation showed (54.8%), TMJ pain showed (64.5%) and tenderness showed (54.8%). radiologically: CT scan showed sensitivity for erosion in TMJ (54.8%), for osteophyte (32.3%), for condylar flattening (16.1%) and for sclerosis (9.7%) while specificity for all were (100%). Association between clinical and radiological changes showed non significant correlation

**Table (1) :Description of patient's age and statistical analysis**

		<i>Psoariatic TMJ</i>		<i>Psoariatic arthritis</i>		<i>P value</i>
		<i>No</i>	<i>%</i>	<i>No</i>	<i>%</i>	
Age (years)	<30	6	19.4	7	10.4	0.645
	30---39	7	22.6	17	25.4	
	40---49	6	19.4	17	25.4	
	=>50	12	38.7	26	38.8	
	Mean±SD (Range)	44.2±13.3 (18-68)		44.5±11.4 (19-61)		
-----		-----		-----		

**Figure (1) :The clinical presentation**

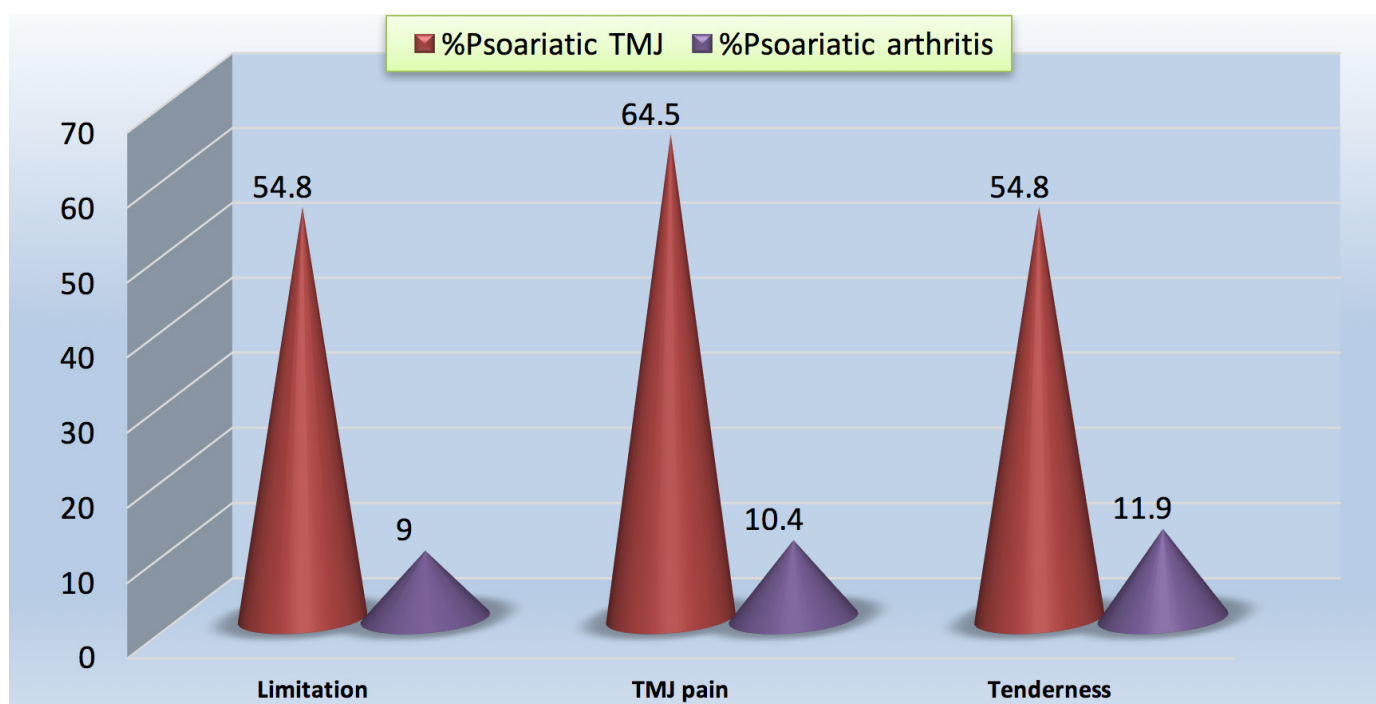


Figure (2) :The percentage of type and associated features.

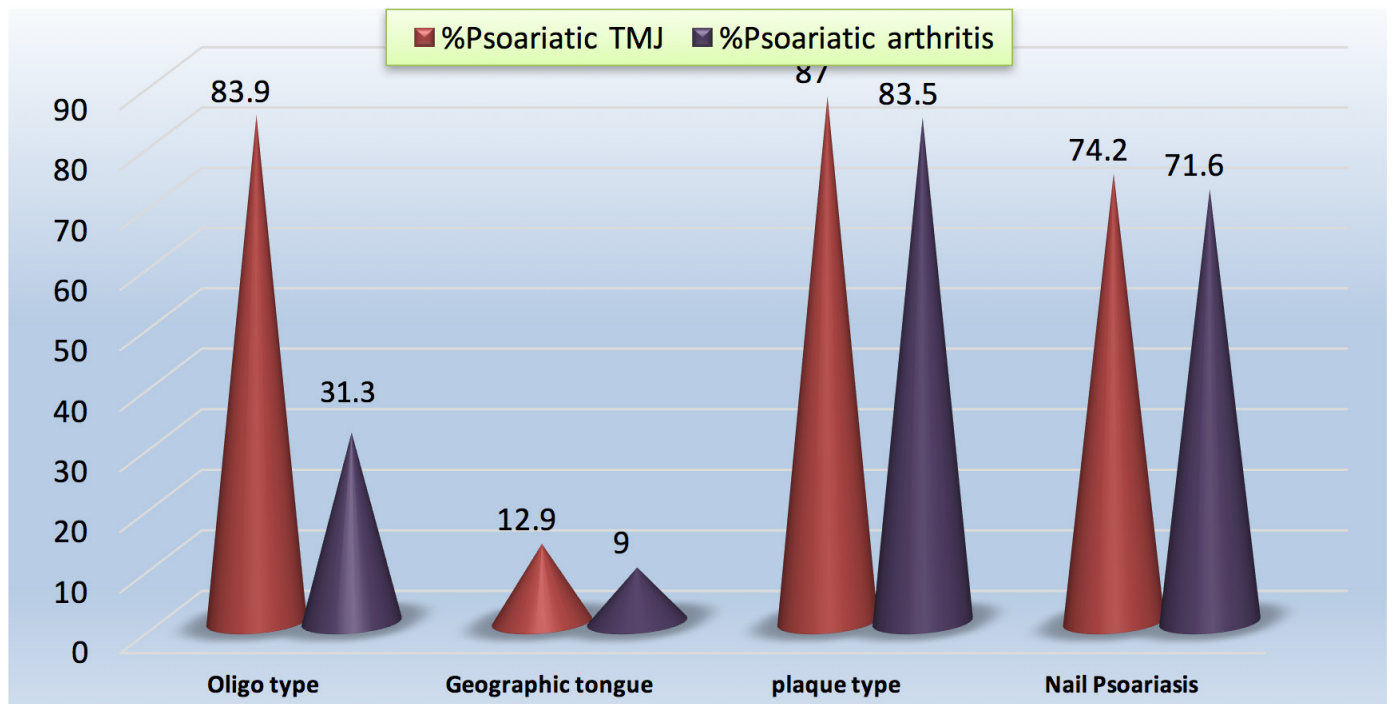


Table (2) :The radiological findings of CT.

CT findings		Psoariatic TMJ	Psoariatic arthritis
		No	No
Erosion	Yes	17	-
	No	14	67
Osteophyte	Yes	10	-
	No	21	67
Flattening	Yes	5	-
	No	26	67
Sclerosis	Yes	3	-
	No	28	67

\*Significant difference in proportions using Pearson Chi-square test at 0.05 level

#### Association between type of psoriatic arthritis and radiological changes.

CT findings		Oligo		Poly		P value
		No	%	No	%	
Erosion	Yes	14	53.8	3	60.0	0.800
	No	12	46.2	2	40.0	
Osteophyte	Yes	7	26.9	3	60.0	0.147
	No	19	73.1	2	40.0	
Flattening	Yes	5	19.2	-	-	-
	No	21	80.8	5	100.0	
Sclerosis	Yes	3	11.5	-	-	-
	No	23	88.5	5	100.0	

\*Significant difference in proportions using Pearson Chi-square test at 0.05 level

## DISCUSSION

In our study we found that the mean age of psoriatic arthritis in TMJ was (44.2), and for control group (44.5) which is very close to the group of

**Wilson**<sup>(8)</sup>. with mean age(43.3),while **Kononen**<sup>(7)</sup> found that the mean age was (51) for both arthritic group and control group ,and **Green**<sup>(9)</sup> found the mean age was (46.08). Sex distribution in our study showed that male (50.7%) and female (49.3%) approximately the same to other studies like **Wilson** in 50% for each one. Other study by **Kononen** showed that male (57.2%) and female (42.8%). While other study by **Green** showed sex distribution (1.2:1) very close to our study.In the view of 43 cases of psoriatic arthritis involving TMJ, there is a male predilection (24:19) and the average age of onset is 43.3. TMJ lesions could occur before, during , or after other joints involvement. **Zhi** <sup>(10)</sup>found the most common clinical findings are pain in the TMJ region (27/43). TMJ tenderness to palpation (22/43), and limited mouth opening was (21/43). The unilateral involvement of TMJ in patients with psoriatic arthritis is more common than bilateral involvement (5:3),these features in agreement with the findings of **Rasmussen and Bakke**<sup>(14)</sup>.Moreover , some clinical studies reported that muscle tenderness on palpation and joint sounds were the most frequent symptoms in TMJ, while restricted mouth opening was not prominent in psoriatic arthritis patients. It could be inferred that the symptoms of psoriatic arthritis affecting the TMJ are nonspecific. While in our study All patients examined clinically and it's highly significant in psoriatic arthritis in TMJ for limitation, TMJ pain and tenderness. Limitation showed (54.8%), TMJ pain showed (64.5%) and tenderness showed (54.8%),very close to the result obtained by **Zhi**. In our study Psoriatic arthritis in TMJ showed highly significant in the unilateral side complaining (100%) and its significant in the positive family history (54.8%) and non significant in the right/left ratio (41.9%). The difference in the studies between our study and (**Zhi** was only the percentages in side complaining, this may be due to the variance in the sample size and the race of affected population. **Wilson et al, 2009** found plaque type with (79%) followed by guttate type , similar to our study in which we found plaque type (87%) followed by guttate type.

Among previous studies **Zhi** found erosion is the most common radiographic changes in psoriatic arthritis in patients with TMJ involvement (37/63) about(58%), which is in accordance with radiologic findings of **Lundberg**<sup>(12)</sup> Other radiographic findings such as bony proliferation (22/63) about (44%), condylar flattening ( 8/63) about (12%) and sclerosis (6/63) about (9.5%), are also typical changes in TMJs of psoriatic arthritis patients. This result seems close to the result obtained in our study that CT scan

showed sensitivity for erosion in TMJ (54.8%), for osteophyte (32.3%), for condylar flattening (16.1%) and for sclerosis (9.7%) while specificity for all were (100%).The radiographic sign score did not correlate significantly with the duration, extent, or severity of PA **Kononen**. The Radiographic score correlated with the subjective symptoms score ( $p < 0.01$ ), and one of the variables of , osteophytes, correlated with pain in the TMJ experienced at the onset of subjective symptoms of craniomandibular disorders ( $p < 0.01$ ). The radiographic score , and especially erosions in the condyle, correlated with the clinical dysfunction score.

## CONCLUSIONS

- Psoriatic arthritis of TMJ is unilateral (oligo type).
- Female/ male ratio is about 1:1.
- About 50% of patients have limitation in mouth opening.
- Most of the patients have positive family history.
- Duration of the disease is related to the development of psoriatic arthritis in TMJ.
- Plaque psoriasis is the most associated type with the psoriatic arthritis in TMJ.
- Most of patients with psoriatic arthritis show psoriatic nail changes.
- The most radiographic findings found in patients with psoritic arthritis was erosion in the condylar head.
- There is association between clinical and radiologic findings for psoriatic arthritis in TMJ.

## REFERENCES

1. Taylor W, Gladman D, Helliwell P, Marchesoni A, Mease P, Mielants H. Classification criteria for psoriatic arthritis: development of new criteria from a large international study. *Arthritis Rheum* Aug 2006;54(8):2665-73.
2. Chandran V, Raychaudhuri SP. Geoepidemiology and environmental factors of psoriasis and psoriatic arthritis. *J Autoimmun* May 2010; 34(3):J314-21.
3. American Academy of Orthopaedic Surgeons, 2012.
4. Klippel JH. Primer on the rheumatic diseases. Springer Verlag. (2008)
5. Todd,Adam,and Suja Elizabeth George(Current and potential new therapies for the treatment of psoriasis) pharmaceutical Journal (vol 284) 2010.
6. American Association for Dental Research. Retrieved 6 June ٢٠١٣ ,Temporomandibular Disorders (TMD) used in (WHO).
7. Könönen M. Craniomandibular disorders in psoriasis. *Community Dent Oral Epidemiol*. 1987;15:108-12.
8. Wilson FC, Icen M, Crowson CS, McEvoy MT, Gabriel SE,Maradit Kremers H. Time trends in epidemiology and characteristics of psoriatic arthritis over 3 decades: a populationbasedstudy. *J Rheumatol*. In press.2009.
9. Green L, Meyers OL, Gordon W, Briggs B. Arthritis in psoriasis. *Ann Rheum Dis* 1981;40:366-9.
10. Zhi Hu Wang, Yan Ping Zhao, Yu Chen Ma: volume 14



- number 1, 2014.
11. Moll JMH, Wright V. Psoriatic arthritis. *Semin Arthritis Rheum* 1973;3:55-78
  12. Lundberg M, Ericson S. changes in Temporomandibular joint in psoriasis arthropathica. *acta derm venereal (stockh)*;1967;47,354-8. ISBN:0387356649.
  13. Dalkiz, :Evaluation of temporomandibular joint dysfunction by magnetic resonance imaging, 2001:80-86.
  14. Rasmussen, O.C.: Semiopaque Arthrography of the Temporomandibular Joint, *Stand. J. Dent. Res.* 88: 521-534, 1982.
  15. Paul Emery, MD, and Zoe Ash, MD, *American College of Rheumatology* September 2012.

# Effect of Systemic Administration of Simvastatin on Dental Implant Stability: A Random Clinical Study

Ali Mohammed Hassan,

B.D.S. =Master Student, Department of Oral and Maxillofacial Surgery, College of Dentistry, University of Baghdad.

Adil Al Kayat,

B.D.S., M.Sc. Sheffield, FDSRCS London. - Assistant Professor, Chairman of the Scientific Council of Oral and Maxillofacial Surgery, Iraqi Council of Medical Specialities.

## ABSTRACT

**Background:** the primary objective for many researches carried out in dental implantology was to reduce the period needed for functional implant loading, simvastatin (cholesterol lowering medication) had many pleiotropic effects, one of which was increasing bone density around titanium implants<sup>(1)</sup> and subsequently establishing faster osseointegrated dental implants<sup>(2,3)</sup>.

**Aim of the study:** this study aims to reduce the period of time needed to establish secondary stability of dental implant measured in ISQ (Implant Stability Quotient) by investigating the effect of orally administered simvastatin on bone.

**Materials and methods:** simvastatin tablets (40mg/day for three months) were administered orally for 11 healthy women aged (40-51) years old who received 15 dental implants (Dentium, Implantium) in the traumatic functional implant zone<sup>(4)</sup>, this is the intervention group, the control group (n=11) received 14 dental implants in the same zone.

3 dental implants in 2 subjects were lost, leaving a total of 26 dental implants in 20 patients with 10 patients in each group. All subjects were radiographed with OPG for preliminary assessment and with CT scan for registering bone density in Hounsfield Units. Different dental implant sizes were used according to optimal patients' needs. An informed consent was obtained from the intervention group and the recommended monitoring protocol was followed.

Dental implant stability ISQ were recorded using RFA by Ostell<sup>TM</sup> ISQ for both groups three times: immediately after implant placement (at surgery) and after 8, 12 weeks respectively.

**Results:** results showed that the mean implant stability for the intervention group was significantly higher  $P = 0.01$  after 12 weeks in comparison to that of the control group.

Simvastatin showed statistically significant effect on implant stability among the intervention group after 8 and 12 weeks ( $P$  value for both times  $< 0.001$ ) with the attributed risk percent was 70.8 and 50 respectively.

**Conclusions:** this study concluded that the intervention group had higher implant stability and was ready for functional loading prior to control group and that simvastatin might enhance and/or accelerate the process of osseointegration.

## KEYWORDS

Implant Stability, Simvastatin, Resonance Frequency Analysis.

## INTRODUCTION

The dental implant is increasingly becoming a popular treatment for replacing missing teeth for partially dentate as well as edentulous patients. In 2011 alone, dentists across the U.S. placed over five million implants, according to the American Dental Association<sup>(5)</sup>.

Osseointegration was first described by Brånemark and co-workers<sup>(6)</sup>. The term was first defined in a paper by Albrektsson et al 1981 as direct contact (at the light microscope level) between living bone and implant<sup>(7)</sup>. Since the histological definitions have some shortcomings, mainly that they have a limited clinical application, another more biomechanically oriented definition of osseointegration has been suggested: "A process whereby clinically asymptomatic rigid fixation of alloplastic materials is achieved, and maintained, in bone during functional loading"<sup>(8)</sup>.

Over the following years attempts have been made by researchers to improve dental implant osseointegration (clinically applicable in terms of dental implant stability) through understanding the factors influencing it and the production of various materials in favor of that concept. One of these newly studied materials is Simvastatin.

Simvastatin is a 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) reductase inhibitor. It is

widely used as a cholesterol-lowering drug and inhibits hepatic cholesterol biosynthesis. Recent studies have shown a beneficial effect of statins on bone mineral density (BMD)<sup>(9,10)</sup>. It has been suggested that several statin drugs, including simvastatin, increase the mRNA expression of bone morphogenetic protein (BMP-2) in osteoblasts, with a subsequent increase in bone formation. Simvastatin has been shown to enhance osseointegration of pure titanium implants in osteoporotic rats.<sup>(11)</sup> Other experimental study shows that locally administered simvastatin was detrimental to the repair of defects in the calvaria of rats<sup>(12)</sup>. The period required generally for an osseointegration to be achieved and for dental implant to be loaded is about 3-6 months which still represent a relatively long period for patients and any efforts focusing on reducing this period are entitled for consideration and scientific research which is the objective of this study.

## MATERIALS AND METHOD

This study was conducted at the dental implant unit in Oral and Maxillofacial Department of College of Dentistry, Baghdad University, from January 2012 to February 2014, where twenty two healthy females aged (40-51) years old received 29 dental implants (Dentium, Implantium) were divided randomly (using alternating randomization method) into two groups, control and intervention group. 3 dental implants in 2 subjects were lost, leaving a total of 26 dental

implants in 20 patients with 10 patients in each group:

The intervention group, this group received 14 titanium screw type endosseous dental implants (Dentium, Implantium) in the traumatic functional implant zone (the area from maxillary right 1st premolar to the maxillary left 1st premolar) along with systemically administered (oral) simvastatin 40mg/day (as an accepted dose for humans)<sup>(13)</sup> post-operatively for three months.

The control group, this group received 12 titanium screw type endosseous dental implants (Dentium, Korea) in the traumatic functional implant zone and submitted to the same procedure of intervention group without the administration of post-operative simvastatin.

#### Exclusion Criteria:

- a. Smokers.
- b. Alcoholics.
- c. Patients with any chronic systemic disease. For example (active liver disease, patients on warfarin and/or antifungal medication and/or cyclosporine... etc.)
- d. Pregnant or lactating females.
- e. Patients with inadequate sub-antral distance, due to maxillary sinus pneumatization<sup>(14)</sup>.
- f. Implant site subjected previously to supplemental surgical procedures (bone graft, ridge augmentation... etc.).

The prospective implant sites were examined clinically and radiographically by two views: Orthopantomogram (OPG) and CT scan for registering bone density (using Hounsfield units and according to Misch classification of bone quality)<sup>(15)</sup> at the target site and also for precise placement of the dental implants through providing information about width of the alveolar bone and proximity to the maxillary sinus. After the patients signed an informed consent expressing their approval for participating in this study the insertion of fixtures is carried out in the traumatic zone for both groups then the primary

stability is measured immediately after dental implant placement by a colleague for each patient by Resonance Frequency Analysis (RFA) using OsstellTM ISQ (Goteborg, Sweden) through inserting the smart peg into the implant and two readings of the ISQ (Implant Stability Quotient) values are recorded; in bucco-palatal direction and the other in mesio-distal one.

Implant stability was measured again after 8 and 12 weeks postoperatively by the same colleague using RFA to compare its values (ISQs) between both groups.

Group A were asked to perform a liver function test (SGPT, SGOT) after 6 and 12 weeks postoperatively as a monitoring for any hepatic side effects and to report any muscular/joint pain, spasm or discomfort for further CPK (Creatine Phosphokinase) evaluation<sup>(16)</sup>.

## RESULTS

### Statistical Analysis

Statistical analyses were done using SPSS version 21 computer software (Statistical Package for Social Sciences) in association with Microsoft Excel 2010. In this study the following statistics were used:

1. Descriptive Statistics: including; mean, standard deviation and standard error.
2. Parametric statistical tests of significance: including; t-test, Paired t-test and Cohen's d (standardized measure of effect size).

### The Difference in Mean Stability between the Two Groups

(Table 1) shows a comparison of dental implant mean stability between both groups through time intervals, where in group A after 8 weeks of surgery there was an increase in the mean stability which was not statistically significant by 1.8 ISQ units compared to the primary stability readings. The changes observed during the 1st 8 weeks of surgery were evaluated as a weak effect (Cohen's D= 0.22).

**Table 1: Mean-ISQ of the 2 perpendicular directions at surgery and after 8 weeks.**

	<i>at surgery</i>	<i>After 8 weeks</i>	<i>Changes compared to baseline</i>	<i>Cohen's d</i>	<i>P (Paired t-test)</i>
Control					
N	12	12	12		
Mean	72.8	66	-6.7	-0.65	0.045
SD	8.2	6.3	10.3		
SE	2.36	1.81	2.97		
Intervention					
N	14	14	14		
Mean	73.5	75.3	1.8	0.22	0.44[N S]
SD	7	6.6	8.3		
SE	1.87	1.78	2.21		

(Table-2) Mean-ISQ of the 2 perpendicular directions after 12 weeks and compared to primary stability

	after 12 weeks	changes compared to baseline	Cohen's d	P (paired t-test)
Control				
N	12	12		
Mean	69.3	-3.5	-0.42	0.17[NS]
SD	5	8.3		
SE	1.44	2.4		
Intervention				
N	14	14		
Mean	79.5	5.9	0.81	0.01
SD	3.9	7.3		
SE	1.05	1.96		

The total change in mean ISQ after 12 weeks of the surgery compared to the primary stability was an average increase of 5.9 ISQ units which is statistically significant (P value = 0.01), and the effect evaluated as a strong effect (Cohen's D= 0.81).

The whole behavior of implant stability for both groups is shown in (figure 1)

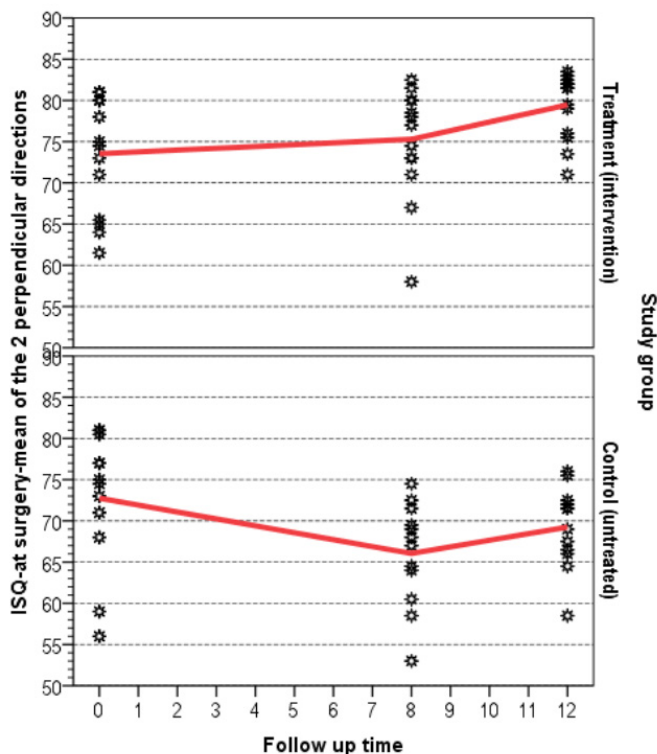


Figure 1: illustrating the mean stability of the dental implants for both control and intervention groups where the left (Y) axis represent the ISQ units and the horizontal (x) axis represent the time measured in weeks.

### Number of Implants achieved 70+ ISQ in Both Groups

The stability of implant at 70 ISQ or more is considered an implant with high ISQ stability (17), statistical test using attribute risk percent was used to compare numbers of dental implants in treated

versus non treated control group that achieved the bench mark of implant stability (70+ ISQ) over time as illustrated in (figure 2) below.

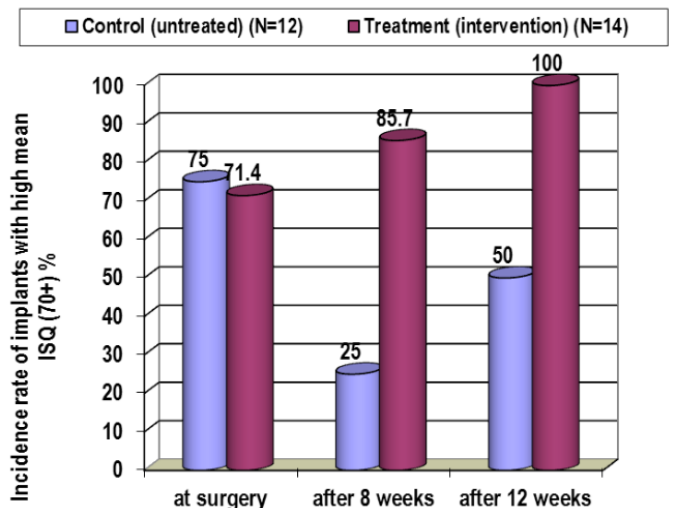


Figure 2: number of implants achieved high stability

In (figure 2) the control group and the intervention group show no difference at surgery but after 8 weeks 25% of the control group reached the high ISQ level while in the intervention group 85.7% achieved the high stability level of 70+ ISQ.

At week 12, 50% of the control group dental implants reached to the bench mark level of high implant stability in comparison to the intervention group where 100% of the dental implants were at high implant stability (all the dental implants in the intervention achieved 70+ ISQ at the end of the study).

### DISCUSSION

To the best of the authors' knowledge; this study is the first and no other comparable studies were available. The sample selection was based on two basic features: gender and age. Females were selected over males because the changes in bone remodeling occur in a faster rhythm, And their age was 40+ years old and not younger because in this range of age most of bone parameters regarding bone regeneration capacity, elasticity, strength and even cell viability



are declining especially in females aged above 40 years old, where 34% had osteopenia and 8% had osteoporosis <sup>(18)</sup>.

Monitoring protocol was applied throughout this study to all intervention group, their liver function tests were negative and within the normal range (SGPT < 34  $\mu$ /l, SGOT < 31  $\mu$ /l).

Members of the intervention group were asked routinely to report any muscular pain, cramps, and weakness. No such reports or complaints were informed.

Among the control group; the mean ISQ after 8 weeks of surgery showed an obvious reduction which re-increased to an obvious increment till 12 weeks. The overall change after 12 weeks compared to primary stability at surgery was still in the negative direction making the final stability still less than the primary one; yet they are loadable. This pattern is well documented in literature especially for mean primary stability of more than 70 ISQ giving the fact that the conventional 2-stage treatment loading protocol used a period of 3-6 months for osseointegration prior to loading <sup>(19,20)</sup>. However, Simvastatin treatment changed the pattern described earlier for the control group. The mean ISQ for the intervention group was obviously higher compared to primary stability (mean ISQ at surgery was 73.5 and at the 8th week was 75.3), although the positive changes observed were less than the level of statistical significance; nevertheless, it contradicted the negative trend observed for ISQ in the first 8 weeks in untreated control group (mean ISQ at surgery was 72.8 and at the 8<sup>th</sup> week was 66).

This positive trend in ISQ change in treatment group continued till the end of the study, making the stability after 12 weeks significantly and noticeably higher than that of primary stability. This finding is opposite to the negative loss in ISQ in relation to primary stability observed in control group within the scope and parameters of the current study.

Another advantage for effectiveness of simvastatin on dental implant stability was the absence of difference in mean ISQ at primary stability between the two groups (control 72.8 ISQ, intervention 73.5 ISQ, the difference was 0.7 ISQ), while after 8 and 12 weeks the intervention group had a significant advantage around 10 ISQ units increase in mean difference over the untreated control group (control group mean ISQ at week 8 and 12 was 66, 69.3 respectively while intervention group mean ISQ at week 8 and 12 was 75.3, 79.5 respectively). Since these numbers represent the mean stability, it doesn't mean that each individual dental implant of the

intervention group is necessarily ready for immediate loading.

To summarize the current study outcome, it is worth mentioning that an almost comparable proportion of subjects had high primary stability (>70 ISQ) in both control (75%) and simvastatin treated group (71.4%). After 8 weeks of surgery the rate of high stability in the intervention group increased to (85.7%) while among those untreated (control group) only (25%) had high stability.

After 12 weeks, all simvastatin treated group achieved the bench mark of implant stability (70+ ISQ) while only 50% of untreated group achieved this favorable outcome which they may reach it eventually on the expense of time.

In conclusion, Simvastatin administration had reduced the generally needed functional loading time in traumatic functional implant zone of dental implants from 3-6 months (12-26 weeks) to almost 2 months (8 weeks) by enhancing osseointegration of dental implant and increasing its stability faster than that in control group. Simvastatin was well tolerated in all healthy subjects as they were submitted for periodic monitoring (liver function test) and all tests were normal and no subject reported muscular pain or weakness.

Further recommendation is to Measure dental implant stability at shorter time intervals (after 7 weeks from primary stability) to detect earlier changes associated with the drug. Inclusion of a larger sample for more conclusive results. A longer period of follow up to evaluate the long term effect on success rate of dental implants.

## REFERENCES

1. Du Z, Chen J, Yan F, Xiao Y. Effects of Simvastatin on bone healing around titanium implants in osteoporotic rats. *Clinical oral implants research* 2009;20(2):145-150.
2. Yang F, Zhao S.F, Zhang F, He F.M, Yang G.L. Simvastatin-loaded porous implant surfaces stimulate preosteoblasts differentiation: an in vitro study. *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology* 2001;111(5): 551-556.
3. Nyan M, Hao J, Miyahara T, Noritake K, Rodriguez R, Kasugai S. Accelerated and Enhanced Bone Formation on Novel Simvastatin-Loaded Porous Titanium Oxide Surfaces. *Clinical implant dentistry and related research* 2013.
4. Tolstunov L. Implant zones of the jaw: Implant location and related success rate. *J Oral Impl* 2007; 33(4), 211-20.
5. <http://www.American dental association.org>. 2014, January.
6. Branemark P, Hansson B, Adell R, Breine U, Lindstrom J, Hallan O, Ohman A. Osseointegrated implants in the treatment of the edentulous jaw. Experience from a 10-years period. *Scand J Plast Reconstr Surg Suppl* 1977; 16, 1-132.
7. Albrektsson T, Brånemark PI, Hansson HA, Lundström

- I. Osseointegrated titanium implants. Requirements for ensuring a long-lasting, direct bone- to-implant anchorage in man. *Acta Orthop Scand*. 1981;52(2):155-70.
8. Mantri, S., Khan, Z. Prosthodontic Rehabilitation of Acquired Maxillofacial Defects. *Head and Neck Cancer. Intech*, 2012; 315-36.
9. Klęcoglu S. S., Erdemli E. New addition to the statin's effect. *Journal of Trauma-Injury, Infection, and Critical Care* 2007; 63(1), 187-191.
10. Uzzan B, Cohen R, Nicolas P, Cucherat M, Perret G.Y. Effects of statins on bone mineral density: a meta-analysis of clinical studies. *Bone*, 2007; 40(6), 1581-1587.
11. Du Z, Chen J, Yan F, Doan N, Ivanovski S, Xiao Y. Serum bone formation marker correlation with improved osseointegration in osteoporotic rats treated with simvastatin. *Clin Oral Imp Res*. 2011
12. Calixto J, Villaboim de Castro Lima C, Frederico L, Pio dos Santos de Castro Lima R, Anbinder A. The influence of local administration of simvastatin in calvarial bone healing in rats. *Journal of Cranio- Maxillofacial Surgery*, 2011; 39(3), 215-220.
13. Park JB. The use of simvastatin in bone regeneration. *Med Oral Patho* 2009; 14(9), 485-8.
14. Calvo L, Gómez-Moreno G, López-Marí L, Ortiz-Ruiz A.J, Guardia-Muñoz J. Atraumatic maxillary sinus elevation using threaded bone dilators for immediate implants. A three-year clinical study. *Med Oral Patol Oral Cir Bucal*, 2010; 15(2), e366-70.
15. Misch C. Contemporary implant dentistry. St Louis, Missouri: Mosby, Elsevier. 3rd edi. 2008; 137.
16. Phillip O, James E, William G. Handbook of clinical drug data. New York: McGraw-Hill Publications, 10 edi, 2002.
17. Sennerby L. 20 years of experience with the resonance frequency analysis. *Implantologie* 2013;21(1):21-33.
18. Unni J, Grag R, Pawar R. Bone mineral density in women above 40 years. *J Midlife Health* 2010; 1(1), 19-22.
19. Atieh M, Alsabeeha N, Duncan W, Silva R, Cullinan M, Schwass D, Payne A. Immediate single implant restorations in mandibular molar extraction sockets: a controlled clinical trial. *Clinical oral implants research*, 2013; 24(5), 484-496.
20. Funato A, Yamada M, Ogawa T. Success rate, healing time and implant stability of photofunctionalized dental implants. *Int J Oral Maxillofac Imp* 2013; 28, 1261-71.



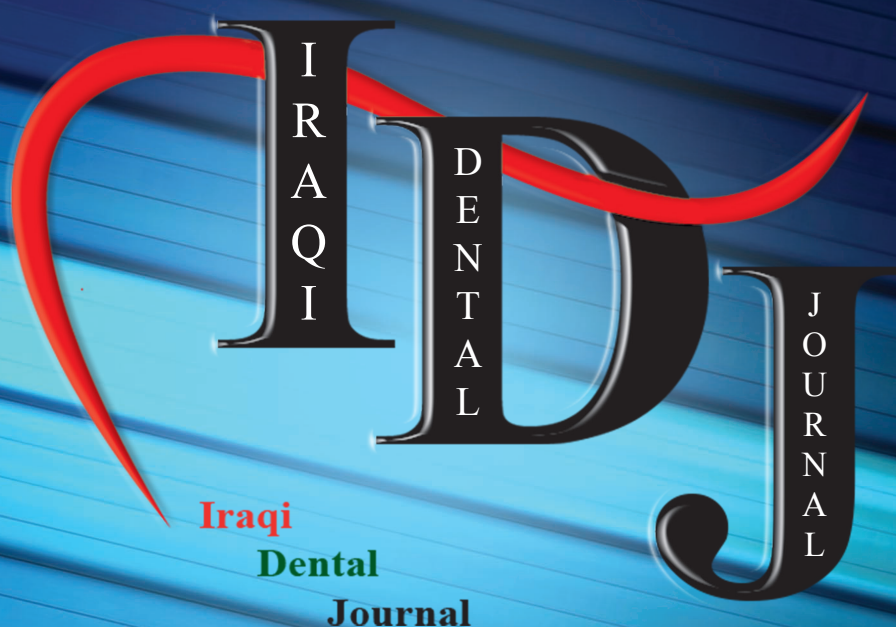




رقم التسجيل: ISSN 2307-4779



# مجلة طب الاسنان العراقية



مجلة علمية محكمة , تصدر عن

نقابة أطباء الاسنان في العراق

المجلد : 38 / العدد : 2 / آب 2016