AJDS LAZHAR Al-Azhar Journal of Dental Science Vol. 21- No. 1- 73:78- January 2018
Record 1110-2624 | the ISSN Portal portal.issn.org

EVALUATION OF MICROBIAL ADHESION AND LEVEL OF SECRETORY IMMUNOGLOBULIN A (SIGA) IN PATIENTS WITH DIFFERENT DENTURE BASE MATERIALS

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ABSTRACT

Objective: The aim of this in vivo and invitro study was to compare the effect of conventional and flexible complete denture bases(Polyamide) on microbial adhesion (streptococcus and candida albicans) and the level of secretory immunoglobulin IGA(SIGA). **Materials and methods**: 10 selected patients were re-habilitated with heat cured mucosa supported complete denture then polyamide flexible denture bases were duplicated. Salivary samples and swaps for microbial adhesion testing were taken. **Results**: The data was collected and statistically analyzed. Flexible denture bases were less in microbial adhesion on their fitting surfaces than conventional denture bases. The levels of salivary immunoglobulin A (SIGA) are higher in conventional denture bases than in flexible denture bases after 7 days. **Conclusion**: flexible dentures are more hygienic and biologically compatible than conventional dentures.

INTRODUCTION

Since Walter Wright first introduced it in 1937, heat cured polymethyl methacrylate (PMMA) polymers denoted as conventional denture base materials and is considered the most popular material for non-metallic denture constructions. Its extensive use was due to low water sorption, solubility and cost, as well as, ease of construction of denture bases by simple processing techniques with acceptable physical and mechanical properties. Despite these favorable properties, a growing number of patients are presenting with hypersensitivity reactions to PMMA which induced by the residual methyl methacrylate monomer[1].The potential alternative materials to PMMA are the polycarbonates and the nylon denture base resins. There are certain types of thermoplastic polymers belonging to the class polyamides. These polyamides are produced by the condensation reaction between a diamine and a dibasic acid[2].

Candida albicans and Streptococcus mutants are important oral pathogens able to form biofilms on different surfaces which may favor the development of diseases such as caries and Denture Stomatitis[3,4]. Epidemiological studies suggest that between 11% and 70% of all patients wearing removable denture prostheses suffer from denture stomatitis[5].Saliva is a complex biofluid comprising of electrolytes, glycoproteins and enzymes that lubricates and cleanses the mucosa, protects it from trauma, aids in digestion and contributes to the sensation of taste. The role of saliva as a lubricant and a buffer is central to the comfort and health of the oral cavity. Any alteration in the quantity or quality of saliva can adversely affect the oral health balance and can lead to various problems[6] . SIgA plays an important role in protection against infections by preventing microbial adherence, neutralizing enzymes, toxins and viruses [7].The intrinsic resistance of IgA to proteolysis, which is reinforced by the presence of

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the SC, preserves the biological functions of the molecule in secretions[8]. This in-vivo and in-vitro study compared the effect of conventional and flexible complete denture bases (polyamide) on microbial adhesion (streptococcus mutants and Candida albicans) and level of secretory immunoglobulin (SIGA).

MATERIALS AND METHODS

Patients Selection

Ten Completely edentulous co-operative male patients were selected from the out-patient clinic, Prosthodontics Department, Faculty of Dentistry AL-AZHAR University.

Criteria for patient's selection

1. The patients' age ranged from 50-60 years.
2. Male patients.
3. All patients had sufficient inter-arch space.
4. All patients had class-I Angle's classification.
5. All patients were free from any systemic diseases that might affect the health of the residual alveolar ridge or might interfere with immunocompetence or salivary gland secretions

Grouping of Patients

The selected patients were re-habilitated with heat cured mucosa supported complete denture. Dentures were constructed following the conventional technique. At the time of denture insertion, two hours, three days and seven days, salivary samples were collected to evaluate the level of salivary immunoglobulin A.

Salivary Sample preparation

1. Five milliliters of patient's saliva have been collected in a plastic clean dry collection tube of 5 cm in length and kept in a small cold ice box immediately after collection.
2. The samples have been centrifuged at 3000 rpm for fifteen minutes then stored at -80 C freezer until the salivary sample supernatants of all the groups were ready for analysis.
3. Concentration of salivary immunoglobulin A (SIGA) has been measured by using Enzyme Linked Immunosorbent Assay (ELISA) Kit (96 wells) for quantitative determination of IgA in human saliva.

Microbial Adhesion Sampling

1. One month after delivery of dentures, swaps were taken from the fitting palatal surface of the upper denture according to 2cm □2cm template delimiting the area to be swabbed; this was done immediately after removal of the denture.
2. Samples were placed in tubes containing 4.5ml of sterile phosphate buffer saline PBS and immediately transported to the laboratory for concentration by centrifugation.
3. They cultured and enucleated on sabouraud dextrose agar plates to detect *Candida albicans* and mitis salivarius agar to detect *streptococcus mutants* then they were incubated at 37 C° for 48 hours
4. After that, the flexible dentures were duplicated from the conventional one, rest of steps were done as before.

RESULTS

TABLE (1) comparison between conventional denture base and flexible denture base regarding colony forming unit of streptococcus mutants in patients

Colony forming unit of Streptococcus mutants	Conventional denture base	Flexible denture base	Independent t-test	
	No.=10	No.=10	t	P-value
Mean±SD	10500000 ± 8868045	107840 ± 194575.9	3.705	0.002
Range	3900000 – 29000000	2900 – 490000		

TABLE (2) show the results of comparison between conventional denture base and flexible denture base regarding colony forming unit of Candida in patients.

Colony forming unit of Candida	Conventional denture base	Flexible denture base	Independent t-test	
	No.=10	No.=10	t	P-value
Mean±SD	3226364 ± 2755682	20127.27 ± 18368.73	3.859	0.001
Range	350000 – 6400000	3800 – 52000		

TABLE (3) show the results of Comparison between conventional denture base group and flexible denture base group regarding SIgA levels.

SIGA μg/ml		**Conventional denture base**	**Flexible denture base**	**Independent t-test**	
		No.=10	**No.=10**	**t**	**P-value**
Pre-operative	Mean±SD	11.23 ± 10.38	10.15 ± 8.28	0.257	0.800
	Range	2.1 – 30.8	3.4 – 25.5		
Post-2hours	Mean±SD	13.97 ± 10.67	10.94 ± 4.77	0.820	0.423
	Range	2.8 – 38.7	3.9 – 19.2		
Post- 3days	Mean±SD	19.77 ± 14.88	17.25 ± 10.48	0.438	0.667
	Range	2.4 – 42.5	7.4 – 40.3		
Post-7days	Mean±SD	31.66 ± 14.02	18.10 ± 12.53	2.280	0.035
	Range	7.2 – 48.8	7.8 – 43.1		

P > 0.05: Non-significant P < 0.05: Significant P < 0.01: Highly significant.

DISCUSSION

As regard to colony forming unit of streptococcus mutants, the results of this study revealed a significant difference between flexible dentures and conventional dentures whereas the conventional one showed higher mean values. The results as regard colony forming unit of Candida albicans revealed also highly significant differences between the two types of dentures where the conventional showed higher mean values. This was attributed to

surface roughness of the fitting surface of the denture. Rougher surfaces can cause discoloration of the prosthesis, be a source of discomfort to patients and it may also contribute to microbial colonization and biofilm formation. Bacterial and fungal species have more of a propensity to adhere to rough denture base materials[9]. These results suggested that the surface roughness of flexible dentures was less than the conventional one. Re-searches demonstrated that material surface roughness and porosity encourage microbial buildup[9, 10]. The present findings were also in accordance with an invitro study made by Patil RA and his colleagues[11]to compare the biofilm development of Candida albicans on two different types of abraded surfaces of heat cure PMMA and flexi denture material. They concluded that Flexible denture material has lesser biofilm development and Candidal count as compared to heat cure acrylic resin. The materials with the roughest surface may serve as reservoir, with surface irregularities providing an increase microorganism retention and protection from shear forces, rough surface has irregularities inducing adhesion of Candida and bacteria, these superficial defect such as voids and micro cracks on surface were possible sites for Candidal adhesion [9]. These results may be attributed to human factor (skills of technician, level of attention) or due to properties of the material. Also these results may be attributed to injection molding system using cartridge which eliminate dosage errors, ensuring long term stability of the shape, reduced contraction, as well as mechanical resistance with aging or it may be due to the recent modification in polyamide which improve the physical properties [12, 13]. Such findings could be confirmed by the previous observation of Aslanimehr et al. who found that significant reduction of Candida albicans adherence to the injection acrylic resin materials makes them valuable for patients with high risk of denture stomatitis[14]. Charged acrylic resins were shown to have a dose-related anti-microbial activity[15]. Bhargava, Ankur, and Sonal Saigal [16] . Suggested that there was significant association between denture type and Candida colony forming unit count. However, the results of this study were in disagreement with Abuzar et al. [17] who evaluated the surface roughness of a polyamide denture base material (Flexi- plast) in comparison with PMMA (Vertex RS), and found that polyamide specimens produced a rougher surface than PMMA, both before and after the polishing process. Another study revealed that the highest Candida species biofilm growth was shown to occur on polyamide resin when compared with PMMA[18].Analysis of the results revealed non-significant difference between the two types of dentures in the pre-operative measures, post 2 hours and post 3 days. while the post 7 days' measure showed statistically significant difference between the two types of denture where the conventional acrylic denture showed higher levels of SIgA mean values. This result may be attributed to the higher level of microbial adhesion in conventional dentures than in flexible ones. There is a direct proportion between microbial invasion and immune system response represented by elevation of the level of salivary immunoglobulin a to face the elevated microbial count [19]. This continuous increase might be attributed to the psychological stress and hyper salivation associated with early patient reaction to the complete denture. This explanation shows some harmony with the opinion of several authors [20, 21]. The results of this study were in disagreement with Dehis [22] who found that wearing complete dentures is immunosuppressive. This difference may be attributed to the patient's factor. In his study, although it has an age range from 50 to 60 years like the current study, he grouped the patients into groups. Each group received one type of denture unlike current study that made the measurements of two types of dentures on the same patients with one-month interval to avoid expected bias resulted from patients grouping.

CONCLUSIONS

Within the limitation of the study it was concluded that:

1) Both the conventional and flexible denture bases have the affinity to support growth of streptococcus and Candida albicans.

2) Flexible denture bases were less in microbial adhesion on their fitting surfaces than conventional denture bases because flexible dentures were less in roughness than conventional dentures, thus flexible dentures are more hygienic and biologically compatible.

3) The levels of salivary immunoglobulin A (SIGA) are higher in conventional denture bases than in flexible denture bases after 7 days. This could be attributed to higher numbers of colony forming unit of both streptococcus mutants and Candida albicans.

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