

## CYTOTOXICITY AND MICROBIOLOGICAL ASSESSMENT OF MAXILLARY ACRYLIC RESIN COMPLETE DENTURES REINFORCED BY GOLD PLATED CR-CO PALATAL PLATE (A RANDOMIZED CONTROLLED CLINICAL TRIAL WITH LABORATORY ASSESSMENT)

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### ABSTRACT

**Background:** Virulence factors of denture stomatitis had been contributed mostly to Staphylococcus aureus and Candida Albicans as an etiological basis in addition to immunodysfunction related to type of denture base and significant Ab-Ag reaction according to individual immune response. This study aimed to evaluate gold electro plating effect on Staphylococcus aureus and Candida Albicans colonization in addition to assessment of its cytotoxicity level. **Subjects and Methods:** Twelve maxillary completely edentulous patients were randomly divided into; Group I (control group): Completely edentulous patients rehabilitated with maxillary complete dentures reinforced with Cr-Co palatal plate and Group II (experimental group): Completely edentulous patients rehabilitated with maxillary complete dentures reinforced with Cr-Co palatal plate which were galvanic electro-plated by gold. Follow up assessment of Staphylococcus aureus, Candida Albicans and cytotoxicity level of Group I and Group II. The collected data were further statistically analyzed using SPSS and GraphPad statistical software. **Results:** Using One Way ANOVA followed by Tukey's post hoc test for multiple comparisons regarding microbiological evaluation, there was a significant difference among follow up periods for both groups. Using Independent t-test for comparison between both groups, was a significant difference especially after two weeks, 30 days and 45 days. While for cytotoxicity level, Gold-Plated experimental group showed insignificant higher cytotoxic effects than Cr-Co control group using Chi-Square test. **Conclusion:** Gold electroplating showed an antibacterial effect and lower rate of Candida Albicans colonization while it showed higher levels of cytotoxicity.

**KEYWORDS:** Denture, Gold, Candida, Staphylococcus, Cytotoxicity

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## INTRODUCTION

An inflammation that mostly affects the palatal mucosa when complete or partial dentures are worn is known as denture-related stomatitis. Patients with denture stomatitis had strong correlations between inflammation and microbiological colonization, according to several researchers<sup>(1)</sup>. Denture stomatitis has been linked to the colonization of *Staphylococcus Aureus*, *Staphylococcus Epidermis*, and *Candida Albicans* as the main cause of its onset, aggravation, and maintenance<sup>(2)</sup>.

Since Cawson and Budtz-Jorgensen's investigations revealed that oral microbiology, particularly *Candida* species, are crucial for the development of denture stomatitis, the significance of oral microbes in the etiology of stomatitis connected to dentures is well acknowledged<sup>(3)</sup>. Different species are thought to cause denture stomatitis by a variety of mechanisms, including the release of irritants, toxins, and yeast antigens from the plaque on the dentures<sup>(4)</sup>.

Furthermore, intraoral prosthesis like acrylic dentures serves as a reservoir for germs. An environment with low oxygen levels and an acidic pH, which is conducive to *C. Albican's* growth, is created when a denture is worn; this may be aided by decreased salivary flow beneath the specially shaped denture base<sup>(5)</sup>.

Micro pits and microporosities are typically visible on the tissue surface of dentures. It is challenging to mechanically or chemically clear out microorganisms that are hiding in these places. According to various *in vitro* investigations, yeasts appear to cling well to denture base materials and the microbial contamination of denture acrylic resin happens extremely quickly. Surface roughness might encourage microbial growth and illness<sup>(6)</sup>. A fine texture and lack of porosity in the dentures prevented plaque from adhering to surface flaws or adhering mechanically to surface irregularities. *In vitro* studies demonstrated that *C. Albicans* entered

the widely used acrylic resin, and the combination of fluorescent dye and *C. albicans* allowed for its confirmation. When in touch with the mucosa, the unpolished surface penetrated more deeply than the polished surface<sup>(7)</sup>.

There has been a lot of discussion in the literature over whether base metal or gold alloys should be used for removable denture frameworks. Although both alloys have excellent qualities, neither is best suited for every part of the framework of the removable denture, according to previous reviews<sup>(8)</sup>.

Inlays were initially electroformed with pure gold in 1961 in conservative dentistry, as previously reported in *Gold Bulletin*. Within the electroplating sector, namely in the creation of jewelry, decorative goods, and watches, the method itself has been well-known for more than 150 years. In the past ten years, restorative and conservative dentists have used the electroforming procedure more frequently<sup>(9)</sup>.

Dental restorations can be electroformed using non-cyanide baths that contain a gold sulfite electrolyte with the chemical formula:  $[\text{Au}(\text{Amin})_2(\text{SO}_3)_2]^{3-} \rightleftharpoons [\text{Au}(\text{Amin})_2]^+ + (2 \text{SO}_3)^{2-}$ <sup>(10)</sup>

To predict a material's biocompatibility, one must be aware of its dissolving characteristics, cytotoxicity, and, regarding metals and alloys in the mouth, information on electrochemical potentials. Saliva has electrochemical potentials for the metals and alloys found in human mouths. When conducting electrochemical testing, an alloy's electrochemical potential is measured against a saturated calomel reference electrode<sup>(11)</sup>.

Between two different alloys, or between a metallic dental restoration and a gold cube used as a reference electrode on the oral mucosa, it is feasible to measure the electrochemical potentials in the mouth. Corrosion is a consequence of low breakdown potentials. A patch test-based epidemiological investigation has been conducted about the potential for an allergic reaction to dental

alloys<sup>(12)</sup>. Following these experiments, an attempt has been made to rank the alloy groups according to their projected “allergenic potential.” There is an “allergenic potential” in every alloy and group of alloys. No statistically significant differences between any individual group, however, have been observed, except the nickel group<sup>(13)</sup>.

It is debatable whether exposure to contact allergens through the oral mucosa might result in sensitization. According to some authors, such exposure only serves to exacerbate pre-existing sensitivity (in those who have already been sensitized by skin contact) or to foster tolerance in those who have not yet been sensitized. However, among people who have dental gold, there has been a large overrepresentation of gold allergy<sup>(14)</sup>.

The development of electroforming technology with pure gold has opened up new research opportunities. This method of electroforming pure gold is still too soft to be used in areas of dental restorations that are subjected to high stresses. The deposition of multilayers by electrochemical structural manipulation may be the subject of further research in electroforming. It should be the goal of both of these research projects to create dense, highly resistant deposits of pure gold<sup>(15)</sup>.

It doesn't matter what the restoration material is; it spends a lot of time close to the oral tissues. Dental materials biodegrade in the oral environment due to bacterial activity, wear and erosion brought on by food and chewing, chemical and physical breakdown by saliva, and other factors<sup>(16)</sup>. The reactivity of the substance in the oral cavity, which is governed by the kinetics of electrochemical reactions and the principles of thermodynamics, must thus be evaluated. This suggests that the saliva-material system is forced toward a state of thermodynamic equilibrium when a substance is placed in the mouth. The substance either transforms to its ionic form at equilibrium or stays stable in its elemental state<sup>(17)</sup>.

## SUBJECTS AND METHODS

### Subjects:

Patients of ages ranging from 40-65 years, having edentulous ridges were chosen among individuals enrolled in the Prosthodontics Department's Outpatient Clinic at Badr University in Cairo and all clinical and laboratory investigations performed at Oral and Dental Research Institute, National Research Centre, Giza, Egypt.

The subjects were chosen using stringent inclusion and exclusion standards. The inclusion criteria included: the edentulous ridges covered by healthy mucosa, acceptable bone status verified by preliminary cone-beam CT, Adequate inter-arch space to accommodate maxillary and mandibular prostheses, normal maxillary-mandibular relationship Angle Class I, and finally good physical and mental health to give informed consent.

The exclusion criteria were: female patients, uncontrolled diabetes, smoking habits, radiotherapy, clinical or radiographic signs of pathologic conditions, osteoporosis, patients on corticosteroids, and bruxism. Only motivated patients were enrolled in the study after signing informed detailed consent.

### Study Design:

This study was conducted as a clinical trial (ClinicalTrials.gov Identifier: NCT05466305). They were randomly categorized following a 1:1 allocation ratio into two groups as follows:

**Group I (control group):** Completely edentulous patients rehabilitated with maxillary complete dentures reinforced with Cr-Co palatal plate.

**Group II (experimental group):** Completely edentulous patients rehabilitated with maxillary complete dentures reinforced with Cr-Co palatal plate which were galvanic electro-plated by gold.

### **Ethical Approval:**

The Medical Research Ethics Committee granted permission for the study to be conducted, which meets at the National Research Centre and operates following the relevant Egyptian laws, the Helsinki Declaration, good laboratory and medical practice guidelines, and WHO regulations regarding the ethics of scientific research. Number of approval: 0127082022

### **Sample Size Calculation:**

With one control for every experimental patient, a study of a continuous response variable was planned using independent control and experimental individuals. In a previous study <sup>(18)</sup>, the response within each patient group was normally distributed with a standard deviation of 0.1467. If the true difference between the experimental and control means is 0.261, then we need to study 6 experimental and 6 control patients (a total of 12 patients) in order to be able to reject the null hypothesis that the population means of the experimental and control groups are equal with a probability (power) of 0.8 with type I error probability of 0.05.

### **Methods:**

#### **a) Chrome Cobalt Palatal Plate Construction:**

Standardized clinical procedures were followed for the fabrication of complete dentures. After taking preliminary alginate impressions (*Cavex, Holland*), an acrylic (*Acrostone, Egypt*) custom tray was fabricated for each patient. Functional impressions were taken using Zn-oxide eugenol impression (*Cavex, Holland*) and master casts (*Type IV dental stone, BMS, Egypt*) were fabricated. Master casts were duplicated using silicone (*Dental duplicate silicone, Dentsil, Netherlands*) for chrome cobalt metal base fabrication. After this procedure, refractory copies of the master casts were provided.

Wax patterns (*Bego, Germany*) were used for framework design. Patterns were then sprued with

an inverted sprue, invested, and cast in Chrome Cobalt alloy (*SYSTEM NH, Adentatec, Germany*) using the conventional lost wax technique. The metal framework was returned to the master cast and checked for proper seating and fit. Then, one layer of base plate wax 1.5 mm in thickness was adapted to the remaining parts of the denture base and the buccal and labial flanges of the denture.

Artificial teeth (*Acrostone Plus, Egypt*) were arranged followed by waxing up the trial denture base, and then the denture was flaked and processed in heat-cured acrylic resin (*Acrostone Egypt*) in the conventional long curing cycle.

After deflasking of the dentures, they were left to bench cool to room temperature. Then, excess material was trimmed, and the dentures were polished with a wet rag and pumice while keeping them on their respective casts to avoid any distortion before measurement of the adaptation accuracy. The dentures were finished and polished before patient delivery.

#### **b) Gold Electro-Plating of Chrome Cobalt Palatal Plate:**

Following the previous standardized laboratory step of Cr-Co palatal plate construction, air abrasion using sandblasting (*Basic Classic; Renfert GmbH, Hilzingen, Germany*) for inner and outer surfaces with 110  $\mu\text{m}$  aluminum oxide. Then, the restoration was cleaned with an ultrasonic cleaner (*Sonica, Soltec, Italy*) and then with a steam cleaner (*Polaris, US*).

Contact ligature wire was attached (0.25 mm) to the metal framework to be gold-plated. A ligature wire (0.25 mm) attached to the surfaces was used to be coated to hang and stabilize the framework on the hanging device. The framework was stabilized horizontally with the wire so that gas bubbles cannot be trapped inside the framework.

The framework was connected to the large plating head with the hanging device. The amount

of gold chloride solution required to gold-plate the surfaces was determined, considering the manufacturer's recommendations. The galvanic electroplating procedures were performed in the electrolysis device following the manufacturer's instructions (2-3 volts with 5.5 minutes required to deposit one-micron thickness gold) then rinsing using tap water was required, figure (1) and (2).

**c) Patient's Instructions and Recall:**

Patients were asked to contact if any pain is experienced, one or two weeks later to perform any needed post-insertion adjustments. Patients were instructed to clean the dentures after every meal under running water only.

They were asked to remove the prosthesis at night to allow tissue rest and immerse it in a cup of tap water in addition not to using any type of denture cleansers. Toothpaste and solutions containing phenol were avoided to prevent abrasion and crazing of the denture. Hot water was avoided to prevent the warpage of acrylic resin denture base material.

Frequent follow-up appointments were scheduled to ensure proper oral and denture hygiene. All patients were recalled after one week, two weeks, 30 days and after 45 days of denture insertion for collection of the samples, evaluation of the prosthesis as well as the condition of the denture-bearing mucosa.



Fig. (1) Galvanic Electro Plating.

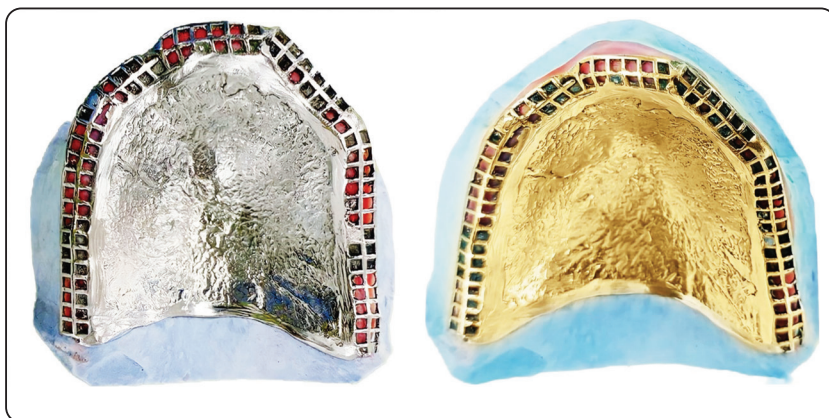


Fig. (2) Chrome Cobalt and Galvanic Gold Plated Palatal Plate.

#### d) Microbiological Assessment:

The protocol for collecting samples was as follows: each patient was asked to rinse their mouths with 20 ml of 1% Betadine mouthwash for 30 seconds, then swab their palatal tissues and denture-fitting surfaces for 30 seconds. To ensure appropriate adherence to the plaque, the swab is rotated three times while being held firmly and laterally against the denture. The vestibular and palate tissues also underwent a comparable procedure. Swabs were immediately placed in a readily available, sterile swab container, kept in the cold box for less than two hours, and then prepared for plating.

The number of Colony Forming Units per ml (CFUs) (ISO 4833, 2003), which was determined using the following procedure, was used to quantify the *Staphylococcus Aureus* colonies: To create culture homogenate, the swab tip of each sample was cut using sterile scissors, placed in 9 ml of sterile 1 percent buffered peptone water, and well mixed for 30 seconds using a vortex (initial dilutions). Up to the fourth dilution, this dilution was successively diluted into further decimal (tenth fold) dilutions. Plate count agar Petri dishes were infected in duplicate by dispersion with 0.1ml. from each dilution to determine the overall bacterial count. The sample was incubated at 30°C for 72 hours. Negative counts for enumeration were made for plates with fewer than three colonies (plates that had 3 to 30 colonies only are to be stated). The total bacterial count (TBC) was assessed as

follows according to the ISO protocol (ISO.18593, 2004): TBC = Colonies count at last positive plate X 10 X dilution (10n).

Colonies of different morphologies were extracted from positive plates, transferred to various sterile buffered peptone water 1 % test tubes, and cultured at 30°C (AOAC, 2000) for 12 hours, for productivity. Loopful is taken out of the peptone test tubes and spread by streaking across particular media surfaces. (**EMB. Eosin Methylene Blue, Rose Bengal Agar, and blood agar media**) to recognize the isolated micro-organisms. Therefore, the isolated colonies were assessed and recognized according to their morphology (ISO.4832, 2006a, 2006b; ISO.16649-2, 2001), figure (3).

For *Candida Albicans* assessment, Chromogenic agar medium was prepared according to manufacturer instructions with 21.36 grams of agar suspended in 500 ml of distilled water. The suspension was boiled in a vacuum dispenser with frequent agitation and the preparation was left to cool at 50° c and the suspension was poured on sterile Petri dishes. The swab was inoculated immediately in a sterile glass tube containing 1 ml of sterile saline and transferred in a dry container to the laboratory within one hour. The sample was placed on a mechanical agitator for at least 60 seconds. The sample was then plated on the Chromogenic agar medium and was spread using a sterile metal loop, whose end was flamed to facilitate the plating and spreading procedure.

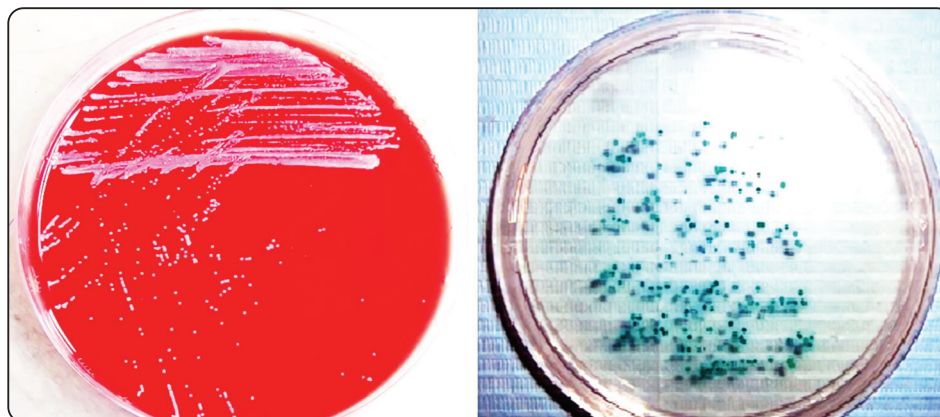


Fig. (3) *Staphylococcus Aureus* and *Candida Albicans* Evaluation.

The plates were covered and left for one minute to dry and then were inverted immediately before being placed in a special incubator for 48 hours at 37°C. Dishes were removed from the incubator after the 48-hour incubation period and examined visually for light green colonies that resembled *Candida albicans* colonies on *Candida* differential chromogenic agar, figure (3).

**e) Cytotoxicity Assessment:**

Before testing, the samples were cleansed ultrasonically in distilled water for 20min and exposed to ultraviolet light for another 20min to kill any microorganisms that may have caused contamination during fabrication. All the following procedures were carried out in a sterile area employing a Laminar flow cabinet Steril GARD class III advance. Normal Fibroblast Cells (BJ1) have been suspended in Dulbecco modified Eagle medium F12 (DMEM F12), 1% antibiotic-antimycotic mixture (10,000U/ml Potassium Penicillin, 10,000µg/ml Streptomycin Sulfate and 25µg/ml Amphotericin B) and 1% L-glutamine at 37 °C under 5% CO<sub>2</sub>. BJ1 Cells have been batch cultured for 10 days, then seeded at a concentration of 10x10<sup>3</sup>cells/well in fresh complete growth medium in 48-well microtiter plastic plates at 37 °C for 24 h under 5% CO<sub>2</sub> utilizing a water jacketed Carbon Dioxide incubator, figure (4).

Media were aspirated then fresh medium (without serum) was added and cells were incubated either alone (negative control) or with different concentrations of agent. After 72 hours of incubation, the medium was aspirated then 40µl MTT salt (2.5µg/ml) was added to each well and incubated for a further four hours at 37 °C under 5% CO<sub>2</sub>. Then 200 µL of 10% Sodium dodecyl sulfate (SDS) in deionized water has been added to each well and incubated overnight at 37 °C. A positive control composed of 100µg/ml was used as a known cytotoxic agent which gives 100% lethality under the same conditions.

The absorbance was then measured by a Microplate multiwell reader at 595nm and a reference wavelength of 620 nm. Dimethyl Sulfoxide (DMSO) is the vehicle utilized for the dissolution of plant extracts and its final concentration in the cells was less than 0.2%. The percentage of change in viability was calculated according to the formula: (Reading of extract/Reading of negative control) x 100.

Following cytotoxic activity investigations against the normal human epithelial cell line BJ1 (normal Skin fibroblast), the following classification was scored; More than 90 percent cell viability: nontoxic. 60-90 percent cell viability: slightly toxic. 30-59 cell viability: moderate cytotoxic. Less than 30 percent cell viability: severely cytotoxic.

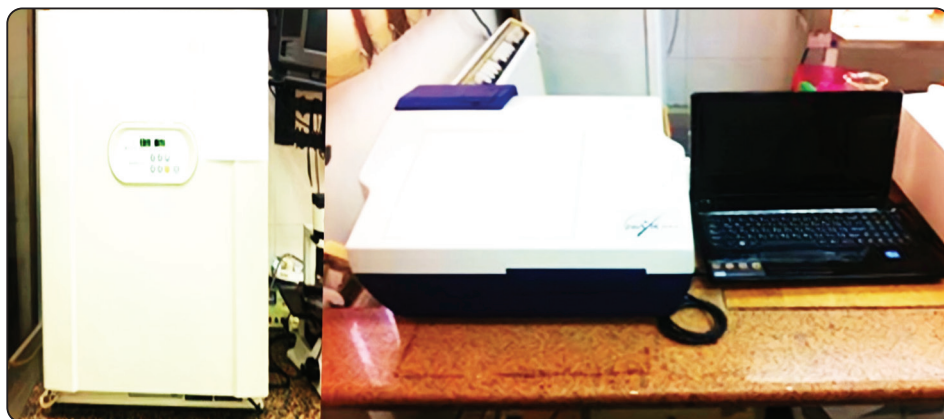


Fig. (4) Cytotoxic Evaluation using Water jacketed Carbon Dioxide Incubator and Microplate Multiwell Reader.

### f) Statistical Analysis:

Statistical analysis of the given data was performed using IBM SPSS software package version 24.0. (Armonk, NY: IBM Corp, US) and Graph Pad Prism (GraphPad Software, Inc, US). Performing analysis for each group separately along time intervals using One Way Analysis of Variance (ANOVA) followed by Tukey's post hoc test for multiple comparisons in addition to independent t-test for significance evaluation between both groups at definite intervals. CFU data were revealed as a mean and standard deviation which were logarithmically transformed into parametric data.

## RESULTS

For Staphylococcus Aureus and Candida Albicans colonization among both groups, log 10 mean  $\pm$  standard deviations of group I (Cr-Co group) showed a significant increase. While for group II (Gold Plated group), log 10 mean  $\pm$  standard deviation showed a significant decrease during study follow-up using the One-Way ANOVA test as P-value < 0.05, listed in table (1) and (2).

Using Tukey's post hoc test for multiple comparisons to reveal the effect of time on CFU log 10 of both groups, it concluded that there was a significant difference between 30 days and 45 days' time intervals (indicated by different letters) for group II (Gold Plated group) as P-value < 0.05. While there was insignificant difference between them in group I (Cr-Co group) as P-value > 0.05, listed in the table (1) and (2).

An independent t-test was performed to reveal the level of significance between group I (Cr-Co group) and group II (Gold Plated group) regarding Staphylococcus Aureus colonization. After one week, there was an insignificant difference between group I (Cr-Co group) and group II (Gold Plated group) ( $1.779 \pm 0.094$  and  $1.791 \pm 0.047$ ) respectively, as P-value > 0.05. While after two weeks, 30 days, and 45 days, there was a significant difference for group I (Cr-Co group) ( $1.814 \pm 0.057$ ,  $1.928 \pm 0.079$ , and  $1.983 \pm 0.083$ ) respectively, as P-value < 0.05. In addition to group II (Gold Plated group), there was a significant difference ( $1.725 \pm 0.028$ ,  $1.432 \pm 0.076$ , and  $1.166 \pm 0.036$ ) respectively, as P-value < 0.05, as in table (1).

TABLE (1) Mean and standard Deviation of Staphylococcus Aureus CFU (Log10) among 45 Days Follow Up in Group I and Group II:

	Total N	Group I (Cr-Co Control group)		Group II (Gold Plated Experimental group)		t	df	95% CI	P-value
		M (log10)	SD	M (log10)	SD				
After One Week	12	1.779 a	0.094	1.791 a	0.047	0.2797	10	-0.08360 to 0.1076	0.7854 (ns)
After Two Weeks	12	1.814 ab	0.057	1.725 a	0.028	3.433	10	-0.1468 to -0.03123	0.0064 *
After 30 Days	12	1.928 bc	0.079	1.432 b	0.076	11.08	10	-0.5957 to -0.3963	<0.0001 *
After 45 Days	12	1.983 c	0.083	1.166 c	0.036	22.12	10	-0.8993 to -0.7347	<0.0001 *
<b>P-value</b>		<b>0.0007 *</b>		<b>&lt;0.0001 *</b>					

N; Number, M; Mean, SD; Standard Deviation, P; Probability Level Df; Degree of Freedom, CI; Confidence Interval at 95% Means with same superscript letter in the same column were insignificant different using Tukey's post hoc test Means with different superscript letter in the same column were significant different using Tukey's post hoc test NS; Insignificant Different \*Significant Different



TABLE (2) Mean and standard Deviation of Candida Albicans CFU (Log10) among 45 Days Follow Up in Group I and Group II:

	Total N	Group I (Cr-Co Control group)		Group II (Gold Plated Experimental group)		t	df	95% CI	P-value
		M (log10)	SD	M (log10)	SD				
After One Week	12	1.598 a	0.0751	1.667 a	0.0514	<b>1.857</b>	<b>10</b>	<b>-0.01378 to 0.1518</b>	<b>0.0929 (NS)</b>
After Two Weeks	12	1.688 a	0.0688	1.245 b	0.0278	<b>14.62</b>	<b>10</b>	<b>-0.5105 to -0.3755</b>	<b>&lt;0.0001 *</b>
After 30 Days	12	1.891 b	0.0439	1.119 c	0.0967	<b>17.81</b>	<b>10</b>	<b>-0.8686 to -0.6754</b>	<b>&lt;0.0001 *</b>
After 45 Days	12	1.906 b	0.0591	1.058 c	0.1071	<b>16.98</b>	<b>10</b>	<b>-0.9593 to -0.7367</b>	<b>&lt;0.0001 *</b>
<b>P-value</b>		<b>&lt;0.0001 *</b>		<b>&lt;0.0001 *</b>					

*N*; Number, *M*; Mean, *SD*; Standard Deviation, *P*; Probability Level *Df*; Degree of Freedom, *CI*; Confidence Interval at 95% Means with same superscript letter in the same column were insignificant different using Tukey's post hoc test Means with different superscript letter in the same column were significant different using Tukey's post hoc test *NS*; Insignificant Different \*Significant Different

While for Candida Albicans Colonization, an independent t-test was performed to reveal the level of significance between group I and group II. After one week, there was an insignificant difference between group I (Cr-Co group) and group II (Gold Plated group) ( $1.598 \pm 0.0751$  and  $1.667 \pm 0.0514$ ) respectively, as  $P$ -value  $> 0.05$ . While after two weeks, 30 days, and 45 days, there was a significant difference for group I (Cr-Co group) ( $1.688 \pm 0.0688$ ,  $1.891 \pm 0.0439$ , and  $1.906 \pm 0.0591$ ) respectively, as  $P$ -value  $< 0.05$ . In addition to group II (Gold Plated

group), there was a significant difference ( $1.245 \pm 0.0278$ ,  $1.119 \pm 0.0967$  and  $1.058 \pm 0.1071$ ) respectively, as  $P$ -value  $< 0.05$ , as in table (2).

Regarding group I (Cr-Co group) and group II (Gold Plated group) cytotoxicity, the viability percent of specimens were (96.5%) and (92%) respectively, as listed in the table (3). Performing the Chi-square test of significance, it revealed an insignificant difference between both groups as  $P$ -value  $> 0.05$ , listed in the table (3).

TABLE (3) Descriptive and Comparative Study of Cytotoxic activity using Cell Viability Test:

	Cytotoxicity %	Viability %	Cytotoxic Level	Chi-Square	P-value
Group I (Cr-Co Control group)	4.5	96.5	Non-Toxic	<b>1.4184</b>	<b>0.2336 (NS)</b>
Group II (Gold Plated Experimental group)	8	92			

*P*; Probability Level

*NS*; Insignificant Different using Chi-Square Test

## DISCUSSION

Numerous factors have been identified as contributors to denture stomatitis in studies, including traumatic occlusion, poor oral and denture hygiene, microbial factors, the age of the denture, allergies to the denture base materials, residual monomer, thermal insulation below the denture, smoking, different types of radiotherapy, xerostomia, systemic conditions, such as diabetes mellitus and immunity malfunctions, diet insufficiencies, and medical drugs.<sup>(19)</sup>

The high rate of *Staphylococcus* colonization in this study, ranging from  $1.779 \pm 0.094$  to  $1.983 \pm 0.083$  for metal denture base, is attributable to several researches that discovered denture wearers had higher levels of monocyte activation than those who didn't wear them but were less likely to experience phagocytosis, suggesting that *Staphylococcus aureus* had stronger virulence characteristics<sup>(7)</sup>. Additionally, according to recent studies, asymptomatic denture wearers and elderly individuals who come with denture stomatitis both frequently and equally have their *Staphylococcus aureus* strains detected<sup>(20)</sup>. Since *Staphylococcus aureus* may attach to prosthetic materials and has a high virulence factor, recent study suggests that it may also play a role in the onset of denture stomatitis.<sup>(21)</sup>

*Staphylococcus* colonization rates decreased in the current study, from  $1.791 \pm 0.047$  to  $1.166 \pm 0.036$  for metal denture bases that were gold-plated, because of the antibacterial properties of the gold particles<sup>(22)</sup>. Gold particles and bacterial cell membranes are electrostatically attracted to one another, which tends to increase particle accumulation on the bacterial cell membrane. This may greatly stress the bacterial membrane allowing the passage of gold particles into the cytoplasm, and ultimately result in cell lysis. Theoretically, DNA replication may be impacted by the interaction of gold-particles with bacterial proteins' thiol groups<sup>(23)</sup>.

The tissue surface of dentures typically exhibits microporosities and micropits that favor the

colonization of *Candida albicans*. According to various in vitro investigations, yeasts appear to stick to denture base materials well and soon get contaminated with denture bases. Rough surfaces may make it easier for bacteria to stick around and cause infection. Plaque adhesion by mechanical fixation to surface irregularities or by penetration of surface defects was not possible with dentures because of their fine texture and lack of porosity. A fluorescent dye and *C. Albicans* were used to demonstrate in vitro that *C. Albicans* permeated the widely used acrylic resin. Compared to the polished surface, the unpolished surface had more penetration into the mucosa<sup>(19)</sup>.

An electroplating bath was developed to improve surface properties and other mechanical properties. The bath contains 10 g/L gold, has a pH of 7.2, and is operated at 0.5 A/dm<sup>2</sup> and 65° C. Gold is deposited at the rate of 118 mg/A-min. Excellent stability and an indirect anti-adhesion property against *Candida albicans* colonization are provided by a particular finishing agent. One or more bath compositions may be used to electrodeposit a metal. Two or more metal ions can be deposited in two or more baths; different metals can be deposited in different baths. Additionally, additives may be added to the mixture for purposes such as surface cleaning, crack reduction, and surface stress elimination.<sup>(24)</sup>

Certain safety measures must be taken when using cyanide solutions. As the gold electrolyte is alkaline and the nickel electrolyte is acidic, they should not be in direct contact. When moving castings from one bath to another, water must be used to rinse them. The casting is extensively rinsed in alternate hot and cold-water rinses and submerged in 10% hydrochloric acid for several hours following gold deposition in order to remove any potential residues of leftover cyanide. Electrolytes without cyanide have been created, which may be useful to the dentistry industry. They have not yet been thoroughly described in detail; however, their deposits are not as hard as brilliant acid gold deposits.<sup>(25)</sup>

In the current study, the cytotoxic properties of elements found in gold alloy and metal-based fabricated prostheses were assessed by examining the impact of ions produced from those materials into human saliva as salt solutions. It has been selected to test the cytotoxicity using salt solutions since metallic and organometallic salts are created when dental materials corrode in the mouth. Additionally, salt solutions made with anions like chloride or sulphate at low concentrations were ineffective.<sup>(11)</sup>

It has been revealed that Zn was the most toxic element released from gold crowns into the mouth. A moderate position in the order of cytotoxicity was also reported for Ag. Following another discovery, which indicated that pure Ag metal had an intermediate cytotoxicity rank between the high toxicity of Cu and the low toxicity of Pd, this moderate position was established.<sup>(26)</sup>

## CONCLUSION

Within the limitations of this study, the following conclusions could be obtained; Denture insertion provides a saprophytic environment, which encourages microbiological growth. The microbial adhesion potential to the gold electroplated chrome cobalt denture base is lower than the non-gold plated one. While for the cytotoxicity effect of gold plating, it reveals a comparable cytotoxic effect rather to non-gold plated.

## Financial Support and Sponsorship

Nil.

## Conflict of Interest

There were no conflicts of interest.

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