

## **“THE ADHERENCE OF CANDIDA ALBICANS ON SURFACE OF DIFFERENT DENTURE BASE MATERIALS (AN IN-VITRO STUDY)”**

<sup>1</sup>Dr. Hemal Agrawal, <sup>2</sup>Dr. Rupal Shah, <sup>3</sup>Dr. Neha Agrawal

<sup>1</sup>Assistant Professor, Dept. of Prosthetic Dentistry,  
Government Dental College & Hospital, Ahmedabad

<sup>2</sup> Professor & Head, Dept. of Prosthetic Dentistry,  
Government Dental College & Hospital, Ahmedabad

<sup>3</sup>Senior Lecturer, Dept. of Conservative Dentistry & Endodontics,  
Karnavati School of Dentistry, Uvarsad, Gandhinagar

**Corresponding author:** Dr. Hemal Agrawal, **Email Id :** drhemal79@gmail.com

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### **ABSTRACT:**

**Introduction:** Over the years researchers have reported on the frequency and distribution of yeast in the oropharynx of apparently normal individuals and those with systemic or mycotic diseases wearing dentures. Hence this study was undertaken with the objective of evaluating the in-vitro adherence of Candida Albicans to different denture base materials namely self cure acrylic resin, heat cure acrylic resin & chrome cobalt alloy.

**Materials and Method :** Thirty set of specimens were made per denture base material (Ashwin self cure acrylic resin, Lucitone 199-Dentsply heat cure acrylic resin and Chrome cobalt alloy-Bego, Germany). The specimen size was 1cm X 1cm (length) and 2mm (width). Culture preparation and growth of Candida Albicans on the specimens was conducted.

**Results & Conclusion :** From our present study, it can be concluded that Chrome cobalt alloy showed less adherence to candidial cells, followed by heat cure acrylic resin and last self cure acrylic resin. The results were statistically highly significant.

**Keywords :** candida albicans, acrylic resin, chrome cobalt alloy

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### **INTRODUCTION:**

The human oral cavity is known to harbor a multitude of organisms. Among them, Candida albicans has lately become a cause of great concern to the dental profession. Candida albicans has been termed as a notorious “opportunistic pathogen” amongst the Candida species.<sup>1</sup> Among the edentulous individuals most particularly the elderly, this pathogen has been observed to harbor the oral cavity.<sup>2-4</sup> An essential prerequisite for successful candidial colonization and infection is the ability of the yeast to adhere to the superficial epithelial cells as well as to the fitting surface of the denture, considering the latter as the reservoir of infection.<sup>5</sup> The purpose of this study is, therefore, to conduct an in-vitro experiment to evaluate the

amount of surface adherence of Candida albicans to self cure acrylic resin, heat cure acrylic resin and chrome cobalt base metal alloy denture base materials and to compare the adherence of the yeast among these materials.

### **MATERIALS AND METHOD:**

Ideal maxillary stone cast, modeling wax, hydraulic bench press, acrylizer, complete set of finishing and polishing burs, type III dental stone, casting wax, phosphate bonded investment material, induction casting machine, ultrasonic cleaner, glass slides-90, sterile petri dishes-30(9cm diameter), sterile test tubes, sterile Sabouraud’s broth-500ml, sterile distilled water, Candida albicans (strain 192887g), light microscope, gram stains, Ashwin self cure acrylic resin,

Lucitone199-Dentsply heat cure acrylic resin and chrome cobalt alloy-Bego, Germany.

**Sample number:** 30 per denture base material.

**Sample size :** 1cm X 1cm (length) and 2mm (width)

**Preparation of acrylic resin specimens :**

Test specimens were fabricated by scribing a 1 cm X 1cm area of incisive papilla and a part of rugae with a 2H black lead pencil using a flexible ruler on an ideal maxillary stone cast. A 2mm thick modeling wax sheet was cut with a sharp BP knife, with a dimension of 1 cm X 1cm and was adapted on the scribed area of the cast with gentle finger pressure after it was softened in warm water. The cast was then invested using type III dental stone to have a good reproduction of the surface. Keeping in mind the sample size and number, the samples were fabricated by proper mixing of powder and liquid, working time and curing time according to the manufacturer instructions for both types of acrylic resin (Ashwin self cure acrylic resin, Lucitone 199-Dentsply heat cure acrylic resin). These acrylized sheets were trimmed for excess. The polished surfaces of the sheets were subjected to finishing & polishing using the standard laboratory finishing & polishing techniques. While the tissue surface was left untouched, all the samples were washed and stored in sterile distilled water.

**Preparation of chrome cobalt alloy specimens :**

Wax patterns with similar tissue topography on one surface were prepared in casting wax. They were then invested in a 9x casting ring with phosphate bonded investment material. The investment was proportioned as per the manufacturer's specifications and then vacuum mixed and invested. The casting rings were subjected to burnout procedure and cast in an Induction casting machine. The metal specimens were then retrieved and

subjected to sandblasting after cutting the sprues. They were then finished, polished, dried and cleaned with an ultrasonic cleaner.

For means of convenience, all the three denture base materials were designated by Groups and then compared. They were

Group I : Ashwin self cure acrylic denture base material

Group II : Lucitone 199 heat cure acrylic denture base material

Group III : Chrome cobalt alloy denture base material

**Culture preparation :**

The culture preparation and the growth of *Candida albicans* on the specimens prepared was conducted in Government Dental College, Ahmedabad, Gujarat. Laboratory isolates of *Candida albicans* (192887g) were used for this study. Before each adhesion experiment, 10ml of Sabouraud's broth was incubated with *Candida albicans* at 37 ° C for 24 hours. This was then added to 90ml of Sabouraud's broth and further incubated at 37 ° C for 24 hours to attain a stationary phase. The 48 hours broth culture with *Candida albicans* (100ml) was dispensed in six sterile petri dishes. To these, 15 specimens in each of the petri dish containing the broth were introduced. These were then incubated at 37 ° C for 48 hours. After completion of the incubation period, the specimens were removed using sterile forceps to avoid any contamination and washed in sterile phosphate buffered saline. The specimens were then fixed using sterile methyl alcohol for 1 min after which the alcohol was drained. Staining was done using Gram's staining technique (Fig. 1).

Specimens were washed in water and the stained smear was allowed to dry in air. A drop of cedar-wood oil was placed over the specimen that was placed on a glass slide and observed under oil immersion lens (1000x) of microscope. Microscopically, yeast cells are dark purple and show characteristic budding (Fig. 2,3). A total of 10 random fields

were viewed under the light microscope for each of 30 samples, and fields that showed 0 cells, were not included in statistical analysis. After 10 random fields, number of cells were counted and were tabulated and used for statistical analysis.



Fig.1 : Gram staining for all specimens

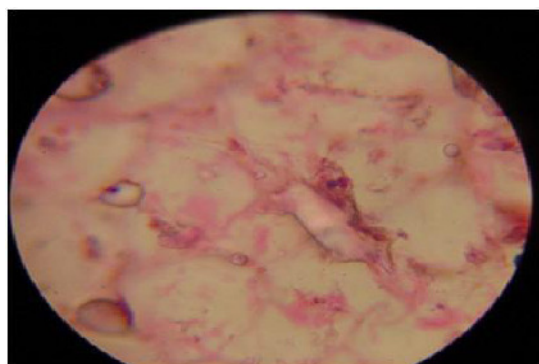


Fig. 2 : Budding cells in self cure acrylic resin specimen

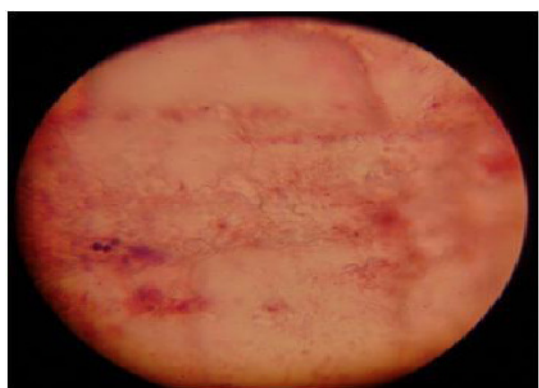


Fig. 3 Candidial cells on Lucitone 199 heat cure acrylic resin specimen

**STATISTICAL ANALYSIS** : The values of the test result were compared using One-way ANOVA (Analysis of Variance) test. The statistical software used for statistical analysis is SPSS Release 6.1.4. From the ANOVA test it was found that the results were highly statistically significant ( $p < 0.001$ ). The adherence of *Candida albicans* was seen to be significantly low in chrome cobalt alloy denture base material, followed by Lucitone 199 heat cure acrylic resin denture base material. Significantly high adherence was noted in Ashwin self cure acrylic resin.

#### **DISCUSSION:**

Edentulousness is not a disease entity in itself, but rather a consequence of pathology. Increasing incidence of edentulousness over the recent years has questioned the adequacy of dental treatment. Yet, the mainstay for the management of the edentulous state till date remains to be an acrylic complete denture. Treatment of these individuals with complete dentures not only rehabilitates them functionally but also esthetically and psychologically. However, prosthetic rehabilitation of the aged has been of great concern. The difficulties that arise may not only be attributed to denture construction but also to associated problems with continuous denture wearing. A commonly occurring condition observed frequently is denture stomatitis secondary to candidial infection. Denture stomatitis is a term applied to an inflammation of the denture bearing mucosa, which may affect as many as two-third of denture wearers.<sup>6</sup> Its incidence has been reported to occur among 11 to 67% of the denture wearers.<sup>7</sup>

The occurrence of an infection is directly related to two major criteria- the virulence of the organism and the number of organisms colonizing the area. These two factors though, independent are related to each other.

**RESULTS :**

The data obtained is shown in Table 1 where the overall totality of cells is shown in all the individual groups.

| <b>Table 1 : Distribution of cells</b> |                             |                             |                             |
|--|-----------------------------|-----------------------------|-----------------------------|
| <b>No. of cells</b>                    | <b>GROUP</b>                |                             |                             |
|  | <b>I</b>                    | <b>II</b>                   | <b>III</b>                  |
| <b>1</b>                               | <b>76</b><br><b>(66.09)</b> | <b>56</b><br><b>(66.67)</b> | <b>58</b><br><b>(81.69)</b> |
| <b>2</b>                               | <b>10</b><br><b>(8.69)</b>  | <b>22</b><br><b>(26.19)</b> | <b>11</b><br><b>(15.49)</b> |
| <b>3+</b>                              | <b>29</b><br><b>(25.22)</b> | <b>6</b><br><b>(7.14)</b>   | <b>2</b><br><b>(2.82)</b>   |
| <b>Total(N)</b>                        | <b>115</b><br><b>(100)</b>  | <b>84</b><br><b>(100)</b>   | <b>71</b><br><b>(100)</b>   |

Table 2 shows a mean  $\pm$  standard deviation values for various groups

| <b>Table 2 : Mean <math>\pm</math> SD of various group</b>  |            |                                   |
|---|------------|-----------------------------------|
| <b>Group</b>  | <b>N</b>   | <b>Mean <math>\pm</math> Sd</b>   |
| <b>Ashwin self cure acrylic denture base material</b>       | <b>115</b> | <b>1.91 <math>\pm</math> 1.39</b> |
| <b>Lucitone 199 heat cure acrylic denture base material</b> | <b>84</b>  | <b>1.42 <math>\pm</math> 1.16</b> |
| <b>Chrome cobalt alloy denture base material</b>            | <b>71</b>  | <b>1.16 <math>\pm</math> 0.43</b> |

Any reason that would cause a fall in the number of organism colonizing would decrease the occurrence of infection. Candida albicans has been demonstrated on the fitting surface of the dentures.<sup>8</sup> This was attributed to the acidic pH prevalent under the fitting surface of the dentures which aided the proliferation of the fungi. LP Samaranayake opined that denture stomatitis is associated with the proliferation of Candida albicans on the fitting surface of the denture

This was attributed to the acidic pH prevalent under the fitting surface of the dentures which aided the proliferation of the fungi. LP Samaranayake opined that denture stomatitis is associated with the proliferation of Candida albicans on the fitting surface of the denture than on the mucosal surface of the palate.

This study was carried out to evaluate the surface adherence of Candida albicans to three commonly used denture base materials. The three denture base materials

used for this study were self cure acrylic resin, heat cure acrylic resin and chrome cobalt base metal alloy. Acrylics and metals have been used in the fabrication of denture bases; however, acrylics have been more widely used. Acrylics though economical and easy to manipulate, show some features such as water sorption and permissive surface in contrast to metallic denture bases.<sup>9</sup> These features may alter the degree of adherence of *Candida albicans* to acrylics, which may be contrasting to the metals. Among the metals, gold has been shown to have better surface topography than acrylics.<sup>10</sup> However, the use of gold is impracticable. Therefore, chrome cobalt, a commonly used metallic denture base material, is been used in this study.

The methodology chosen here was based on a study by John F. Miner(1973) with following modifications. An incubation period of 48 hours was considered for all the specimen groups. Therefore, the adherence of the cells was observed only for the tissue surface of the specimens, keeping in mind of the fact that *Candida albicans* is mainly seen thriving in this surface.<sup>11</sup> The polished surface of a denture on the other hand, presents a smooth and glossy texture, which is not much capable of harboring fungi and bacteria as compared to the rough surface.<sup>12</sup> Moreover, the unpolished fitting surface is more capable of absorbing infected oral fluids than the polished surface, thus creating a suitable environment for the microorganisms to grow.<sup>13</sup>

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## CONCLUSION :

The following conclusions were drawn from our present study :

1. Chrome cobalt alloy denture base material showed less adherence of candidial cells compared to heat cure and self cure acrylic resin denture base materials.
2. Among the acrylics, heat cure acrylic resin showed comparatively less adherence of candidial cells than self cure acrylic resin.

Above findings suggest that the metallic denture bases are far better than acrylic denture bases in terms of incidence of denture stomatitis due to candidial infection. Further, they are seemed to be more biocompatible with the oral tissues with minimal tissue reactions if any.<sup>14</sup> However, usage of these base metal alloys as denture bases may not be practicable in regard to their cost, difficulty in relining the necessary set up for their fabrication. Thus, acrylic resins still remain the materials of choice answering the requisites of economy and ease of fabrication. However, patient's oral hygiene should be given prime importance irrespective of the denture base materials used.

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Date of Submission: 22 December 2012

Date of Provisional acceptance: 08 January 2013

Date of Final acceptance: 16 February 2013

Date of Publication: 05 March 2013

Source of Support: Nil Conflict of Interest: Nil