This finding emphasizes the importance of maintaining oral environmental stabilization procedures, which are commonly employed in dentistry. The aims of these procedures include prevention of the progression of oral infections, resistance against commonly used antifungal agents, and the potential to adhere to host surfaces, such as teeth or mucosa, and to artificial, nonbiological surfaces, such as restorative dental materials. This study compared the susceptibility of six dental restorative materials to the adhesion of the major pathogen in oral and systemic candidosis, C. albicans.

Aims: To evaluate the susceptibility of six dental restorative materials to the adhesion of C. albicans.

Materials and methods: Cylindrical samples of each material were made and put in plates with sterile Mueller Hinton and Sabouraud dextrose agar previously seeded with C. albicans. The antifungal effect of the samples was determined with the disc-diffusion method. The samples were then incubated for 72 hours. The amount of candidal growth was significantly lower on the resin-modified glass ionomer and glass-ionomer cement samples. The compomer and the composite samples revealed the lowest quantity of biofilm formation. Within the SEM examination, samples showed more candidal adhesion. The median XTT values, significantly higher than the other materials (< 0.001), were found for the composite and the compomer samples.

Results: The results showed that the resin-modified glass ionomer and glass-ionomer cement samples had a lower amount of candidal growth. The compomer and the composite samples revealed the lowest quantity of biofilm formation. Within the SEM examination, samples showed more candidal adhesion. The median XTT values, significantly higher than the other materials (< 0.001), were found for the composite and the compomer samples.

Conclusion: The study concluded that resin-modified glass ionomer and glass-ionomer cement samples had a lower amount of candidal growth. The compomer and the composite samples revealed the lowest quantity of biofilm formation. The SEM examination showed more candidal adhesion. The median XTT values, significantly higher than the other materials (< 0.001), were found for the composite and the compomer samples.
several diseases by *C. albicans* is widely acknowledged. Several studies have suggested that the initial stage of various microbial diseases involves microorganisms adhering to the target tissue.\(^6\)\(^7\) Thus, interest in using dental materials that might inhibit plaque formation and adhesion of oral microorganisms to restorative materials is increasing. Generally, it is important to obtain information on how biofilm may be influenced by various dental restorative materials, because microorganisms adhering to restorative materials can colonize other oral surfaces and eventually cause oral infections in predisposed individuals. Various kinds of supporting materials release diverse substances\(^8\), which exert an antibacterial action against oral microorganisms *in vitro*.\(^9\) Thus, studies concerning the adhesion of *C. albicans* to biomaterials have focused on the denture base and denture relining materials;\(^10\)-\(^12\) however, fungi effectively adhere to all kinds of resin, glass, and even metal surfaces.\(^13\) Although dental restorative materials are known to be a potential source of fungal infections, fewer investigations have been carried out on these materials.\(^12\),\(^14\),\(^15\) In general, to ensure the clinical relevance of microbiological adhesion studies, materials should be used that are appropriate to clinical dentistry.\(^11\)

Against this background, we compared the susceptibility of six dental restorative materials (two compomers, two composites, two glass ionomer cements, three resin modified glass ionomer cements, one giomer, glass ionomer cement, and one self-etching bonding agent) to *C. albicans* adhesion.

### Materials and Methods

#### Preparation of the Samples

In the present setup, six commercially available dental restorative materials (compomer, light curing glass ionomer cement, nanohybrid composite, amalgam, giomer, glass ionomer cement) were assessed. All materials, manufacturers, and material information are presented in Table 1. All materials were handled in strict compliance with their manufacturers’ instructions. For each test material, 10 samples were prepared. Cylindrical samples (7 mm in diameter, 2 mm in height) were made, using a custom Teflon mold with calibrated circular holes. The materials were inserted into the mold, and their surfaces were covered with a transparent strip and pressed with two glass slides from the top and bottom. Light-activated materials were activated for 40 s of overlapping surface exposure using a polymerization light T-LED (Anthos, Italy, 1900 mW/cm\(^2\); 2 cm distance from the tip). The samples were then removed from the molds, mechanically polished sequentially with coarse, medium, fine, and superfine aluminum oxide abrasive discs (Sof-Lex, 3M, St Paul, MN, USA), and stored in distilled water before further processing.

#### Fungal Growth Conditions

The *C. albicans* clinical strain SC5314 was used. Cells were grown for 24 h at 37°C in the yeast nitrogen base (YNB; Sigma, St Louis, MO, USA) supplemented with 50 mM dextrose. After the incubation period, the cells were harvested, washed with phosphate-buffered saline (PBS; Cellgro, Media-tech, Herndon, VA, USA), and standardized to \(1 \times 10^7\) cells/ml spectrophotometrically at 492 nm for the biofilm formation experiments.

### Table 1: Details of the Dental Materials

<table>
<thead>
<tr>
<th>Restorative Materials</th>
<th>Type</th>
<th>Composition</th>
<th>Manufacturer</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Dyract Extra</td>
<td>Polyacid modified resin composite (compomer)</td>
<td>UDMA, TCB resin, TEGDMA, trimethacrylate resin, camphorquione, ethyl-4-dimethylaminobenzoate, BHT, str-alumino-sodium-fluro-phosph-silicate glass, strontium fluoride, iron oxide and titanium dioxide pigments</td>
<td>Dentsply Detrey GmbH, Konstanz, Germany</td>
</tr>
<tr>
<td>2 Ionolux AC Capsule</td>
<td>Resin modified glass ionomer cement</td>
<td>Polycrylic acid solution, 2-hydroxyethyl methacrylate, glycerin dimethacrylate, urethane dimethacrylate, tartaric acid, initiators</td>
<td>Voco GmbH, Cuxhaven, Germany</td>
</tr>
<tr>
<td>3 Grandio SO</td>
<td>Universal nanohybrid composite</td>
<td>BisGMA, BisEMA, TEGDMA, camphorquione, butylated hydroxytoluene (BHT)</td>
<td>Voco GmbH, Cuxhaven, Germany</td>
</tr>
<tr>
<td>4 ANA 2000 Capsule Amalgam</td>
<td></td>
<td>Ag 43.1%, Sn 30.8%, Cu 26.1% Multifunctional glass filler and S-PRG filler based on fluoroboroaluminosilicate glass, BisGMA, TEGDMA resin</td>
<td>Nordiska Dental, Angelholm, Sweden</td>
</tr>
<tr>
<td>5 Beautifil II</td>
<td>Giomer</td>
<td></td>
<td>Shofu Inc., Kyoto, Japan</td>
</tr>
<tr>
<td>6 Ionofil Molar Capsule</td>
<td>Glass ionomer cement</td>
<td>Polycrylic acid, tartaric acid, aluminofluorosilicate glass</td>
<td>Voco GmbH, Cuxhaven, Germany</td>
</tr>
</tbody>
</table>
**Quantitative measurement of C. albicans biofilms**

Metabolic activity of *C. albicans* biofilms was assessed using a colorimetric assay (XTT). To evaluate biofilm formation by *Candida* isolates, the samples were washed with PBS, placed in 24 well-culture plates with 2-ml standardized cell suspension (1 × 10⁷ cells/ml), and incubated for 72 h at 37°C on a rocker. Biofilms were quantified using a tetrazolium XTT [2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-2H-tetrazolium-5-carboxanilide] assay as described previously.[16]

**Scanning electron microscopy**

Three samples of each material were used for scanning electron microscopy (SEM) investigation. The samples with the adhering fungi were rinsed in PBS, fixed with ethanol, and air-dried. The test samples were then mounted on aluminum stubs and sputter-coated with gold. Samples were examined with a scanning electron microscope (magnification ×3000 and ×5000; EVO LS 10, Carl Zeiss Microscopy, LLC, NY / United States).

**In vitro antifungal study**

The antifungal effect of the restorative materials on *C. albicans* was determined with the disc diffusion method. Cylindrical samples of each material were put in plates with sterile Mueller Hinton agar (with 2% glucose) and Sabouraud dextrose agar previously seeded with *C. albicans*. The agar plates were incubated at 37°C for 24 h and then the inhibition zone was evaluated around each sample. The entire operation was carried out under aseptic conditions in duplicate.

**Statistical analysis**

A multiple significance test with the Duncan correction was used to compare the XTT and biofilm vitality values of *C. albicans* biofilm formation various restorative materials. Data are presented as the mean ± standard deviation (SD), and the level of statistical significance was set at 5% for all analyses. The statistical analyses were performed using a computerized statistical software program SPSS 11.5 for Windows, (SPSS Inc., Chicago, IL, USA).

<table>
<thead>
<tr>
<th>Restorative Materials (n)</th>
<th>Type</th>
<th>OD value (mean ± SD)</th>
<th>Vitality (%) (mean ± SD)</th>
<th>F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dyract Extra (10)</td>
<td>Compomer</td>
<td>0.1790 ± 0.020a</td>
<td>55.0 ± 6.07a</td>
<td></td>
</tr>
<tr>
<td>Ionolux AC (10)</td>
<td>Resin modified Glass ionomer cement</td>
<td>0.1559 ± 0.016b</td>
<td>48.4 ± 4.59b</td>
<td></td>
</tr>
<tr>
<td>Grandio SO (10)</td>
<td>Composite</td>
<td>0.1764 ± 0.010a</td>
<td>54.1 ± 2.98a</td>
<td></td>
</tr>
<tr>
<td>ANA 2000 Capsule (10)</td>
<td>Amalgam</td>
<td>0.1205 ± 0.010c</td>
<td>37.2 ± 3.07c</td>
<td></td>
</tr>
<tr>
<td>Beautifil II (10)</td>
<td>Giomer</td>
<td>0.1399 ± 0.019d</td>
<td>43.1 ± 5.83d</td>
<td></td>
</tr>
<tr>
<td>Ionofil Molar (10)</td>
<td>Glass ionomer cement</td>
<td>0.1408 ± 0.019d</td>
<td>43.3 ± 5.85d</td>
<td></td>
</tr>
</tbody>
</table>

*F, Duncan test frequency; Means followed by the same letter are not significantly different at P = 0.05.*

**RESULTS**

In general, the *C. albicans* biofilms adhered firmly to the restorative materials. No inhibition zone was observed around any sample; thus, none of the tested restorative materials reduced the amount of fungal growth. In addition, no differences were observed in the gross morphology and adhesion of biofilms formed by this pathogen on various types of restorative material [Figure 1].

During the scanning electron micrograph examination, a fine biofilm layer of *C. albicans* species was found on the surfaces of all examined materials. However, the quantity of adhering microorganisms varied among the materials. The amount of candidal growth was significantly lower on the surfaces of the resin-modified glass ionomer cement (Ionolux AC) and glass ionomer cement (Ionofil Molar).
To our knowledge, the current work is the first to compare differences in Candida albicans adhesion on dental materials. This property affects the attachment of biofilm bacteria, as close as possible for the minimal roughness surface. The tested materials were prepared and polished plane as possible. Therefore, in the present study, the tested materials exhibited antifungal properties. In agreement with previous studies, our results confirm that different fungal species have different adhesion potential. Theoretically, and as a consequence, dental materials are known to be involved. The human mouth presents various surfaces to which microorganisms can adhere. The human denture base and relining material have focused on the denture base and relining material are known to be involved. Surface roughness is well documented to play a role in achieving long-term success of oral healing and protection from oral diseases. Although other species of Candida are found on rough surfaces than on polished, smooth surfaces. This study compared the susceptibility of six dental materials with statistically lower XTT values than the other materials (Table 2). Taken together, these results demonstrate that the ability of Candida albicans to adhere directly to the surface of restorative materials such as amalgam, resin composite, compomer, and glass-ionomer cement plays an important role in achieving long-term success of oral healing and protection from oral diseases.

When the adhering fungi were investigated, the amalgam samples (ANA 2000) showed more biofilms formed (median XTT value, 0.1205) of all the materials. The compomer (Dyract Extra) and glass-ionomer cement (Ionofil molar) samples showed more biofilms formed than the other materials (median XTT value, 0.1205). The highest median XTT values were significantly higher than those of the other materials. The highest median XTT values were found for the composite (Grandio SO) and compomer (Dyract Extra) samples. The glass ionomer cement (Ionofil molar) and the giomer (Beatifill II) had low XTT values with similar values. The highest median XTT values were significantly higher than those of the other materials. The lowest quantity of biofilms formed of all the materials was found on Grandio SO, Ionolux AC, and ANA 2000 Capsule; 5, Beautifil II; 6, Ionofil Molar.

Candidal adhesion was not evaluated in the present study. Therefore, the results cannot be related to specific surface characteristics. Moreover, the correlation between surface roughness and candidal adhesion was not evaluated in the present study. However, since we used only one type of culture strain of Candida albicans, so that a surface as close as possible for the minimal roughness surface.

Studies concerning the adhesion properties of Candida albicans preferred to provide as simple a test model as possible, for dental material surfaces. Different fungal species have different adhesion potential. The genus Candida is the major microbiological factor in oral candidosis. Therefore, the results cannot be related to specific surface characteristics. Moreover, the correlation between surface roughness and candidal adhesion was not evaluated in the present study. However, since we used only one type of culture strain of Candida albicans, so that a surface as close as possible for the minimal roughness surface.

Various material characteristics such as surface composition, hydrophobicity, electrostatic forces, filler composition, composition of the material, type of matrix, size of fillers, and configuration of fillers are relevant factors affecting attachment of organisms to the surface to form a biofilm.

Scanning electron micrographs of C. albicans biofilm layers revealed the lowest quantity of biofilms formed of all the materials. The XTT values of the amalgam (ANA 2000) samples revealed the lowest quantity of biofilms formed of all the materials. The XTT values of the amalgam (ANA 2000) samples revealed the lowest quantity of biofilms formed of all the materials. The highest median XTT values were significantly higher than those of the other materials. The highest median XTT values were significantly higher than those of the other materials. The highest median XTT values were significantly higher than those of the other materials. The highest median XTT values were significantly higher than those of the other materials. The highest median XTT values were significantly higher than those of the other materials.
Belduz, et al.: Candida albicans adhesion on dental materials

(Dyract Extra) samples. These important findings confirm the hypothesis that different types of material and their specific chemistry and composition (matrix and fillers) interfere considerably with *C. albicans* adherence.[20,26,27] Heavy metals have antimicrobial properties on biofilm formation,[28] and amalgam liberates ions that kill bacteria in the adhering biofilms.[28,29] Although the amalgam samples showed a very high percentage of surface coating within the scanning electron micrograph examination, low vitality values of the adhering *C. albicans* biofilm were found on the amalgam samples. This might be due to the metallic ions released from the surfaces. The low quantity of adhering *C. albicans* on glass ionomer cements in this investigation may be because glass ionomer cements are known to release significant quantities of fluoride, and fluoride components are associated with antimicrobial effects.[30,31] However, it was not within the scope of the present investigation to analyze these diverse chemical entities (i.e., different metallic cations, fluoride ions, and organic molecules), so such conclusions must be regarded as rather speculative.

Bürgers *et al.*[32] reported that the composites Compoglass F and Dyract eXtra and the ormocer Admira revealed lower amounts of adhering fungi than the conventional hybrid composites and a novel silorane-based restorative. This finding is not in agreement with our findings. In our study, the highest XTT values were found for the composite (Grandio SO) and compomer (Dyract Extra) samples.

To prevent *C. albicans* biofilms from accumulating and to reduce adhesion, several promising inventions have been introduced.[33-35] As the etiology of Candida-associated stomatitis is multifactorial with numerous influencing parameters, a better understanding of the essentials of fungal adhesion *in vitro* methods to study these adhesion processes is needed. In general, conclusions from this *in vitro* investigation and other related studies may not be transferred to the clinical situation without restriction of any kind, and results have to be interpreted carefully because only a limited number of parameters can be simulated outside the oral cavity. Considering the limitations of this study, the amalgam and glass ionomer cements revealed lower amounts of adhering fungi than the composites and compomers.

These findings emphasize the use of glass ionomer restorative cements and amalgam regarding reducing *C. albicans* adhesion to dental restorative materials especially in people with weakened immune systems, neutropenia, and cancer.

**Conclusion**

These findings emphasize the use of glass ionomer restorative cements and amalgam regarding reducing *C. albicans* adhesion to dental restorative materials especially in people with weakened immune systems, neutropenia, and cancer.

**Financial support and sponsorship**

Nil

**Conflict of interest**

None

**References**