RESEARCH ARTICLE

AN IN VITRO COMPARATIVE EVALUATION OF COMMERCIAL AVAILABLE MOUTH RINSES (ALCOHOL CONTAINING AND ALCOHOL FREE) ON THE MICROHARDNESS OF A NANOFILLED COMPOSITE RESIN RESTORATIVE MATERIAL

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Abstract

Aim: The purpose of this study was to compare the effect of commercially available mouth rinses (alcohol containing and alcohol free) on the micro hardness of nanofilled composite restorative material.

Materials and Methods: 75 discs of nanofilled composite resins were prepared, measuring 4 mm in diameter and 3 mm in thickness. The composite resin specimens prepared were kept in artificial saliva for 24 hours to simulate oral environment. The specimens were divided randomly into three groups with two subgroups in Group A and Group B each containing 15 specimens (n=15). Samples were immersed according to groups. In Group-A subgroup 1, in alcohol-containing mouthwash- Listerine, Group-A subgroup 2, in alcohol-containing mouthwash- Colgate Plax. Group-B subgroup 1, in non alcohol-containing mouthwash- Everfesh. Group B subgroup 2, in non alcohol-containing mouthwash- Hiora. Group C, The control group consists of distilled water. Samples were immersed for 24 hrs in respective test groups. The post-immersion microhardness testing was done using Vickers microhardness Tester.

Results: Statistical analysis was done using one-way ANOVA followed by Student ‘t’ test. Significant reduction in the microhardness was observed in both the groups after immersion in the mouth rinses compared to the control group. Group A showed statistically significant difference when compared to Group B.

Conclusion: Both the mouthrines showed a reduction in the microhardness of nanofilled composite resins with Listerine group.
(Group A subgroup 1) showing the highest reduction in microhardness value.

Introduction:
Composite resins, due to their esthetic features and improved properties, are largely used in anterior and posterior restorations. The introduction of composite-based resin technology to restorative dentistry was one of the most significant contributions to dentistry in the last century. As a polymer-based material, resin composite may suffer degradation when exposed to the oral environment. As a result, there may be a decrease in properties such as hardness, flexural strength, and elastic modulus.

Nanofilled materials are believed to offer excellent wear resistance, strength, and ultimate esthetics due to their exceptional polishability, polish retention, and lustrous appearance. The technology of such resins may also improve the continuity between the dental structure and the nanoparticles, providing more balance between the mineralized hard tissue of teeth and those of the advanced restorative biomaterials.

In recent years, clinicians have become increasingly interested in the potential of antimicrobial mouth rinses in the control of plaque in patients for effective prevention and control of dental caries and periodontal disease. Mouth rinse solutions have various components such as detergents, emulsifiers, organic acids, dyes, and alcohol (predominantly ethanol). Alcohol is used in mouth rinses as a solvent for other ingredients, to dissolve plaque and as an antiseptic. Since maintaining the dental restorations in the mouth is also important for oral health, it is necessary to know the effect of mouth rinses on these composite resin restorations. There are reports in the literature stating that the alcohol in the mouth rinses may soften composite restorations. However, both alcohol-containing and alcohol-free mouthrinses could affect the hardness of the restorative materials. As the hardness is related to material's strength and rigidity, it has great implication on the clinical durability of restorations.

Hence, the aim of the study is to compare the effect of commercially available mouth rinses (alcohol containing and alcohol-free) on the microhardness of nanofilled composite restorative material.

Materials and Method:
75 Disc shaped specimens of nanofilled composite material measuring 4 mm in diameter and 3 mm in thickness were prepared by using a plastic ring mould. The mould was placed on a transparent glass slide and the mould space was filled with composite resin. The mould space was packed with composite to fill it to a slight excess using a plastic filling composite instrument. Another glass slide was used to pack the composite inside the mould and gently pressed for 30 seconds to extrude the excess material to obtain a smooth surface. Each composite resin specimen was cured for 40 seconds from top and another 40 seconds from bottom of the slide using a visible light curing unit. The composite resin specimens thus prepared were kept in artificial saliva for 24 hours to simulate oral environment. Base line microhardness was recorded using a Vickers Hardness Tester. Before immersion, the pH of the commercial mouth rinses was recorded using a digital pH meter.

Details of Mouthwashes used In The Study:

| SL NO | MOUTH RINSE | pH | COMPOSITION | MANUFACTURER |
|-------|-------------|----|-------------|--------------|
| 01    | Group A, subgroup 1 (Listerine) | 3.69 | Thymol – 0.06% w/v, Eucalyptol-0.09% w/v, Menthol-0.04% w/v, Ethanol-21.6%v/v | 0.04% w/v, Ethanol-21.6%v/v Johnson and Johnson Ltd, Kolhapur, India. |
| 02    | Group A, subgroup 2 (colgate plax) | 5.13 | Glycerin, Ethyl Alcohol-7.2% v/v, Propylene Glycol, Sorbitol, Polysorbate 20, Sodium Benzoate, Cetylpyridinium Chloride, Sodium Fluoride, Sodium Chloride | Chlorhexidine gluconate- 0.12% w/v, Ethyl alcohol-11.6% v/v Colgate Palmolive Ltd, Mumbai. |
The composite resin specimens was immersed in 20 ml respective mouth rinses and distilled water while maintaining the temperature of 37⁰c in an incubator for 24 hours. The composite resin specimens was then washed in distilled water, blotted dry and evaluated for post immersion micro hardness using the same micro hardness tester previously mentioned for base line values.

**Statistical Analysis:**
The intra group (pre and post immersion values) comparison of the mean Value of micro hardness of the composite resin specimens was done using Wilcoxon signed rank test. For inter group comparison Kruskal-wallis test followed by Mann-Whiteny U test was used.

**Results:-**
For each subgroup mean microhardness and standard deviation was calculated that showed that
Group A subgroup 1 (Listerine group) - The baseline mean microhardness value was 60.31 ± 1.81 and after treatment with Listerine it was 52.76 ± 1.8 and the mean change in microhardness in Listerine group was 7.5 ± 2.4.

Group A subgroup 2 (Colgate plax group) - The baseline mean microhardness value was 59.17 ± 3.09 and after treatment with Listerine it was 57.79 ± 1.80 and the mean change in microhardness in Colgate plax group was 1.38 ± 0.39.

Group B subgroup 1 (Ever fresh group) - The baseline mean microhardness value was 59.77 ± 1.16 and after treatment with Listerine it was 59.46 ± 1.01 and the mean change in microhardness in Everfresh group was 0.31 ± 0.83.

Group B subgroup 2 (Hiora group) - The baseline mean microhardness value was 60.19 ± 1.49 and after treatment with Listerine it was 59.66 ± 1.52 and the mean change in microhardness in Hiora group was 0.53 ± 0.3.

Group C (Distilled group) - The baseline mean microhardness value was 60.14 ± 1.13 and after treatment with Listerine it was 59.96 ± 1.17 and the mean change in microhardness in Listerine group was 0.18 ± 0.19.

**Table1:-** Showing Mean change in microhardness score among all the subgroups.

| Sl no | Group            | Mean+SD change in Microhardness |
|-------|------------------|---------------------------------|
| 1     | Listerine        | 7.5 ± 2.4                       |
| 2     | Colgate Plax     | 1.38 ± 0.39                     |
| 3     | Everfresh        | 0.31 ± 0.83                     |
| 4     | Hiora            | 0.53 ± 0.3                      |
| 5     | Distilled Water  | 0.18 ± 0.19                     |
The One way ANOVA used to compare the mean microhardness difference between all the subgroups that showed statistical significant result at 5% significance level.

Table 2: Showing one way ANOVA used to compare the mean microhardness difference between all the subgroups.

| Variable                  | Group       | Mean+sd change in microhardness | F value | P value |
|---------------------------|-------------|---------------------------------|---------|---------|
| Mean Microhardness difference | Listerine   | 7.5 ± 2.4                       |         | <0.001  |
|                           | Colgate Plax| 1.38 ±0.39                      |         |         |
|                           | Everfresh   | 0.31 ±0.83                      |         |         |
|                           | Hiora       | 0.53 ±0.3                       |         |         |
|                           | Distilled Water | 0.18 ±0.19                     |         |         |

Pair wise comparison done with Scheffe Post-Hoc Test showed statistical significant difference between Listerine and other sub groups, but when other sub groups were compared amongst each other, there was no statistical significant difference between them.

Table 3: Showing Scheffe Post-Hoc Test to compare the pair wise mean microhardness among the subgroups.

| Sl No | Group A         | Group B       | P value |
|-------|-----------------|---------------|---------|
| 1     | Listerine       | Colgate Plax  | <0.001  |
| 2     | Listerine       | Everfresh     | <0.001  |
| 3     | Listerine       | Hiora         | <0.001  |
| 4     | Listerine       | Distilled Water | <0.001 |
| 5     | Colgate Plax    | Everfresh     | 0.200   |
| 6     | Colgate Plax    | Hiora         | 0.430   |
| 7     | Colgate Plax    | Distilled Water | 0.112  |
| 5     | Everfresh       | Hiora         | 0.992   |
Discussion:-
Today, due to their improved mechanical properties, composites have become the material of choice for esthetic restorations involving both the anterior and posterior teeth. As a polymer-based material, resin composites may suffer degradation when used in the oral environment.

Under oral conditions, resin based composites may be exposed either intermittently or continuously to chemical agents found in saliva, food, beverages and more recently, mouth rinses. Consequently, the leaching of composite fillers and the disintegration of filler-resin interface (silane coupling agent) can also occur under oral conditions. Therefore, in the case of resin based composites, degradation typically occurs because of the following two reasons.

Hydrolytic breakdown of the bond between the silane & filler particles and the resin matrix, finally resulting in debonding.

The softening of dental resins through the plasticizing action of water. As for the effects of solvents on dental composites, many factors come into play, such as the hydrophilicity of polymers and the cross linking density of the network.

In an in vivo situation, it is reported that food components and beverages may affect resin-composites. Amongst the factors that affect the properties of the composite restoration, mouth rinse is one that has recently gained significant attention. The popularity of mouth rinses has grown considerably not only because of their effectiveness in caries and gingivitis control but also because of social and cosmetic reasons. Literature on the effect of mouth rinses on the microhardness of newly introduced composites is limited, hence this motivated us to fill this lacuna in literature and to conduct the present study using commonly used mouth rinses in the Indian market.

Hardness is considered as the test parameter, as it is an important property for the restorative materials to have long-term durability in the oral cavity. Hardness has also been used to predict the wear resistance of a material and its ability to abrade or be abraded by opposing dental structures and materials. So a decrease in the hardness of a material may result in premature failure of a restoration requiring its replacement.

In the present study, all the mouth rinses irrespective of the presence or absence of alcohol resulted in significant reduction in the micro hardness of the tested resin composite material compared to baseline values. This may be because of the acidic pH of the mouth rinses which would have caused acid erosion of the resin composite by acid etching and leaching of the principle matrix forming cations. This is in accordance with the observations by Diab et al in 2007 who reported that mouth rinses with low pH are detrimental to the hardness of resin composites.

Basically, the low pH of mouth rinses may have acted in the polymeric matrix of the resin composites used in the study through catalysis of ester groups from dimethacrylate monomers present in the composition (Bis-GMA, Bis-EMA, UDMA and TEGDMA). The hydrolysis of these ester groups may have formed alcohol and carboxylic acid molecules that may have accelerated the degradation of the resin composite.

Water sorption in the composite resin is a controlled diffusion process that occurs primarily in the resin matrix. Here in this study comparison of different sub-groups shows that Listerine, containing alcohol 21.6% w/v, caused statistically more reduction in microhardness as compared to all other mouthrinses. There was statistically no significant difference in the reduction in the microhardness when Colgate Plax, Everfresh, HiOra and Distilled water were compared amongst each other. This observation is in accordance with the observations by Penugonda et al who reported that the higher percentage of alcohol in the mouth rinses causes more reduction in the hardness of restorative materials.

In the present study the softening effect of alcohol in Listerine on the resin composite may be due to susceptibility of Bis-GMA and UDMA based polymers present in them and irreversible leaching of the components. Filtek Z 350 primarily consists of Bis-GMA and TEGDMA as its polymeric matrix and this explains the significant reduction in microhardness caused by Listerine.
When comparing the sub-groups (Listerine, Colgate Plax, Ever fresh, Hiora and Distilled water) Pair wise comparison done with Scheffe Post-Hoc Test showed statistical significant difference between Listerine and other subgroups (p<0.001), but there was no statistical significant difference between other subgroups when compared amongst each other (p>0.005).

According to Villalta et al, the low pH and alcohol concentration of solutions affect the hardness of composite resins\textsuperscript{13}. Miranda et al demonstrated that Colgate Plax has phosphoric acid in its composition, which might alter the polymer matrix of composites by catalysis of the ester groups present in the dimethacrylate monomers\textsuperscript{14}.

The action of mouthrinses on hardness depends on the composition of the restorative material, which can be attributed to different chemical compositions and the composition of the organic matrix. Several factors related to polymer chemical structure and molecular chains are important in determining how these materials will be affected by an aqueous environment\textsuperscript{15}. It is known that there is a difference in hydrophilicity between the matrix monomers and the degree of difference is presented in the following order: TEGDMA>Bis-GMA>UDMA>HMDMA. TEGDMA is more susceptible to enzymatic hydrolysis than Bis-GMA or Bis-EMA.

In the present study mouthwashes used were both alcohol containing [Listerine (21.6%) and Colgate Plax (7.2%)] and the non alcohol-containing mouthwash [Ever fresh and Hiora]. The nanofilled composite is more prone to absorbing fluids and softening by alcohol in the mouth rinses. The use of alcohol-containing mouth rinses with a low pH may increase the sorption and solubility of resin composites. Upon loading, the load applied through the Vickers indenter on the microhardness tester would degrade the resin composite structure with lower energy for the specimens immersed in the alcohol mouthwash than those immersed in the non alcohol one.

Figures:

![Figure No: 1:- Materials Used.](image1)

![Figure No: 2 :-Packing Of Composite Material In To The Mould.](image2)
Figure No: 3:- Placement Of Glass Slides On The Composite Mould.

Figure No: 4:- Light Curing Of Composite Mould.

Figure No: 5:- The Composite Specimens In Mouthrinses Placed In Incubator.
Conclusion:-
Within the limitations of this study, it can be concluded that:
1. All the mouth rinses used in the study, irrespective of the presence or absence of alcohol reduced the micro hardness of the tested resin-composite material.
2. Reductions in microhardness values were lower in alcohol-free mouth rinses than in alcohol containing mouth rinses.
3. Alcohol content is not the only factor in mouth rinses that can degrade composite materials.
4. Alcohol-free mouth rinses may be preferable to alcohol containing mouth rinses.
5. However, the results of this in vitro study may not be directly related to the clinical situation where saliva may dilute or buffer the mouth rinses. Hence further in vivo studies are recommended to substantiate the results of this present study.

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