TO STUDY THE EFFECT OF FOUR ALCOHOLIC BEVERAGES ON THE SURFACE PROFILE AND COLOUR STABILITY OF NANO-FILLED RESIN COMPOSITE : AN IN VITRO STUDY

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LIST OF ABBREVIATIONS

Sr. No.	Abbreviations	Full form
01.	SEM	Scanning Electron Microscope
02.	Ml	Milli litres
03.	SPSS	Statistical Package for the Social Sciences
04.	µm	Micro meter
05	ANOVA	Analysis of Variance
06.	HSD	Honest Significant Difference
07	SD	Standard Deviation
08	S	Significant
09	NS	Not Significant
10	HS	Highly Significant
11	N	Total Number of specimens
12.	n	Samples divided in groups
13.	p-value	Probability of obtaining a test statistic at least as extreme as the one that was actually observed
14.	Max.	Maximum
15.	Min.	Minimum
16.	No.	Number
17.	CI	Confidence Interval
18.	CAD	Computer-aided design
19.	CAM	Computer-aided Manufacturing.

Sr. No.	Abbreviations	Full form
20.	mW/cm2	Mililiwatts per centimeter- square
21.	˚C	Degree celcius
22.	mm	milimeter
23.	s/sec	seconds
24.	min	minute
25.	Ra	Average roughness
26.	Mm/sec	Milimeter per second
27.	QTH	Quartz Tungsten Halogen
28.	v/v	Volume per volume
29.	vol%	Volume Percent
30.	ABV	Alcohol by volume
31.	GIRD	Gastro esophaseal reflux disease

INTRODUCTION

"The possession of knowledge does not kill the sense of wonder and mystery. There is always more mystery" – Anais Nin

One of the quintessential challenges in restorative and aesthetic dentistry is to produce a natural looking restoration that blends harmoniously with the oral environment. Apart from the important parameters of shape and morphology, the colour of a restoration is often regarded as being another essential building block necessary for constructing an aesthetic restoration.[1]

Composite resins are presently among the most popular aesthetic restorative materials used in dentistry and have become one of the most commonly used esthetic restorative materials because of their adequate strength, excellent esthetics, moderate cost compared with that of ceramics, and ability to be bonded to tooth structure.[2] The use of these materials has been on the rise for the past several years and their popularity will only increase as manufacturers introduce stronger composite materials.[3]

For a lasting impression of the esthetic outcome, “colour stability” or the ability of the material to retain the matched shade over a long period of time is crucial. “Colour selection” is nevertheless often considered one of the most fascinating and frustrating areas in dentistry. Therefore, an understanding of colour science and its parameters are important tools for optimizing the esthetic outcome of restorations.[4]

Colour and surface roughness are very important properties in aesthetics, characterizing a smile. Since the introduction of composites in 1960, efforts have been made to increase the longevity of composite restorations.[4]

Surface quality of dental restorations is an important factor in determining the success of the restorations. The major disadvantage of resin composites is their colour instability, which may be a major reason for the replacement of restorations due to esthetic failure.[3]

Dental Composites are compounds by an organic polymerizable matrix, inorganic fillers, borosilicate, silica, and a silane-coupling agent.[5,6] The organic phase (resin phase) of composite resins is constituted by monomers such as Bis-GMA (bisphenol A diglycidyl dimethacrylate) and/or UDMA (urethane dimethacrylate), co-monomers such as TEGDMA (triethylene glycol dimethacrylate) and/or Bis-EMA (ethoxylated bisphenol A glycol dimethacrylate), additives, initiator (canphoroquinone), co-initiator (dimethyl-aminobenzoic acid-ester), a polymerization inhibitor, and a photostabilizer (benzophenone). The inorganic phase is composed by fillers of different types and particle sizes.[7]

Polymer degradation is classified as thermal, mechanical and passive hydrolysis. Degradation process is associated with erosion of composite resin that causes the material losses. Several problems like abrasion, corrosion and fatigue cause the wear phenomenon.[5] The dental erosion has intrinsic and extrinsic factors. Acidic foods, alcoholic beverages, soft and energy drinks are the extrinsic factors that causes dental erosion.[8] Intrinsic factors include chronic GIRD, anorexia and bulimia where the regurgitation and persistent act of vomiting are common reducing pH in the oral environment.[9,10] The effects of such factors in restorative materials are softening, surface roughness increases, and tooth erosion, making them more susceptible to wear.[11,12]

Surface roughness contributes to exterior discoloration.[13] It is closely related to the type of composite material and the polishing and finishing systems used. During wear of dental resin composites, inorganic fillers debond from the resin matrix and leave a void, increasing the surface roughness and forming a surface susceptible to exterior stain. The average particle size of inorganic fillers in nanofilled dental composites has been reduced to around 5-20 nm or less so that the polished restoration can achieve adequate gloss and, during long-term service, the wear of the restoration does not create a rough surface.[2]

In addition, the surface roughness of composites can reduce some mechanical properties such as increase the wear of restorations and susceptibility to extrinsic stain. Thus, polished and smooth composite resin restorations present a better esthetic appearance and greater longevity.[14]

Colour changes in resin composites can occur from intrinsic and extrinsic factors. Intrinsic factors such as chemical composition of the material alteration in the resin matrix, filler loading and particle size distribution, type of photoinitiator and percentage of remaining C=C bonds, which is directly influenced by the duration of the light exposure and the type of curing device have a considerable influence on colour stability.[15] The resin monomers also have an important role in the colour stability of the composites.[16]

Extrinsic tooth discolouration can be caused by surface staining from the adsorption or absorption of colorants, cigarette smoking have been shown to influence the colour stability of restorative material. The structure of the resin composite and the characteristics of the particles have direct impact on the surface smoothness and the susceptibility to extrinsic staining. This is usually attributed to chemical degeneration of the filler resin bond and solubility of the resin matrix.

Also, in today's world consumption of beverages and drinks has dramatically increased. According to the National Health Portal, alcohol use is quite common in India both in rural and urban areas with prevalence rates as per various studies varying from 23% to 74%. The increased consumption of alcoholic beverages in young and adult population has raised questions about the erosive potential and discoloration caused by them on the composite restorations.[17] The alcoholic beverages are known to have an acidic nature as they diffuse into the resin, plasticizing the polymer matrix[17] and reducing its physical properties. It has been demonstrated that surface discolorations in composite resins are related to hygiene, eating habits and smoking. The maintenance of the esthetics of a restoration is therefore related to the patients' habits and lifestyle.[14]

It is known that composite resins exposed to ethanol exhibit lower micro hardness values compared to non-exposed materials.[2] In addition, ethanol can reduce bonding between resin matrix and inorganic cause staining of resin matrix.[18]

Previous studies concerning colour stability have shown that beverages such as coffee, tea, red wine, and coke and mouth rinses have varied degrees of staining effect on self and light-cured composite resin. **Park JK et al. (2017)**[19] suggested that consumption of beverages such as coffee, tea and soft drinks causes discoloration of composite restoration.

Extrinsic dental stains caused by alcoholic beverages i.e beer, red wine, whiskey, vodka are also a well-observed and studied phenomenon. The staining potential of these drinks vary according to their percent composition of alcohol.

Several composite resins have been the subjects of colour stability and surface roughness studies, but very few studies have evaluated the changes in surface profile and colour stability of nano-filled resin composites FILTEK™ Z350 XT due to alcoholic drinks.

Thus, the purpose of this in vitro study was to evaluate the surface roughness and colour stability of nano-filled composite resins after exposure to four commonly consumed alcoholic beverages i.e beer, red wine, whiskey, vodka at baseline and after 30 days.

The null hypothesis was that there is no change in surface roughness and colour stability of nano-filled resin composite after exposure to alcoholic beverages i.e beer, red wine, whiskey, vodka after 30 days.

AIMS AND OBJECTIVES

AIM:

To evaluate and compare the surface roughness and colour stability of nano-filled resin composite after exposure to four different alcoholic beverages i.e beer, red wine, whiskey and vodka after 30 days

PRIMARY OBJECTIVE:

1) To evaluate the surface roughness and colour stability of nano-filled resin composites after exposure to four different alcoholic beverages i.e beer, red wine, whiskey, vodka at baseline and after 30 days.

2) To compare the surface roughness of nano-filled resin composites after exposure to different alcoholic beverages i.e beer, red wine, whisky, vodka and artificial saliva after 30 days.

3) To compare the colour stability of nano-filled resin composites after exposure to different alcoholic beverages i.e beer, red wine, whisky, vodka and artificial saliva after 30 days.

4) To compare the co-relation between surface roughness and colour stability after exposure to alcoholic beverages.

REVIEW OF LITERATURE

The current strategies for achieving predictable aesthetic restoration are based on the evolution of different dental restorative materials and techniques combined with the ever-growing expertise and understanding of the clinician. Therefore, it is imperative to understand the mechanism, principles and strategies of restorative materials that have evolved over time; and the challenges encountered by researchers and clinicians that are associated with this technique sensitive procedure.

De Gee AJ, ten Harkel-Hagenaar E, Davidson CL (1984)[20], conducted a study on 'colour dye for identification of incompletely cured composite resin.' Cylindrically shaped (5X5) samples were prepared, fractured it into axial halves by diametral tensile loading, and exposed to a solution of Astra blue for at least 2 min. The results showed that for light-activated systems, the curing depth is more limited for microfilled than for conventional or hybrid composites. In the deeper areas of some light-activated microfilled composite resins, insufficient coupling between prepolymerized filler particles and the matrix can be demonstrated. They concluded that Visualization with Astra blue proved to be an easy method to locate areas of incomplete polymerization in composite resin.

Lu H, Roeder LB, Lei L, Powers JM (2005)[4], compared four different resin composites for their colour stability and observed that coffee had significant influence on discoloration of dental composite resins. They tested four resin composites i.e; Filtek Supreme (nanocomposite), Filtek A110 (microfilled composite), Filtek Z250 (microhybrid composite), and Filtek P60 (microhybrid composite). Coffee solution had a significant effect on these composite resins. Discoloration increased as surface roughness increased for the composites tested, except with Filtek A110.

Bagheri R, Burrow MF, Tyas M (2005)[21], studied six different composite resins i.e; a light cured microfilled RBC (Durafil, Kulzer), a light-cured microglass RBC (Charisma, Kulzer), a polyacid-modified RBC (F2000, 3M/ESPE), a conventional GIC (Fuji IX, GC) and two resin-modified GICs (Fuji II LC; Photac Fil, 3M/ESPE) and total 576 samples were made and randomly divided into groups. All specimens were immersed in testing solution for 1 week, 2 week and 3 week. Colour coefficients were calculated with the spectrophotometer. They noted that all materials are more prone to staining by coffee, red wine and tea. Fuji IX showed the least susceptibility and F2000 the greatest to staining.

Catelan A, Briso AL, Sundfeld RH, Dos Santos PH (2010)[22], evaluated the effect of artificial aging on the surface roughness and microhardness of sealed microhybrids and nanofilled composites. In their study, one hundred disc-shaped specimens were made for each composite. After 24 hours, all samples were polished and surface sealant was applied to 50 specimens of each composite. Surface roughness was determined with a profilometer and Knoop microhardness was assessed with a 50-g load for 15 seconds. Ten specimens of each group were aged during 252 hours in a UV-accelerated aging chamber and immersed for 28 days in cola soft drink, orange juice, red wine staining solutions, or distilled water. They reported that artificial aging decreased microhardness values for all materials, with the exceptions of Vit-l-escence and Supreme XT sealed composites and surface roughness values were not altered. Water storage had less effect on microhardness, compared with the other aging processes.

Wasilewski Mde S, Takahashi MK, Kirsten GA, de Souza EM (2010)[23], studied the effect of cigarette smoke and whiskey on the colour stability of dental composite and stated that immersion in whiskey showed less colour changes. Total 50 disc shaped samples were made from five different composite resin and were randomly divided into groups. Specimens then immersed in whiskey for 24 hrs and then subjected to cigarette smoke for 10 min , four times. Author concluded that when samples were exposed to whiskey and smoke both it increases the discolouration significantly. Thus, the effect of smoke exacerbated due to softening of the resin surface, causing an intense discolouration of all the composite evaluated.

Elizabeth S (2012)[3], conducted a study to evaluate the colour change of five aesthetic dental materials, before and after immersion in distilled water and blue food colour solution for 7 and 21 days, and to study the effect of finishing the surfaces on any colour change. Disc shaped samples of five types of light curing composite (A2) were prepared and were cured with a Plasma Arc light cure unit for 10 sec. One side of each sample disc was finished and polished with a Super-Snap system. After 24 h, colour measurements were conducted using a digital spectrophotometer. 5 sample discs from each composite group were immersed in food colour solution for 7 and 21 days, while the remaining 5 sample discs were immersed in 30 ml of distilled water as a control. Colour measurements were repeated for all samples at 7 and 21 days after immersion. The study concluded that finished composite surfaces showed less coloration after 7 days, but all surfaces were highly coloured after 21 days.

Bansal K, Acharya SR, Saraswathi V (2012)[13], evaluated the effect of alcoholic and non-alcoholic beverages on colour stability and surface roughness of a methacrylate-based nano-filled composite and a silorane-based microhybrid composite after immersion into four beverages; whiskey, Coca-Cola, nimbooz, and distilled water in each beverage for 10 minutes each day for 56 days. They concluded that coca-cola, caused the highest discoloration and surface roughness change in both the tested resin composites.

Lepri CP, Palma-Dibb RG (2012)[24], studied the surface roughness and colour changes of a composite resin under influence of beverages and tooth brushing. 120 disc samples were prepared with Z250 and randomly divided into 4 groups(red wine, soft drinks, sugarcane spirit and artificial saliva) and 3 subgroups(without brushing, brush with Colgate or closeup. After 30 days values were evaluated using rugosimeter and spectrophotometer. They observed that beverages increased discolouration in composite resin however both beverages and brushing were responsible for increase in surface roughness.

Reddy PS, Tejaswi KS, Shetty S, Annapoorna BM, Pujari SC, Thippeswamy HM (2013)[25]**,** studied the effects of three beverages i.e. coke, coffee and tea on surface roughness of nano-filled, microhybrid, and hybrid composites after immersion period of 30 days. observed that coke causes more surface roughness than coffee and tea. Also, coffee has shown more tendency for causing colour change than tea and coke.

Bagis B, Tuzuner T, Turgut S, Korkmaz FM, Baygın O, Bagiş YH (2014)[26]**,** performed a study to determine the effect of protective resin coating on the surface roughness and colour stability of resin based restorative material and noted that surface roughness and colour discolouration value of resin based restorative material increased after exposure to ultraviolet aging.

Tanthanuch S, Kukiattrakoon B, Siriporananon C, Ornprasert N, Mettasitthikorn W, Likhitpreeda S (2014)[27], investigated the effects of five beverages (apple, cider, orange juice, Coca-Cola, coffee, and beer) on microhardness and surface characteristic changes of nanohybrid resin composite and giomer. Ninety-three specimens of each resin composite and giomer were prepared. Before immersion, baseline data of Vicker's microhardness was recorded and surface characteristics were examined using scanning electron microscopy (SEM). Five groups of discs (n = 18) were alternately immersed in 25 mL of each beverage for 5 s and in 25 mL of artificial saliva for 5 s for 10 cycles for 28 days. They found that Microhardness values of all groups were decreased from the initial week of immersion until the end of the 28 days period and the greatest change in hardness shown occurred within the first 7 days and Coca-Cola produced the roughest surface as compared to other beverages.

de Alencar E Silva Leite ML, da Cunha Medeiros E Silva FD, Meireles SS, Daarte RM, Andrade AK (2014)[28], studied the effect of drinks on colour stability and surface roughness of nanocomposites. Total 40 disc shaped samples prepared with two composite resin Filtek Z350 XT and Evolu-X and randomly divided into four groups- distilled water, acai juice, grape juice and red wine. Initial and final colour change and surface roughness value were calculated using spectrophotometer and profilometer resp. based on the results, author stated that red wine caused more discolouration in nanocomposite than by grape juice and acai juice. Colour changes in nano composites can be reduce by repolishing.

Maganur P, Satish V, Prabhakar AR, Namineni S (2015)[29], conducted study on effect of soft drinks and fresh fruit juice on surface roughness of flowable composite and resin-modified glass ionomer cement. Flowable composite specimens showed an increased tendency of surface roughness as compared to resin-modified glass ionomer cement.

Tan BL, Yap AU, Ma HN, Chew J, Tan WJ (2015)[30], conducted a researched on effect of beverages on colour and translucency of new tooth coloured restoratives. Total 250 samples were fabricated with six composite resin: two resin based composite, Filtek Z 350 XT and filtek z 350 XT flowable, two Giomer shofu beautifil II beautifil flow plus and two glass inomer cement:- ketac nano (N100) and photac fil. Samples then randomly divided into 7 groups i.e cola, orange juice, red wine, vodka, black coffee, green tea and distilled water. Initial and final value were calculated using spectrophotometer. Author concluded that for most materials, coffee followed by red wine and tea was the most prominent staining beverage than vodka, green tea and produced the largest decrease in translucency.

Tantanuch S, Kukiattrakoon B, Peerasukprasert T, Chanmanee N, Chaisomboonphun P, Rodklai A (2016)[31], studied the effect of red and white wine on the surface roughness and erosion of nanohybrid and nano-filled resin composites. And evaluated that Red and white wine significantly increased the surface roughness and erosion of nano-filled and nanohybrid resin composites after evaluation at the end of the 14 days' immersion period.

DA Silva MA, Vitti RP, Sinhoreti MA, Consani RL, Silva-Junior JG, Tonholo J (2016)[32]**,** studied the effect of alcoholic beverages on surface roughness and microhardness of dental composites. Total 120 samples were made from Durafill, Z250 and Z350XT composite resin and were randomly divided into 4 groups. Artificial saliva used as a control group. Samples were immersed in alcoholic beverages i.e beer, whiskey and vodka for 30 days. He stated that, alcoholic beverages increase the surface roughness of the dental composite after immersion for 30 days. Whiskey and beer produced higher changes on surface profile of the dental composite than other alcoholic beverages.

Sadeghi M, Deljoo Z, Bagheri R (2016)[33]**,** studied effects of distilled water, coffee, and cola on the roughness of nanohybrid and microhybrid resin composites. After 7 days of immersion microhybrid composite showed a greater Ra than nanohybrid in all solutions. Specimens immersed in coffee exhibited significantly greater surface roughness than that of distilled water and cola. They concluded that nano-hybrid composite showed a significantly smoother surface than microhybrid.

Baglar S, Keskin E, Orun T, Es A (2017)[34]**,** conducted a study on discolouration effect of traditional Turkish beverages like pomegranate juice, ottoman's syrup, tamarind syrup and turnip juice on different composite restorations. Total 175 disk shaped samples were made from five different methacrylate based composite resin and were randomly divided into five groups. Samples were immersed in solution for 12 days and values were calculated with spectrophotometer. They stated that turnip juice caused highest discoloration on all the nano-filled composite resins.

Ceci M, Viola M, Rattalino D, Beltrami R, Colombo M, Poggio C (2017)[35]**,** compared microfilled flowable composite, nano-filled composite, nanohybrid composite, microfilled composite and nanohybrid Ormocer based composite for colour stability after immersion in coffee and red wine. A colorimetric evaluation was performed at 7, 14, 21, 28 days. Coffee demonstrated a higher staining potential as compared to red wine. Among the different materials tested, nanohybrid composites reported the lowest colour variations.

Chakravarthy Y, Claerence S (2018)[36]**,** studied the effect of red wine on colour stability of three different esthetic restorative materials. Total 30 specimens were made from three different composite and randomly divided into three groups. Specimens were immersed in 25 ml of red wine for 20 min a day for 28 days. Before immersion and after immersion values were calculated with spectrophotometer device. Author concluded that red wine caused more colour changes after 28 days which was due to the physical and chemical composition of the restorative materials and properties of staining pigments in the red wine.

Tavangar M, Bagheri R, Kwon TY, Mese A, Manton DJ (2018)[37]**,** evaluated the degree of surface roughness of resin composites (RC) after immersion in three beverages; soft drink, distilled water and coffee for 1 week. They have reported that the polishing techniques, storage solutions, as well as material compositions were found to be important in the surface roughness (Ra) of all composite resins. After 7 days of immersion, the Ra of almost all materials significantly decreased.

Karadaş M, Demirbuga S. (2018)[38], Studied the effects of coffee, tea, grape juice, orange juice and strawberry fruit punch on surface roughness four bulk- fill resin composites (SonicFill, Filtek Bulk Fill Flowable, X-tra fil, Filtek Bulk Fill Posterior) and three nanocomposites (G-aenial Universal Flo, Herculite XRV Ultra, Filtek Ultimate). They stated that surface roughness after aging increased significantly for SonicFill, Filtek Bulk Fill Flowable, X-tra fil, Filtek Bulk Fill Posterior. Xtra Fil showed highest surface roughness.

Shree Roja RJ, Sriman N, Prabhakar V, Minu K, Ambalavanan P (2019)[39], evaluated the effect of chlorhexidine mouthrinse on the colour stability of three different types of composites i.e. Nanoceramic composite, nano-filled composite and microhybrid composite. 30 samples were randomly divided into 3 groups and were immersed in 20 ml of 0.2% chlorhexidine mouthwash. Baseline colour values and after 24 hr immersion colour values were evaluated with spectrophotometer. Statistical significant difference were observed between the 3 groups. They concluded that nano-filled composites had higher colour stability.

Elwardani G, Sharaf A.A and Mohmoud A (2020)[40]**,** evaluated and compared the surface roughness and colour change of microhybrid and nanocomosite after exposure to beverages commonly used by children. Total 30 disc shaped specimens were fabricated with filtek Z250 and Filtek Supreme composite and randomly divided into 3 groups: distilled water, orange juice and Coca Cola. Colour and surface roughness were measured at baseline and on days 15 and 30. Author concluded that there was significant increase in surface roughness and colour change in all immersion solutions tested over time.

Lee JH, Kim SH, Yoon HI, Yeo IL, Han JS (2020)[41]**,** conducted a study on 'colour stability and surface properties of high translucency restorative materials for digital dentistry after simulated oral rinsing.' Total 200 samples from five high translucency restorative materials, LAVA Ultimate, VITA Enamic, VITA BLOCS Mark II, IPS e maxCAD, and Rainbow Shine T were prepared and randomly divided into 4 groups: listerin cool, listerin healthy white natural lemon & salt and hexamedian, and distilled water. Author concluded revealed that rising with certain mouthwashes significantly affects the colour stability and surface properties of high translucency CAD/CAM restorative materials.

Ozkanoglu S, Akin EG (2020)[42]**,** investigated the effects of distilled water, tea, coffee, cola on the color stability and microhardness of two direct composite resins (Filtek Z250, Filtek Z550); one indirect composite resin (Solidex); and one high viscosity glass ionomer cement (Equia Forte Fil) . After one week samples were tested for microhardness and they reported that the highest values of hardness were observed in the Z550 group. The highest levels of hardness change were detected in the coffee and cola groups and nanohybrid composite resins are resistant to external coloration and hardness change.

Bahbishi N, Mzain W, Badeeb B, Nassar HM (2020)[43] studied the effects of tea ,coffee, berry juice and distilled water on surface microhardness and colour stability of five different bulk fill composites namely Filtek Z350, Filtek Bulk-Fill, Tetric N-Ceram Bulk-Fill, Sonic Fill 2, and SDR after 10days, 30 days, 60 days , 90days they reported that all bulk fill composites have lower surface microhardness compared to universal composite resin and there were no major differences between Bulk-Fill tested brands regarding colour change.

Liliany D, Violetta V (2020)[44]**,** studied effect of soft drink on surface roughness of preheated and non -preheated nanohybrid composite after 15days. In their study they fabricated cylindrical samples of nanohybrid composite resin Filtek™ Z 250 XT (10 mm in diameter and 2 mm in height) were prepared and randomly divided into two groups: preheated and non – preheated and they reported that soft drinks significantly increased the surface roughness of preheated nanohybrid composite resins after 15 days of immersion.

Vaidya N, Kumar P, Pathak K, Punia SK, Choudhary A, Patnana AK (2020)[45] evaluated the surface roughness of three flowable esthetic restorative materials after exposure to sports/energy drinks and alcoholic beverages. Total of 210 specimens of dimension (2cm diameter and 2 mm thickness) with giomer, compomer, and composite (70 samples with each esthetic material) were made with the help of plastic rings. The prepared samples were tested in six experimental sports/energy drinks (beer, whiskey, vodka, Gatorade, Red Bull, and Sting) and distilled water was considered as the control group. Profilometric analyses of all samples were recorded before immersing into the experimental and control solutions. Then, the samples were stored in the experimental and control group solutions for 5min for 30 days and they reported that Flowable composite showed the minimum surface roughness, whereas the flowable compomer showed the maximum surface roughness and When the erosive potential of the test solutions was evaluated, surface roughness values were more for sports/energy drinks when compared to that of alcoholic beverages.

Meenakshi CM, Sirisha K (2020)[46], evaluated the effect of artificial saliva, orange juice, and Coca-Cola on surface roughness and colour stability of Filtek Bulk-Fill posterior restorative composite in comparison with Filtek P60 posterior restorative composite. The surface roughness and colour change of both composites increased significantly in acidic beverages and more in Coca cola. Surface roughness bulk-fill composite in artificial saliva is lowest and microhybrid in Coca-Cola highest among all the groups. There was a significant surface degradation in all the acidic beverages and microhybrid composite exhibited more roughening than bulk-fill composite.

Camilotti V, Mendonça MJ, Dobrovolski M, Detogni AC, Ambrosano GMB, De Goes MF (2021)[47], s conducted a study on impact of dietary acids on the surface roughness and morphology of three composite resins 4 Seasons, Z250, after immersion into the solutions G1: distilled water; G2, Coca-Cola, and G3: orange juice for period of 180 days and they stated that 4 Seasons and Z250 had statistically similar roughness values for all the solutions and evaluation periods. With the exception of 180-day immersion in Coca-Cola, 4 Seasons showed significantly higher values than Z250.

MATERIALS AND METHOD

ARMAMENTARIUM

INSTRUMENTS USED

- Glass slabs (PLATE I)
- Mylar Matrix Strips (Golden Matrix Strips, India)
- Teflon Mold (PLATE I)
- Composite finishing and polishing kit (3M ESPE Sof-Lex Polishing discs)
- Beakers (PLATE I)
- Composite Manipulation Instrument (GDC)
- Digital Vernier Calliper ((Workzone Tools, Germany)

EQUIPMENTS USED

- Light Emitting Diode (LED) Curing Unit (Bluephase, Ivoclar Vivadent, Schaan, Liechtenstein)

- Micromotor (Marathon)
- Contra angle handpiece (Being Foshan, Being Foshan medical equipment ltd, China)
- Incubator
- Surface Roughness tester (Mitutoyo-Japan, Model: SJ 210)
- Spectrophotometer (SPECTRASCAN 5100)

MATERIALS USED IN THE STUDY

- Artificial saliva (Control Group)- ICPA HEALTH PRODUCTS LTD
- Beer – (United Breweries Ltd, INDIA)
- Red Wine - (Sula Vineyard, INDIA)
- Whiskey – (Jack Daniel Distillery Lynchburg, Tennessee)
- Vodka – (Radico Khaitan, INDIA)
- Filtek^{TM} Z350 XT-Shade A2(3M-ESPE, St. Paul, MN, USA)

METHODOLOGY

Approval from the institutional ethics committee was taken for the study.

A Nano-filled resin composite (Filtek Z-350 XT) of shade A2 was selected for this study. And total 130 disc shaped specimens were fabricated.

SAMPLE SIZE ESTIMATION

Referring to the study by DA SILVA MA et al. (2016)[32], the authors evaluated the micro hardness and surface roughness of three composite resins immersed in various alcoholic beverages. The initial and after 30 days values were obtained for the parameters and compared. Assuming that the mean initial surface roughness of the resins were insignificantly different across beverages, for each resin, the final data after 30 days was used for estimating the sample size for the proposed study. The effect size for the three resins in the reference study ranged between 0.354 to 1.186.

The proposed study uses five different beverages and a single material. The surface roughness coefficient was referred for sample size estimation. Considering a stringent effect size of 0.3 for the study, a sample of **130** (**i.e**.**26 per group**) will be needed to obtain the desired effect with 95% confidence and 80% power.

The formula used for estimation was:

Null hypothesis:

H_0: $\mu_1 = \mu_2 = \mu_3 = \mu_4 = \mu_5$

i.e. the four mean surface roughness across groups is same

against the alternative that

H_1: $\mu_i \neq \mu_j$ ($i \neq j$ =1,2,3,4,5)

Accordingly, the sample size was obtained using formula:

$$n = \frac{(z_{1-\alpha/2\tau} + z_{1-\beta})^2}{ES^2}$$

where τ is the number of possible comparisons (5_{C_2} = 10 in the present case), $z_{1-\alpha/2\tau}$ (2.5758) is the standardized value for 5% error and for 10 paired comparisons, $z_{1-\beta}$ (0.842) is the value for 80% power

and ES is the effect size.

SPECIMEN PREPARATION

1. Composite disc preparation:

A Teflon mould of 12 mm diameter and 2mm thickness was used for sample preparation. A glass slab was used on which mylar strip was placed over which teflon mould was placed. The resin composite was inserted into the mould cavity in a single increment and covered with mylar strip.

Another glass slab was placed over the resin and mould assembly to remove excess material and provide smooth and highly flat surface. After 30 seconds, the second glass slab was removed and resin composite light cured for 40 seconds each from both upper and lower sides using Light Emitting Diode (LED) light curing unit (Bluephase N, Ivoclar Vivadent) at a distance of 1 mm using continuous mode for curing at an intensity of 1200 mW/cm2.

2. Finishing and polishing of composite discs

The specimens were stored in 37° C ± 1˚C in distilled water for 24 hours in an incubator before finishing and polishing in order to replicate the oral conditions following polymerization. Each sample were polished from both sides using aluminium oxide finishing and polishing discs (3M ESPE Sof-Lex Polishing discs). Standardized protocol was followed according to manufacturer's instructions. In sequential order according to manufacturer instruction, the coarse, medium, fine and super fine used for 20 sec each and the number of strokes were standardized for each specimen.

3. Distribution of Samples

The 130 samples were randomly divided into five groups with 26 samples in each.

The alcoholic beverages viz. Beer, Red Wine, Whiskey, Vodka were the four test solutions and artificial saliva was used as a control group. The Distribution of composite resin samples in different alcoholic beverages are described in Table 1.

1) Group 1 - Artificial saliva (control group)

2) Group 2 – Beer

3) Group 3 –Red Wine

4) Group 4 –Whiskey

5) Group 5 –Vodka

Before immersion into the beverages baseline values of surface roughness was calculated using Surface profilometer (Ra) and colour (ΔE) of specimens were measured using a Spectrophotometer.

IMMERSION OF COMPOSITE DISCS IN ALCOHOLIC BEVERAGES:

The samples were immersed in 10 ml of test groups for 15 minutes, 3 times a day for 30 days.[32] For the remaining part of the day the samples were individually stored in artificial saliva. The test solutions were changed after every immersion. Before measurements, composite samples were rinsed with tap water for 10 seconds and blotted dried with paper towels.

After immersion into beverages final values after 30 days of surface roughness was calculated using Surface profilometer (Ra) and colour change (ΔE) of specimens were measured using a Spectrophotometer.

TESTING OF THE SAMPLES

SURFACE ROUGHNESS TEST

The surface roughness of each sample was performed with the Surface Roughness Tester (Mitutoyo-Japan, Model: SJ 210) with a standard cut off value of 0.8 mm, a transverse length of 8 mm, and a stylus speed of 0.1 mm/s. The "Ra" of a specimen was defined as the arithmetic average height of roughness component irregularities from the mean line measured within the sampling length. Three profilometric tracings were made near the center of the specimen and a numeric average was determined for each specimen. Surface roughness changes were calculated by the differences between mean values obtained before and after immersion.

COLOUR STABILITY

Before immersion, colour reading of the test specimens was performed according to the CIE (Commission Internationale L'Eclairage) L*a*b* system, against a white background. This equipment is specific for colour reading and has 30 led lamps with 10 different colour arranged in a circle which directs a light bundle at 45o with the material surface. This light bundle was reflected 0° back to the equipment, which captured and recorded the L*, a*, b* values of each specimen. The L* axis referred to the lightness coordinate and its value ranges from zero (black) to 100 (white). The axis a* and b* were chromaticity coordinates in the red green axis and the yellow-blue axis, respectively. Positive a* values indicated a shift to red and negative values indicate a shift to green. Similarly, Positive b* values indicated the yellow colour change and negative values indicated the blue colour range.

After immersion for 30 days in alcoholic beverages, specimen's colour was measured by the spectrophotometer, as previously described. Based on the L*, a*, b* values, colour variation (ΔE) was determined using the following equation: $\Delta E=[(\Delta L^*)^2 +(\Delta a^*)^2 +(\Delta b^*)^2]$ ½

ΔE values ranging from 1 to 3 are perceptible to the naked eye but clinically acceptable. Values of $\Delta E > 3.3$ were considered clinically unacceptable.

ALGORITHM OF METHOD

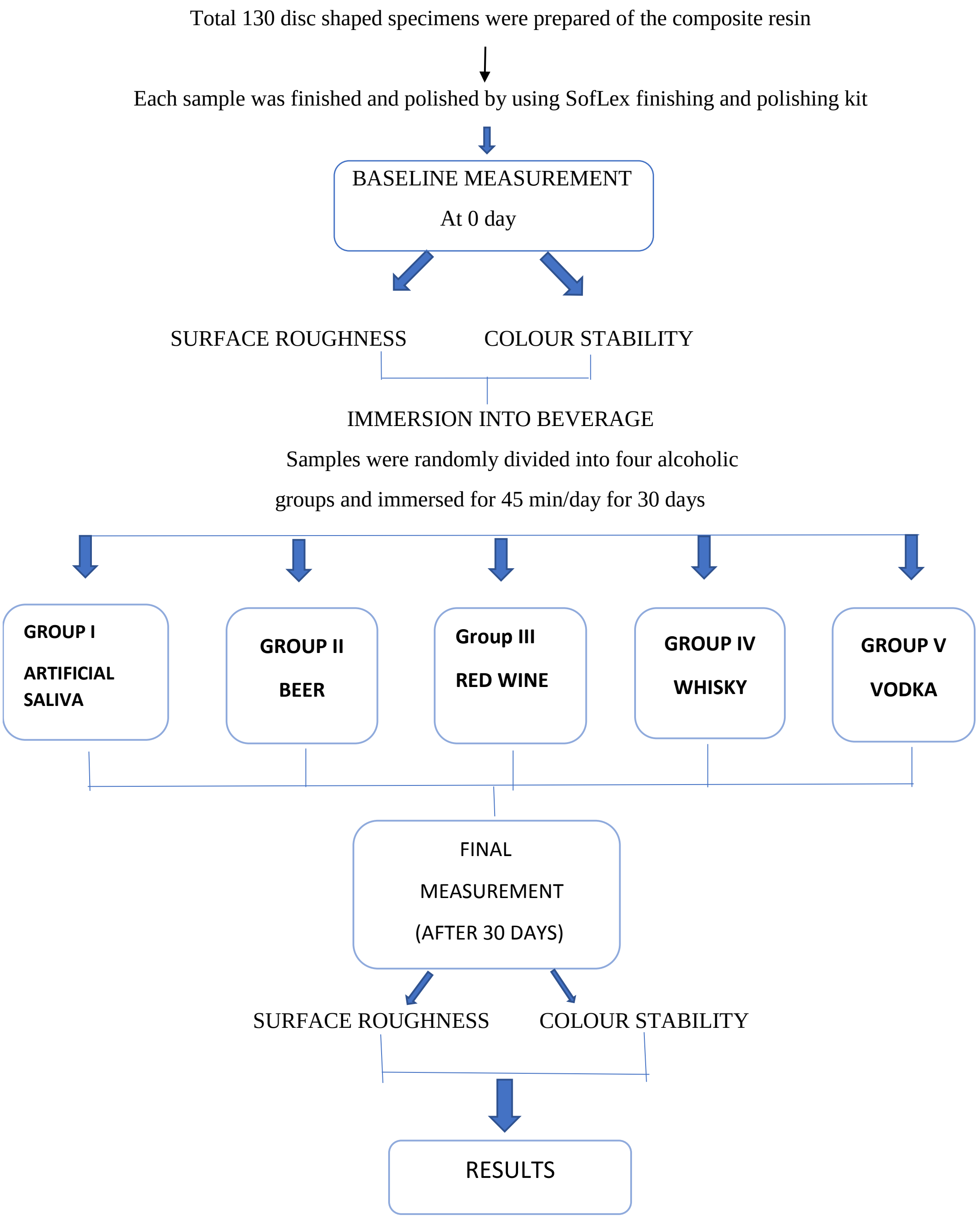
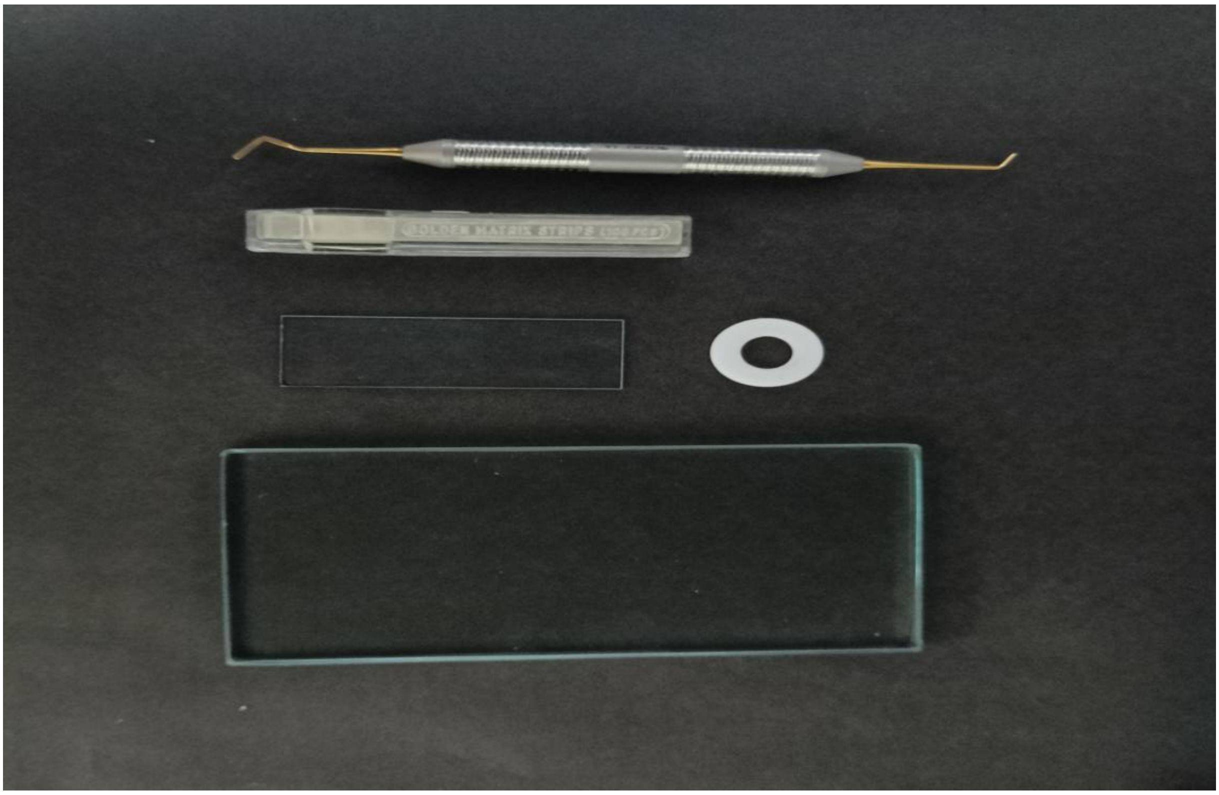


PLATE -I

ARMAMENTARIUM



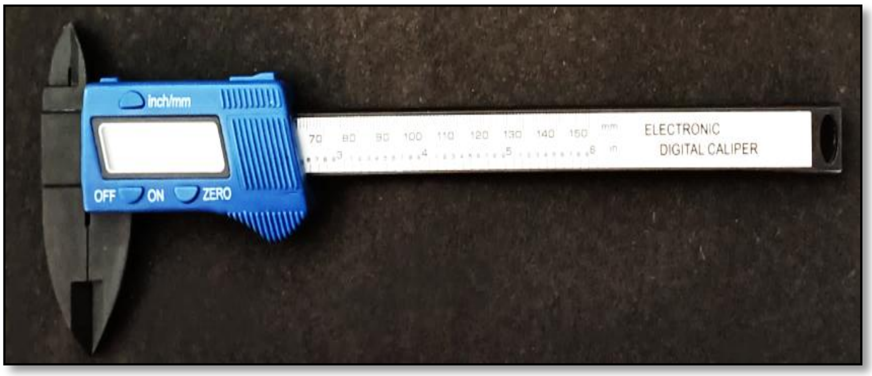
Glass Slab, Matrix Stips, Teflon Mould And Composite Instrument.

Glass beaker

PLATE -II

ARMAMENTARIUM

Composite finishing and polishing kit



Digital Vernier Caliper (Workzone Tools,Germany)

PLATE -III

MATERIALS

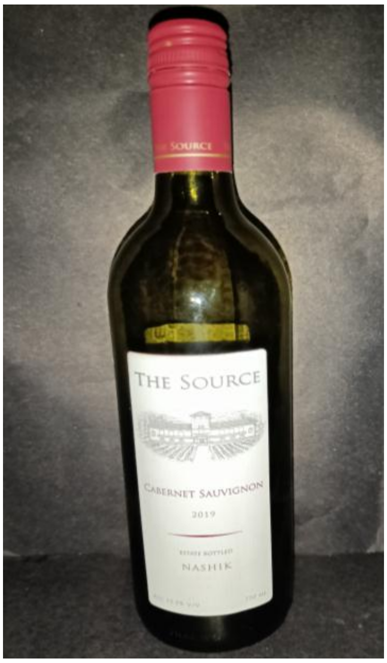
Filtek™ Z350 XT-Shade A2(3M-ESPE, St. Paul, MN, USA)

ARTIFICIAL SALIVA

BEER

PLATE -VI

MATERIALS



RED WINE

WHISKY

VODKA

PLATE - V

EQUIPMENTS

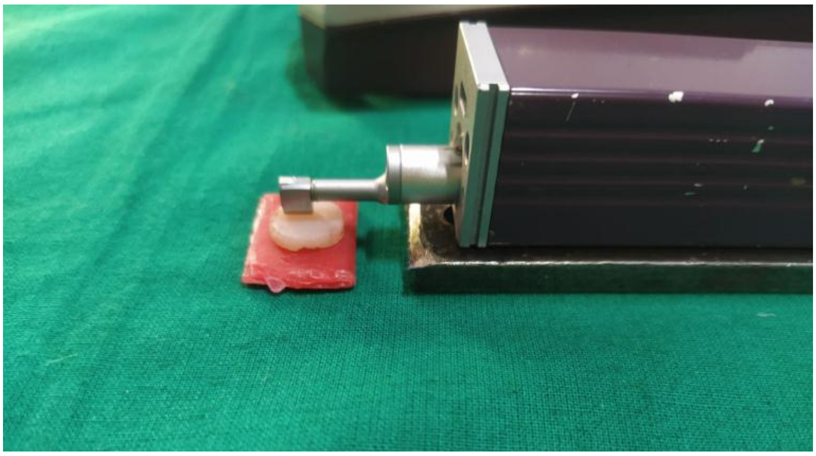
MICROMOTOR (MARATHON)

CONTRA ANGLE HANDPIECE (NSK, JAPAN)

LIGHT EMITTING DIODE (LED) CURING UNIT (BLUEPHASE G2, IVOCLAR VIVADENT, SCHAAN, LIECHTENSTEIN)

PLATE -VI

EQUIPMENTS

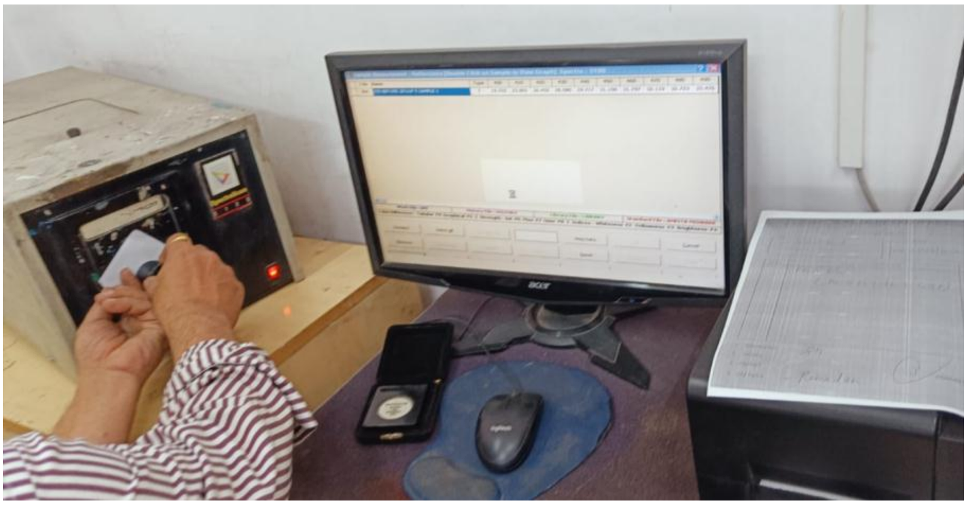


SURFACE ROUGHNESS TESTER

(MITUTOYO-JAPAN, MODEL: SJ 210)

PLATE -VII

EQUIPMENTS

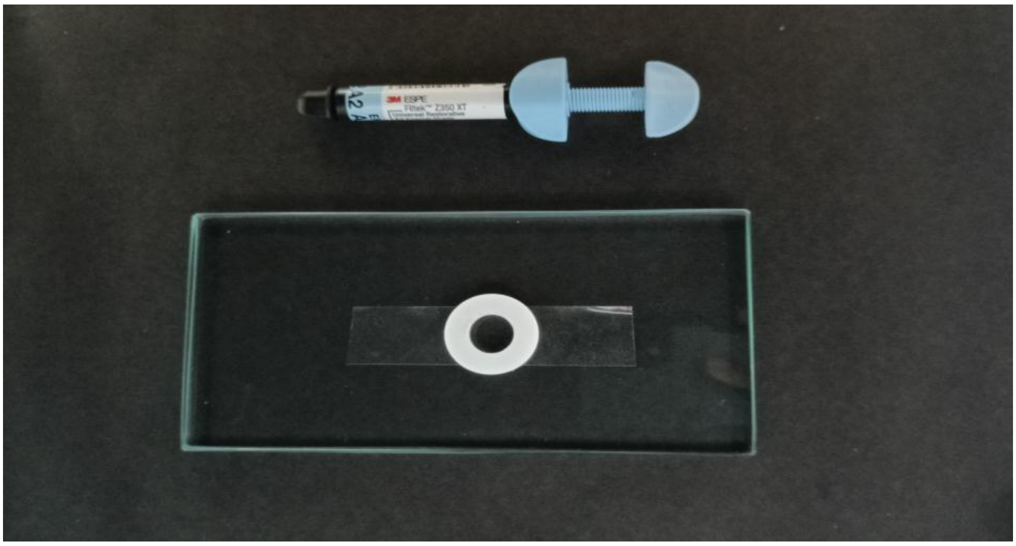


SPECTROPHOTOMETER
(SPECTRASCAN 5100)

PLATE -VIII

METHODOLOGY

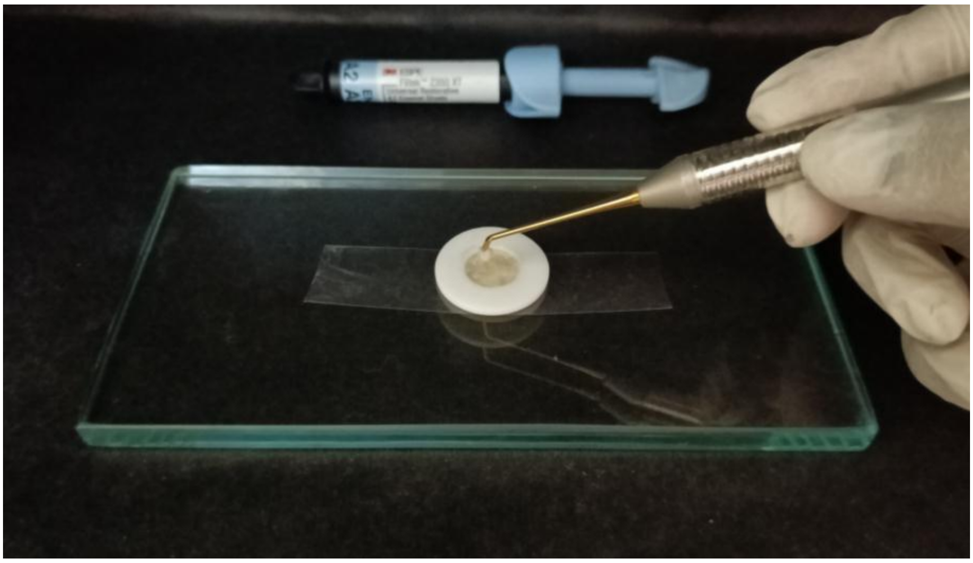
MOULDS OF 12 MM X 2MM DIAMETER WERE TAKEN



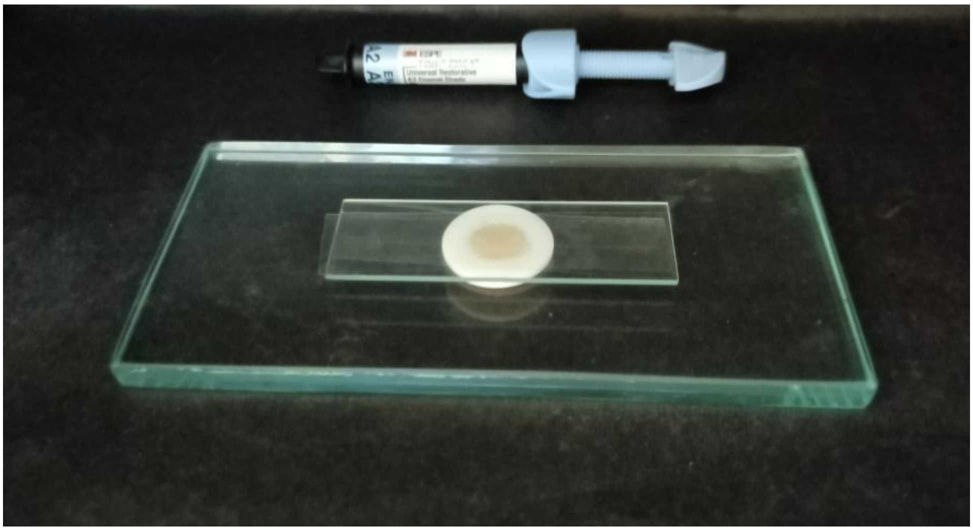
TEFLON MOULD WAS PLACED ONTO THE GLSAA SLAB

PLATE -IX

METHODOLOGY



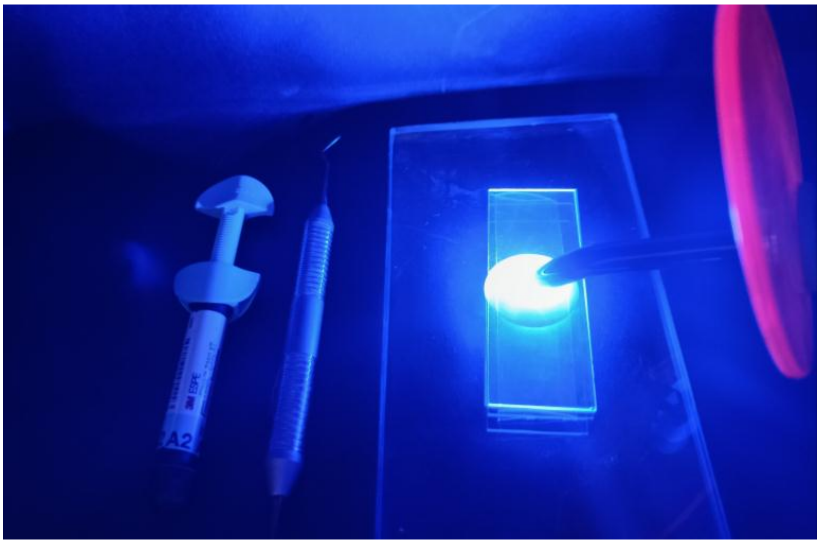
INCREMENTS OF COMPOSITE WERE TAKEN IN
INCREMENTS INTO THE MOULDS



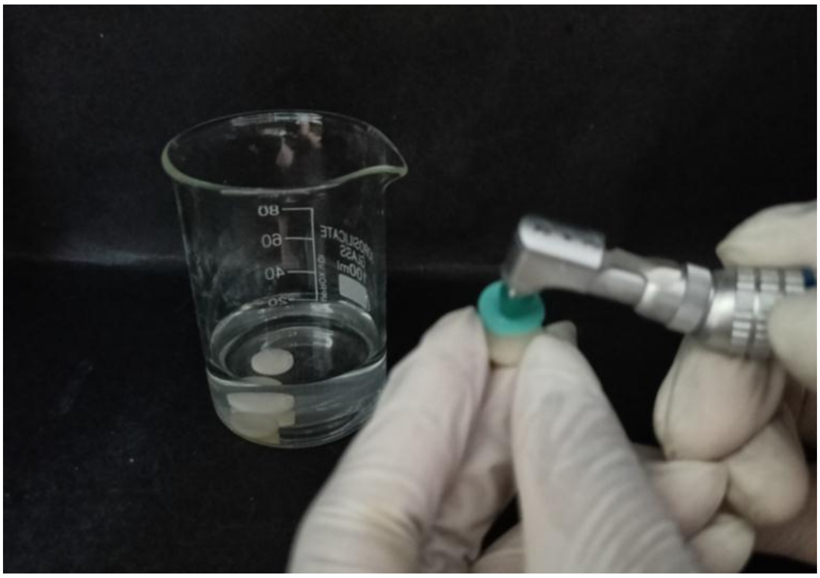
ANOTHER GLASS SLAB WAS PLACED
OVER THE RESIN

PLATE -X

METHODOLOGY



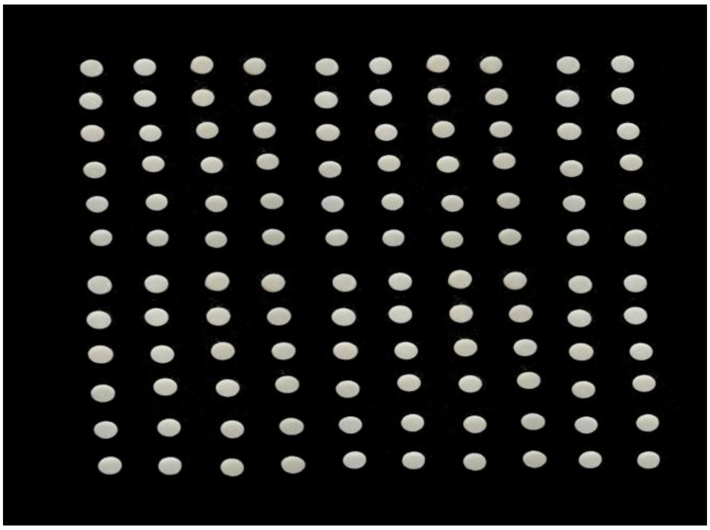
SPECIMENS ARE CURED USING AN LIGHT ACTIVATED LED DEVICE



SPECIMENS WERE POLISHED USING SUPER SNAP POLISHING SYSTEM

PLATE -XI

METHODOLOGY



SAMPLE SIZE N= 130

SPECIMENS WERE STORED IN
DISTILLED WATER FOR 24 HOURS

PLATE -XII

METHODOLOGY

GROUP I – ARTIFICIAL SALIVA

GROUP 2 – BEER

GROUP 3 – RED WINE

PLATE -XIII

METHODOLOGY

GROUP 4- WHISKEY

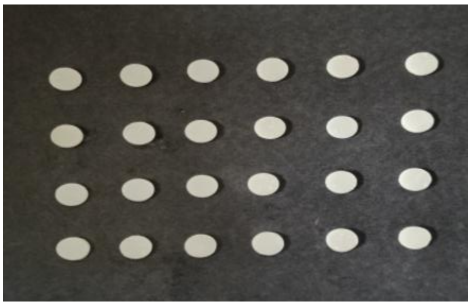
GROUP 5 – VODKA

PLATE -XIV

Composite Resin Specimens After 30 Days Of Immersion In Various Alcoholic Beverages.

GROUP - I

ARTIFICIAL SALIVA



GROUP – II

BEER

GROUP - III

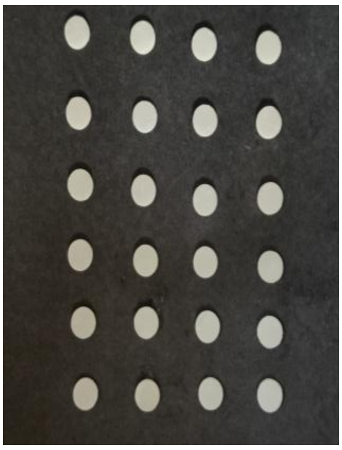
RED WINE

GROUP - IV

WHISKEY

GROUP - V

VODKA



RESULTS

The present in vitro study was conducted to evaluate the effect of four different alcoholic beverages (beer, red wine, whiskey and vodka) on the surface profile and colour stability of nanofilled composite resin (filtek Z 350 XT) after 30 days.

Depending on type of material used the samples were randomly divided into five groups as follow:

1) Group 1 - Artificial saliva (control group)

2) Group 2 – Beer

3) Group 3 –Red Wine

4) Group 4 –Whiskey

5) Group 5 –Vodka

STATISTICAL METHODS

The descriptive statistics like mean, standard deviation, median, minimum and maximum were obtained for surface roughness at day 30, change in surface roughness from baseline to day 30 and colour change from baseline to day 30. The comparison of these parameters across exposure groups was performed using one-way analysis of variance, and the paired comparisons between groups were done using Tukey's post-hoc test. The correlation between change in surface roughness and colour change was determined for each exposure group using Pearson's correlation coefficient. All the analyses were performed using SPSS ver 26.0 (IBM Corp, USA) and the statistical significance was evaluated at 5% level.

The formulations used in the study are as under:

1. Measures of central tendency

If x_1, x_2,…x_n are the observations on a random variable X, then following measures of central tendency can be obtained:

- **Mean** for a set of observations is given by

$$\overline{x} = \frac{1}{n}\sum_{i=1}^{n} x_i$$

- **Median**: It is the middle value of a set of values when arranged in the increasing order of magnitude.

2. Measures of dispersion

- **Standard deviation** for a set of observations in given by

$$s = \sqrt{\frac{1}{(n-1)}\sum_{i=1}^{n}(x_i - \overline{x})^2}$$

where x_i = observation on each object

n = number of objects

- **Range** is the difference between maximum and minimum value of the variable.

- **One-way Analysis of variance**

Analysis of variance (ANOVA) is used to test the significance of difference in the mean of three or more groups. The basic assumption is that the variable of interest is normally distributed in the population under study.

Here the interest is to test the null hypothesis that the population means are same, $H_0 : \mu_1 = \mu_2 = ...\mu_m$ i.e.

against the alternative H_1 that they are not same.

Some of the statistics computed to test the hypothesis are as below:

i) **Grand mean**: It is the mean of set of all observations in the studied groups and is given by:

$$\overline{x}_{GM} = \frac{1}{N}\sum_{i=1}^{N} x_i$$

ii) **Total sum of squares:** It is the sum of squares of each observation from the grand mean and is given by:

$$TSS = \sum_{i=1}^{N} (x_i - \overline{x}_{GM})^2$$

Total sums of squares is the sum of two components i.e., variation between groups and within groups.

iii) Between group sum of squares

$$SSB = \sum_{j=1}^{m} n_j (\overline{x}_j - \overline{x}_{GM})^2$$

iv) Within group sum of squares

$$SSW = \sum_{j=1}^{m} \sum_{i=1}^{n} (x_{ij} - \overline{x}_j)^2$$

The mean sum of squares is obtained by dividing the above sum of squares with the respective degrees of freedom, i.e. N-1, p-1 and $p(n$-1).

v) F-statistic: It is the ratio of between and within mean sum of squares

$$F = \frac{MS_{Between}}{MS_{Within}}$$

If the p-value based on F-statistic is greater than 0.05, H_0 is accepted, otherwise H_1 is accepted.

vi) Tukey's post-hoc test

After performing ANOVA, if alternative hypothesis H_1 is accepted, then the subsequent interest is to determine the pair wise significance of difference in the means of study groups. This could be carried using Tukey's post-hoc test. The difference between the means of all groups are determined and compared with this critical difference called the honest significant difference (HSD). It is given by:

$$HSD = q\sqrt{\frac{MS_{within}}{n}}$$

where, q is the studentized range statistic derived from the tables, n is the sample size and the mean square value is from the ANOVA analysis. If the critical difference exceeds the absolute difference between any two sample means, then the corresponding means differ significantly.

- **Pearson's correlation**

Pearson's correlation coefficient quantifies the relationship between two measurable variables. It measures the linear relationship between two variables. Thus, if X and Y are two variables taking values $x_1, x_2, \ldots x_n$ and $y_1, y_2, \ldots y_n$, then the correlation coefficient (r)between the two variables is given by:

$$r = \frac{n(\Sigma xy) - (\Sigma x)(\Sigma y)}{\sqrt{[n\Sigma x^2 - (\Sigma x)^2][n\Sigma y^2 - (\Sigma y)^2]}}$$

The value of r lies between -1 to +1, with -1 indicating perfect negative correlation and +1 indicating perfect positive correlation.

Overall Results:

The mean values and standard deviations of surface roughness and colour stability of composite resin after immersion in four different alcoholic beverages after 30 days have been described in the Table 2 to table 9 respectively.

The mean change in surface roughness (Ra) of composite resin after 30 days of immersion into alcoholic beverages was found to be highest **(0.326 µm)** in group 5 vodka with standard deviation (SD) 0.029 as compared to other groups and the least change in surface roughness **(0.034 µm)** observed in the control group of artificial saliva where ethanol concentration is 0%.

The mean colour change **(ΔE)** of composite resin after 30 days of immersion in alcoholic beverages was found to be highest **(ΔE= 3.694±1.356)** in group 3 red wine with standard deviation (SD: 1.356) as compared to other groups and the least colour change **(ΔE)** of composite resin was observed in the control group of artificial saliva (**(ΔE=1.415±2.084)** with standard deviation (SD: 2.084) after 30 days when compared to the baseline.

I) ANALYSIS FOR SURFACE ROUGHNESS :-

Table 3 provides the descriptive statistics for surface roughness of samples immersed in five different alcoholic beverages at day 30. The mean for control i.e. artificial saliva group was 0.286 (SD: 0.026) µm and ranged between 0.209 µm to 0.339 µm. In the Beer group, the mean surface roughness was 0.179 (SD: 0.012) µm with a minimum of 0.155 µm and a maximum of 0.199 µm. In the Red wine group, the mean was 0.309 (SD: 0.027) µm and a minimum of 0.272 µm, while a maximum of 0.375 µm. In the Whiskey group, the mean was 0.363 (SD: 0.019) µm, with a minimum of 0.329 µm and a maximum of 0.395 µm. In the Vodka group, the mean was 0.494 (SD: 0.023) µm, with a minimum of 0.449 µm and a maximum of 0.535 µm.

The difference of means across groups was statistically significant as indicated by a p-value < 0.0001.

The pairwise comparison of surface roughness between groups is provided in Table 4. The mean difference of surface roughness for samples exposed to artificial saliva as compared to samples exposed to Beer, Red wine, Whiskey and Vodka, was statistically significant. The mean surface roughness was higher for Red wine, Whiskey and Vodka as compared to artificial saliva. Further, the mean difference between Beer and Red wine, Whiskey and Vodka was statistically significant (p < 0.0001). The means were higher for Red wine, Whiskey and Vodka as compared to Beer. The samples exposed to Red wine showed significantly lower surface roughness as compared to Whiskey and Vodka (p < 0.0001). The samples exposed to Whiskey showed significantly lower roughness as compared to Vodka (p < 0.0001).

Table 5 provides the descriptive statistics for the change in surface roughness of samples exposed to five different treatments from baseline to day 30. The mean change for control i.e. artificial saliva group was 0.034 (SD: 0.024) µm and ranged between -0.072 µm to 0.058 µm. In the Beer group, the mean change in surface roughness was 0.048 (SD: 0.021) µm with a minimum of 0.005 µm and a maximum of 0.086 µm. In the Red wine group, the mean change was 0.229 (SD: 0.032) µm and a minimum of 0.190 μm, while a maximum of 0.305 μm. In the Whiskey group, the mean change was 0.187 (SD: 0.02) μm, with a minimum of 0.125 μm and a maximum of 0.207 μm. In the Vodka group, the mean was 0.326 (SD: 0.029) μm, with a minimum of 0.290 μm and a maximum of 0.404 μm.

The difference of mean change in surface roughness across groups was statistically significant as indicated by a p-value < 0.0001.

Table 6 shows the pairwise comparison of change in surface roughness between groups. The mean difference of change in surface roughness for samples exposed to artificial saliva as compared to samples exposed to Red wine, Whiskey and Vodka, was statistically significant ($p < 0.0001$). The mean change was higher for Red wine, Whiskey and Vodka as compared to artificial saliva. Further, the mean difference of change between Beer and Red wine, Whiskey and Vodka were statistically significant ($p < 0.0001$). The means were higher for Red wine, Whiskey, and Vodka as compared to Beer. The samples exposed to Red wine showed significantly higher change as compared to Whiskey ($p < 0.0001$), while significantly lower change as compared to Vodka ($p < 0.0001$). The samples exposed to Whiskey showed significantly lower roughness as compared to Vodka ($p < 0.0001$).

II) ANALYSIS FOR COLOUR STABILITY :-

Table 8 provides the descriptive statistics for the colour change of samples exposed to five different treatments from baseline to day 30. The mean change for control i.e. artificial saliva group was 1.415 (SD: 2.084) and ranged between 0.021 to 9.06. In the Beer group, the mean colour change was 2.084 (SD: 0.731) with a minimum of 0.715 and a maximum of 3.671. In the Red wine group, the mean change was 3.694 (SD: 1.356) and a minimum of 1.187, while a maximum of 6.395. In the Whiskey group, the mean change was 2.535 (SD: 0.545) μm, with a minimum of 1.839 and a maximum of 3.681. In the Vodka group, the mean was 1.712 (SD: 1.013), with a minimum of 0.453 and a maximum of 5.117.

The difference of mean colour change across groups was statistically significant as indicated by a p-value < 0.0001.

Table 9 shows the pairwise comparison of colour change between groups. The mean difference of colour change for samples exposed to artificial saliva as compared to samples exposed to Red wine and Whiskey, was statistically significant with p-values < 0.0001 and 0.01 respectively. The mean change was higher for Red wine and Whiskey as compared to artificial saliva. Further, the mean difference of colour change between Beer and Red wine was statistically significant ($p < 0.0001$). The mean was higher for Red wine as compared to Beer. The samples exposed to Red wine showed significantly higher colour change as compared to Whiskey ($p=0.007$) and Vodka ($p < 0.0001$).

III) CORRELATION OF CHANGE IN SURFACE ROUGHNESS AND COLOR CHANGE

Figure 5: Bar diagram showing correlation between change in surface roughness and colour change for samples exposed to artificial saliva, beer, red wine, whiskey and vodka.

The bar diagram shows the relationship between change in surface roughness and colour change for samples exposed to artificial saliva. The correlation coefficient was 0.046 suggesting a negligible correlation between the two parameters, which was statistically insignificant (p=0.823)

Figure 5 shows the relationship between the change in surface roughness and colour change for samples exposed to beer. The correlation coefficient was -0.373 suggesting a low negative correlation between the two parameters, which was statistically insignificant (p=0.061).

Figure 5 shows the relationship between the change in surface roughness and colour change for samples exposed to red wine. The correlation coefficient was -0.033 suggesting a negligible correlation between the two parameters, which was statistically insignificant (p=0.874).

Figure 5 shows the relationship between the change in surface roughness and colour change for samples exposed to whiskey. The correlation coefficient was 0.082 suggesting a negligible correlation between the two parameters, which was statistically insignificant (p=0.692).

Figure 5 shows the relationship between the change in surface roughness and colour change for samples exposed to vodka. The correlation coefficient was -0.113 suggesting a weak negative correlation between the two parameters, which was statistically insignificant (p=0.584).

DISCUSSION

The use of resin based-based dental resorative materials has increased because of their good esthetic, easy-handling, and ability to establish a bond to dental hard tissue.[32]

The growing demands for improved esthetics in dentistry has led to an increase in the use of tooth coloured resin composites. It has become one of the most commonly used esthetic restorative materials because of their adequate strength, excellent esthetics, moderate cost compared with that of ceramics, and ability to be bonded to tooth structure. With the improvement of restorative materials and the demand for aesthetic restorations, the composite resin has become the material of choice for esthetic restorations.

It was developed in the mid of 1960's by combining dimethacrylates (epoxy resin and methacrylic acid) with silanized quartz powder. Composites are composed of three major components: resin matrix (organic content), fillers (inorganic part) and coupling agents. The resin matrix mainly consists of Bis-GMA (bisphenol-A-glycidyldimethacrylate), in which TEGDMA is added to reduce the viscosity of the resin matrix.

Clinically acceptable hue match is an imperative thought in an aesthetic dentistry. Surface irregularity, surface lustre, and colour are the most important aesthetic factors of a restorations.[11] It is testified that saliva, constituents of food and beverages might have an effect on the reliability and aesthetics of composite resin. Change in colour of composite resin occur due to the external factors or internal factors. The resin's empathy for external stains is controlled with adaptation percentage and physical-chemical physiognomies, and water absorption rate being of specific importance. Inside the mouth, for the reason that of apparent deprivation or a slim diffusion and adsorption of staining agents at the shallow layer of the composite resin, the superficial or sub-surface staining of composite resin is seen. Furthermore, extrinsic stains is associated to surface coarseness, surface veracity and is affected by finishing and polishing.

Several factors, including composite resin type, content of alcohol and immersion cycle had a significant influence on colour stability, surface roughness and microhardness of composites.

According to National Health Portal, alcohol use is quite common in India both in rural and urban areas with prevalence rates as per various studies varying from 23% to 74%. The increased consumption of alcoholic beverages in young and adult population has raised questions about the erosive potential and discoloration caused by them on the composite restorations.[17] The alcoholic beverages are known to have an acidic nature as they diffuse into the resin, plasticizing the polymer matrix and reducing its physical properties. Very few studies have evaluated the changes in surface profile and colour stability of resin composites due to alcoholic beverages.

Hence, the following study was conducted to evaluate the effect Four different alcoholic beverages i.e beer, red wine, whiskey and vodka on surface profile and colour stability of nanofilled composite resins using surface Profilometer and spectrophotometer.

More recently, nano composite have been introduced to a class of new materials with nano scale inorganic filler particles in the range of 0.1-100 nm dispersed within the resinous matrix. In comparison with microhybrid composites, these materials have been reported to have improved properties, such as, elasticity modulus, mechanical strength and colour stability. Furthermore, these improvements are achieved at low concentrations of the inorganic filler particles. This fact contrast with conventional filled composites, which generally require high loadings within the range of 60%.

The filler particles are responsible for spreading the light, which in turn, provides the opacity of the restorative resin materials, which is a principal factor in aesthetic maintenance of non-particulate composite. The light spread is greater when the filler particle size is larger and consequently, the greater the opacity. The opacity of composites increases as the difference between refraction indexes of resinous matrix and filler particles also increases.[6]

Enone LL et al. (2020)[48] evaluated the clinical performance of nanohybrid composites in Nigerian adult population and stated that resin composites with nanoparticles are characterised by low incidences of roughness and wear after finishing and polishing.[48]

Methacrylate based nanofilled composite resin Filtek™ Z 350 XT (3M-ESPE, St. Paul, MN, USA) was used in this study. The composite was selected as these are recomandable for anterior resorations. Nanofillers present in them enhance their properties by providing reduction in shrinkage and relatable stresses. High radiopacity, wear resistance and excellent polishability with high gloss are the other added advantages of this material. There is a combination of a nonagglomerated/ nonaggregated (20nm nanosilica filler) and loosely bound agglomerated zirconia/silica nanocluster with the particle size of 5-20 nm fillers and a cluster particle size of 0.6-1.4 μ, In addition, the smaller the filler particle, the smaller the amount of water absorbed by the polymer network, which results in lower degradation of the interface matrix/particle, and consequently, presented a lower surface roughness after polishing and lower colour changes for the composites.[49]

Setty A et al (2019)[50], comparatively evaluated the surface roughness of Cention and filtek Z 350 XT and conclude that filtek Z350 XT exhibited lowest surface roughness value than other novel resin composite used in the study.

Long term clinical performance and colour stability of the nanofilled composites are yet to be known and proven. Also, it has not been adequately researched on its degradation under acidic conditions.

In this study, shade A2 of composite resin was selected to examine colour change. **According to Uchida et al (1998)**[51]**,** the selected shade was a universal colour for all materials.

In accordance to the study by **DA Silva MA et al. (2016)**[32], a sample size of 130 was obtained. Assuming that the mean initial surface roughness of the resins were insignificantly different across beverages, for each resin, the final data after 30 days was used for estimating the sample size for the proposed study.

The proposed study uses five different beverages and a single material. The surface roughness coefficient was referred for sample size estimation. Considering a stringent effect size of 0.3 for the study, the estimated sample size that could provide 80% power and 95% confidence interval, 26 samples were allocated per group. Therefore, the total sample size of 130 was taken for the current study.

The formula used for estimating the sample size was:

Null hypothesis:

H_0: $\mu_1 = \mu_2 = \mu_3 = \mu_4 = \mu_5$

i.e. the four mean surface roughness across groups is same

against the alternative that

H_1: $\mu_i \neq \mu_j$ ($i \neq j$ =1,2,3,4,5)

Accordingly, the sample size was obtained using formula:

$$n = \frac{(z_{1-\alpha/2\tau} + z_{1-\beta})^2}{ES^2}$$

where τ is the number of possible comparisons (5_{C_2} = 10 in the present case), $z_{1-\alpha/2\tau}$ (2.5758) is the standardized value for 5% error and for 10 paired comparisons, $z_{1-\beta}$ (0.842) is the value for 80% power and ES is the effect size.

To minimize the edge loss effect, the diameter of the specimens prepared in the present study were 12 mm, greater than the aperture size of the instrument (3 mm × 8 mm).

Thickness and smoothness of the specimen surface also affect colour.[52] Hence in the present study, the thickness of composite resin was prepared as 2 mm as it is the accepted thickness for the incremental technique of composite application.[53]

A matrix strip was used in this study to mimic the clinical procedures for restoring anterior teeth, as it provides the smoothest surface when compared to other finishing and polishing procedures.

Equal irradiance of material was applied using a led light cure unit, regardless of instructions from the manufacturers.[1] In the present study, the same principle was applied. The sample was light cured using LED curing unit (Bluephase N, Ivoclar Vivadent) for 40 seconds. The tip was kept in close contact and perpendicular to the surface of composite specimen. During curing of composite specimen, the specimens were carefully centred at the tip of curing gun.

Kramer Norbert et al. (2008)[54]**,** considered LED curing unit superior to QTH because of longer lasting bulbs with fewer maintenance concerns, emit radiation only in blue part of visible spectrum, they do not require filters, are less energy consuming and require less wattage, generates no heat, produces higher power intensity, thus reducing polymerization time. **Price et al. (2003)**[55]**,** compared two irradiation time 20sec and 40 sec by using LED curing light and they have reported that 40 sec exposure can polymerised all the composite resin. Thus, in the present study, Specimens were irradiated for 40 second as it gives better results.

The specimens were stored in Artificial saliva at 100% humidity for 24 hours at 37°C in an incubator to stimulate oral condition.[23] Addition to this, Artificial saliva also used as a control group in this study which composed of Carboxymethylcellulose, glycerine, minerals like phosphorous, calcium and fluoride, Xylitol, other ingredients.[23]

Smoother resin composites are more comfortable for the patient[56] and increased surface roughness leads to staining in vitro studies.[57]

Chandhok et al (2017)[58], stated that Finishing and polishing system with aluminium oxide discs (Soflex) provided the smoothest surface followed by Mylar strip and Fine & extrafine grit aluminium oxide paste along with polishing discs and cones (Enhance) for both Nanofilled and Microhybrid composite resins.[58] For these reasons, in this study the finishing and polishing process was performed with Sof-Lex finishing and polishing system as specified by manufacturers, which have a greater hardness than the majority of particles found in the formulation of composite resins.[53] In sequential order according to manufacturer instruction, the coarse, medium, fine and super fine used for 20 sec each and the number of strokes were standardized for each specimen.

In the present study after composite disc preparation block randomisation was done into five groups containing 26 samples in each group as follows:

1) Group 1 - Artificial saliva (control group)

2) Group 2 – Beer

3) Group 3 –Red Wine

4) Group 4 –Whiskey

5) Group 5 –Vodka

According to **Villalta et al (2006)**[59]**,** the alcohol content and low pH of solutions affect the surface roughness of composite resins and causing stating. Thus, in the present study, the alcoholic bevereges were used according to their presence of Alcohol by volume percent ABV (%).

The most commonly consumed alcoholic beverages are beer, red wine, whiskey and vodka. Many previous studies used this alcoholic beverages to evaluate the surface roughness, microhardness and colour stability of composite resins after immersion for a period of 7 days, 15 days and 30 days.[31,60] Therefore, in the present, different alcoholic beverages according to their alcohol by volume percent i.e beer, red wine, whiskey and vodka were used as a testing samples.

Beer contains ethanol 5% to 8% ABV (%). Also it composed of Water (90-94%), higher alcohol, aldehydes and organic acids ect.[61] Red wine is a type of wine made from dark-coloured grape varieties consumed worldwide. Generally wine is composed of water (86%), ethanol (12-14%), glycerol, higher alcohol, polysaccharides and organic acids etc.[62]

Whiskey is a type of distilled alcoholic beverage made from fermented grain mash. It composed of ethanol 38-44 %, whiskey lactone, Aldehydes, esters, phenolic compounds and other ingredients. Vodka is a clearless alcoholic beverage which mainly composed of ethanol (ABV 40-44%) and water. Additives i.e citric acid and glycerol, Impurities are added to this.

Artificial saliva was kept as a control to simulate the in-vivo conditions as performed in the study by **Badra VV et al. (2005)**.[63]

An accelerated lab test was given by **Asmussen (1983)**[64] to stimulate the clinical discolouration potential of the composite, which stated that the colour changes produced in composite resins by storing for one month (30 days) was well correlated with colour change obtained after 12 months at 37°C in oral cavity. In many in vitro studies, the immersion period is typically four weeks or more in order to achieve a cumulative staining effect and obtain distinct results.[64,65]

According to **Silva D et al (2016)**[32]**, t**he 30 days period of immersion in alcoholic beverages was enough to produce significant differences in surface roughness and other physical properties of the dental composite resin. Groups immersed in alcoholic beverages were able to cause chemical degradation in the polymer of all dental composites tested.[32]

In the study conducted by **Chakravarthy et al (2018)**[36], they stated that the effect of red wine on the colour change of composite resin depend on the physical and chemical composition of the material and adsorption or absorption of staining pigments in the red wine, which significantly causes the colour change in nonohybrid composites after 28 days.[36]

Therefore, in the current research, the assay was performed maintaining the samples immersed for 15 min, three times a day, for 30 days period as performed in the study by **Silva D et al (2016)**[32]**.**

The surface quality or texture of a restoration is an important factor determining their clinical success in the oral environment. The surface roughness (Ra) value is described as the arithmetic mean value of movement of profile above and below the centerline of the surface. Mechanical profilometer, Scanning Electron Microscopy (SEM), optical 3-D profilometer, etc. are some of the methods used for measuring the surface roughness (Ra) value.[66]

Surface profilometers have been used for years to measure surface roughness in laboratory investigations. There are two types of profilometers available which are contact profilometer and non-contact profilometer. The contact profilometer can be further classified as manual or digital.

Average surface roughness was measured with the help of profilometer. It is a device that uses a diamond stylus of 2µ diameter to trace a fixed linear distance over the surface of the prepared sample. The profilometer produces a tracing using digital and analogue hardware and software, and calculates the average surface roughness (Ra) value for the resultant tracing.[67] It provides a quantitative recording of surface irregularities. Hence, in this study, Profilometer (**Mitutoyo-Japan, Model: SJ 210**) readings were made at the center of each specimen, and the numerical averages were determined for each group.

According to Whitehead SA et al. 1989[68], it is a more desirable and accurate way of detecting and assessing surface roughness than scanning electron microscopy images. Also, minor difference in surface roughness could not be corroborated by SEM evaluation but that could be easily done with the help of profilometer readings.

In the present study, surface roughness assessment was chosen because surface micromorphology would affect the staining susceptibility.

Another parameter tested in the present study was colour stability, which can be evaluated both visually and by specific instruments. The methodology used in the present study is according to the previous studies,[2,3,23] that used spectrophotometry and the CIE L*a*b system was chosen to evaluate colour stability (ΔE) because it is appropriate for small color changes determination and have advantages such as repeatability, sensitivity and objectivity.[69] Spectrophotometer **(SPECTRA SCAN 5100)** was the device used to evaluate the colour stability of the composite resin.

I) Effect of different alcoholic beverages on surface roughness :-

Higher surface roughness (Ra) values (> 0.2μm) have been reported as a risk factor for extensive plaque accumulation on dental materials and as the main contributor to the multifactorial discolouration of resin restoration.[2,22,26,70]

The pH is a very important factor to determine the erosive potential of a solution[71]. Low pH and higher ethanol concentration may affect the surface integrity of composite resins. Alcohol, propionic and acetic acids in beverages causes significant increases in surface roughness and erosion in resin composite because alcohol is also thought to act as a plasticizer of the polymer matrix to soften and dislodge filler particles, resulting in a rapid increase in surface roughness and erosion.[17,31,72,73] The softening effect of alcohol on the RBCs may be due to the susceptibility of bisphenol A-glycidyl methacrylate (Bis-GMA) and urethane dimethacrylate (UDMA)-based polymers.[74] This explains the change in surface roughness of the dental composites immersed in alcoholic beverages.[75]

The current research indicated significant changes on the surface of composite resin when exposed for 30 days in alcoholic beverages when compared to the baseline.

The descriptive statistics for the change in surface roughness of samples exposed to five different treatments at day 30 and from baseline to day 30 are provided in the table 4 and table 5 respectively:

In the present research, the difference of mean change in surface roughness (Ra) for Filtek Z350 immersed in an artificial saliva as compared to filtek Z350 immersed in Beer, Red wine, Whiskey and Vodka, was found to be statistically significant after 30 days as indicated by **p-value <0.001.**

Maximum roughness was observed in the following order:

Vodka > Red Wine > Whiskey > Beer > Artificial Saliva.

The mean change in surface roughness (Ra) of composite resin after 30 days of immersion into alcoholic beverages was found to be highest **(0.326 µm)** in group 5 vodka with standard deviation (SD) 0.029 as compared to other groups and the least change in surface roughness **(0.034 µm)** observed in the control group of artificial saliva where ethanol concentration is 0%.

These results are in accordance with findings of **Sarrett DC et al (2000)**[17], who found that a likely reason for the increased wear caused by the higher ethanol concentration i.e 9 vol% compared to that the lower ethanol concentration i.e 5 vol%.

In study conducted by **Münchow EA et al. (2014)**[76], they stated that acidic solutions increase the roughness of composites, probably because they soften their surface, leading to the leachability of resin components and, consequently displacement of filler particles which contribute to the formation of a rough surface.

In the present study, vodka showed significant change in surface roughness than other beverages after 30 days when compared to baseline. (**p-value < 0.0001)** The reason behind this could be the presence of higher concentration of ethanol in the range of 38- 43% v/v and acids than other beverages. It also contains trace amount of other compounds such as esters, aldehydes, higher alcohols, methanol, acetates, acetic acid and fusel oil.[77–79]

In the current research, for group beer, mean change in surface roughness after 30 days was found to be statistically significant to the baseline with mean difference 0.048 (SD: 0.021) µm and p-value< 0.0001. it causes more surface roughness than group artificial saliva. As it contains 5-8% of ethanol which increases surfaces roughness than.

The mean difference of change in surface roughness after 30 days in the current research for the group 4 whiskey, was found to be statistically significant to the baseline with mean difference value 0.187 (SD: 0.02) µm and p-value 0.0001. Whiskey showed higher degradation than beer and artificial saliva because it contain higher ethanol concentration. (40% v/v)

Omer Geha et al (2021)[80]**,** observed in his study that the surface roughness after chemical degradation of Filtek Z 350 was reported to nearly similar in alcohol solution (0.047) and citric acid solution (0.044).

In the study conducted by **Saijai Tantanuch et al (2016)**[31], red wine (ethanol content 13.5 % v/v) caused greater surface roughness than white wine (ethanol content 12.5% v/v) because red wine has a higher ethanol concentration.

In the present research, for the group of red wine, the mean difference of change in surface roughness after 30 days was statistically significant to the baseline with mean difference 0.229 (SD: 0.032) and p-value< 0.0001 than artificial saliva, beer and whiskey. Also, red wine showed lesser surface roughness than vodka because the ethanol content is less in red wine than vodka.[17]

This difference can probably be explained due to variations in the factors such as ethanol concentration and low pH affecting on the surface roughness of composite resin after immersion in different alcoholic beverages like beer, red wine, whiskey and vodka after 30 days. The penetration capability of alcoholic beverages into composite resin may be closely dependent on their ethanol concentration, pH of the beverages, presence or absence of inorganic fillers and hydrophilic or hydrophobic resin monomers.

This may possibly explain the significant difference found in the surface roughness of composite resin when immersed in the different alcoholic beverages for 30 days.

II) Effect of different alcoholic beverages on Colour stability:-

The colour change in composite resin usually occurs due to three factors. The first factor for colour change is that external discolouration from accumulation of stains and plaque. The second factor is that surface alteration of composite that promote surface roughness, absorption and adsorption of staining agent on the surface of composite. The last factors is intrinsic discolouration from physiochemical reaction of composite.[70,31,81]

Also, the staining of composite is related to resin matrix, the percentage of fillers and size of the filler.[82] **Tonetto MR et al (2012)**[83], stated that wine has tendency to produce more colour change.[83] Previous studies, demonstrated that the ethanol-containing solution was readily absorbed by the resin monomer of the nanofilled composite, i.e., bisphenol glycidyl methacrylate (bis-GMA), ethoxylated bisphenol-A dimethacrylate (bis-EMA), urethane dimethacrylate (UDMA), and TEGDMA.[63,84,85] That could result in the softening of the composite resin surface and contribute to staining, which could explain the results obtained for red wine solution.[22,59,86]

Susceptibility of stain to composite resin can be a result of the type of resin matrix and water absorption of the resin matrix which could decrease the longevity of composite resin by expanding and plasticizing the resin matrix, hydrolyzing the silane coupling agent, and producing microcrack formations. Consequently, the microcracks at the interface between filler particles and the resin matrix permit surface acid degradation, increase surface roughness and penetration of staining solution.[72]

In the current research, the colour change of nanofilled composite resin Filtek Z350 XT when immersed in different alcoholic beverages was evaluated at the baseline and after 30 days. ΔE^* values ranged between the lowest 1.41 ± 2.08 and the highest 3.69 ± 1.35.

Several authors have reported that ΔE values ranging from 1 to 3 are perceptible to the naked eye but clinically acceptable and ΔE values greater than 3.3

are clinically unacceptable.[2,87–89] Considering these concepts; the composite resins tested in the present study demonstrated acceptable colour stability (ΔE) when stored in different alcoholic beverages (beer, whiskey, vodka) except red wine which demonstrated clinically unacceptable colour difference.

In the present study, Maximum colour change was observed in the following order:

Red Wine > Whiskey > Beer > Vodka > Artificial Saliva.

In the current research, the mean colour change **(ΔE)** of composite resin after 30 days of immersion in alcoholic beverages was found to be highest **(ΔE= 3.694±1.356)** in group 3 red wine with standard deviation (SD: 1.356) as compared to other groups and the least colour change **(ΔE)** of composite resin was observed in the control group of artificial saliva (**(ΔE=1.415±2.084)** with standard deviation (SD: 2.084) after 30 days when compared to the baseline.

In the present study, the mean difference of colour change for Filtek Z350 XT when immersed in artificial saliva as compared to samples immersed in Red wine and Whiskey, was statistically significant with p-values < 0.0001 and 0.01 respectively. higher surface roughness causes penetration of extrinsic stains into the composite resin which could be a possible reason for the colour change of composite resin in red wine. Another explanation for the above result may be elucidated by findings of **villalta et al (2006)**[59], who stated that the staining capacity of the composite resin is related to extrinsic factors, such as the pigment agent is subjected. **Fei He et al (2011)**[90], stated that Originating in the grapes, anthocyanins and their derivatives are the crucial pigments responsible for colour of the red wine.[90] Tannin, anthocyanin and its pigments in the red wine may have a significant effect on colour change.[31]

The pH of the beverages reproduces the strength of acidity, low pH can negatively affect the surface integrity by softening the matrix, causing a loss of structural ions and affecting the wear resistance of dental materials.[21,22,59,91,92] Red wine had pH (3.32±0.02). Thus, red wine caused significantly higher colour change (^E >3.3) than white wine and deionised water. He also stated that red wine has higher ethanol concentration i.e 13.5 vol% than white wine i.e 12.5 vol% that might be a etiology of colour change. The red wine promoted a marked colour change of composite resin, probably due to the the higher concentration of pigments, cause greater staining.[21,22,83]

Hwang S et al (2018)[60], stated that the pH solely did not significantly affect the colour change of the composite resin, but the degeneration of composite resin caused by ethanol was accelerated by the acidic solution of pH 3.0–4.0. He reported that when the ethanol concentration was 20% without pigment at a pH of 3.0 or a pH of 4.0, there was a clinically noticeable colour change, while there was no clinically noticeable colour change was seen in the pH 2.0 and pH 5.5 solution.[60]

Thus, another potential explanation for the colour change in red wine could be a low pH that causes surface degradation, resulting in penetration of coloured pigments into the resin matrix.[36]

The composite resin immersed in Red wine showed significantly higher colour change after 30 days and when compared to Whiskey ($p=0.007$) and Vodka ($p < 0.0001$).

In the study conducted by **Meira IA et al (2020)**[92], on the erosive potential of different types of alcoholic beverages, concluded that whiskey were not potentially erosive on dental hard tissue.[92] This could justify the result of the present study that colour change of whiskey is less than the red wine because the surface roughness is more in red wine than whiskey.

According to **Mortensen A et al (2006)**[93], plain caramel pigment which is a natural colour which is used for beverages with high alcohol content like whiskey, while Ammonia caramel is used in beer.[93]

Further, the mean difference of colour change between Beer and Red wine was statistically significant as indicated by **($p < 0.0001$)**. The mean difference of colour change **(ΔE)** composite resin when immersed in Red wine was observed to be higher as compared to composite resin immersed in Beer **($\Delta E = 2.084 \pm 0.731$)**. Thus, in the present study, although beer and whiskey produced colour changes in composite resin, its magnitude of colour change was comparably lower than that of red wine. Their colorants might have been readily taken up to produce colour change, but the colourants were comparatively lighter in colour than the darker colourants of red wine.[30,60,83]

This result are in accordance with the result of study **by James V et al (2019)**[94], who reported that red wine causes more colour changes than beer in composite resin. Author stated that the intense staining by red wine is probably due to the presence of large amounts of these colourant species present in the beverages.[94]

In the present study, the mean colour change **(ΔE)** of composite resin after 30 days of immersion in vodka was found to be i.e **$\Delta E = 1.712 \pm 1.013$** to the baseline which is perceptible through close observation. Vodka caused maximum surface roughness but vodka is a clearless. Stainability is a combination of absorption and adsorption of extrinsic stains and surface profile. In case of vodka though it is showing maximum surface roughness, the stainability was lesser due to its colour and mimic water. This is in accordance with the study by **Tan BL et al in (2015)**[30], who depics that vodka produced least amount of colour change (**ΔE= 2.29 ±0.73)** than red wine **(ΔE= 12.96±0.63)** in filtek Z350 XT, because vodka is a colourless beverage that does not consist colorant pigments. They also stated that vodka with an alcohol content of 40% v/v might produce colour changes by degrading the resin matrix of the composite resin. The current result is in vitro condition and thus, further clinical studies and co relation is required.

In this study, the artificial saliva also promoted a slight colour change in the specimens (**ΔE =1.415±2.084**), classified as slightly perceptible. **Omata et al (2006)**[65], who observed that artificial saliva causes slight colour changes due to presence of mucin in its composition.[65]

In the current research, the mean colour change (**ΔE)** of composite resin Filtek Z350 XT when immersed in different alcoholic beverages after 30 days was found to be statistically significant in the red wine group and whiskey group.

3. Correlation between surface roughness and colour stability:

In the present study, statistically insignificant differences were observed between correlation of surface roughness and colour stability of both the composite resins after immersion in four different alcohol beverages for 30 days. There was negligible correlation found among the parameters, which was statistically insignificant.

In the present study, there was statistically significant difference showed in the surface roughness for samples immersed in control group (artificial saliva) as compared to samples immersed in beer, red wine, whiskey and vodka. The probable reason for this is higher alcohol content and low pH than artificial saliva. Also, the significant difference in the colour change showed in the samples that immersed in beer, red wine and whiskey as compared to samples immersed in artificial saliva and vodka.

In the current research, results showed low pH and higher ethanol concentration leads to increase surface roughness and decrease colour stability. Also, another factor affected on the colour change is presence of pigments in the beverages which increase the colour change of composite resin. This occurs due to absorption and adsorption of pigments by the composite resin when surface roughness is higher.

The result of the present research are in accordance with the study conducted by **Ozyurt E et al (2021)**[95]**,** there was no correlation found between colour changes and water absorption exhibited better surface properties in the composite resin following immersion in the different beverages[37,95]

Thus, in the present study, there was no correlation found between the surface roughness and colour stability, which was statistically insignificant. There is limited research available on the affect of low pH, alcohol content and presence of pigments on the surface roughness and colour stability of nanofilled composite resin and furthermore dedicated research is needed to be carried out in this area of correlation between surface roughness and colour stability when exposed to different beverages.

Thus, the null hypothesis of this study that there is no significant change in surface roughness and colour stability of nano-filled resin composite after exposure to alcoholic beverages i.e beer, red wine, whiskey, vodka after 30 days was rejected.

Clinical significance of this current research is that though in vitro, this research will still be able to give both clinicians and patients the ability to evaluate the effect of patient's lifestyle habits on dental restorative treatment procedures and motivate them to change alcoholic beverage consumption behaviours.

LIMITATIONS

The findings of the present study have to be seen in light of several possible limitations despite stringent adherence to research protocols:

1. Saliva could dilute or buffer pH of mouth rinse solutions, thus reducing the effect of resinous matrix plasticization and forming a pellicle that could have a protective effect on the composite surface, thus, decreasing material staining

2. It is difficult to extrapolate the results of this study to in vivo conditions. However, the results of the study can give an insight into how different resin composites may behave when exposed to different alcoholic beverages, thus affecting the clinician's choice of material and the patient's control of dietary habits.

3. Based on such factors, further studies are needed to determine the effects of alcoholic beverages on these properties of composites.

Colour stability (ΔE) and Surface roughness (Ra) are may not in all cases primarily & solely responsible for longevity of the composite restoration.

SUMMARY AND CONCLUSION

In the past decade, several experimental and clinical studies have been carried out on the consumption of alcoholic beverages that have shown to alter the surface properties and colour stability of composite restorations. Despite all the precautions has taken and guidelines being scrupulously followed by clinicians, surface alteration and colour change of the composite resin is a regularly faced challenge in a clinical practice.

The present in vitro study was thus performed to evaluate the effect of four different alcoholic beverages (beer, red wine, whiskey and vodka) and artificial saliva as control on the surface roughness and colour stability of nanofilled composite resin after 30 days.

In the present study, total 130 disc shaped specimens of dimensions 12 mm X 2 mm of nanofilled composite resin Filtek Z350 XT of shade A2 were made from a custom made teflon mould.

Composite resin samples were randomly divided into Five subgroups i.e 26 samples in each groups (artificial saliva, beer, red wine, whiskey and vodka).

Composite material.	Groups	Testing solutions.	No of samples.
Nono-filled Filtek Z350 XT (N=130)	Group 1	Artificial saliva (control group)	n=26
	Group 2	Beer	n=26
	Group 3	Red wine	n=26
	Group 4	Whiskey	n=26
	Group 5	vodka	n=26

Before immersion into the beverages baseline values of surface roughness was calculated using Surface profilometer (Ra) and colour (ΔE) of specimens were measured using a Spectrophotometer.

Time of immersion was calculated according to Silva DA et al (2016).[32] To simulate the period of 12 months exposure of restorative material to the beverages in oral condition, time of immersion for samples is calculated as 15 min, 3 times a day for 30 days.

After immersion into beverages final values after 30 days of surface roughness was calculated using Surface profilometer (Ra) and colour change (ΔE) of specimens were measured using a Spectrophotometer.

The results of the present study indicated that there is significant difference in the surface roughness and colour stability of the nanofilled composite resin after immersion into different alcoholic beverages when compared to the baseline. (p-value<0.001)

But there was no statistical significant correlation found between the surface roughness and colour stability of composite resin after 30 days when immersed in four different alcoholic beverages i.e beer, red wine, whiskey and vodka.

Within the limitations of the present study, following conclusions can be drawn:

1. There is significant difference in the surface roughness and colour stability of the nanofilled composite resin after immersion into different alcoholic beverages when compared to the baseline.

2. The surface roughness and colour stability of nanofilled composite resin is affected by the type of beverages, low pH, ethanol concentration and pigments present in the beverages.

3. Surface roughness for the composite resin significantly increased after immersion into beverages in the following order:

   Artificial Saliva < Beer < Whiskey < Red Wine <Vodka.

4. Colour stability for the composite resin significantly reduced after immersion into beverages in the following order:

   Red Wine> Whiskey > Beer> Vodka > Artificial Saliva

5. There was a no correlation between surface roughness and colour stability, which was statistically insignificant.

Taking into consideration the findings of the present research, it can be concluded that the need for periodic finishing and polishing of composite resins after every 6 months to enhance the clinical performance and to improve longevity of the restorations. However, further clinical investigations which could give a conclusive remark on the long-term effect of alcoholic beverages on mechanical properties of composite resins in oral cavity are needed.

Clinical significance of this current research is that though in vitro, this research will still be able to give both clinicians and patients the ability to evaluate the effect of patient's lifestyle habits on dental restorative treatment procedures.

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TABLES

Table No 1: Distribution of composite resin samples in different alcoholic beverages.

Composite material.	Groups	Testing solutions.	No of samples.
Nono-filled Filtek Z350 XT (N=130)	Group 1	Artificial saliva (control group)	n=26
	Group 2	Beer	n=26
	Group 3	Red wine	n=26
	Group 4	Whiskey	n=26
	Group 5	vodka	n=26

Table 2: Descriptive statistics for surface roughness at baseline (day 0) and end of follow up (day 30) for samples in different exposure groups.

	Exposure groups									
	Artificial Saliva (Control)		Beer		Red wine		Whiskey		Vodka	
Time	Day 0	Day 30	Day 0	Day 30	Day 0	Day 30	Day 0	Day 30	Day 0	Day 30
N	26	26	26	26	26	26	26	26	26	26
Mean	0.252	0.286	0.131	0.179	0.08	0.309	0.176	0.363	0.168	0.494
Standard Deviation	0.024	0.026	0.015	0.012	0.011	0.027	0.018	0.019	0.016	0.023
Median	0.248	0.284	0.129	0.179	0.079	0.301	0.178	0.362	0.17	0.497
Minimum	0.206	0.209	0.101	0.155	0.048	0.272	0.124	0.329	0.115	0.449
Maximum	0.298	0.339	0.172	0.199	0.099	0.375	0.214	0.395	0.19	0.535

Table 3: Descriptive statistics for surface roughness of samples exposed to five different treatment after 30 days

	Exposure groups				
	Artificial Saliva (Control)	Beer	Red wine	Whiskey	Vodka
N	26	26	26	26	26
Mean (µm)	0.286	0.179	0.309	0.363	0.494
Standard Deviation (µm)	0.026	0.012	0.027	0.019	0.023
Median (µm)	0.284	0.179	0.301	0.362	0.497
Minimum (µm)	0.209	0.155	0.272	0.329	0.449
Maximum (µm)	0.339	0.199	0.375	0.395	0.535

P-value < 0.0001 (Significant) using one-way analysis of variance.

Table 4: Pairwise comparison of surface roughness of samples exposed to five different treatments

Group (I)	Group (J)	Mean Difference (I-J)	P-value	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Artificial Saliva (Control)	Beer	0.107	< **0.0001**	0.090	0.124
	Red wine	-0.023	**0.002**	-0.040	-0.006
	Whiskey	-0.077	< **0.0001**	-0.094	-0.060
	Vodka	-0.208	< **0.0001**	-0.225	-0.191
Beer	Red wine	-0.130	< **0.0001**	-0.147	-0.113
	Whiskey	-0.184	< **0.0001**	-0.201	-0.167
	Vodka	-0.315	< **0.0001**	-0.332	-0.298
Red wine	Whiskey	-0.054	< **0.0001**	-0.071	-0.037
	Vodka	-0.185	< **0.0001**	-0.202	-0.168
Whiskey	Vodka	-0.131	< **0.0001**	-0.148	-0.114

*Obtained using Tukey's post-hoc test; Bold p-values indicate statistical significance

Table 5: Descriptive statistics for change in surface roughness of samples treated with five different exposures

	Exposure group				
	Artificial Saliva (Control)	Beer	Red wine	Whiskey	Vodka
N	26	26	26	26	26
Mean (μm)	0.034	0.048	0.229	0.187	0.326
Standard Deviation (μm)	0.024	0.021	0.032	0.020	0.029
Median (μm)	0.038	0.049	0.225	0.199	0.325
Minimum (μm)	-0.072	0.005	0.190	0.125	0.290
Maximum (μm)	0.058	0.086	0.305	0.207	0.404

P-value < 0.0001 (Significant) using one-way analysis of variance

Table 6: Pairwise comparison of the change in surface roughness of samples exposed to five different treatments

Group		Mean Difference (I-J)	P-value	95% Confidence Interval	
(I)	(J)			Lower Bound	Upper Bound
Artificial Saliva (Control)	Beer	-0.014	0.314	-0.033	0.006
	Red wine	-0.195	< **0.0001**	-0.215	-0.176
	Whiskey	-0.153	< **0.0001**	-0.173	-0.134
	Vodka	-0.291	< **0.0001**	-0.312	-0.272
Beer	Red wine	-0.182	< **0.0001**	-0.201	-0.162
	Whiskey	-0.139	< **0.0001**	-0.160	-0.120
	Vodka	-0.278	< **0.0001**	-0.298	-0.259
Red wine	Whiskey	0.042	< **0.0001**	0.022	0.062
	Vodka	-0.097	< **0.0001**	-0.116	-0.077
Whiskey	Vodka	-0.138	< **0.0001**	-0.158	-0.119

*Obtained using Tukey's post-hoc test; Bold p-values indicate statistical significance

Table 7: Descriptive statistics for parameters at day 0 and day 30 for samples exposed to different treatments

Group	Parameter	Day 0			Day 30		
		L	a	b	L	a	b
Artificial Saliva (Control)	Mean	70.465	2.079	13.401	70.513	2.110	13.819
	SD	1.653	0.403	0.973	1.639	0.396	1.762
	Minimum	67.253	1.067	10.996	67.250	1.112	11.012
	Maximum	73.112	3.252	14.917	73.099	3.262	21.043
Beer	Mean	67.681	1.231	11.623	66.220	2.003	12.636
	SD	1.961	0.592	1.651	1.998	0.682	1.722
	Minimum	64.781	0.282	7.895	63.198	1.102	9.012
	Maximum	72.050	2.351	14.819	70.091	3.090	15.239
Red wine	Mean	68.129	1.217	11.214	66.043	2.292	13.731
	SD	1.999	0.502	0.000	1.654	0.777	1.597
	Minimum	64.991	0.317	11.214	62.863	0.851	10.936
	Maximum	72.315	2.619	11.214	69.997	3.819	16.896
Whiskey	Mean	69.451	1.577	12.557	68.028	2.540	14.331
	SD	2.721	0.443	1.251	2.849	0.563	1.272
	Minimum	62.013	0.823	10.510	60.310	1.543	11.671
	Maximum	73.110	2.319	14.316	71.981	3.301	16.319
Vodka	Mean	68.400	0.984	11.379	67.728	1.477	12.726
	SD	1.778	0.329	1.379	1.712	0.478	1.131
	Minimum	64.781	0.282	7.895	63.993	0.430	11.097
	Maximum	71.321	1.632	14.363	70.104	2.767	15.432

Table 8: Descriptive statistics for colour change of samples exposed to five different treatments

Parameters	Exposure group				
	Artificial Saliva (Control)	Beer	Red wine	Whiskey	Vodka
N	26	26	26	26	26
Mean	1.415	2.084	3.694	2.535	1.712
Standard Deviation	1.910	0.731	1.356	0.545	1.013
Median	0.842	1.981	3.697	2.390	1.405
Minimum	0.021	0.715	1.187	1.839	0.453
Maximum	9.060	3.671	6.395	3.681	5.117

P-value < 0.0001 (Significant) using one-way analysis of variance

Table 9: Pairwise comparison of colour change in samples exposed to five different treatments

Group		Mean Difference (I-J)	P-value	95% Confidence Interval	
(I)	(J)			Lower Bound	Upper Bound
Artificial Saliva (Control)	Beer	-0.669	0.277	-1.599	0.262
	Red wine	-2.278	< **0.0001**	-3.209	-1.348
	Whiskey	-1.119	**0.010**	-2.050	-0.189
	Vodka	-0.296	0.903	-1.227	0.634
Beer	Red wine	-1.609	< **0.0001**	-2.540	-0.679
	Whiskey	-0.450	0.667	-1.381	0.480
	Vodka	0.372	0.803	-0.559	1.303
Red wine	Whiskey	1.159	**0.007**	0.229	2.090
	Vodka	1.981	< **0.0001**	1.051	2.912
Whiskey	Vodka	0.822	0.110	-0.108	1.753

*Obtained using Tukey's post-hoc test; Bold p-values indicate statistical significance

GRAPHS

Figure1: Column chart showing mean surface roughness at day0 and day30 for different exposures

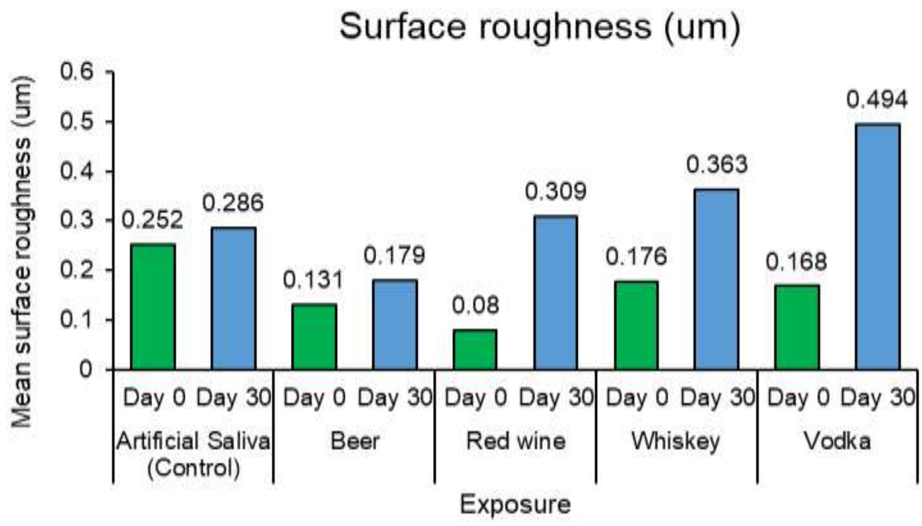


Figure 2: Column chart showing mean surface roughness at day 30 for samples in different exposure groups

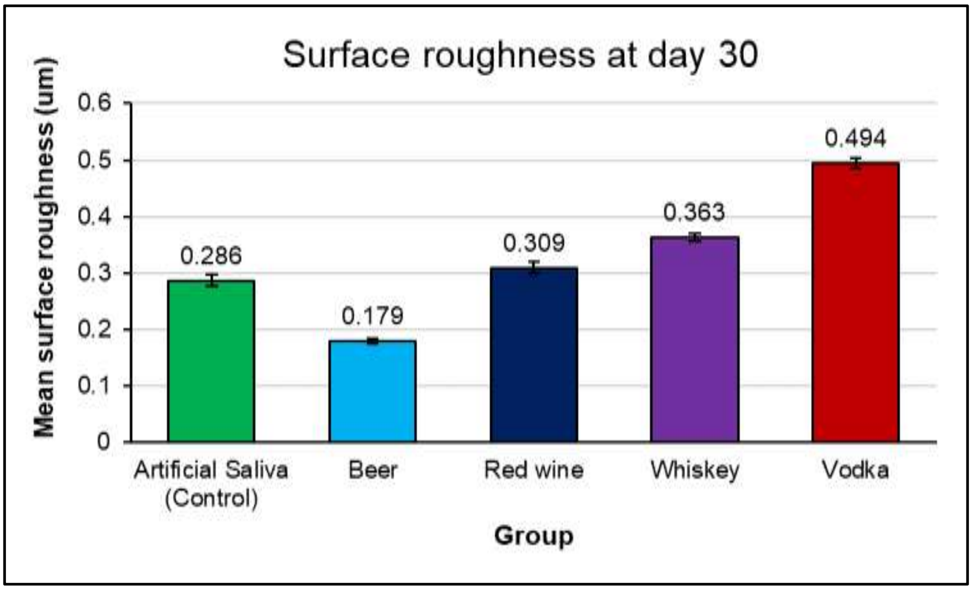


Figure 3: Column chart showing mean change in surface roughness for samples in different exposure groups

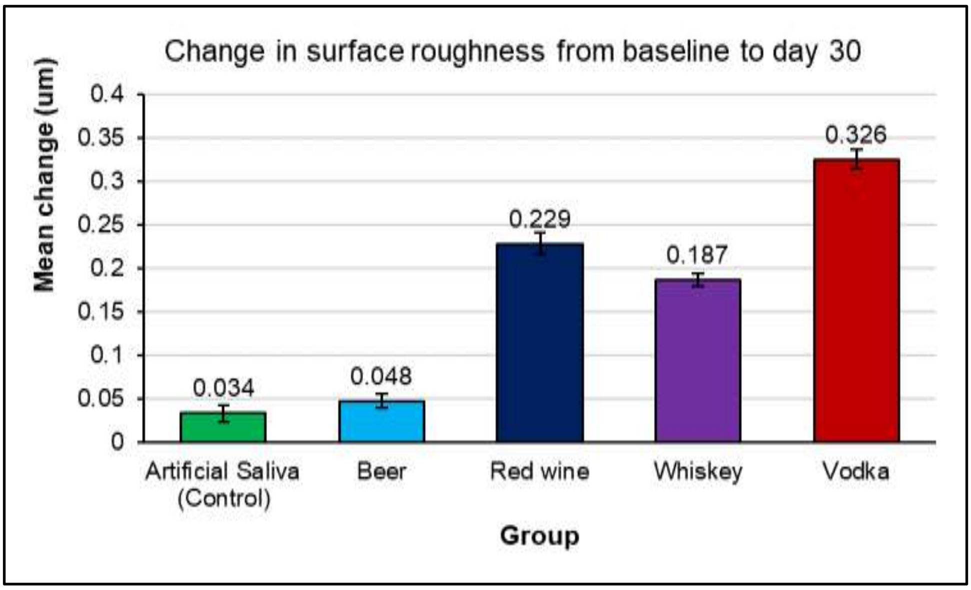


Figure 4: Column chart showing mean colour change for samples in different exposure groups.

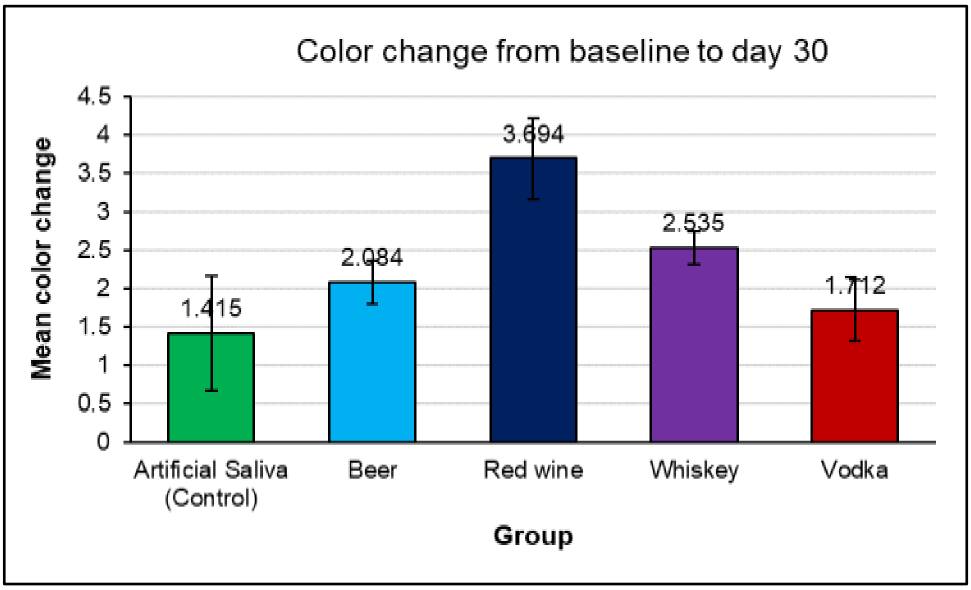
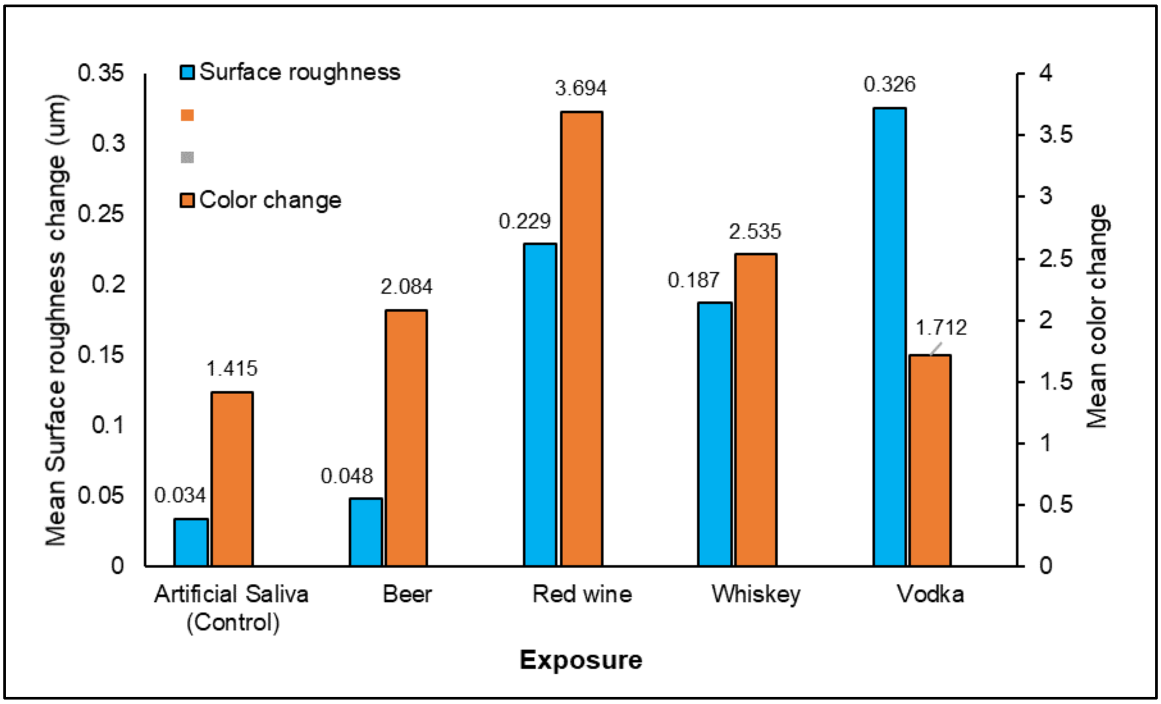


Figure 5: Column chart showing mean surface roughness change and mean colour change for different exposures


Surface roughness
Color change
0.35
0.3
0.25
0.2
0.15
0.1
0.05
0
Mean Surface roughness change (um)
4
3.5
3
2.5
2
1.5
1
0.5
0
Mean color change
0.034
1.415
0.048
2.084
0.229
3.694
0.187
2.535
0.326
1.712
Artificial Saliva (Control)
Beer
Red wine
Whiskey
Vodka
Exposure

ANNEXURE I

Surface Roughness in Group I – Artificial saliva

Sample No	Baseline Value At 0 Day µm	Final value After 30 Days µm
1	0.261 µm	0.305 µm
2	0.248 µm	0.274 µm
3	0.253 µm	0.297 µm
4	0.269 µm	0.303 µm
5	0.281 µm	0.209 µm
6	0.252 µm	0.276 µm
7	0.236 µm	0.261 µm
8	0.248 µm	0.285 µm
9	0.268 µm	0.292 µm
10	0.235 µm	0.278 µm
11	0.286 µm	0.309 µm
12	0.231 µm	0.273 µm
13	0.241 µm	0.288 µm
14	0.279 µm	0.315 µm
15	0.234 µm	0.271 µm
16	0.298 µm	0.339 µm
17	0.221 µm	0.265 µm
18	0.241 µm	0.286 µm
19	0.292 µm	0.319 µm
20	0.206 µm	0.253 µm
21	0.215 µm	0.268 µm
22	0.246 µm	0.281 µm
23	0.231 µm	0.277 µm
24	0.271 µm	0.329 µm
25	0.259 µm	0.294 µm
26	0.244 µm	0.283 µm
Average	0.252 µm	0.286 µm

ANNEXURE II

Surface Roughness in Group II – Beer

Sample No	Baseline Value At 0 Day µm	Final value After 30 Days µm
1	0.14 µm	0.173 µm
2	0.113 µm	0.199 µm
3	0.124 µm	0.166 µm
4	0.13 µm	0.184 µm
5	0.125 µm	0.178 µm
6	0.138 µm	0.165 µm
7	0.101 µm	0.187 µm
8	0.123 µm	0.196 µm
9	0.129 µm	0.155 µm
10	0.148 µm	0.176 µm
11	0.14 µm	0.192 µm
12	0.126 µm	0.179 µm
13	0.172 µm	0.19 µm
14	0.151 µm	0.177 µm
15	0.125 µm	0.189 µm
16	0.12 µm	0.165 µm
17	0.132 µm	0.191 µm
18	0.157 µm	0.162 µm
19	0.143 µm	0.175 µm
20	0.113 µm	0.159 µm
21	0.128 µm	0.164 µm
22	0.12 µm	0.186 µm
23	0.146 µm	0.19 µm
24	0.122 µm	0.174 µm
25	0.131 µm	0.185 µm
26	0.115 µm	0.193 µm
Average	0.131 µm	0.179 µm

ANNEXURE III

Surface Roughness in Group III – Red Wine

Sample No	Baseline Value At 0 Day µm	Final value After 30 Days µm
1	0.124 µm	0.329 µm
2	0.169 µm	0.361 µm
3	0.175 µm	0.345 µm
4	0.186 µm	0.383 µm
5	0.147 µm	0.348 µm
6	0.165 µm	0.354 µm
7	0.179 µm	0.378 µm
8	0.162 µm	0.362 µm
9	0.185 µm	0.355 µm
10	0.173 µm	0.375 µm
11	0.149 µm	0.349 µm
12	0.195 µm	0.395 µm
13	0.163 µm	0.364 µm
14	0.186 µm	0.389 µm
15	0.214 µm	0.339 µm
16	0.191 µm	0.39 µm
17	0.18 µm	0.381 µm
18	0.187 µm	0.362 µm
19	0.177 µm	0.346 µm
20	0.167 µm	0.331 µm
21	0.178 µm	0.369 µm
22	0.181 µm	0.344 µm
23	0.193 µm	0.393 µm
24	0.166 µm	0.367 µm
25	0.201 µm	0.351 µm
26	0.17 µm	0.377 µm
Average	0.175 µm	0.363 µm

ANNEXURE IV

Surface Roughness in Group IV – whiskey

Sample No	Baseline Value At 0 Day µm	Final value After 30 Days µm
1	0.074 µm	0.274 µm
2	0.08 µm	0.33 µm
3	0.072 µm	0.342 µm
4	0.068 µm	0.299 µm
5	0.09 µm	0.293 µm
6	0.048 µm	0.328 µm
7	0.086 µm	0.286 µm
8	0.063 µm	0.313 µm
9	0.082 µm	0.332 µm
10	0.079 µm	0.329 µm
11	0.095 µm	0.295 µm
12	0.07 µm	0.375 µm
13	0.076 µm	0.346 µm
14	0.089 µm	0.319 µm
15	0.073 µm	0.323 µm
16	0.094 µm	0.297 µm
17	0.086 µm	0.286 µm
18	0.079 µm	0.339 µm
19	0.075 µm	0.275 µm
20	0.098 µm	0.298 µm
21	0.083 µm	0.303 µm
22	0.072 µm	0.272 µm
23	0.099 µm	0.289 µm
24	0.085 µm	0.285 µm
25	0.074 µm	0.324 µm
26	0.078 µm	0.278 µm
Average	0.079 µm	0.309 µm

ANNEXURE V

Surface Roughness in Group V – Vodka

Sample No	Baseline Value At 0 Day µm	Final value After 30 Days µm
1	0.187 µm	0.487 µm
2	0.181 µm	0.535 µm
3	0.172 µm	0.472 µm
4	0.168 µm	0.468 µm
5	0.19 µm	0.51 µm
6	0.148 µm	0.478 µm
7	0.186 µm	0.486 µm
8	0.163 µm	0.503 µm
9	0.182 µm	0.512 µm
10	0.149 µm	0.449 µm
11	0.165 µm	0.525 µm
12	0.17 µm	0.47 µm
13	0.179 µm	0.509 µm
14	0.172 µm	0.492 µm
15	0.169 µm	0.459 µm
16	0.185 µm	0.515 µm
17	0.176 µm	0.476 µm
18	0.167 µm	0.507 µm
19	0.155 µm	0.521 µm
20	0.177 µm	0.477 µm
21	0.162 µm	0.502 µm
22	0.115 µm	0.519 µm
23	0.183 µm	0.483 µm
24	0.149 µm	0.526 µm
25	0.165 µm	0.465 µm
26	0.159 µm	0.501 µm
Average	0.168 µm	0.494 µm

ANNEXURE VI

Colour Stability in Group I – Artificial Saliva

Sample No.	Baseline Value At 0 Day			Final value After 30 Days			ΔL	Δa	Δb	ΔE
	L0	a 0	b 0	L30	a30	b30				
1	69.587	1.918	13.887	69.581	1.928	13.991	-0.006	0.01	0.104	0.1046518
2	72.01	3.252	14.037	72.001	3.262	14.131	-0.009	0.01	0.094	0.09495789
3	67.499	1.978	14.065	67.491	1.981	14.099	-0.008	0.003	0.034	0.0350571
4	67.253	1.988	14.1933	67.25	1.997	14.218	-0.003	0.009	0.0247	0.02645921
5	69.991	2.899	12.751	69.987	2.912	12.783	-0.004	0.013	0.032	0.03477068
6	70.582	2.441	13.784	70.577	2.456	13.798	-0.005	0.015	0.014	0.02111871
7	68.003	1.964	10.996	67.994	1.984	11.012	-0.009	0.02	0.016	0.02714774
8	71.02	2.411	14.318	71.013	2.432	14.411	-0.007	0.021	0.093	0.09559812
9	70.183	1.945	12.007	69.531	1.963	21.043	-0.652	0.018	9.036	9.05951014
10	69.538	1.939	14.539	68.96	1.951	14.581	-0.578	0.012	0.042	0.57964817
11	68.964	1.901	12.361	72.103	1.98	12.391	3.139	0.079	0.03	3.14013726
12	72.118	2.029	13.945	69.475	2.089	13.984	-2.643	0.06	0.039	2.64396861
13	69.482	1.926	11.799	70.617	1.983	11.833	1.135	0.057	0.034	1.13693887
14	70.628	2.189	14.917	72.32	2.201	14.991	1.692	0.012	0.074	1.69365994
15	72.331	2.19	11.995	70.67	2.232	11.999	-1.661	0.042	0.004	1.66153574
16	70.687	1.92	12.999	67.66	1.963	13.101	-3.027	0.043	0.102	3.02902327
17	67.692	1.067	13.567	71.101	1.112	13.585	3.409	0.045	0.018	3.40934451
18	70.112	1.897	13.73	72.545	1.921	13.743	2.433	0.024	0.013	2.4331531
19	72.558	1.89	13.699	70.93	1.899	13.701	-1.628	0.009	0.002	1.62802611
20	70.931	1.899	13.384	71.461	1.913	13.411	0.53	0.014	0.027	0.53087192
21	71.472	2.598	13.803	70.042	2.62	13.83	-1.43	0.022	0.027	1.43042406
22	70.05	2.182	12.201	71.998	2.221	12.211	1.948	0.039	0.01	1.94841602
23	72.001	1.771	13.478	71.728	1.813	13.499	-0.273	0.042	0.021	0.27700903
24	71.738	1.909	13.879	71.099	1.952	13.901	-0.639	0.043	0.022	0.64082291
25	73.112	2.048	14.392	73.099	2.11	14.423	-0.013	0.062	0.031	0.07052659
26	72.558	1.89	13.699	72.102	1.997	14.632	-0.456	0.107	0.933	1.04397031
Average	70.46538	2.0785	13.401	70.5129	2.110462	13.819	0.0475	0.032	0.41833	1.41525953

ANNEXURE VII

Colour Stability in Group II– Beer

Sample No.	Baseline Value At 0 Day			Final value After 30 Days			ΔL	Δa	Δb	ΔE
	L0	a 0	b 0	L30	a30	b30				
1	66.427	0.288	10.094	63.198	1.102	11.638	-3.229	0.814	1.544	3.67055486
2	66.451	0.809	11.804	63.895	1.204	12.012	-2.556	0.395	0.208	2.5946917
3	66.859	0.652	11.218	63.678	1.651	12.09	-3.181	0.999	0.872	3.44632355
4	70.022	0.606	13.104	67.645	1.248	13.581	-2.377	0.642	0.477	2.50795175
5	64.781	0.955	8.733	63.518	1.33	9.329	-1.263	0.375	0.596	1.4460325
6	65.999	0.282	10.842	64.319	1.562	11.54	-1.68	1.28	0.698	2.22441093
7	66.262	0.575	7.895	66.144	1.254	9.012	-0.118	0.679	1.117	1.31249914
8	67.11	1.31	9.991	66.991	2.001	10.132	-0.119	0.691	0.141	0.71520836
9	68.91	1.419	11.813	67.321	2.091	13.918	-1.589	0.672	2.105	2.72167779
10	69.21	2.001	13.191	68.101	2.401	14.789	-1.109	0.4	1.598	1.98582099
11	69.232	1.189	11.319	67.99	1.299	12.384	-1.242	0.11	1.065	1.63978322
12	65.138	0.993	11.819	64.998	1.531	12.561	-0.14	0.538	0.742	0.92715047
13	66.323	0.981	10.139	64.321	1.437	11.562	-2.002	0.456	1.423	2.49817313
14	66.913	1.931	10.991	65.819	2.321	11.432	-1.094	0.39	0.441	1.24234335
15	69.139	1.413	11.231	68.101	2.918	11.979	-1.038	1.505	0.748	1.97534124
16	72.05	1.321	14.156	70.091	2.871	15.239	-1.959	1.55	1.083	2.72269536
17	64.999	2.013	14.321	63.878	3.018	15.132	-1.121	1.005	0.811	1.71008392
18	67.899	1.813	11.917	65.645	3.09	12.678	-2.254	1.277	0.761	2.70006778
19	66.319	0.918	10.999	65.001	1.561	12.119	-1.318	0.643	1.12	1.84525689
20	68.139	0.701	11.543	66.918	1.998	12.981	-1.221	1.297	1.438	2.28929989
21	65.328	0.831	13.513	64.831	1.519	14.99	-0.497	0.688	1.477	1.70349112
22	67.139	2.351	12.219	65.983	3.012	13.318	-1.156	0.661	1.099	1.72657406
23	69.692	1.917	10.139	66.987	2.876	11.329	-2.705	0.959	1.19	3.10689652
24	68.135	1.819	11.377	67.329	2.319	13.319	-0.806	0.5	1.942	2.16124964
25	70.23	2.01	14.819	69.101	2.854	15.105	-1.129	0.844	0.286	1.43832298
26	71.005	0.901	13.019	69.918	1.621	14.369	-1.087	0.72	1.35	1.87682418
Average	67.68119	1.23073	11.6233	66.22	2.003423	12.636	-1.4611	0.7727	1.01277	2.08418174

ANNEXURE VIII

Colour Stability in Group III – Red Wine

Sample No.	Baseline Value At 0 Day			Final value After 30 Days			ΔL	Δa	Δb	ΔE
	L0	a 0	b 0	L30	a30	b30				
1	66.002	0.754	11.214	63.761	0.851	10.936	-2.241	0.097	-0.278	2.26025972
2	65.575	0.491	11.214	65.191	1.154	12.402	-0.384	0.663	1.188	1.4136368
3	69.158	0.926	11.214	67.994	1.003	10.993	-1.164	0.077	-0.221	1.18729356
4	66.79	0.317	11.214	64.788	1.047	14.749	-2.002	0.73	3.535	4.12760572
5	70.198	0.993	11.214	66.019	1.679	15.691	-4.179	0.686	4.477	6.16264278
6	71.319	1.31	11.214	69.386	2.54	15.399	-1.933	1.23	4.185	4.77112293
7	66.639	1.691	11.214	65.713	2.997	13.01	-0.926	1.306	1.796	2.40597756
8	69.143	0.932	11.214	66.31	1.876	12.819	-2.833	0.944	1.605	3.39014012
9	70.831	0.819	11.214	68.197	2.01	14.999	-2.634	1.191	3.785	4.76263184
10	68.513	0.764	11.214	67.319	1.989	14.101	-1.194	1.225	2.887	3.35574582
11	65.169	1.321	11.214	63.669	2.923	13.21	-1.5	1.602	1.996	2.96655018
12	64.991	1.908	11.214	62.863	3.003	12.069	-2.128	1.095	0.855	2.54134492
13	68.319	1.354	11.214	65.997	2.876	15.005	-2.322	1.522	3.791	4.69891998
14	69.96	1.213	11.214	66.319	2.769	12.819	-3.641	1.556	1.605	4.27247493
15	69.997	0.998	11.214	67.001	3.11	11.667	-2.996	2.112	0.453	3.69347655
16	66.697	0.897	11.214	65.103	1.765	13.697	-1.594	0.868	2.483	3.07563798
17	67.328	1.113	11.214	65.916	2.13	14.919	-1.412	1.017	3.705	4.09329427
18	69.946	1.678	11.214	66.319	2.316	14.762	-3.627	0.638	3.548	5.11375371
19	66.713	1.532	11.214	65.321	2.106	12.857	-1.392	0.574	1.643	2.22858453
20	68.321	2.619	11.214	66.001	3.151	11.992	-2.32	0.532	0.778	2.50413818
21	66.561	1.131	11.214	65.101	2.643	13.102	-1.46	1.512	1.888	2.82529432
22	68.103	0.954	11.214	67.006	1.545	14.699	-1.097	0.591	3.485	3.70106944
23	66.324	1.218	11.214	65.201	2.813	15.187	-1.123	1.595	3.973	4.42604598
24	69.654	0.997	11.214	66.306	2.31	12.897	-3.348	1.313	1.683	3.97058711
25	66.785	1.691	11.214	64.316	3.156	16.132	-2.469	1.465	4.918	5.69463871
26	72.315	2.019	11.214	69.997	3.819	16.896	-2.318	1.8	5.682	6.39517381
Average	68.12888	1.21692	11.214	66.0428	2.291577	13.731	-2.0860	1.0747	2.51712	3.69377082

ANNEXURE IX

Colour Stability in Group IV – Whiskey

Sample No.	Baseline Value At 0 Day			Final value After 30 Days			ΔL	Δa	Δb	ΔE
	L0	**a 0**	**b 0**	**L30**	**a30**	**b30**				
1	69.589	1.316	14.033	68.135	1.998	16.319	-1.454	0.682	2.286	2.79374945
2	62.013	1.157	14.065	60.532	1.897	15.365	-1.481	0.74	1.3	2.1049848
3	67.499	1.59	14.193	66.231	2.301	15.32	-1.268	0.711	1.127	1.83942219
4	69.819	0.823	12.751	68.234	1.791	14.378	-1.585	0.968	1.627	2.46908445
5	68.003	1.132	13.784	66.897	2.432	15.654	-1.106	1.3	1.87	2.53182464
6	71.02	1.869	11.456	70.31	3.21	14.81	-0.71	1.341	3.354	3.68126296
7	69.538	2.319	14.316	68.21	3.301	15.676	-1.328	0.982	1.36	2.13951116
8	72.118	0.996	10.91	70.879	1.767	12.346	-1.239	0.771	1.436	2.0473539
9	70.628	2.001	11.319	69.34	3.234	12.105	-1.288	1.233	0.786	1.94859667
10	69.554	1.876	10.51	68.432	2.894	11.671	-1.122	1.018	1.161	1.90869825
11	62.819	1.548	13.564	60.31	2.99	15.73	-2.509	1.442	2.166	3.61469238
12	67.692	1.329	12.997	66.543	2.547	14.456	-1.149	1.218	1.459	2.22090207
13	72.543	2.087	11.132	69.993	3.086	13.001	-2.55	0.999	1.869	3.31566916
14	70.931	0.998	12.001	69.943	1.717	14.569	-0.988	0.719	2.568	2.84389328
15	70.183	2.098	11.819	69.154	3.161	14.631	-1.029	1.063	2.812	3.17744457
16	72.543	1.348	13.013	71.32	2.418	15.131	-1.223	1.07	2.118	2.66956045
17	70.931	1.569	10.997	69.148	2.143	13.887	-1.783	0.574	2.89	3.44393162
18	70.738	1.783	13.108	69.32	2.878	14.343	-1.418	1.095	1.235	2.17599954
19	73.11	1.908	14.113	71.981	2.997	15.81	-1.129	1.089	1.697	2.31092427
20	67.692	1.367	12.719	65.719	2.476	14.569	-1.973	1.109	1.85	2.92320201
21	72.558	1.897	11.949	71.349	2.679	13.369	-1.209	0.782	1.42	2.02227718
22	69.991	1.867	13.341	68.543	2.99	14.71	-1.448	1.123	1.369	2.28735524
23	68.861	2.319	14.181	67.32	3.139	15.432	-1.541	0.82	1.251	2.14757584
24	66.543	0.987	11.21	65.431	1.543	13.469	-1.112	0.556	2.259	2.57851915
25	67.869	1.831	10.989	66.523	2.731	12.31	-1.346	0.9	1.321	2.08967868
26	70.931	0.998	12.001	68.943	1.717	13.543	-1.988	0.719	1.542	2.61665225
Average	69.45062	1.57742	12.5566	68.0285	2.539885	14.331	-1.4221	0.9625	1.77435	2.53472178

ANNEXURE X

Colour Stability in Group V – Vodka

Sample No.	Baseline Value At 0 Day			Final value After 30 Days			ΔL	Δa	Δb	ΔE
	L0	**a 0**	**b 0**	**L30**	**a30**	**b30**				
1	66.427	0.288	10.094	66.201	0.43	11.254	-0.226	0.142	1.16	1.19031088
2	67.451	0.809	11.804	67.205	1.13	12.009	-0.246	0.321	0.205	0.45341151
3	66.845	0.652	11.21	65.947	1.004	12.132	-0.898	0.352	0.922	1.33431331
4	70.022	0.606	13.104	69.873	0.99	13.989	-0.149	0.384	0.885	0.97615675
5	64.781	0.955	8.733	63.993	1.23	11.731	-0.788	0.275	2.998	3.11200466
6	65.999	0.282	10.842	64.839	0.63	11.942	-1.16	0.348	1.1	1.63606357
7	66.262	0.575	7.895	65.89	0.99	12.982	-0.372	0.415	5.087	5.11743862
8	69.732	0.979	12.304	69.32	1.32	12.705	-0.412	0.341	0.401	0.66845045
9	68.453	1.101	11.413	68.002	1.301	12.673	-0.451	0.2	1.26	1.35314486
10	67.321	1.32	11.693	66.987	1.73	12.32	-0.334	0.41	0.627	0.82023472
11	66.991	1.632	12.301	66.543	1.903	14.979	-0.448	0.271	2.678	2.72870464
12	68.351	0.876	13.376	67.83	1.34	13.987	-0.521	0.464	0.611	0.92739312
13	69.982	1.31	10.254	69.34	1.789	11.543	-0.642	0.479	1.289	1.51760535
14	70.413	1.038	11.693	69.987	1.679	12.53	-0.426	0.641	0.837	1.13706904
15	71.321	0.899	12.701	70.104	1.53	13.305	-1.217	0.631	0.604	1.49802069
16	66.87	1.32	10.321	66.32	1.23	11.432	-0.55	-0.09	1.111	1.24294851
17	69.543	0.994	11.432	69.004	1.458	12.642	-0.539	0.464	1.21	1.40353732
18	68.376	1.23	9.976	68.004	1.89	11.097	-0.372	0.66	1.121	1.35300591
19	67.321	0.88	11.543	66.732	2.767	13.432	-0.589	1.887	1.889	2.73422951
20	69.831	1.154	12.532	69.304	1.758	13.987	-0.527	0.604	1.455	1.66119535
21	70.462	0.764	14.363	69.32	1.79	15.432	-1.142	1.026	1.069	1.87072205
22	67.354	1.41	10.79	66.43	1.701	11.809	-0.924	0.291	1.019	1.4059936
23	71.043	1.362	11.432	67.889	1.946	12.676	-3.154	0.584	1.244	3.44039358
24	69.543	1.104	10.693	69.104	1.876	11.783	-0.439	0.772	1.09	1.40598898
25	67.305	1.005	11.67	66.769	1.32	13.979	-0.536	0.315	2.309	2.39123441
26	70.413	1.038	11.693	69.987	1.679	12.53	-0.426	0.641	0.837	1.13706904
Average	68.40046	0.98396	11.3793	67.7278	1.477346	12.726	-0.67262	0.4934	1.34685	1.71217848