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Susceptibility of repigmentation of enamel surface to red wine and brandy staining after 35% hydrogen peroxide bleaching

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Abstract
Aim and objectives: To investigate the susceptibility of enamel surface for repigmentation by red wine and brandy stains after 35% hydrogen peroxide bleaching.
Methodology: 40 freshly extracted human permanent maxillary central incisor teeth were collected for the study. Incisors were stored in 0.1% thymol solution and were used within 1 month of extraction. The teeth were cleaned with ultrasonic scaler, polished with pumice paste. The samples were than divided into two groups of 20 incisors teeth each. Both groups of 20 teeth each were than bleached using 35% hydrogen Peroxide as per the manufacturer's instructions, i.e. 15 minutes three times a day, every alternate day for four days. Group 1 were than immersed in 200 ml of red wine and group 2 were immersed in 200 ml of brandy for 15 minutes, 6 hours, 1 week and 1 month each. Advance spectrophotometer was used to measure the color shade of the teeth pre bleaching, after 15 minutes, after 6 hours, after 1 week and after 1 month of red wine and brandy immersion. The colour differences (ΔE) at each time interval were calculated accordingly.
Results: Spectrometer analysis showed mean ΔE values ± standard deviation of group 1 and group 2 at different time intervals. Color change due to repigmentation was lowest for group 1 teeth from baseline to 15 minutes time interval. Mean ΔE value ± standard deviation was 4.04045±1.308029. However color change value was highest in group 2 teeth at 1 week to 1 month time interval and was 13.45717±1.098210. Group 2 teeth had higher ΔE value than group 1 at any point of time interval.
Conclusions: It was concluded that low pH values causes more repigmentation of bleached enamel surface as it was found more with brandy in comparison to red wine. The bleaching treatment with 35% hydrogen peroxide enabled the teeth to become clearer; nevertheless, the intake of food and beverages containing colorants or an acidic pH can negatively affect the effectiveness of bleaching.

Keywords: Repigmentation, Brandy, Red wine, Spectrometer, Hydrogen Peroxide, Bleaching

1. Introduction
One of the main reasons for which patient seek esthetic dental treatment is for real or perceived discoloration of anterior teeth. Smiling is one of the most important factors in social relationships. Nowadays the desire to have white teeth and a pleasant smile is an important esthetic need of patients. White teeth increase self-confidence and improve social eminence of people. One of the greatest assets a person now can have is smile that shows beautiful and natural teeth. Improving the patient's smile often contribute to an improved self-image and enhanced self-esteem [1]. Esthetics by definition is the science of beauty, it is derived from the greek word 'perception' which deals with beauty as an art. In the modern civilized cosmetically conscious world well-contoured and well-aligned white teeth, set the standard for beauty. The goal in the creation of the esthetic dental restorations is to stimulate, or improve upon, the appearance of the natural dentition. The successful esthetic restorations must integrate harmoniously with the face, not just with the surrounding teeth. Such teeth are not only considered attractive, but also indicative of nutritional health, self-esteem, hygienic pride and economic status [2].
There are several methods available to treat discolored dentitions such as laminates, porcelain jacket crowns and bleaching, the advantages of laminates are they are aesthetically good, typically two appointment process but disadvantages are like it involves removal of very

minimal enamel, but is irreversible. Currently bleaching as a treatment modality, continues to hold its century old place as the simplest, most common, least invasive and least expensive means to lighten discolored teeth. Nowadays, dental bleaching is one of the most popular esthetic treatments in dentistry due to its fast satisfactory results and is safe when performed by a trained professional [3]. Tooth discoloration varies in aetiology, appearance, localization, severity, and adherence to tooth structure. It may be classified as intrinsic, extrinsic and a third category of 'Stain internalization ' has recently been described to include those circumstances where extrinsic stain enters the tooth through defects in the tooth structure. Intrinsic discoloration is caused by incorporation of chromogenic material into dentin and enamel during odontogenesis or after eruption. Exposure to high levels of fluoride, tetracycline administration, inherited developmental disorders, and trauma to the developing tooth may result in pre-eruptive discoloration. Extrinsic discoloration arises when external chromogens are deposited on the tooth surface or within the pellicle layer [4].

The technique of bleaching or whitening of teeth was first described in 1877 by Chapple. It was in the year 1916 that Dr. Walter Kaine used hydrochloric acid to successfully remove the fluorosis stains. In 1918, Abbot pioneered the dental effects of superoxol and found that chemical was suitable for bleaching teeth, its action could be greatly enhanced by the addition of heat and light. In the year 1937 Ames reported an alternative for removing fluorosis using hydrogen peroxide instead of hydrochloric acid. Later it was McInnes who reported a technique where hydrogen peroxide, hydrochloric acid and ethyl ether were used. This technique has been found to be successful for bleaching the teeth of patients with endemic fluorosis. In 1965 Stewart introduced thermocatalytic technique in which Pellet saturated with superoxyl was inserted into pulp chamber and heated with hot instrument for non-vital teeth. In1989 Haywood and Heyman developed Night-guard Vital Bleaching using 10% carbamide peroxide in a tray for all stains of vital and non-vital teeth. Reyto in 1996 developed Laser tooth whitening for vital teeth. Present day we have Plasma arc and light activated bleaching techniques, Power gels for-in-office bleaching, Laser activated bleaching and home bleaching available in different concentrations and flavours [5].

The development of various bleaching agents which uses hydrogen peroxide in high concentrations (35% to 45%) has made in-office bleaching procedures easier. The advantage of this technique is the favorable immediate results achieved without the need for further patient cooperation. The efficacy of bleaching agents is validated by *in vitro* and *in vivo* studies. But the adverse effects to dental hard tissues must be carefully evaluated in order to use them safely [6]. A study conducted by Sulieman M *et al*. concluded that the bleaching gel with a higher hydrogen peroxide concentration needed fewer applications to produce bleaching effect and the relationship between peroxide concentration and the number of applications was not linear but exponential [7].

A major drawback of using bleaching agents is its direct effect on the organic content of the tooth. Dentists have advised patients against drinking some beverages and smoking particularly after bleaching session with 35% hydrogen peroxide, since some studies have reported superficial enamel alterations promoted by bleaching products. Hydrogen peroxide can promote varying/assorted degrees of surface porosity, structural and permeability changes, depending on the bleaching agent [8-11]. Changes on enamel surface would be attribute to oxidative and demineralization processes produced by the hydrogen peroxide agents [12-14].

Coffee, tea, juices, wines and cola-based soft drinks are potential dark or coloring beverages, which could stain or discolor the bleached enamel surface. Some of them are acidic solutions that can increase the demineralization, while others contain ethanol and/or pigments. In addition, certain beverages, artificial food colorations and smoking used with a high frequency are considered responsible for primary staining, dark and discoloration of teeth [15, 16]. As already mentioned that regardless of the bleaching protocol employed, clinicians often ask patients to stop drinking coffee, red wine, coke, or any type of pigmented foods and beverages. This caution has been taken as some *in vitro* studies have suggested that the concomitant exposure of this colored diet may jeopardize bleaching efficacy [17-20]. On the other hand, the available clinical trials do not support this hypothesis. Indeed, it was shown that exposure to coffee and tea during bleaching does not have a negative impact on color change [21, 22]. Also, the consumption of coke during in-office bleaching treatment did not affect color change [23] hence, pigment containing diets may not have any detrimental effects on color change [24].

However, it is uncertain if other types of colored diet components can affect bleaching results. Brandy and Red wine are alcoholic drinks in which red wine is considered as soft liquor whereas, brandy is considered as hard liquor and is made by distillation in comparison to red wine which is made by fermentation. Red wine is an acidic drink largely consumed worldwide with lower alcohol content approximately 9-12% in comparison to brandy which has a higher alcohol content of 35-60%. Till date no study suggests the effect of brandy in pigmentation of bleached enamel whereas there are number of researches which studied red wine effect on bleached enamel. As we know that red wine gained popularity after the identification of anti-inflammatory and antioxidant compounds, such as polyphenols, in its composition [25]. These are suggested to protect against cardiovascular and inflammatory bowel diseases [26, 27]. Red wine extracts were also found to have antimicrobial activities against periodontal pathogens [28] and to decrease macro phage-mediated inflammatory responses [29, 30]. Interestingly, it was demonstrated that tooth bleaching agents can induce crevicular inflammation and damage, a response largely associated with nitric oxide (NO) synthesis and leukocyte activation [31].

Therefore, the purpose of this study was to evaluate the influence of 35% hydrogen peroxide bleaching agents on the enamel surface susceptibility to red wine and brandy staining.

Materials and Methods

Preparation of Specimen

40 freshly extracted human permanent maxillary central incisor teeth were collected for the study. Incisors were stored in 0.1% thymol solution and were used within 1 month of extraction. Dental calculus and periodontal membrane remnants were removed using hand tools. The teeth were cleaned with ultrasonic scaler, polished with pumice paste, and then stored in saline. The apical foramina were sealed with glass ionomer cement. (GC Fuji) The roots were sectioned from the dentino-enamel junction using a diamond disc. Each specimen, with the labial surface exposed, were individually submerged in chemically cured acrylic resin moulds, through which light passes. After preparation, the

samples were polished, using a prophylaxis paste administered via a polishing brush and washed. The samples were than divided into two groups of 20 incisors teeth each.

Group 1.
20 incisor teeth were bleached using 35% hydrogen Peroxide as per the manufacturer's instructions, i.e. 15 minutes three times a day, every alternate day for four days and then immersed in 200 ml of red wine for 15 minutes, 6 hours, 1 week and 1 month.

Group 2.
20 incisors teeth were bleached using 35% hydrogen Peroxide as per the manufacturer's instructions, i.e. 15 minutes three times a day, every alternate day for four days and then immersed in 200 ml of brandy for 15 minutes, 6 hours, 1 week and 1 month.

Inclusion criteria
Non carious, sound mature incisor teeth with no anomalies were selected for this study.

Exclusion criteria
Teeth with following criteria were not included in the study.
1. Fractured teeth
2. Grossly destructed teeth
3. Teeth with non-carious lesion
4. Teeth with fluorosis
5. Previously restored teeth

Procedure
Removal of external residual tissues: The collected teeth were cleaned of any visible debris and calculus using hand scalers mechanically, following extraction and stored in 0.1% thymol solution and were used within 1 month of extraction.

Sample Preparation: Decoronation of the teeth using the carborundum disc (Fig: 1) was done at the level of cemento-enamel junction. Coronal portion of each tooth was impregnated in cold cure acrylic resin (Fig: 2) in such a way that the labial surface of the crown was levelled on top lying flat and parallel to the horizontal plane. Working window was marked at the centre of the teeth using digital vernier calipers of 5mm x 5mm dimensions on the labial surface of the teeth. The labial enamel surface was polished with fine grit (240, 400, 600 size) silicon carbide paper of the outermost enamel layer to prepare a flat surface. Nail varnish was applied on the surface of the teeth except for a 5mm× 5mm working window. Pre-bleaching (baseline) color shade measurements were evaluated of the sample teeth through spectrometer. All the samples of group 1 and 2 were than subjected to bleaching agent on the working window. The samples were then placed in distilled water.

Bleaching procedure
Pola office (SDI) is a chemically activated power liquid whitening gel. The kit contains
1- Pola office liquid syringe which contains 35% hydrogen peroxide and 65% water
2- Pola office powder pot contains 73.26% thickeners, 26.2 % catalyst, 0.04% Dye, 0.5% desensitizing agents.

However the kit also contains gingival barrier which is a combination of 83.95% methacrylic ester, 16% silica, 0.04% pigment, 0.01% butylated hydroxy toluene. This gingival barrier is used in in-office bleaching technique to protect the patient's gingiva from harmful effects of bleaching agents. This gingival barrier is of no use in this study as the study was done in-vitro, therefore, no patient protection is needed.
Pola office powder pot is opened. Tip is attached firmly to pola office syringe and carefully all the contents of syringe liquid is extruded in powder pot. Mixing of the powder and liquid was done using a brush applicator until gel is homogenous than a layer of activated bleaching gel is applied on the prepared specimen. The powerful 35% hydrogen peroxide were activated just prior to application, ensuring that every dose of bleaching agent were fresh and effective. The activated bleaching agents were applied on all the samples of group 1 and group 2 teeth for approximate thickness with applicator tip for 3 times (each time for 15 minutes) every alternate day for four days. After completion of bleaching procedure irrigation of samples with distilled water were done. The samples were stored in distilled water in between the bleaching procedures.
After bleaching, the teeth of group 1 and group 2 were immersed in 200 ml of red wine and brandy respectively for 15 minutes, 6 hours, 1 week and 1 month respectively.

Tooth color measurements
Advance spectrophotometer was used to measure the color shade of the teeth pre bleaching, after 15 minutes, after 6 hours, after 1 week and after 1 month of red wine and brandy immersion. The spectrophotometer was calibrated according to the manufacturer's instructions before taking each reading and then carefully placed at right angle to buccal surface of the crown. The values were selected when two consecutive, identical readings were generated for each area. The resulting shades were taken directly from the digital screen of spectrophotometer device and CIE L*a*b* readings were taken. All color measurements were taken three times at different places on the middle third of each sample surface using the inbuilt synchronized image program. Pre-bleaching spectrometeric color measurements of the buccal surface were taken and considered as baseline data to which the subsequent readings of immersion in red wine or brandy at 15 minutes, 6 hours, 1 week and 1 month were compared.
The colour difference (ΔE) at each time interval was calculated according to the following formula:

$$\Delta E=[(\Delta L^*)^2+(\Delta a^*)^2+(\Delta b^*)^2]^{1/2}$$

Where ΔL is the difference in lightness calculated from differences in the L* readings between two periods.
Δa and Δb refer to the difference in chroma and are also obtained in the same manner as for ΔL. ΔE value equal or larger than 3.5 is considered as a clinically perceptible colour change.
Statistical analysis was done using analysis of variance (ANOVA) which is a collection of statistical models, and their associated procedures, in which the observed variance is partitioned into components due to different explanatory variables.

Results
Mean ΔE values ± standard deviation of group 1 at different time intervals shows that color change due to repigmentation of sample teeth was lowest from baseline to 15 minutes time interval. Mean ΔE value ± standard deviation was 4.04045±1.308029. However color change value was highest in 6 hours to 1 week time interval and was

11.19205±1.706909. Mean ΔE value ± standard deviation in 1 week to 1 month time interval was 11.12707±2.538020 and was lower than mean ΔE value of 6hours to 1 week time interval.

Mean ΔE values ± standard deviation of group 2 at different time intervals shows color change due to repigmentation was lowest from 15 minutes to 6 hour time interval. Mean ΔE value ± standard deviation was 6.00537±1.098081. However color change value was highest in 1 week to 1 month time interval and was 13.45717±1.098210. Mean ΔE value ± standard deviation in 6 hours to 1 week time interval was 12.99540±2.500035 and was lower than mean ΔE value of 1 week to 1 month time interval.

When comparison of mean ΔE values ± standard deviation of group 1 and group 2 at different time intervals was done (Table:1). It was found that color change due to repigmentation was lowest for group 1 teeth from baseline to 15 minutes time interval. Mean ΔE value ± standard deviation was 4.04045±1.308029. However color change value was highest in group 2 teeth at 1 week to 1 month time interval and was 13.45717±1.098210. Group 2 teeth had higher ΔE value than group 1 at any point of time interval.

Discussion

The study assessed the influence of different staining agents on the color change of tooth enamel, which may occur during the immediate bleaching treatment. The demand for conservative aesthetic treatments is increasing. Tooth bleaching is a treatment that improves the appearance of teeth and is considered as safe procedure when well indicated and performed [32]. Tooth bleaching is considered the easiest and most cost effective procedure for treating tooth discoloration and has gained popularity in clinical set up because of its superficial nature, easy manipulation and its quality of being less expensive. Oxidation-reduction reaction of bleaching agents produce hydrogen ions (H+) that can create an acidic environment and lead to dissolution of the organic and inorganic enamel. There is a ingress of oxidizers and oxygenating molecules via enamel micro pores along a diffusion gradient. These reduce or cleave pigment molecule double bonds either to break down pigments to small enough molecules that diffuse out of the tooth, or to those that absorb less light and appear lighter. Hydrogen peroxide forms a loose association with urea to produce urea peroxide which is easily broken down in the presence of water to release free radicals that penetrate through the enamel pores and into the dentine to produce the bleaching effect [33]. Changes in the chemical and physical structure of enamel must be of great concern to a dentist who utilizes bleaching techniques as a treatment for lightening teeth [34].

Human teeth are exposed to a different point-to-point pressure during mastication. Therefore, the study and analysis of their hardness is very important. Mechanical properties of enamel depend largely on the degree of mineralization [35]. Cuy *et al*. [36] hypothesized that there is a strong correlation between degree of repigmentation of enamel and its mechanical properties. Dental enamel is composed of a huge number of highly mineralized prisms, and its hardness is due to a high percentage of inorganic matrix (95%) made of hydroxyapatite crystals (calcium phosphate) and low percentage of organic protein nature matrix (0.36 to 2%) together with polysaccharides.

Many agents have been used in the past, and a number of new methods have continued to be introduced. It was oxalic acid first by Chappel in 1877 which was used as bleaching the agent it was followed by various forms of chlorine until H_2O_2 was first used by Harlan in 1884.

In-office procedure mainly utilizes hydrogen peroxide usually in concentrations ranging from 35% to 50%. Carbamide peroxide in the concentration ranging from 10% to 22% is the most common for at home treatment/night guard vital bleaching/matrix bleaching/dentist prescribed/home applied bleaching. “Power bleaching” uses 30-35% H_2O_2 solution in conjunction with heat or light to increase the kinetics of stain removal.

Tooth bleaching therapy might negatively affect the tooth structure due to the oxidative action, pH or the composition of the bleaching agent in varying degrees, depending on the type of bleaching system.[37] Thus, the long-term performance of bleaching treatments is debatable, and in many cases, some degree of rebound effect has been observed within days or weeks following the bleaching procedure [38]. Oxidative action by hydrogen peroxide can cause structural and permeability changes and surface porosity in the enamel surface [10, 20]. Furthermore, coloring pigments might accumulate on the rough surface, and a rough enamel surface with pores or superficial defects might discolor easily [39].

It has been reported in a number of articles that bleaching of enamel increases its susceptibility to extrinsic stains [19, 40, 41]. 35 % hydrogen peroxide was used in this study as because in a previous *in vitro* study, 35% hydrogen peroxide was found to cause greater tendency for staining compared to low concentration peroxide [42]. It has also been revealed that surface morphological alterations increase with higher concentrations of hydrogen peroxide and longer treatment times [1, 43].

Hydrogen peroxide can result in a decrease in microhardness of enamel. Hydrogen peroxide may cause significantly more loss of calcium from the enamel surface than carbamide peroxide. Following H_2O_2 application, a decline in mechanical properties and fracture toughness and an increase in enamel surface dissolution by phosphoric acid have been reported.

In a recent clinical study, home bleaching treatment with 10% carbamide peroxide was found more effective, more acceptable to patients and required less chair time to inoffice bleaching with 35% H_2O_2 (Zekonis *et al*.). Carbamide peroxide is suggested as a safer alternative to hydrogen peroxide-based systems.

A number of methods are available for evaluating tooth shade changes after bleaching, following immersion in coloring solutions. These can be classified as subjective, such as the use of a standard tooth shade guide, and objective, such as use of spectrophotometers [44], colorimeters and computer digitization [45]. Spectrophotometers differ from colorimeters in that they measure reflected light within the entire visible spectrum, whereas colorimeters measure reflected light at only three wavelengths [46]. Although colorimeters provide reproducible results, such results can be affected by tooth translucency, contour and texture [47]. Some conditions interfere with the measurement of tooth color, such as a rough surface and non-uniform surface geometry. The spectrophotometer in a diffuse reflectance mode minimizes edge losses at the side of the sample tooth and maximizes the collection of reflected light in all directions, all of which minimize the disadvantages of sample characteristics. In this study, in order to achieve more consistent and accurate results, a spectrophotometer was used. Spectrometer allows completely accurate evaluation of spectral data, unaffected by light sources in other ambient light. The surfaces of the

samples were not ground flat before the experiment, as to investigate the teeth under natural conditions. However, this might have led to a greater variation among the specimens, with respect to the adsorption of stain and the determination of the color, because of some irregularities in the surface composition of the samples. All the samples were thoroughly cleaned and polished before the experiment. This is common practice, and it is recommended that the teeth are thoroughly cleaned prior to bleaching.

We stored the samples in 0.1 percent thymol solution throughout the experiment, as we do not wanted to simulate both the remineralization of the bleached specimens and the impact of saliva as being significant in the formation of tooth staining. It is debatable as to whether microstructural defects may be repaired by remineralization of saliva.

Numerous previous studies have evaluated the adverse effects of peroxide containing bleaching products on tooth enamel, with conflicting results. Some studies have reported no significant deleterious effects on the surface microstructure of the enamel and dentine after bleaching treatment [48, 49]. However, others have actually shown a deleterious effect on the enamel and/or dentine, such as alteration of surface morphology [12-50]. Staining susceptibility cannot be related to surface roughness alone, but to enamel composition, water absorption rate, due to permeability alterations, and irregularities left on bleached enamel surfaces, which could facilitate the accumulation of dye [11, 12, 14, 51, 52].

Coffee, tea, juices, wines and cola-based soft drinks are beverages with the potential to darken, discolor or stain bleached enamel surfaces. Some of these beverages are acidic solutions that can increase demineralization and others contain ethanol or pigments [16, 20]. Certain beverages, artificial food colorations and frequent smoking are responsible for primary staining, darkening and discoloration of teeth. The ΔE values in Table I and II suggest that bleached teeth are more susceptible to staining, particularly by brandy and red wine, which are acidic, colored and alcoholic beverage. Although both brandy and red wine were effective in staining, brandy had a greater capacity for staining than red wine. These results are consistent with those of Liporoni *et al*., [19] who reported that alcoholic beverages effectively interfered with the reflectance values on the bleached enamel surface. However, they also concluded that wine discolored the enamel compared to that of the control group. Although some authors have observed that bleached enamel is not susceptible to staining by other pigments, such as tea, after saliva storage [16]. Therefore in this study 0.1 % thymol was used instead of saliva so as to negative the effect of saliva. However, the predisposition of enamel to repigmentation after bleaching was previously observed, and this tendency was even greater with wine storage [18-20].

Tooth discoloration or repigmentation after bleaching is dependent on a variety of factors, such as the pH value of the staining solution [53]. In this study, brandy had the lowest pH and may have affected the surface of the samples greater than red wine; it showed the highest ΔE* value 1 month and 1 week after immersion in comparison than red wine. In one study statistically significant differences between teeth treated with wine after bleaching [25]. However, another study**19** demonstrated that bleached enamel was susceptible to red wine staining after bleaching procedures. In this study, statistically significant differences were observed between red wine and brandy group at any point of time.

Teeth exposed to a pH < 5.5 for enamel and dentin for 6.0 for an extended period of time can lead to demineralization [54] and erosion of enamel [55]. Brandy and red wine used in this study are highly acidic solutions, showing that the low pH of those solutions may have had a major effect on the structure of the teeth bleached. The erosive loss of enamel is higher with brandy, besides having a low pH that is constituted of phosphoric acid that has a high erosive power [56]. Red wine has a low pH and is also an alcoholic beverage, which may have contributed to the higher enamel demineralization of this solution.

Color change of the teeth submitted to bleaching and aging in staining solutions was evaluated by the CIE L*A*B* method in this study. In principle, if the color of a material is completely stable, no difference will be detected after exposure to the environment tested (ΔE =0). In addition, ΔE values between 3 and 8 will already be moderately visible, and values >8 would be extremely perceptible [57]. After bleaching, the specimens in both group 1 and group 2 were stored for 15 minutes, 6 hours, 1 week and 1 month in brandy and red wine. The ΔE values of the bleached group 1 and group 2 were considered moderately perceptible between baseline and 15 minutes as well as between 15 minutes and 6 hours of immersion in brandy and red wine whereas, extremely perceptible between 6 hours and 1 week as well as between 1 week and 1 month of immersion in brandy and red wine. It is known that the larger the surface roughness, the greater the absorption of more pigments, which helps to explain the result obtained for the brandy and red wine group [10]. Additionally, the low pH combined with alcohol molecules of brandy and red wine could provide a surface change by the mineral loss, thus leaving the enamel surface more susceptible to staining by pigments of brandy and red wine.

In teeth, an inorganic mineral (calcium phosphate in the form of hydroxyapatite) is combined with an organic protein matrix. Only the chemical and structural interplay between these two components leads to the extraordinary mechanical properties of teeth with respect to hardness and fracture toughness. Thus, teeth are not simply inorganic materials but highly optimized and complex organic-inorganic biocomposites. If aggressive bleaching agents like hydrogen peroxide are applied in high concentrations, it will also damage the organic matrix in the tooth, especially in dentin. We note that enamel contains about 1% organic matrix and dentin contains about ca. 20% organic matrix, mainly collagen. This could lead to a mechanical weakening of the tooth due to a decreasing integration of the calcium phosphate crystals. Fearon discussed a number of studies of different whitening procedures and concluded that changes in the tooth surface structure and increased tooth sensitivity can occur, especially if highly concentrated hydrogen peroxide solutions are applied. There are also reports about a structural damage of enamel surface prisms after application of 35% carbamine peroxide. and the risk increases with increase concentration.

Finally, the modes of action of many whitening agents *in vivo* are still unknown. Thus, mechanistic studies are required to understand the mechanisms of action from a chemical and biological viewpoint, which is an important requirement for the development of more efficient teeth whitening formulations.

For whitening, two major approaches can be distinguished, as follows: Chemical bleaching by peroxides and mechanical cleaning by toothpaste abrasives. Chemical bleaching leads to good results, especially when it is performed with high peroxide concentrations in a controlled environment, i.e., in the dental practice. Mechanical cleaning relies on suitable

abrasives that are harder than stains but less hard than enamel. Considerable progress into this direction has been achieved with silica toothpaste formulations in the last years (optimized RDA/PCR ratio), but current formulations always represent a compromise between desired cleaning efficiency and unwanted tooth abrasion.

Studies like these are important to understand the post bleaching complications and how to inform the patient in regard to the same, more effective ways of tooth whitening need to be formulated so that the integratiy, of the tooth structure is not compromised and the patient can enjoy his normal life style and drinking choices.

There was a trend of potential dye substances, such as brandy or red wine, to exert influence in the final color; therefore, the consumption of highly colored food and beverages should be decreased. Moreover, high mineral loss was observed when the enamel was exposed to acidic drinks that could decrease the staining resistance by superficial changes of the dental enamel, making extremely important the guidance to avoid intake of acidic substances, especially during bleaching treatment.

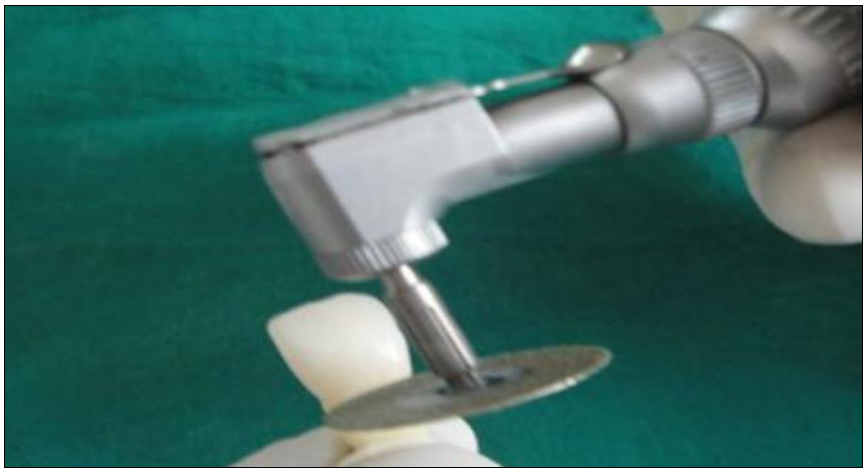


Fig 1: Decoronation of tooth

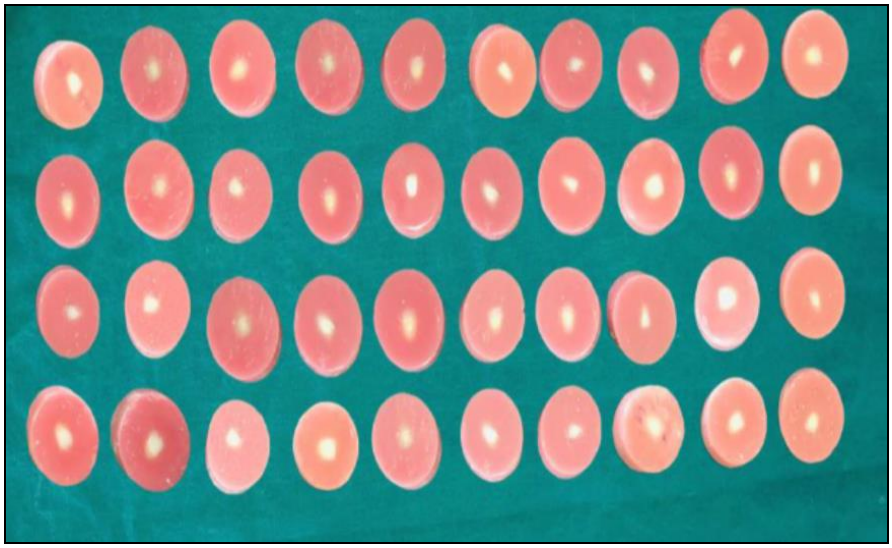
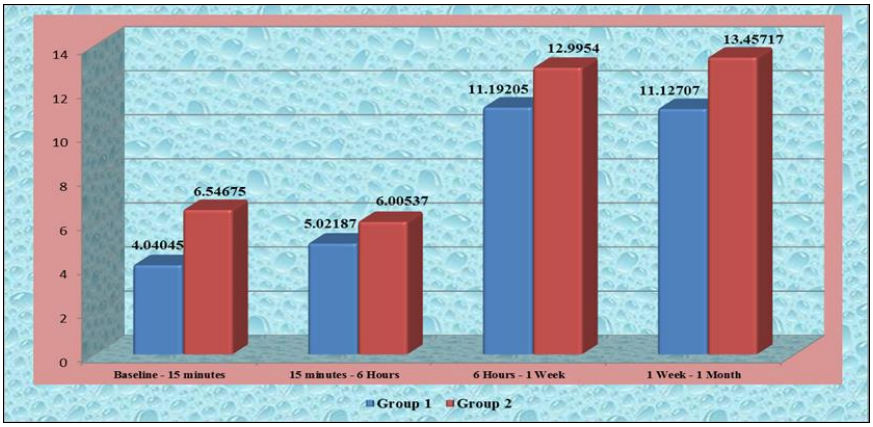


Fig 2: Mounted enamel samples



Graph 1: Comparision of mean ΔE value recorded of group 1 and group 2 at different time intervals

Table 1: Mean ΔE value recorded of group 1 and group 2 at different time intervals

Group	Baseline-15 minutes	15 minutes-6 hours	6 hours-1 week	1week-1month
1	4.04045±1.308029	5.02187±2.688029	11.19205±1.706909	11.12707±2.538020
2	6.54675±1.289078	6.00537±1.098081	12.99540±2.500035	13.45717±1.098210

Conclusions

The present study was conducted to investigate the susceptibility of enamel surface for repigmentation by red wine and brandy stains after 35% hydrogen peroxide bleaching. In our study the results shows that;

1- The ΔE values of the bleached group 1 were considered moderately perceptible between baseline and 15 minutes as well as between 15 minutes and 6 hours of immersion whereas, extremely perceptible between 6 hours and 1 week as well as between 1 week and 1 month of immersion.
2- The ΔE values of the bleached group 2 were considered moderately perceptible between baseline and 15 minutes as well as between 15 minutes and 6 hours of immersion whereas, extremely perceptible between 6 hours and 1 week as well as between 1 week and 1 month of immersion.
3- When comparing group 1 and group 2 it was found that group 2, ΔE values were more than group 1 ΔE values indicating low pH values cause more repigmentation of bleached enamel surface as it was found more with brandy in comparison to red wine.
4- The bleaching treatment with 35% hydrogen peroxide enabled the teeth to become clearer; nevertheless, the intake of food and beverages containing colorants or an acidic pH can negatively affect the effectiveness of bleaching.

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