EFFECT OF POLISHING PROTOCOL AND EXPOSURE TO STAINING SOLUTIONS ON THE COLOUR STABILITY OF DENTAL COMPOSITE MATERIALS

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I, Dr. Chamunorwa Marufu, student number V60/88169/2016, of College of Health Sciences, School of Dental Sciences, Department of Conservative and Prosthetic Dentistry, declare that this thesis titled "**EFFECT OF POLISHING PROTOCOL AND EXPOSURE TO STAINING SOLUTIONS ON THE COLOUR STABILITY OF DENTAL COMPOSITE MATERIALS"** is my original work.

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DEDICATION

This work is dedicated to my wife, Fortunate, and our daughter, Rujeko for their love and support.

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LIST OF ABBREVIATIONS

a*	Chromaticity coordinates defining a colour in the red-green axis
b*	Chromaticity coordinates defining a colour in the yellow-blue axis
Bis-EMA	Bisphenol A polyethylene glycol diether dimethacrylate
Bis-GMA	Bisphenol glycidyl methacrylate
BP	Benzoyl peroxide
CIE-Lab	Commission International de l' Eclairage L*a*b colour system
CQ	Camphorquinone
DCF	Soflex disc finished – Filtek specimens in the control
DCV	Soflex disc finished – Vit-l-escence specimens in the control
df	Degrees of freedom
DK1F	Soflex disc finished – Filtek specimens in khat 1
DK1V	Soflex disc finished – Vit-l-escence specimens in khat 1
DK2F	Soflex disc finished – Filtek specimens in khat 2
DK2V	Soflex disc finished – Vit-l-escence specimens in khat 2
DMAB	4-(dimethylamino) benzoic acid ethyl ester
DMAEM	Dimethylaminoethyl methacrylate
DTF	Soflex disc finished – Filtek specimens in tea
DTV	Soflex disc finished – Vit-l-escence specimens tea
DRF	Soflex disc finished – Filtek specimens in red wine
DRV	Soflex disc finished – Vit-l-escence specimens in red wine
LCU	Light curing unit
L*	Characterizes the lightness of the colour
M	Mean
MCF	Mylar finished – Filtek specimens in the control
MCV	Mylar finished – Vit-l-escence specimens in the control
MK1F	Mylar finished – Filtek specimens in khat 1
MK1V	Mylar finished – Vit-l-escence specimens in khat 1
MK2F	Mylar finished – Filtek specimens in khat 2
MK2V	Mylar finished – Vit-l-escence specimens in khat 2
MTF	Mylar finished – Filtek specimens in tea

MTV	Mylar finished – Vit-l-escence specimens in tea
MRF	Mylar finished – Filtek specimens in red wine
MRV	Mylar finished – Vit-l-escence specimens in red wine
n	Sample size
NIR	Near infra-red
n	Sample size
P	p value
PPD	1-Phenyl-1, 2- Propanedione
SEM	Scanning electron microscope
SCF	White polishing stone finished – Filtek specimens in the control
SCV	White polishing stone finished – Vit-l-escence specimens in the control
SD	Standard deviation
SK1F	White polishing stone finished – Filtek specimens in khat 1
SK1V	White polishing stone finished – Vit-l-escence specimens in khat 1
SK2F	White polishing stone finished – Filtek specimens in khat 2
SK2V	White polishing stone finished – Vit-l-escence specimens in khat 2
STF	White polishing stone finished – Filtek specimens in tea
STV	White polishing stone finished – Vit-l-escence specimens in tea
SRF	White polishing stone finished – Filtek specimens in red wine
SRV	White polishing stone finished – Vit-l-escence specimens in red wine
TEGDMA	Triethylene glycol dimethacrylate
TPO	2,4,6-trimethylbenzoyldiphenylphosphine oxide
UDMA	Urethane dimethacrylate
UV	Ultra violet
VL	Visible light
WHO	World Health Organisation
Δa*	Change in the a* coordinate
Δb*	Change in the b* coordinate
ΔE	Colour difference
ΔL*	Change in the L* coordinate
µm	Micro-meter

DEFINITION OF TERMS

Dental resin composite: Tooth coloured filling materials made of resin reinforced with inorganic fillers

Intrinsic discolouration: Permanent discolourations related to the materials' composition, that is, the matrix, filler type and amount, photo-initiator system and percentage of remaining carbon double bonds.

Extrinsic discolouration: Discolouration due to superficial degradation or slight penetration and adsorption of dyes from foods, drinks or habitually chewed items

Hue: The quality that distinguishes one family of colour from another

Value: The value indicates the lightness of a colour

Chroma: The degree of saturation of the hue

Nanofill composite: A composite material with nanometer size filler particles 1-100nm in size

Microhybrid composite: A composite material composed of two types of fillers blended together. They are composed of fine particles are of an average size of 0.04 to 1µm and 5 to 15% microfine particles of 0.04 to 0.2µm size. The distribution of filler particles allows for efficient packing and increased filler loading

Finishing composite restorations: Finishing is the process of removing surface defects or scratches created during the contouring process through the use of cutting or grinding instruments or both.

Polishing composite restorations: Polishing is the process of providing lustre or gloss on a material surface

Colour difference: A calculated figure which quantifies the change in colour from an initial measurement. Derived using the CIE system which uses the three dimensionless colorimetric measurements. L*a*b* colour) system

Spectrophotometer: An instrument used to evaluate colour via via full spectrum colour measurement

ABSTRACT

Background: Colour instability of dental resin composite materials can result in their failure and subsequently need for replacement. The discolouration may be intrinsic or extrinsic. Intrinsic discolouration is dependent on the materials' composition while extrinsic discolouration can be due to adsorption of stains which make contact with the restorations. Finishing and polishing is one of the most crucial steps in the placement of dental resin composite restorations. Apart from being essential for oral health and function by ensuring smooth surfaces that prevent accumulation of plaque, allow food to glide more freely over tooth surfaces during mastication and minimise wear to opposing and adjacent dentition, finishing and polishing is also an important determinant of extrinsic discolouration of composite restorations.

Broad objective: To evaluate the effect of finishing and polishing protocol and exposure to staining solutions on the colour stability of dental resin composite restorative materials.

Study design: A laboratory based experimental study.

Study area: School of Dental Sciences, Prosthetics laboratory and School of Pharmacy, Pharmaceutical Chemistry laboratory, University of Nairobi.

Materials and methods: Two commercial dental resin composite materials were evaluated for colour stability: a nano-fill composite, *Filtek Z 350* (3M ESPE St Paul, MN, USA) and a micro-hybrid composite, *Vit-l-escence* (Ultradent Inc, South Jordan, UT, USA). The material specimens were prepared from perspex moulds with dimensions of 8mm diameter x 2mm width. A total of 150 specimens were prepared, 75 from *Vit-l-escence* and 75 from *Filtek Z 350 XT*. Three finishing and polishing protocols were evaluated, namely, mylar strip finish (Maquria industries, Brazil), *Soflex* polishing discs (3M ESPE St Paul, MN, USA) and white polishing stone (Prime dental, UK), while the staining solutions were tea, red wine and khat extract (diluted to 2 different concentrations). Distilled water was the control for staining solutions. The method specified in International for standardisation ISO 7496:2000 for determination of colour stability of dental materials was used. There were 5 test specimens for each finishing protocol and staining solution for both materials. Baseline colour measurements were taken before the specimens were placed in the staining solutions. A digital spectrophotometer (*Vita Easyshade*, Vita Zahnfabrik, Bad Sackingen, Germany) was used for all colour measurements which were

done using the CIE-Lab-colour (Commission International de l' Eclairage L*a*b colour) system. The colour measurements were taken at 6hrs, 1, 2, 4, 7, 10 and 14days against a white background and the mean calculated. An Independent-Samples t-test was used to determine the difference in colour at baseline and at the end of staining between the two materials while Two-Way Analysis of Variance (ANOVA) was used to determine the difference in mean colour change within or between the groups of finishing protocols and staining solutions followed by Tukey's HSD post hoc test at an alpha level of 0.05.

Results: Total colour difference (ΔE) above 2.6 units is considered to be clinically perceptible while ΔE of 5.5 is considered clinically unacceptable. At the end of the staining period all specimens, except Soflex disc finished – microhybrid specimens (*Vit-l-escence*) in the khat 1 and khat 2 staining solutions, demonstrated a clinically unacceptable ΔE.

In the microhybrid group (*Vit-l-escence),* S*oflex* disc finished specimens recorded the least mean ΔE whilst mylar finished specimens demonstrated the highest ΔE. Within the Soflex finish – microhybrid group, red wine produced the highest ΔE of 8.2±0.67 units while khat 2 (khat diluted at 1:3) demonstrated the least ΔE of 5.02±0.72 units. Red wine was the only staining solution to produce a statistically significant ΔE, $p=0.023$ in this group. The pattern in the white polishing stone finish – microhybrid group was similar to the Soflex group with red wine producing the highest ΔE of 9.47±0.85 units and khat 2 producing the lowest ΔE of 8.18±1.62 units. All the 4 staining solutions produced statistically significant ΔE with $p<0.001$ in red wine, $p=0.004$ in tea, $p=0.008$ in khat 1 and $p=0.011$ in khat 2. In the mylar finish - microhybrid group, khat 2 produced the highest ΔE of 18.3±4.76 units while red wine produced the least ΔE of 10.66±1.02 units. All the 4 staining solutions produced statistically significant ΔE with $p<0.001$ for all of them.

In the nanofill group (*Filtek Z 350XT*), white polishing stone demonstrated the highest ΔE in all the staining solutions while Soflex discs demonstrated the least ΔE in all the staining solutions with the exception of the red wine staining group where mylar finished specimens demonstrated the least ΔE. In the white polishing stone group, khat 2 produced the highest ΔE of 15.66±1.3 units and red wine produced the least ΔE of 11.81±0.68 units with all 4 staining solutions producing statistically significant ΔE $p<0.001$. In the mylar finish group tea produced the highest ΔE of 13.65±1.89 while red wine produced the least ΔE of

7.82±0.79 units. All 4 staining solutions again produced statistically significant ΔE, p=0.002 with wine and p<0.001with the other 3 solutions. In the Soflex group red wine produced the highest ΔE of 8.17±1.27 while tea produced the least ΔE of 5.96±1.21. In this group only red wine produced a statistically significant ΔE p=0.002.

Comparing the two materials, the microhybid composite showed better colour stability compared to the nanofill when finished with Soflex discs and white polishing stone, while the nanofill had better colour stability with the mylar finish in the staining solutions used in the study. The staining solutions did not show any clear pattern as to which produces the highest ΔE.

Conclusion: There is a difference in colour stability of resin composites depending on the type of material and the finishing protocol. Overall, Soflex disc finished specimens demonstrated the least ΔE for both materials while white polishing stone finished specimens demonstrated the highest ΔE for the nanofill composite and mylar finished specimens demonstrated the highest ΔE for the microhybrid composite. Clinicians should select the recommended finishing protocol for various materials as guided by the filler content.

CHAPTER ONE: INTRODUCTION

1.1 Dental resin composites

Dental resin composites are reinforced polymer systems which can be used to restore tooth structure or to alter tooth colour and morphology to enhance aesthetics. Since their introduction in the 1960s, they have become increasingly popular for both anterior and posterior restorations.[1,2] This has been due to growing aesthetic demands from patients and change of philosophies in operative dentistry, moving away from GV Blacks' principles to current minimum intervention concepts.[3] They are currently the materials used most for restoration in the aesthetic region.[4] Various types of resin composite materials are available for different clinical applications. Some have been designed for use in aesthetic regions, while others are indicated for use in high stress bearing areas.[5] the most recent innovation are nano-composite materials, which are optimized for use in both aesthetic restorations and high stress bearing areas.[6]

Various researchers have evaluated the clinical longevity of dental composite restorations and have reported an average longevity of 6 to 10 years.[7] The failure behaviour is however different for anterior and posterior composites. For posterior composites the chief contributor of failure are secondary caries and restoration fracture,[8] while factors related to aesthetics such as colour alterations are more common for anterior restorations.[9,10,11] Other factors influencing longevity include patient related factors such as caries risk and clinician factors such as clinical experience.[1]

1.2 Importance of aesthetic dentistry

The impact of dentofacial appeal in regards to an individual's psychosocial wellbeing is well documented. An important element of dentofacial appeal is an aesthetic smile of which the appearance of teeth has an impact. Individuals who are not happy with the appearance of their teeth tend to avoid smiling or cover their mouths when they smile.[12] Aesthetic restorations can have a positive impact on these patients' self-confidence and mental health.

To achieve imperceptible aesthetic restorations, the optical properties of the restorations should be in harmony with those of the adjacent natural teeth. This is in part determined by the colour matching of the restoration on placement and colour stability in the longrun.[13] In the mouth, resin composite restorations are subjected to moisture, stain and mechanical wear which can cause discolouration.[14,15]

1.3 Comparison of available aesthetic dental restorations

Currently materials being used for definitive aesthetic restorations are dental porcelains and resin composites.[16] Dental porcelains are regarded as the ultimate aesthetic dental material due to their natural appearance, good wear resistance and colour stability.[17] They however have a number of undesirable characteristics which include susceptibility to brittle fracture,[18] technically demanding and time consuming fabrication requiring expensive processing equipment and abrasion of opposing natural teeth.[19] Some of these draw backs are eliminated by use of composite resin materials.

Compared to porcelain, composite resin restorations are less technically demanding, cheaper, do not cause wear to opposing dentition and can be easily repaired.[16] In addition composites can be bonded to tooth structure allowing for conservative preparations and resin systems can be cured on demand allowing for control of working time.[20] For aesthetic restorations, resin composites are available in a number of different shades and translucencies to allow for the replication of the combined optical properties of dentine and enamel. Opaque composites can be used to mask discoloured tooth structure or mimic dentinal opacity before application of a more enamel like translucent composite.

Disadvantages of resin composites include polymerization shrinkage which results in micro-leakage from gap formation, colour instability, incomplete polymerisation with the degree of conversion ranging from 55% to 65% only and limited curing depth for light cured systems necessitating the material to be placed incrementally when restoration exceeds 2-3mm.[18,21]

1.4 Factors that influence colour stability of dental resin composites

Discolouration of composite resin restorations can be intrinsic or extrinsic.[17,22] Intrinsically induced discolourations are permanent[23] and are related to the materials' composition, that is, the matrix, filler type and amount, photo-initiator system and percentage of remaining carbon double bonds.[11] Extrinsic discoloration can be due to accumulation of plaque and stain, low degree of polymerisation, exposure to environmental factors including heat, water, food colourants, ambient and ultraviolet light. More than one factor may be responsible for the colour change. An important determinant of extrinsic discolouration is the finishing and polishing protocol used on the composite restoration.[24]

1.5 Finishing and polishing of composite restorations

Finishing is the process of removing surface imperfections which are left on the restoration after the contouring process using cutting or grinding instruments and polishing is the process of providing lustre on a material surface. This provides three benefits, oral health, function and aesthetics.[18,25] Finishing and polishing are essential for oral health because smoother surfaces prevent the accumulation of plaque and are also easier to maintain clean by the patient with routine preventive oral home care. In terms of function smoother surfaces allow food to glide more freely over tooth surfaces during mastication. The smooth surfaces also reduce wear to opposing and adjacent dentition. In terms of aesthetics, finishing and polishing allow restorations in aesthetic areas to be made compatible with adjacent teeth.[18] Smooth surfaces also improve colour stability of restorations.[10,26,27]

Unfortunately the proper sequence of polishing steps is often overlooked and this may result in restorations being left rough and susceptible to plaque retention and staining. The need for replacement of restorations results in patient dissatisfaction, inconvenience and added cost. Hence the objective of the study is to evaluate effect of polishing protocol and exposure to staining solutions on colour stability of dental resin composite restorations.

CHAPTER TWO: LITERATURE REVIEW

2.1 Introduction

This section details the composition of dental resin composites, the importance of finishing and polishing composite restorations, factors which impact their colour stability and colour measurement in dentistry. A review of the staining materials used in this study is also included.

2.2 Dental resin composites

Currently there is a wide variety of dental composite materials on the market for various applications. This market continues to evolve with emphasis shifting from aiming to produce materials with optimum mechanical properties and polishability retention to addressing the issues of polymerisation shrinkage and its accompanying stresses which have a negative impact on the restoration-tooth bond.[28]

Despite the large number of new materials available, they all have a similar composition. They are all combinations of filler particles coated with a coupling agent mixed with monomers such as "bisphenol-A-glycidyl methacylate (Bis-GMA), urethane dimethacrylate (UDMA) and triethylene glycol dimethacrylate (TEGDMA)."[18] Figure 1 shows the chemical formulae for these monomers.

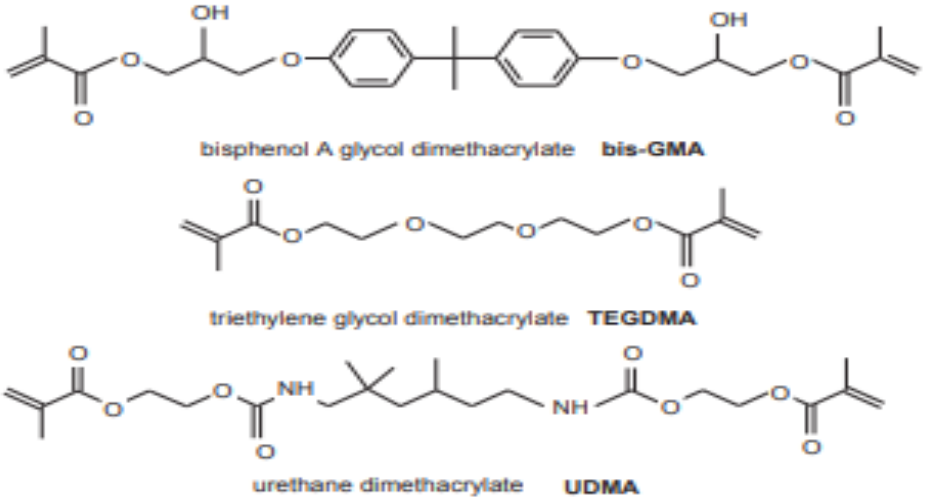

Figure 1: Chemical formulae of monomers commonly used in composites[29]

These monomers polymerise by addition polymerisation reactions initiated by free radicals. These free radicals can be produced by chemical activation or external energy activation in the form of

heat, light or microwave. Chemically activated composite resins are supplied as two pastes, one containing the initiator (benzoyl peroxide, BP) and the other containing an aromatic tertiary amine activator (e.g N, N-dimethyl-p-toluidine). When the two pastes are mixed, the activator reacts with the initiator to form free radicals which initiate the polymerisation reaction. The disadvantages of chemically activated composites include incorporation of air during mixing, forming pores which trap oxygen which inhibits polymerisation and lack of command cure. To overcome these problems light cured dental composites were developed.[18]

In light activated resins systems, the monomers are polymerised by light in the presence of photoinitiators. The photoiniators can be differentiated according to the spectral region in which they absorb light as ultra violet (UV), visible light (VL) and near infrared radiation (NIR).[30] The radicals that initiate the monomer polymerisation are either formed by bond fission or by the transfer of a hydrogen atom from a second compound, the coinitiator.[31] Most photoinitiators contain carbonyl groups as light absorbers. Lucirin TPO (2,4,6-trimethyl benzoyl diphenyl phosphine oxide) is an example of a carbonyl compound that forms radicals by bond fission. Camphorquinone (CQ) forms radicals by transfer of hydrogen atoms from an amine co-initiator such as dimethylaminoethyl methacrylate (DMAEMA) and 4-(dimethylamino) benzoic acid ethyl ester (DMAB). This produces a reactive free radical based on the co-initiator and an unreactive radical based on camphorquinone. This reaction also eliminates the molecule responsible for the yellow shade of CQ (photo-bleaching effect).[30] Aliphatic amines can be used in photo curing systems in place of aromatic amines used with chemical curing which enhances the material's colour stability.[18]

Due to strong light absorption properties and low toxicity CQ containing photo-initiators have been used almost exclusively in dentistry. CQ-amine photo-initiators can also be combined with other photo-initiators such as 1-phenyl-1.2-propanedione (PDD) to effect more reliable curing on the resin composites. CQ-amine photo-initiator systems however have a number of shortcomings. In acidic compositions such as self-adhesive cements containing acidic monomers, an acid base reaction of the monomer with the amine based co-initiator may negatively affect the formation of radicals.[30] If in these systems the curing is inadequate, the unconverted CQ can cause a yellowish discolouration.[23]

The filler particles are either barium silicate glass, quartz or zirconium silicate which is usually mixed with colloidal silica particles.[32] They make up the major portion of the material by volume or weight. The functions of the filler include, reinforcement of the polymer, control of polymerisation shrinkage, control of thermal expansion, improving workability, increased radiopacity and diagnostic sensitivity by incorporation of heavy metals, reduction in water sorption,[18] and to achieve the required level of translucency.[5]

Dental resin composites can be classified based on the size and shape of the filler particles and the classification is as follows[5]:

2.2.1 Macrofills

These are also referred to as traditional composites. They had comparatively large filler particles resulting in opaque composites with low resistance to wear. They are no longer widely used.

2.2.2 Hybrid and microhybrid composites

These are composed of two different sizes of filler particles mixed together. Hybrid composites are composed of larger fillers particles with an average size of 2 to 4 μm mixed with smaller filler particles of an average size of 0.04 to 0.2μm. In microhybrid composites the larger particles are of an average size of 0.04 to 1μm. This variation of filler particles size allows for increased filler loading. These materials have superior mechanical properties and can be used for restoration in high stress bearing areas. However they tend to lose their surface lustre with time and become rough and dull with time.[5]

2.2.3 Microfills

Traditional microfills are made from fumed silica filler particles with an average size of 0.04μm. The silica particles tend to aggregate and the structure of the aggregates result in a relatively low filler loading. To increase filler loading, prepolymerised filled resin particles are added.[33] The major shortcoming of this is that the bond of the prepolymerised filler particles to the resin is relatively weak and breakdown frequently occurs at the interface and so this material is not suitable for stress bearing surfaces. Microfills produce the smoothest finish and are preferred for class III and class V restorations.[18]

2.2.4 Nanocomposites

These represent the latest advancement in composite technology. There are two types of composite materials in this group, nanofills and nanohybrids. Nanofills contain only nanometer size filler particles 1-100nm in size. These can be in the form of nanomers or nanoclusters. Nanohybrids consist of nanometer size particles with larger particles 0.4 to 5 μm added. The size of the nano-filler particles is less than that of visible light making it possible to create highly translucent materials as light can pass directly through the composite resin. Larger filler particles in conventional composites tend to scatter light resulting in an opaque appearance.[34] The sizes of the smallest nanoparticles is comparable to that of the polymer molecules with which they can form molecular scale interaction. Nanocomposites have the mechanical properties similar to microhybrids and retain surface smoothness during service similar to microfills. Nanohybrids however lose surface lustre with time because the wear of composites is determined by the largest filler particles.[5]

The composition of a dental resin composite is an important determinant of colour stability.

2.3 Colour stability of resin composites

Discolouration of composite resin restorations can be intrinsic or extrinsic. [17,22]

2.3.1 Intrinsic discolouration

Intrinsic discolourations are permanent[23] and are dependent on the composition of the material, that is, the resin matrix, filler type and amount, photo-initiator system and percentage of remaining carbon double bonds.[11,35] The resin plays an important role in discolouration of dental resin composites.[36] The resin's affinity for stains is determined by the degree of polymerisation and its chemical characteristics, with water affinity being a major determinant.[35] Resin matrix hydrophilicity determines the degree of water sorption. The fact that a composite resin can absorb water, means it also has affinity for other fluids which might cause discolouration.[37] Incorporation of TEGDMA can increase water sorption in Bis-GMA based resins [38]. This is because of hydrophilic groups in TEGDMA such as ethoxy groups increase affinity for water by hydrogen bonding to oxygen.[39] Kalachandra and Turner[38] reported that an addition of TEGDMA to Bis-GMA resin to 1% can double water uptake from 3 to 6%. UDMA has lower water affinity compared to Bis-GMA.[40]

Water sorption results in expansion and plasticising of the composite resin material leading to hydrolysis of silane bonds and formation of micro cracks. These micro cracks allow penetration of stains causing discolouration.[23] Water sorption also promotes degradation of the polymer network releasing by products such as methacrylic acid and formaldehyde which result in a colour change.[41]

The degree of water sorption is also influenced by the resin filler content and the integrity of the resin filler interface. Fillers do not absorb water but they contribute to water adsorption on the surface of the material. Composite resins with larger filler particles are more susceptible to water aging compared to composites with smaller filler particles because of the hydrolytic degradation of the matrix filler interfaces.[4] They are also more difficult to finish and polish and the rough finish increases extrinsic discolouration.[42]

The concentration of the photoinitiator should be the minimum possible to achieve a photo-curing reaction with a high degree of conversion. The substitution of Bis-GMA for Bis-EMA is known to increase the degree of conversion on curing and decrease water sorption. In camphorquinone-amine photo-initiator systems, un-reacted molecules will return back to their ground state causing discolouration of the final polymer. The aromatic or aliphatic amines, which are accelerators, can cause a yellow discolouration when exposed to light or heat.[43] Other chemical additives such as ultraviolet filters can also degrade into coloured compounds.[40]

Intrinsic discolouration can also occur due to influence of various physico-chemical conditions for example, humidity, thermal changes, visible light and ultra violet radiation.[44] Except due to improper light curing, intrinsic discolouration depends on manufacturer's formulation.[43]

2.3.2 Extrinsic discolouration

Extrinsic discolouration can result from adsorption of colourants on the surface if the restorations. The material affinity for extrinsic stains is determined by the degree of polymerisation and its physico-chemical characteristics. The more unconverted double bonds the material has, the more susceptible it is to staining.[45]

A number of studies have shown that dental resin composites can discolour when exposed to various staining solutions including coffee,[46] tea,[35] red wine, [11] fruit juices, [47,48] carbonated drinks [49] and mouth rinses.[50] An important determinant of extrinsic discolouration is the surface finish of the restoration which is determined by the finishing and polishing protocol used.[24]

2.4 Finishing and polishing

The nature of composite materials, being composed of a soft resin matrix and hard filler particles make it difficult to finish and polish them to a high lustre because of selective grinding associated with the soft material and harder particles. In addition the final finish of resin composite depends on the filler particle size, morphology, loading and type.[51,52] Instruments used for finishing and polishing tooth restorations include aluminium oxide coated abrasive disks, abrasive strips, and polishing pastes.[25] Finishing and polishing removes the oxygen inhibited surface layer on composite restorations, but also leaves surface defects.[25] The integrity of the surface finishing and polishing can be evaluated by measuring the surface roughness using a profilometer.[18] Different studies have come up with different conclusions as to the best instruments for smooth surface restorations.[10]

The polishing method and the instruments used, including the curing lights for polymerization which must produce a minimum light output of 475mw/cm^2, are also important.[18,51] Therefore the finishing and polishing procedures are both material and technique sensitive. Min *et al*.[53] investigated the surface roughness of two composite resins after different finishing methods and reported that there was a significant correlation between type of composite used, polishing method and surface roughness. Uctasli *et al.*[54] and Ryba *et al*.[55] also reported the same conclusion that surface roughness after polishing depends on inorganic filler component size, size distribution, and loading.

Finishing and polishing composite restorations requires a stepwise approach, with the gradual use of finer instruments in order to methodically remove deeper scratches. Generally it consists of three steps. The first step involves contouring the restoration with either a 3 to 100μm diamond burs, 12 flute carbide burs or course abrasive coated disc. The second step involves finishing with fine or extra fine diamond burs, 16 to 30 flute carbide burs, white stones (aluminium oxide), white

Arkansas stones or medium and fine abrasive coated discs. The final step is polishing which aims to provide an enamel-like lustre. This can be achieved by use of fine and extra fine polishing paste (aluminium oxide or diamond), extra-fine abrasive coated discs, silicon or diamond impregnated rubber polishing disks, cups or points.

It is recommended that the finishing and polishing instruments be used in the proper sequence and operator should use one system from start to finish.[18] The main reason of finishing and polishing restorations is to define anatomical contours and attain a smooth surface. Surface defects on the restoration surface, may increase surface staining, plaque accumulation, gingival irritation.[51,56,57]

Investigators have recommended different polishing techniques to improve colour stability.[58] However, it is has been reported that the surface finish of restorative materials can show significant differences even when they were polished under the same conditions.[59] Curing composite restorations in contact with matrix strips is known to provide smooth surfaces.[60] However this leaves a resin rich surface layer that can be easily worn away exposing the rough inorganic filler.[54] This resin rich layer has a high affinity for water and also staining. [61,62]

Yew *et al*.[63] did a study to evaluate the colour stability of a nano-filled and a micro-filled composite materials on exposure to spices and reported that microhybrid and nanofilled resin composites stained differently when either finished with plastic strip or polished with Soflex discs. Plastic strip finished specimens displayed more colour change compared to Soflex disc finish. This is in agreement with results from studies by Kumari *et al*.[25] and Ergucu *et al*.[64] It has been speculated that this is due to the surface beneath the strip having a lower degree of polymerisation compared to the deeper layers which has not been exposed to oxygen.[25]

2.5 Review of staining solutions for the study

The staining agents used for the study were khat extract (at 2 different concentrations), tea, red wine and distilled water (control).

2.5.1 Khat

The use of khat is prevalent in some parts of the world including the East African countries of

Djibouti, Ethiopia, Kenya, Tanzania and Uganda.[65,66] It is estimated that worldwide over 20 million people regularly use it.[67] It is known by different names in different countries such as qat in Yemen, eschat in Ethopia and miraa in Kenya.[68] There are at least 44 different types of khat available from different geographic regions.[69] The use of khat varies, the plant can be chewed and then swallowed, which is the most common form of usage. Alternatively the dry leaves can be ground to make Abyssinian, African or Arabian tea or the powder can be mixed with honey and eaten as a paste.[65] The psychoactive effects of khat are mainly attributed to cathinone, a phenylpropylamine which is similar in structure and function to amphetamine and cocaine.[70] Cathinone is estimated to have one third the potency of amphetamine.[71] The effect on the user include increased energy and self-esteem, excitement, increased libido.[72]

Khat is considered a drug of abuse by the World Health Organization (WHO) but with less addictive potential compared to alcohol and tobacco. Its use is illegal in some parts of the world but it is legal in countries including Kenya, Uganda, Ethiopia and Eritrea.[68] The estimated prevalence of khat use in Kenya is 3.7% among 16-65 year olds. There is substantial regional variation though, the estimated prevalence in the North Eastern region of Kenya is 28%, for Nairobi its 7.2%, Coast 6.2% and Eastern 5.4%.[73] In total an estimated 1.6 million people use khat. It is the third most consumed substance after alcohol and tobacco. Use is predominantly among males.[74] Khat is grown in the Meru and Mbeere districts in the Eastern regions of Kenya.[75]

A study to assess the prevalence of chewing khat by college students in Gondar town, Ethiopia, found that the most common frequency of khat chewing was once a day, reported by 33.1% of the respondents. Most of the respondents, 53.8% , would spend 1-4 hrs in a khat chewing session.[68] A study on use of khat by Somalis living in the United Kingdom reported an average of three days a week.[66] In Yemeni, around 90% of the adult male population chew khat for 3 to 4 hours a day.[65] Usually a person consumes 100-200 grams per chewing session.[66] Substances used together with khat include alcohol[68] as well as beverages such as cola and cold water.[69] In Kenya khat is customarily wrapped into small bundles which can be purchased at local markets.

The use of khat has been linked to a range of health issues including liver toxicity, gastritis, ischaemic heart disease, oral cancer, hypertension, spermatorrhoea and haemorrhoids.[72] Oral

conditions which have been reported to be associated with khat include periodontal disease, dry mouth, dental caries, oral mucosal lesions and temporomandibular disorders.[66,76]

Staining of teeth from khat chewing has been documented by a number of investigators. This has been attributed to direct staining by chemicals (tannins) in the khat leaves and a change in the oral flora caused by the khat favouring chromogenic bacteria.[77,78] A single study on the effect of khat on colour stability of dental resin composites was available. This was by Dubais[79] who investigated the its effect of khat on nanofill and microhybrid composite materials. The study concluded that khat had a significant effect on colour stability of the resin composite materials and the nanofill was more colour stable compared to the microhybrid.[79] The study however did not address the effect of different polishing protocols and differential concentrations to simulate the role of saliva or other beverages taken with the khat.

2.5.2 Tea

Tea is the second most consumed beverage after water. It is usually prepared by mixing hot water with cured leaves of the plant Camellia sinesis. Kenya is one of the oldest tea growing countries in Africa with tea being introduced in 1903 and commercialized in 1924.[80] Currently Kenya consumes only 5% of its total tea production with the rest being exported mainly to Arabic countries where it is highly consumed.[81]

Types of tea differ depending on the plant with which the tea is made from and the processing of the harvested tea leaves. Based on the processing, they can be classified into black (fermented), green (non-fermented) and oolong (semi-fermented). A number of researchers have reported a wide range of pharmacological, physiological and biochemical effects of different types of tea attributed to their different compositions.[82] Tea consumption has been shown to have positive medical benefits which include prevention of cardiovascular diseases, particularly coronary heart diseases and atherosclerosis.[83] Tea also has anti-aging, anti-diabetic, antibacterial, anticancer, anti-obesity and anti-dental caries effects.[84]

Guler *et al*.[85] investigated the effect of various beverages on colour stability of interim dental composite materials using tea and tea with sugar as two of the staining solutions. They concluded

that tea had a significant effect on the colour stability of resin composite material and the presence of sugar increased the colour difference compared to tea without sugar. Omata *et al*.[86] reported that tea stained dental composites by a mechanism that depended on external conditions such as the presence of chlohexidine which increased the degree of staining.

2.5.3 Red wine

The term "FRENCH PARADOX" has been used to describe the low prevalence of heart disease in the French population even though they consume a diet rich in saturated fats.[87] Many experts suspect this to be due to a high intake of red wine within this population.[88] Red wines are made from dark coloured grape families. Their colour ranges from violet, for young wines, to red and brown for mature wines. The red colour comes from anthocyan pigments found in the skin of the grapes.[89]

In a number of studies, red wine has reported to cause the most significant colour change compared with other solutions.[11] A study by Kisumbi[46] on the colour stability of glass ionomer cements, compomers, and resin composites after exposure to coffee, red wine and coca cola, concluded that red wine caused the most significant change in colour. The colour change of all the specimens surpassed the acceptability tolerance ($\Delta E > 5.5$). This is consistent with results from other studies.[43,85,90, 91]

2.6 Staining technique

A number of techniques have been used in in-vitro experiments to assess colour stability of dental materials such as dental composites, acrylic resins and porcelain. The most commonly used are immersion of specimens in staining solution and subjection to accelerated aging conditions.[82] No studies comparing the two seem to be available.

2.6.1 Immersion in staining solutions

A number of researchers have used immersion into staining solutions over a measured time interval to evaluate colour stability of dental materials. Some authors have used intermittent immersion into the staining solutions alternating with immersion in distilled water or artificial saliva to try imitate oral conditions,[92] while others have used continuous immersion technique with the

specimens being removed from the staining solutions only for colour measurement.[93,94] There is however a wide variation in the time periods in which the specimens are immersed in the staining solutions. A number of investigators who have used continuous staining technique have reported that most of the staining occurs in the first week of immersion[27,95] and so a two weeks immersion period was chosen for this study. The type of staining solution used seems to affect the degree of colour change with coffee and red wine often causing the most severe colour changes to the materials being tested.

2.6.2 Accelerated aging

The accelerated aging process has also been used to determine colour stability of dental materials by a number of investigators.[41,96,97] The process, which initially was designed to evaluate commercial products such as paints, simulates long term exposure to environmental conditions It is estimated that 300 hours of accelerated aging simulates one year of service though it is not certain how such an outdoor simulation relates to intra-oral conditions. [20] Accelerated aging has been used to evaluate colour stability of dental materials since 1978.

2.7 Colour measurement

Colour measurement of a specimen is not only dependant on the colour of the material but also on other factors including the type of incident light in which the colour is measured, dimensions of the specimens, and surface finish of the specimen.[98,99] Colour in dentistry can be measured either by visual or instrumental techniques.[100] Visual evaluation of colour is highly subjective even however a study has shown visual evaluation can be used to determine colour stability using a shade guide.[101,100]

The system commonly used for visual determination of colour is the Munsell colour system. In this system three parameters which are the value, chroma and hue are used to describe the colour. The value is the lightness of a colour, the hue is the quality of the colour that distinguishes one family of colour from another and the chroma is intensity of the hue. Figure 2 is an illustration of the Munsell colour system. Factors such as the type of incident light, fatigue, experience and age, can lead to discrepancies of colour measurements made using this method. [35,94,102]

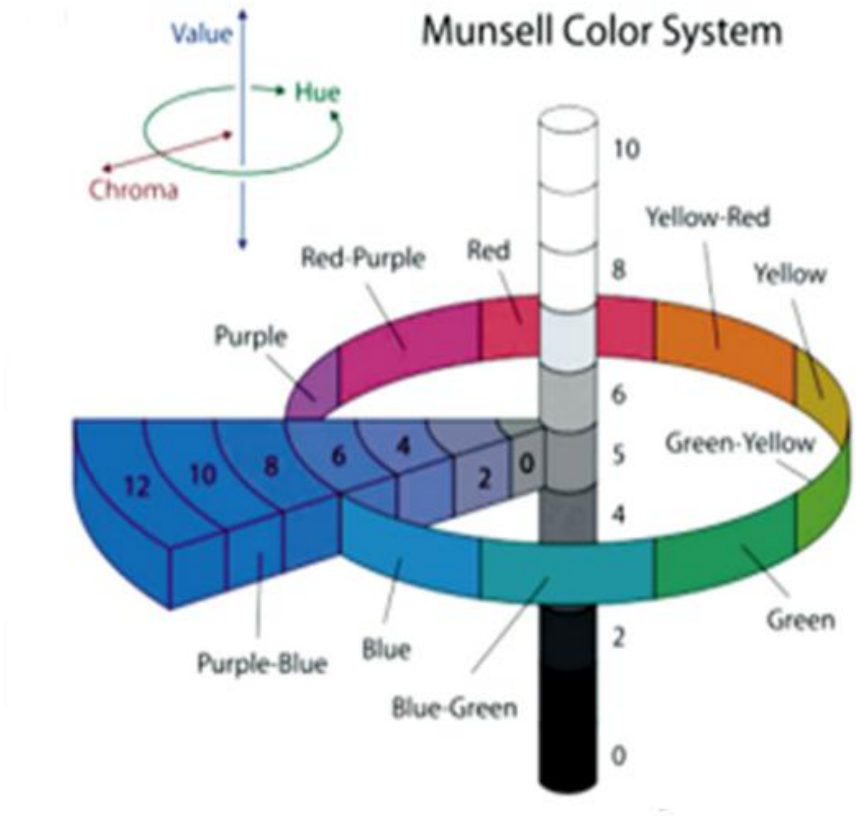

Figure 2: Munsell colour system[103]

In research projects in dentistry, commercial colour measurement instruments or clinical intraoral measuring devices have been used. Studies in which these instruments have been used include comparison between visual and instrumental colour assessment, evaluation of measurement uncertainties and evaluation of colour stability of dental materials. Clinical colour measuring devices are designed to simplify colour assessment in dentistry and can provide additional information such as the matching shade tab, and information to assist with tooth shade communication, replication and confirmation. Colour measuring instruments include spectrophotometers, colorimeters, and imaging systems (digital cameras).[104]

The first colour measuring instruments to be made available were colorimeters. They can be used to measure colour on transparent (transmission) and opaque (reflectance) specimens. They measure tristimulus values and in the red, blue and green region of the visible spectrum.[104] They consist of a light source that is transmitted/reflected from a specimen which passes through filters

and then detected by a photodetector.[105] Digital cameras can also be used to evaluate colour. To get a colour image, sensors on digital cameras capture colour as a product of the three primary colours, red, blue and green, in way similar to the colorimeter. Various approaches can then be used to convert this data into colour information.

Spectrophotometers are considered to be more accurate compared to the above two for colour measurement in dentistry. Unlike colorimeters which measure tristimulus values spectrophotometers evaluate colour via full spectrum colour measurement producing more precise data.[104] Spectrophotometers consist of a source of light, a monochromator and a photodetector.[105] An examples of a spectrophotometer is the Vita Easyshade Compact (Vita Zahnfabrick, Bad Sackingen Germany), which is a hand held, cordless, battery operated, contact type spectrophotometer. Figure 3 illustrates the components of a spectrophotometer.

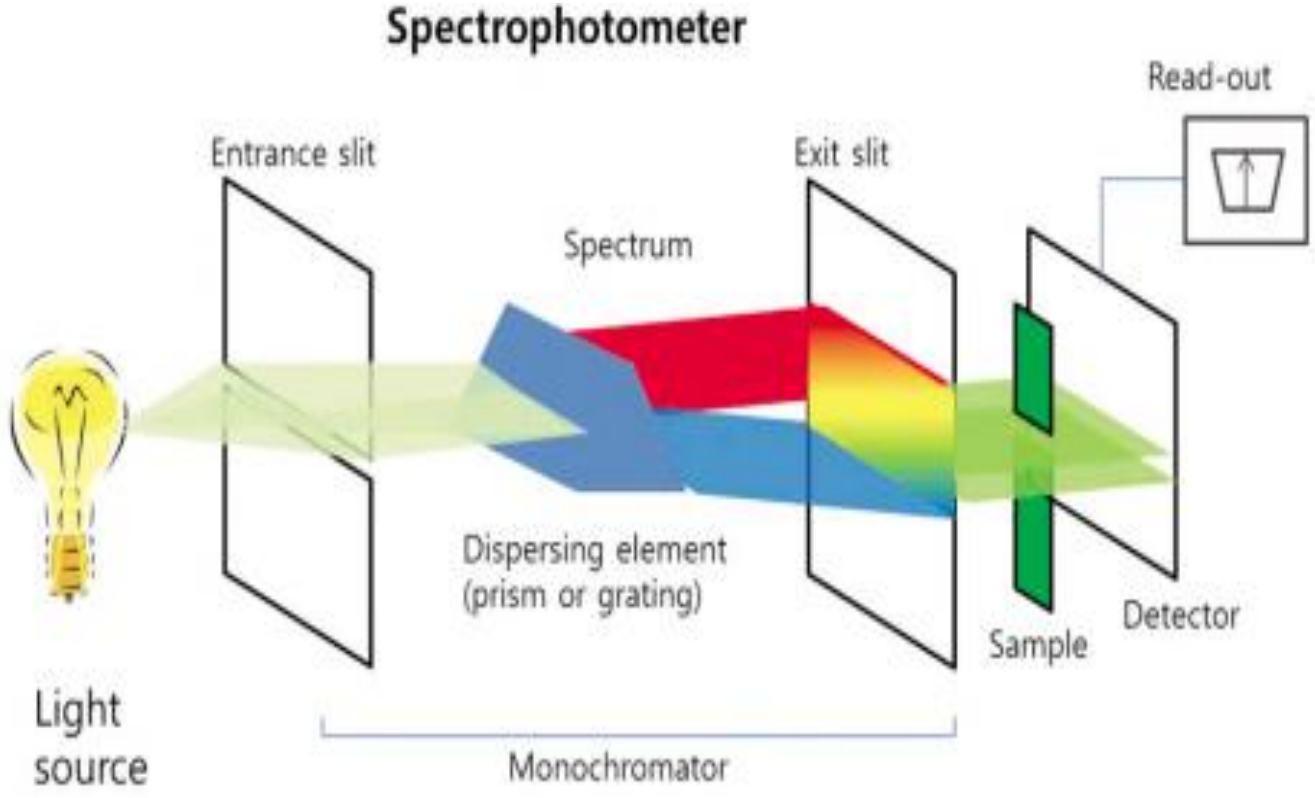

Figure 3: Components of a spectrophotometer [103]

In this study a spectrophotometer will be used to measure the colour change using the CIE-Lab-colour (Commission International del' Eclairage L*a*b colour) system. The CIE system

quantifies colour using three colorimetric measurements. L* characterizes the lightness of the colour and can be ranged between 0 for perfect black and 100 for perfect white while a* and b* are chromaticity coordinates defining a colour in the red-green axis and yellow-blue axis respectively. A positive a* value reflects the red colour range while a negative value indicates the green colour range. In the same manner positive b* values shows the yellow colour range and negative values shows the blue colour range. The colour difference (ΔE) can be calculated from ΔL*, Δa*, Δb* values using the formula:

$$\Delta E = \sqrt{((\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2)}.$$ [100]

Figure 4 is an illustration of the CIE-Lab colour system.

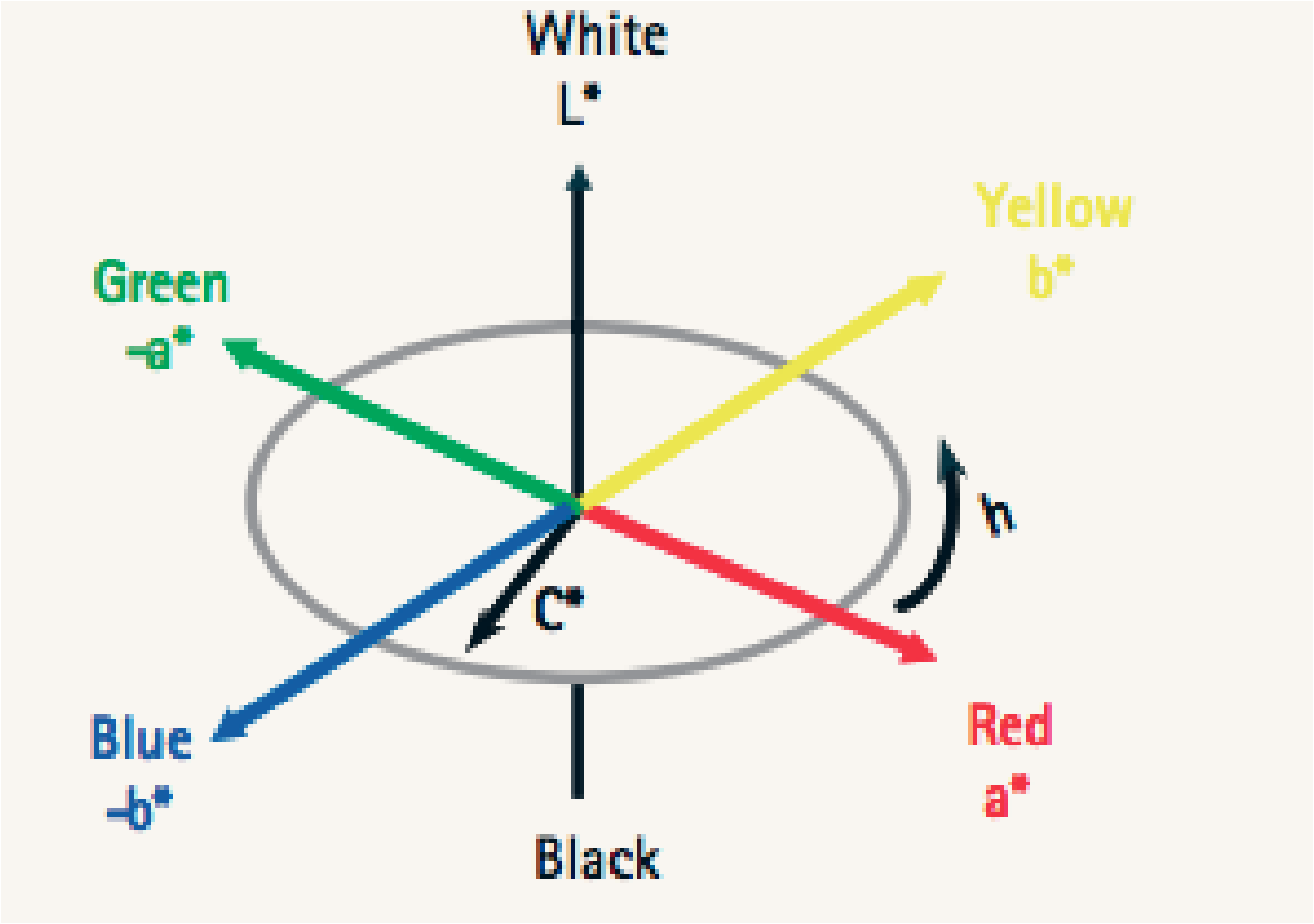

Figure 4: Representation of CIEL*a*b colour system. [105]

Several studies have investigated the perceptibility and acceptability tolerances for differences in shades of dental restorative materials. Kuehni and Markus[106] reported a perceptibility tolerance of 1 ΔE unit for half of sample and Seghi *et al*.[107] reported that a colour difference of 2 ΔE units was correctly judged by all the sample subjects. On acceptability tolerances, Ruyter et al[108] considered ΔE to be unacceptable at approximately 3.3 units. Ragain and Johnstone[109] reported that at a ΔE of 2.72 units, observers had a 50% chance of accepting or rejecting the colour mismatch. These studies were however done under in vitro conditions and have limited clinical applications. The colour matching tolerances considered for this study were determined in an in vivo study by Douglas *et al.*[110] They reported that the ΔE at which half of the dentist sample may imply a colour mismatch was 2.6 units and the ΔE at which the colour mismatch was considered clinically unacceptable was 5.5 units. Colour evaluation after staining beverages has been done, however the effect of khat and polishing protocols is lacking hence the aim of this study was to evaluate the effect of surface finishing protocol and exposure to staining solutions on the colour stability of dental resin composite restorative materials.

CHAPTER THREE: PROBLEM STATEMENT, STUDY JUSTIFICATION AND OBJECTIVES

3.1 Problem statement

Restorative dentistry is an invasive and irreversible procedure and consequently failed restorations need to be replaced. This places the tooth in a restorative cycle that continues throughout the life of the patient. The restorative cycle consists of three major events; first there is the initial loss of hard tooth tissue due to trauma or initial disease. Next there is further tooth tissue loss from preparing the tooth for the restoration and finally more tooth tissue is lost when the restoration fails and has to be replaced. With need for replacement of the restoration the cycle is repeated.[7]

In the oral environment, restorations are exposed to combined effects of moisture, stain and mechanical wear which can cause discolouration and restoration failure.[14] A number of investigators have evaluated the clinical longevity of dental composite restorations and these have reported an average longevity of 6 to 10 years. [3,7] A study by Mjor *et al*.[111] on the reasons for replacement of composite restorations in permanent teeth reported that bulk discolouration was the third most common reason.[112] A survey on "initial placement and replacement of restorations in clinical practice" by Deligeorgi et al.[113] showed a ratio of ranging from 1:1.1 to 1:3.8 and discolouration was among the main reasons for failure of aesthetic restorations.

One of the important factors, which influence colour stability of resin composite restorations, is the finishing and polishing. When the proper technique is followed, finishing and polishing enhances the aesthetic result and maximizes patient's oral health. Unfortunately the proper sequence of polishing steps is often overlooked.[114] This is because clinicians are often in a hurry to complete the procedure or fail to invest in proper polishing kits. This may shorten the longevity of the restoration and increase expenses to the patient by necessitating repeat restorative procedures.

3.2 Justification of the study

Colour instability can result in the restoration failure and consequently acceleration of the restoration cycle. At the same time, progressive generations of restorations would have increasingly shorter restorative cycles due to an increase in the size of the restorations and

weakening of the tooth.[3] As colour instability can be due to extrinsic factors, it is important to consider the type of diet of a particular population when evaluating discolouration potential.[115] Need for restoration replacement affects the patients' image and psychology. It also results in inconvenience and additional costs to the patients.

In order to realize the full benefits of dental resin composite restorations, it is important for dentists to appreciate the importance of proper finishing and polishing techniques and to incorporate these into everyday practice.[116]

3.3 Objectives

3.3.1 Broad objectives

To evaluate the effect of surface finishing protocol and exposure to staining solutions on the colour stability of dental resin composite restorative materials.

3.3.2 Specific objectives

1. To determine the effect of surface finishing protocol on colour stability of nanofill and micro-hybrid dental resin composite restorative material.
2. To determine the effect of immersion in staining solutions on colour stability of nanofill and micro-hybrid dental resin composite restorative material.
3. To compare the colour stability of micro-hybrid and nanofill dental resin composite materials subjected to different finishing protocols in different staining solutions.

3.4 Hypothesis

3.4.1 Null hypothesis

1. There is no significant difference in the colour stability of nanofill and micro-hybrid dental resin composite restorative materials with different surface finishing treatments.
2. There is no significant difference in the colour stability of nanofill and micro-hybrid dental resin composite restorative materials when exposed to different staining solutions.
3. There is no significant difference in the colour stability of micro-hybrid and nanofill dental resin composite materials subjected to different polishing protocols when exposed to different staining solutions.

3.4.2 Alternative hypothesis

1. There is a significant difference in the colour stability of nanofill and micro-hybrid dental resin composite restorative materials with different surface finishing treatments.
2. There is a significant difference in the colour stability of nanofill and micro-hybrid dental resin composite restorative materials when exposed to different staining solutions.
3. There is a significant difference in the colour stability of micro-hybrid and nanofill dental resin composite materials following different finishing protocol when exposed to different staining solutions

3.5 Variables

Table 1: Variables

Independent variables	Measurements
Staining solutions	Khat extract (at 2 different concentrations) Tea Red wine
Polishing protocols	Mylar finish Aluminium oxide coated polishing discs White polishing stone
Resin composite materials	Nano-fill composite material Micro-hybrid resin composite material
Dependant variable	
Colour change	Total colour difference (ΔE) within and between polishing and staining groups.

CHAPTER FOUR: MATERIALS AND METHODS

4.1 Study design

The study was a laboratory based experimental study which was conducted at the University of Nairobi, School of Dental Sciences, Dental Prosthetics laboratory and School of pharmacy, Pharmaceutical chemistry laboratory. A pilot study with 10% of the samples was done.

4.2 Study sample

The effect of polishing protocol and three staining solutions on the colour stability of two resin composite materials marketed for aesthetic restorations was assessed. The resin composite materials that were used for the study were a nanofill composite, *Filtek Z350 XT* (3M ESPE St Paul, MN, USA) and a microhybrid composite, *Vit-l-escence* (Ultradent Inc, South Jordan, UT, USA). The composite materials and the polishing techniques investigated are the ones commonly used at the University of Nairobi, Dental School, Conservative dentistry clinic and also thought to be commonly used among dental clinics in Nairobi which has the largest number of dentists practicing restorative dentistry in Kenya. The immersion solutions were obtained from common foods with the highest potential for staining, as has been shown in similar studies.

4.3 Sample size calculation

International Organisation for Standardisation (ISO) 7491:2000 for determination of colour stability of dental materials was used for the study. However it does not specify the sample size per evaluation. Therefore the Sample size was based on similar studies evaluating colour stability of dental resin composite materials.[2,43,96,95] In these studies, 5 specimens where used for each test group. The study sample was n =150 specimens, 75 from *Vit-l-escence* and 75 from *Filtek Z 350 XT*. Table 2 summarizes the sample size calculation.

Table 2: Summary of the sample size

Material	Polishing protocol	Khat (K1)	Khat (K2)	Tea (T)	Red wine (R)	Distilled water (C)	No. of Specimens
Filtek Z350 XT	Mylar strip (M)	5	5	5	5	5	25
	Soflex discs (D)	5	5	5	5	5	25
	Arkansas stone (S)	5	5	5	5	5	25
Vit-l-escence	Mylar strip (M)	5	5	5	5	5	25
	Soflex discs (D)	5	5	5	5	5	25
	White polishing stone (S)	5	5	5	5	5	25
Total		30	30	30	30	30	150

4.4 Preparation of resin composite specimens

The same batch of resin composite materials, Table 3, were used to fabricate 75 specimens of each material. The composition of these resin composites materials are also presented in Table 3. A single shade A2 was utilized to assure constancy of shade at baseline. The curing light used (Blueluxcer M385, Monitex Industrial Co. Ltd, Taiwan) for photo-polymerisation was calibrated using a radiometer before use and was determined to have an output of 1000 mW/cm^2.

Table 3: Types and composition of composite materials used for the study

Material	Composite resin type	Manufacturer	Monomer matrix	Filler: size, proportion (volume/weight)	Batch number and expiry date
Filtek Z350 XT	Nanofill Composite	3M ESPE St Paul, MN, USA.	Bis-GMA, UDMA, BIS-EMA , TEGDMA	Nanofillers of silicon(5-75nm), zircon/silicon nanoclusters (0.6-1.4µm). 63.3% vol	N972732 28/05/2021
Vit-l-escence	Micro-hybrid composite	Ultradent Inc, South Jordan, UT, USA	Bis-GMA and diluents	Barium aluminoborosilicate (0.4-0.6 µm) including silica filler particles 0.04 to 0.1 µm, 52% by vol	BG77R 30/05/2023

Figure 5: Filtek Z350 XT (3M ESPE) composite used for the study

Figure 6: Vit-l-escence (Ultradent Inc) composites used for the study

4.5 Specimen fabrication

A perspex mould with dimensions of 8mm diameter and 2mm width was used to fabricate the specimens. A single layer of the uncured composite material was put into a mould with a segment of dental floss attached. Care was taken during placement of the material in the mould to avoid air entrapment that may cause porosities. The light curing unit was held at 90^0 approximately 1mm away from the specimen and cured on alternate sides twice for 20 seconds. The dental floss segment, secured at the edge of the specimen, was used to suspend the specimens in the staining solutions and to attach a piece of adhesive tape to label each specimen, Figure 7. The specimens for the mylar finish where cured with mylar strips on both sides of the specimen. Those for the two other polishing groups had a mylar strip on one side and the other side left exposed during curing. These specimens were then divided into the 2 polishing groups and finished on the side left exposed.

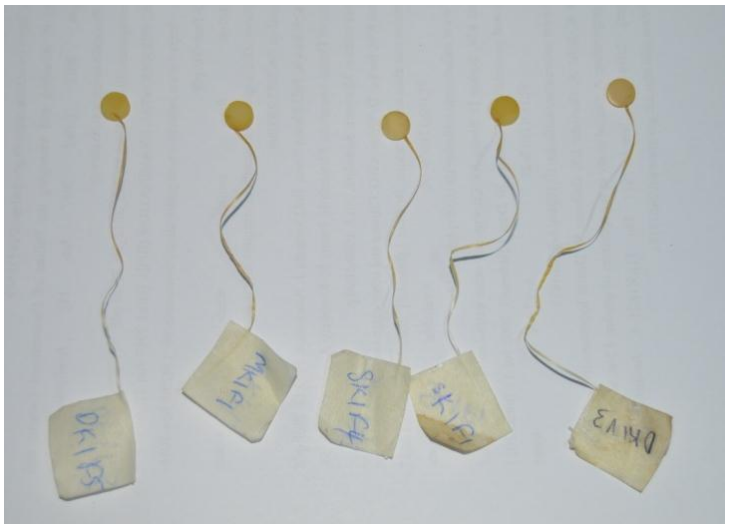

Figure 7: Prepared specimens

4.6 Polishing groups and procedures

Three finishing and polishing protocols were evaluated: mylar strip finish, *Soflex* contouring and polishing disks (3M ESPE St Paul, MN, USA) and white polishing stones (Prime dental, UK), Table 4. Four staining solutions were used, namely tea, red wine and khat plant extract (diluted to 2 different concentrations). Distilled water was used as the control.

Table 4: Polishing materials used for the study

Polishing material	Manufacturer	Product information
Mylar strip	Maquira industries, Brazil	Mylar polyester film.
Soflex contouring and finishing disks	3M ESPE St Paul, MN, USA	Flexible aluminium oxide coated polishing discs.[117]
White stone burs	Prima Dental, UK	Pieces of micro-crystalline quatz attached to a metal shank.

4.6.1 Mylar strip finish

This group was left unpolished after curing with a mylar strip over the surface layer of the composite. Figure 8 is a picture of the mylar strips used in the study.

Figure 8: Mylar strips (Maquira Industries, Brazil)

4.6.2 Soflex contouring and polishing discs finish

Course, medium, fine and superfine grit sequence held with a slow speed hand piece were used for 30 seconds each. After use of each polishing disc, the specimens were rinsed and air-dried for 10 seconds before the next step. Figure 9 is a picture of the Soflex disc kit used in the study.

Figure 9: Soflex polishing discs (3M ESPE)[118]

4.6.3 White polishing stone burs finish

The burs were used for 2 minutes with a slow-speed hand piece with minimal pressure. Figure 10 is a picture of the white polishing stones used in the study.

Figure 10: White polishing stone

After the finishing treatments, all the specimens were placed in distilled water at room temperature for 24 hours to allow for rehydration and completion of polymerization,[95,92] after which baseline colour measurements were recorded.

4.7 Preparation of staining solutions

Table 5 shows details of the three staining solutions for the study. Distilled water was the control solution. A pH meter (Jenway 3510 pH meter, Keison products, UK), Figure 11, was used to determine the pH of the staining solutions. The pH meter was calibrated before use.

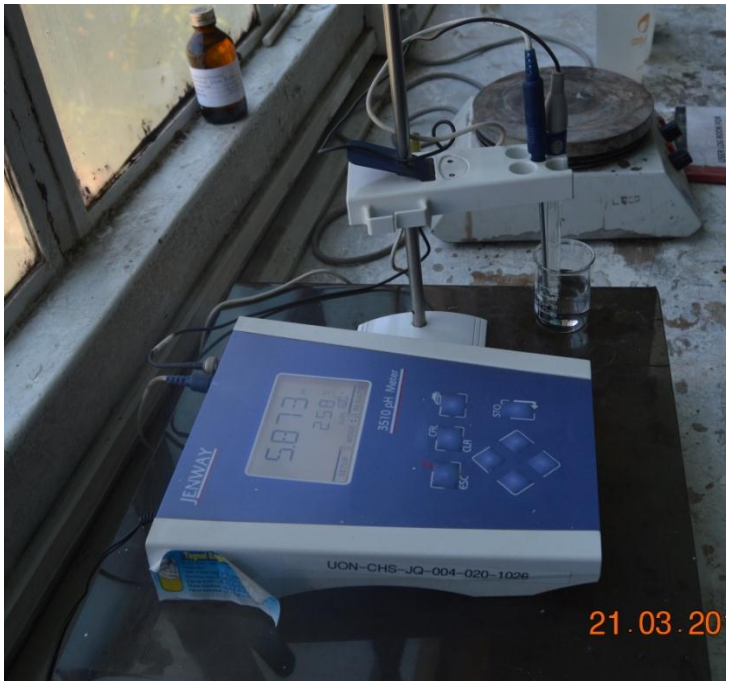

Figure 11: Calibration of Jenway 3510 pH meter (Keison products, UK)

Table 5*:* Staining solutions used and their manufacturers

Staining solution Brand name	Manufacturer/region of origin	pH
Khat extract	Meru County, Kenya -	5.1
Ketepa tea	Kenya Tea Packers Ltd, Nairobi, Kenya	5.5
Red wine	Robertson's red wine, Robertson Winery, South Africa.	3.4

The specimens were then suspended in the solutions with the aid of the embedded floss for the specified amount of time, Figure 12. The test solutions were refreshed once a week. The solutions to be used for the experiment were prepared as follows.

Figure 12: Specimens storage

4.7.1 Tea

The tea was prepared in accordance with the manufacturer's instructions. 250 ml of water was brought to a boil and 1 tea bag of Ketepa tea (2g), Figure 13, was added and the tea solution left to brew for four minutes. The tea was allowed to cool before being used. No sugar was added. The presence of sugar in some drinks increases colour difference and this is thought to be due to the sugar making the staining drink sticky.[10]

Figure 13: Ketepa tea

4.7.2 Khat extract

The extraction protocol for khat used was the same protocol used by Dimba[72] in an in vitro study for the cytotoxicity of khat. This extraction protocol is a modification of the methanolic extraction protocol described by Lee[71] excluding the alkaloid purification, so as to minimize acid or basic residues in the extract which might act as confounders. Fresh khat shots were chopped into small pieces of approximately $5mm^2$ in size and methanol was added to cover the plant material. The mixture was agitated in a sonicator at room temperature and filtered through an 11µm filter, Figure 15. The green methanolic extract was then placed into a rotorvapor vacuum drier at 337 millibar until all the methanol evaporated. Figure 16. The extract was then refrigerated awaiting testing.

In total 870 grams of khat, Figure 14, was processed and it produced 10 grams of khat extract, Figure 17, giving a dilution ration of 87. This dilution factor was used to formulate the khat solution designated K1, Figure 18. To mimic the effect of saliva when the khat plant is being chewed, a second solution of khat (K2) was made at 1:3 dilution of K1.

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Figure 14: Khat plant

Figure 15: Methanolic extract

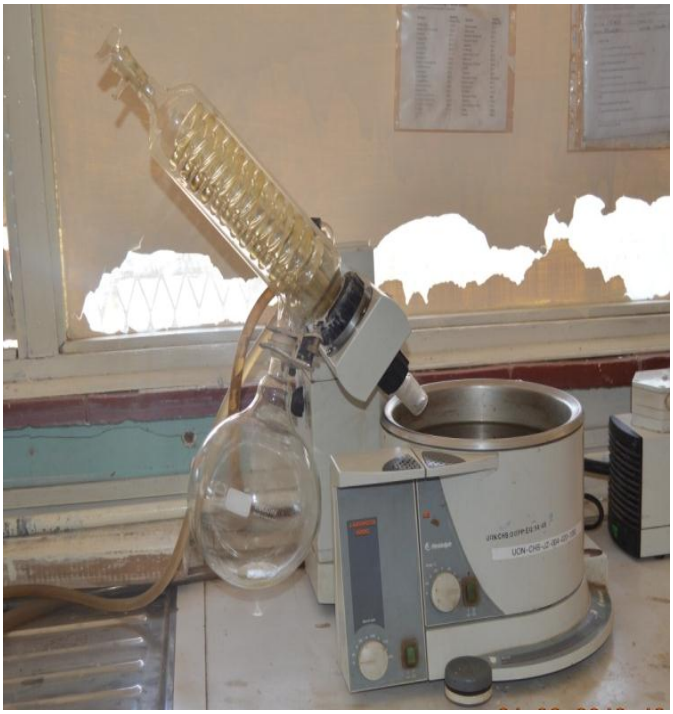

Figure 16: Rotorvapour used in study

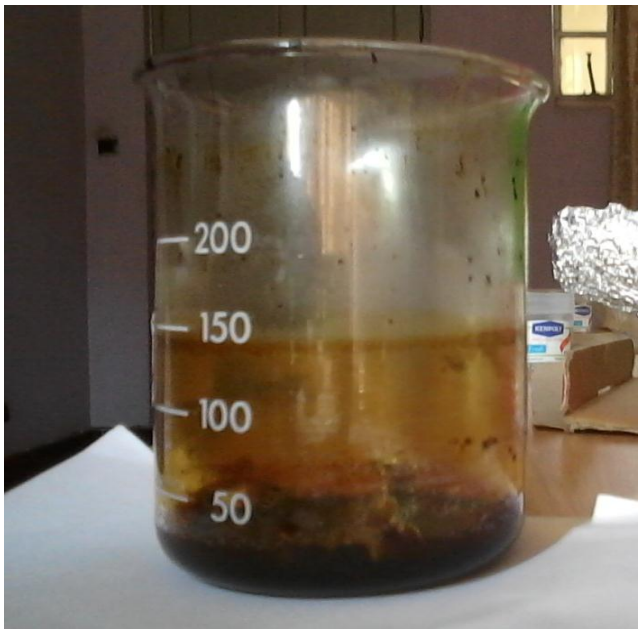

Figure 17: Khat extract

Figure 18: Khat extract diluted with distilled water

4.7.3 Red wine

The red wine which was used for this study was Robertsons' red wine, Figure 19. It had an alcohol content of 7.5% and was used as is.

Figure 19: Red wine (Robertson winery)

4.7.4 Distilled water: control group

Distilled water was the control for staining solutions.

4.8 Codes for experimental groups

The experimental groups were coded as illustrated in Tables 6 and 7 and organized as illustrated in Table 8.

Table 6: Codes for the Vit-l-escence (V) experimental groups

	Khat (K1)	Khat (K2)	Tea (T)	Red wine (R)	Distilled water (C)
Mylar strip (M)	MK1V	MK2V	MTV	MRV	MCV
Soflex discs (D)	DK1V	DK2V	DTV	DRV	DCV
White polishing stone (S)	SK1V	SK2V	STV	SRV	SCV

Table 7: Codes for the Filtrek (F) experimental groups

	Khat (K1)	Khat (K2)	Tea (T)	Red wine (R)	Distilled water (C)
Mylar strip (M)	MK1F	MK2F	MTF	MRF	MCF
Soflex discs (D)	DK1F	DK2F	DTF	DRF	DCF
White polishing stone (S)	SK1F	SK2F	STF	SRF	SCF

Table 8*:* Experimental groups for each material

Surface finishing	Staining	Number of specimens	Staining durations
Mylar strip/Soflex discs/white polishing stone	Tea specimens	5	6 hours
			24 hours (1 day)
			48 hours (2 days)
			96 hours (4 days)
			168 hours (7 days)
			240 hours (10 days)
			336 hours (14 days)
	Red wine	5	6 hours
			24 hours (1 days)
			48 hours (2 days)
			96 hours (4 days)
			168 hours (7 days)
			240 hours (10 days
			336 hours (14 days)
	Khat extract (K1)	5	6 hours
			24 hours (1 days)
			48 hours (2 days)
			96 hours (4 days)
			168 hours (7 days)
			240 hours (10 days)
			336 hours (14 days)
	Khat extract diluted 1:3 (K2)	5	6 hours
			24 hours (1 days)
			48 hours (2 days)
			96 hours (4 days)
			168 hours (7 days)
			240 hours (10 days)
			336 hours (14 days)
	Distilled water	5	6 hours
			24 hours (1 day)
			48 hours (2 days)
			96 hours (4 days)
			168 hours (7 days)
			240 hours (10 days)
			336urs (14 days)

4.9 Colour measurement

At each of the colour measurement stages, the specimens were rinsed with under running water for 5 minutes water and bloated dry with a paper towel. Colour reading was taken using a digital spectrophotometer (Vita Easyshade, Vita Zahnfabrik, Bad Sackingen, Germany), Figure 20. Colour measurement was done by placing the samples on a white background. This was to avoid absorption effects on any colour parameters.[37] The colour change for all the specimens were measured and the average calculated. The probe tip of the spectrophotometer was 6mm in diameter. One reading was taken from the center of each specimen. The spectrophotometer measured the colour using the L, a and b coordinates according to the CIE-Lab-colour (Commission International de l' Eclairage L*a*b colour) system. The colour difference (ΔE) was then calculated from the formula $\Delta E = \sqrt{((\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2)}$, where ΔL*, Δa* and Δb* are the differences of the measured coordinates from the baseline readings of each specimen. After this, the specimens were re-suspended into their respective solutions until the 14th day. The embedded floss was placed at the edge of the specimens so as not to interfere with the surface analysis

Figure 20: Vita easy shade digital spectrophotometer. (Vita Zahnfabrik)

4.10 Statistical analysis

The data was entered into a computer using the IBM Statistical Package for Social Sciences (SPSS), Version 22. All statistical tests were considered significant at an alpha level of 0.05. Two-Way Analysis of Variances (ANOVA) test was used to determine the statistical significance amongst the finishing protocols and staining solution groups and Tukey HSD post hoc test was used to detect the differences within each group.

An Independent-Samples t-test was used to determine the statistical significance between colour at baseline and at the end of staining between the two different composite resin materials.

The results were presented using text, charts, tables and figures using American Psychological Association (APA) format.

4.11 Ethical considerations

The research proposal was submitted to the Kenyatta National Hospital-University of Nairobi Ethics and Research Committee for approval. Approval number P574/08/2018, Appendix 9.

4.12 Limitations of the study

- The experiment was done at room temperature which is different from the oral environment which could influence the colour stability of the materials.
- The specimens were flat whereas clinically, restorations have irregular shapes
- The specimens were exposed to the staining solutions continuously will in reality exposure is intermittent.
- In the tea staining solution the effect of sugar which is typically taken with tea was not evaluated.
- Surface roughness of the specimens was not analysed after finishing and polishing the specimens.

CHAPTER FIVE: RESULTS

At the end of the staining period all specimens, except Soflex disc finished – Vit-l-escence specimens in the khat 1 and khat 2 staining solutions, demonstrated a clinically unacceptable ΔE. All the control specimens demonstrated a ΔE below the perceptible level.

5.1 Comparison of type of resin composite and colour stability

In this section, the mean ΔE of the specimens from the two materials were compared for all the finishing protocols in all of the staining solution. An Independent-Samples t-test was used to determine the statistical significance between colour at baseline and at the end of staining between the two composite resin materials.

5.1.1 Comparison of mylar finished specimens

In this section, the mean ΔE of mylar finished specimens of the two materials were compared in the four different staining solutions.

5.1.1.1 Effect of red wine on mylar finished specimens

The mean ΔE of mylar finished specimens for Vit-l-escence and Filtek in red wine compared to the control is illustrated in Figure 21. Filtek specimens were more colour stable for the entire duration of the staining. The mean ΔE between the two composite materials at the end of two weeks was statistically significant, p=0.001, Table 9.

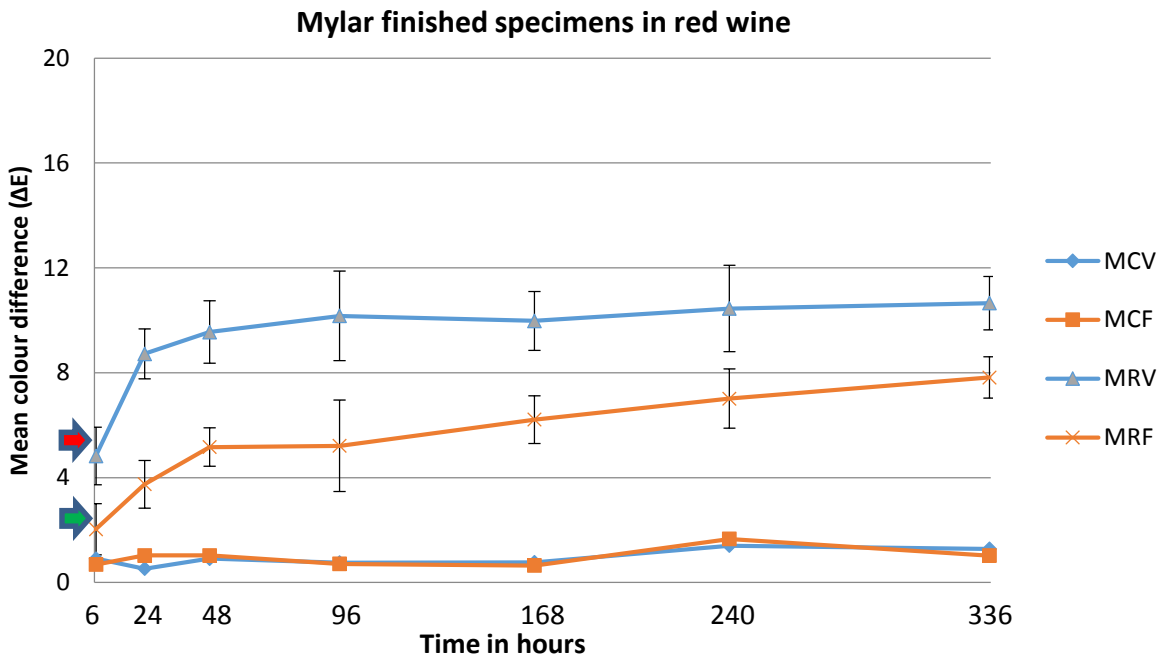

Figure 21: Mean colour difference (ΔE) for mylar finished specimens of the two materials in red wine showing the standard errors compared to the controls. (M-mylar finish, C-control, R-red wine, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 9: Statistical results of the mean colour difference at the end of two weeks comparing mylar finished Vit-l-escence and Filtek specimens in red wine using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*P*
				Lower Bound	Upper Bound			
MRV	5	10.66	1.02	1.50	4.57	8	4.902**	0.001
MRF	5	7.82	0.79					

**p<0.01

5.1.1.2 Effect of tea on mylar finished specimens

The mean ΔE of mylar finished specimens for Vit-l-escence and Filtek in tea and the controls are presented in Figure 22. Vit-l-escence specimens had less mean ΔE with the exception of readings taken at 240 hours. Table 10 shows the results of statistical analysis using independent samples t-test. The mean ΔE between the two materials was not statistically significant p=0.144.

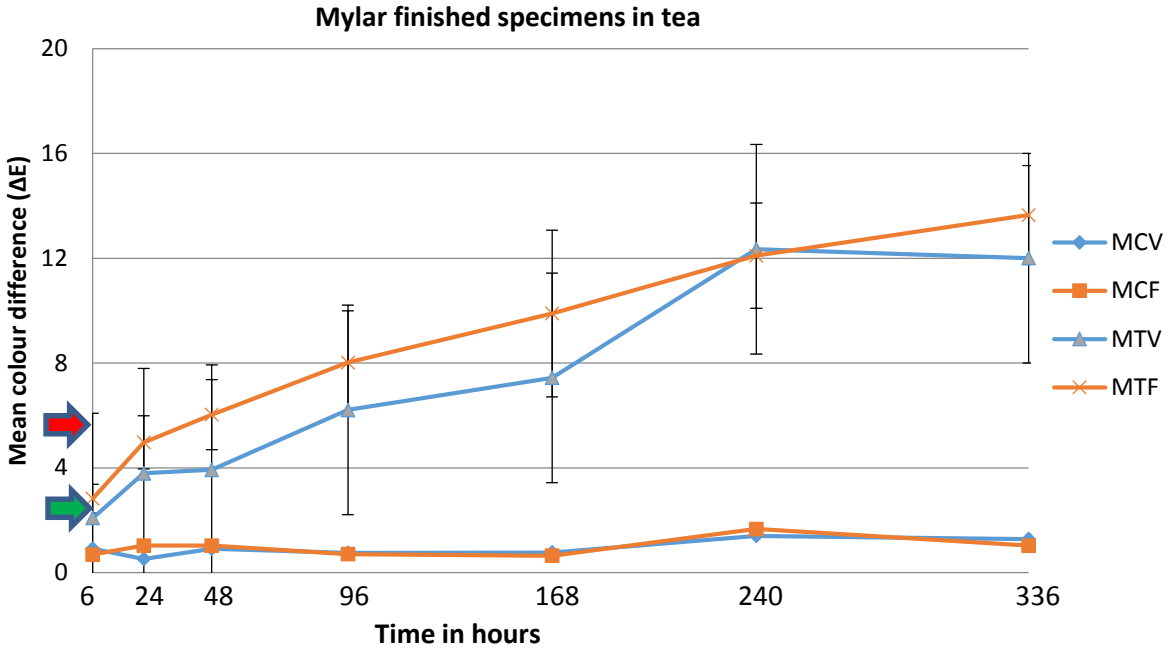

Figure 22: Mean colour difference (ΔE) of mylar finished specimens in tea showing the standard errors compared to the controls. (M-mylar finish, C-control, T-tea, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 10: Statistical results of the mean colour difference at the end of two weeks comparing mylar finished Vit-l-escence and Filtek specimens in tea using Independent-Sample t test

				95% Confidence Interval				
Specimen	*n*	*M*	*SD*	Lower Bound	Upper Bound	*df*	*t* test	*P*
MTV	5	12.00	1.27	-3.99	0.70	8	1.618	0.144
MTF	5	13.65	1.89					

5.1.1.3 Effect of khat 1 on mylar finished specimens

The mean ΔE of mylar finished specimens for Vit-l-escence and Filtek in khat 1 compared to the controls are presented with line graphs in Figure 23. Filtek specimens were more colour stable for the duration of the staining period. Results of statistical analysis for this group using independent t tests are shown in table 11. The mean ΔE between the two materials was statistically significant p=0.012.

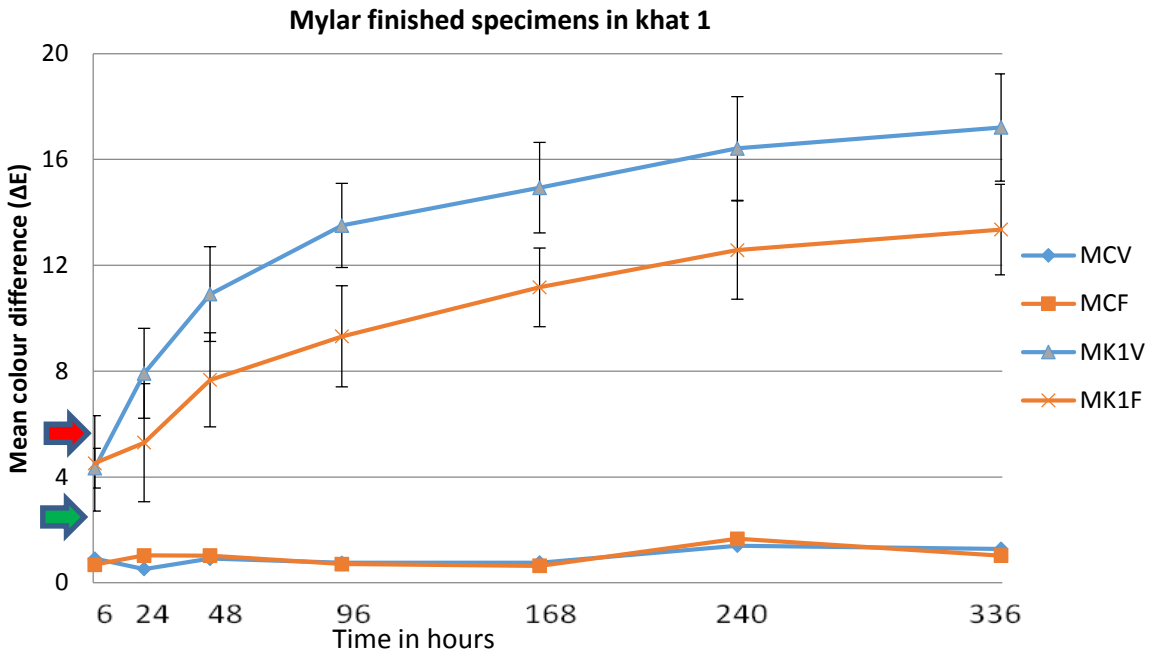

Figure 23: Mean colour difference (ΔE) for mylar finished specimens in khat 1 showing the standard errors compared to the controls. (M-mylar finish, C-control, K1-khat 1, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 11: Statistical results of the mean colour difference at the end of two weeks comparing mylar finished Vit-l-escence and Filtek specimens in khat 1 red wine using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*P*
				Lower Bound	Upper Bound			
MK1V	5	17.21	2.03	1.12	6.59	8	3.252*	0.012
MK1F	5	13.35	1.71					

*p<0.05

5.1.1.4 Effect of khat 2 on mylar finished specimens

The mean ΔE of mylar finished specimens for Vit-l-escence and Filtek in khat 2 compared to the controls are presented with line graphs in Figure 24. Filtek specimens were more colour stable for the duration of the staining period. Results of statistical analysis for this group using independent t tests are shown in table 12. The mean ΔE between the two materials was not statistically significant p=0.087.

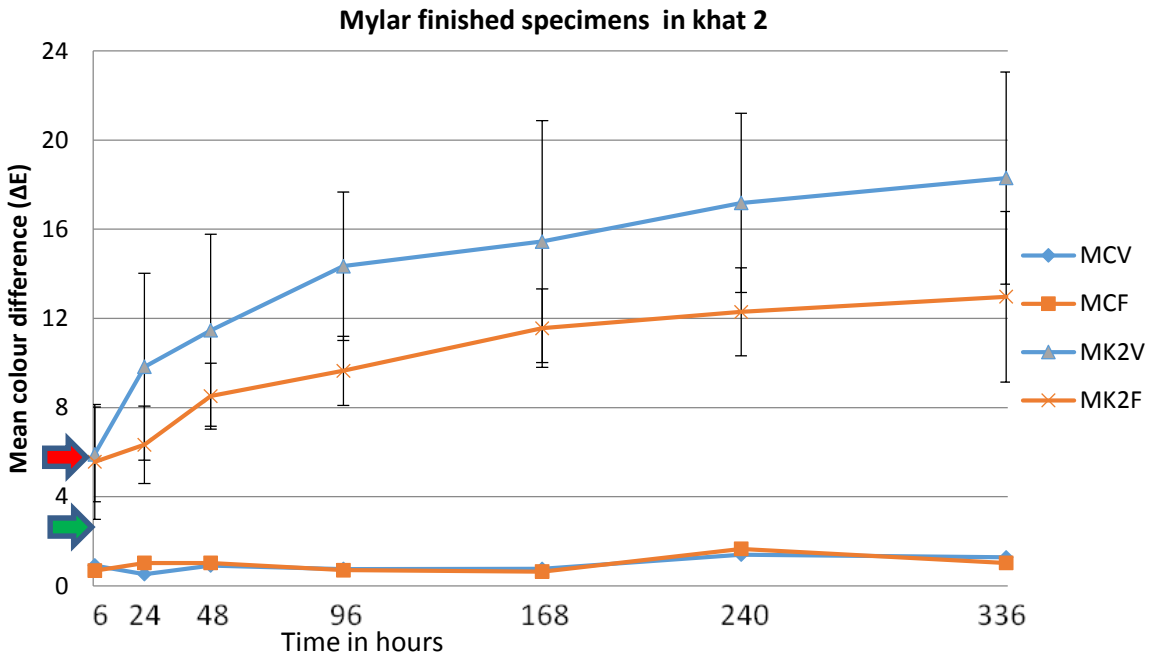

Figure 24: Mean colour difference (ΔE) for mylar finished specimens in khat 2 showing the standard errors compared to the controls. . (M-mylar finish, C-control, K2-khat 2, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 12: Statistical results of the mean colour difference at the end of two weeks comparing mylar finished Vit-l-escence and Filtek specimens in khat 2 using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*P*
				Lower Bound	Upper Bound			
MK2V	5	18.30	4.76	-0.97	11.63	8	1.950	0.087
MK2F	5	12.97	3.83					

5.1.2 Comparison of Soflex disc finished specimens

In this section Soflex disc finished specimens for both materials were compared in the four different staining solutions, that is, tea, red wine, khat 1 and khat 2.

5.1.2.1 Effect of tea on Soflex disc finished specimens

The mean ΔE of Soflex disc finished specimens for Vit-l-escence and Filtek in tea compared to the controls are presented with line graphs in Figure 25. The two materials had comparable colour

difference. Results of statistical analysis for this group using independent t tests are shown in table 13. The mean ΔE between the two materials was not statistically significant, p=0.53,

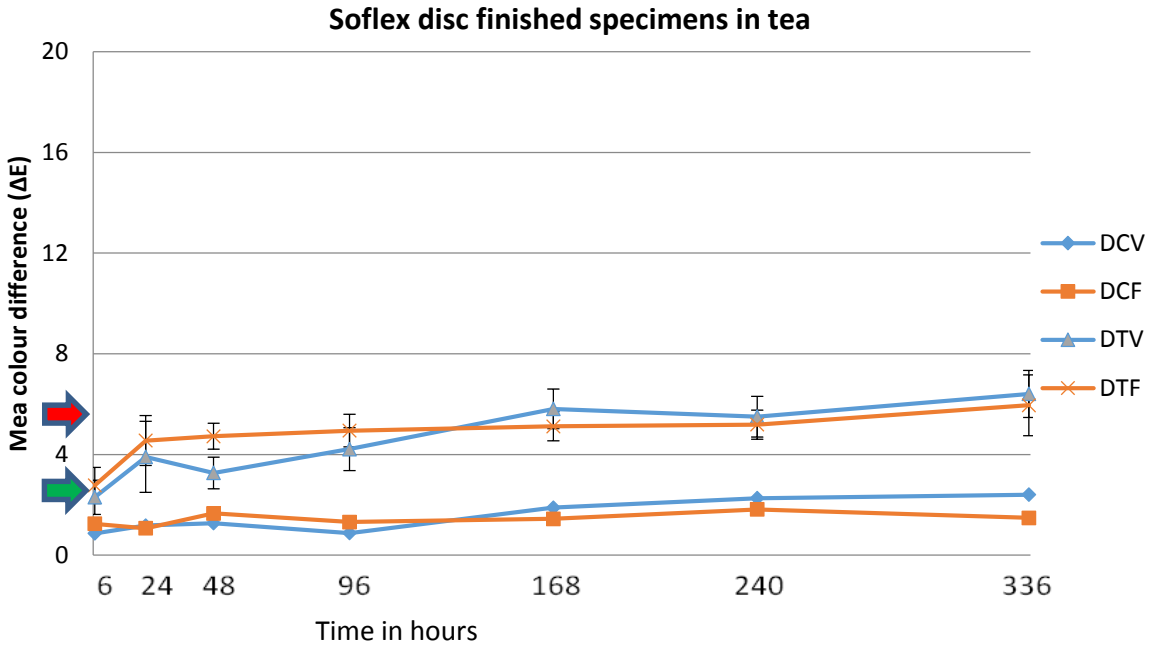

Figure 25: Mean colour difference (ΔE) for Soflex disc specimens in tea showing the standard errors compared to the controls (D-Soflex disc finish, C-control, T-tea, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 13: Statistical results of the mean colour difference at the end of two weeks comparing Soflex disc finished Vit-l-escence and Filtek specimens in tea using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*p*
				Lower Bound	Upper Bound			
DTV	5	6.41	0.93	-1.13	2.02	8	0.657	0.530
DTF	5	5.96	1.21					

5.1.2.2 Effect of red wine on Soflex disc finished specimens

The mean ΔE of Soflex disc finished specimens for Vit-l-escence and Filtek in red wine compared to the controls are presented with line graphs in Figure 26. The mean ΔE of the two materials were comparable. Results of statistical analysis for this group using independent t tests are shown in

table 14. The mean ΔE between the two materials was not statistically significant, p=0.971.

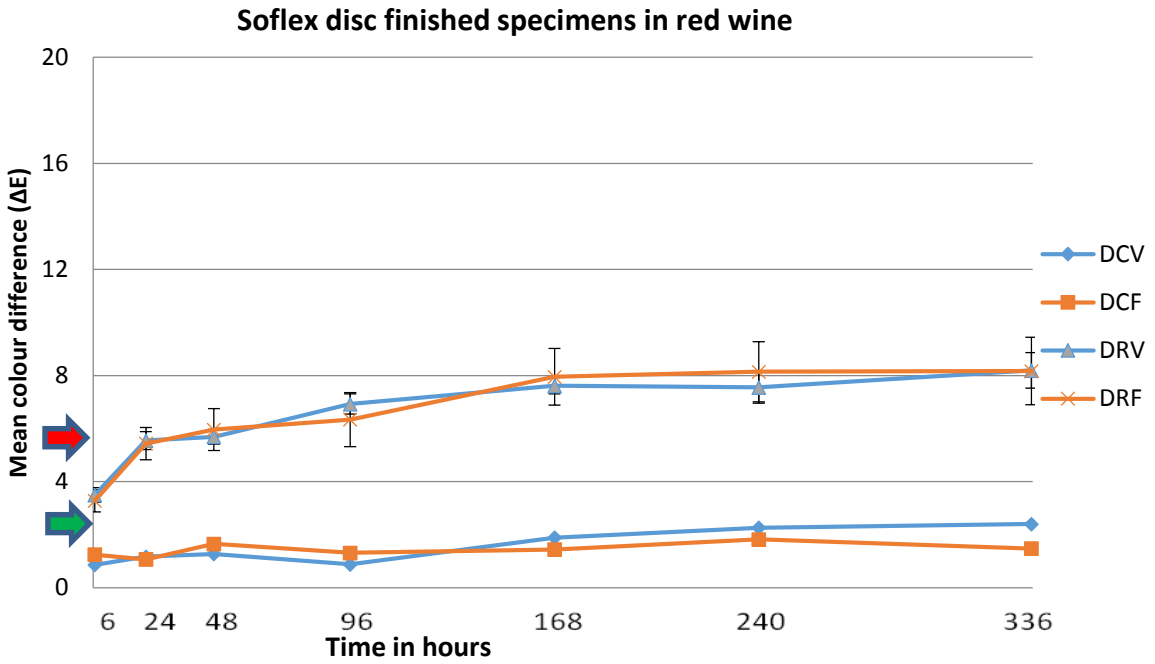

Figure 26: Mean colour difference (ΔE) for Soflex disc finished specimens in red wine showing the standard errors compared to the controls (D-Soflex disc finish, C-control, R-red wine, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 14: Statistical results of the mean colour difference at the end of two weeks comparing Soflex disc finished Vit-l-escence and Filtek specimens in red wine using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*p*
				Lower Bound	Upper Bound			
DRV	5	8.20	0.67	-1.45	1.50	8	0.038	0.971
DRF	5	8.17	1.27					

5.1.2.3 Effect of khat 1 on Soflex disc finished specimens

The mean ΔE of Soflex disc finished specimens for Vit-l-escence and Filtek in khat 1 compared to the controls are presented with line graphs in Figure 27. Vit-l-escence specimens had better colour stability for the durations of the staining period. Results of statistical analysis for this group using independent t tests are shown in table 15. The mean ΔE between the two materials was not statistically significant, p=0.247.

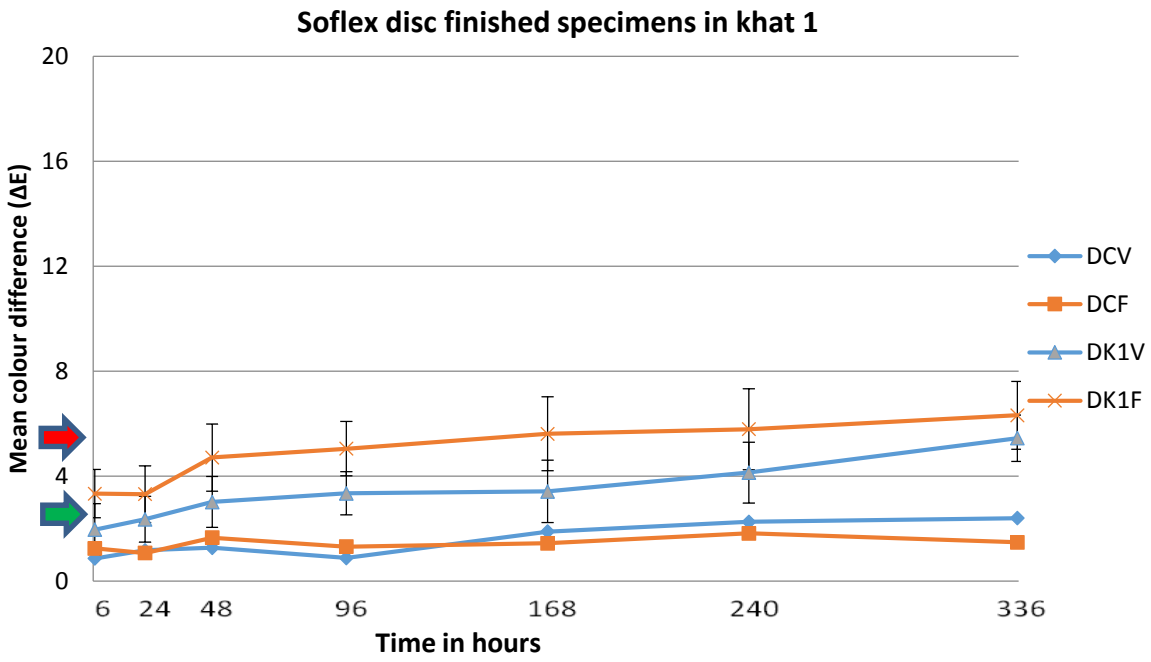

Figure 27: Mean colour difference (ΔE) for Soflex disc finished specimens in khat 1 showing the standard errors compared to the controls. (D-Soflex disc finish, C-control, K1-khat 1, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 15: Statistical results of the mean colour difference at the end of two weeks comparing Soflex disc finished Vit-l-escence and Filtek specimens in khat 1 using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*p*
				Lower Bound	Upper Bound			
DK1V	5	5.45	0.88	-2.48	0.74	8	1.249	0.247
DK1F	5	6.32	1.29					

5.1.2.4 Effect of khat 2 on Soflex disc finished specimens

The mean ΔE of Soflex disc finished specimens for Vit-l-escence and Filtek in khat 2 compared to the controls are presented with line graphs in Figure 28. Vit-l-escence specimens had better colour stability for the durations of the staining period. Results of statistical analysis for this group using independent t tests are shown in table 16. The mean ΔE between the two materials were statistically significant, p=0.011.

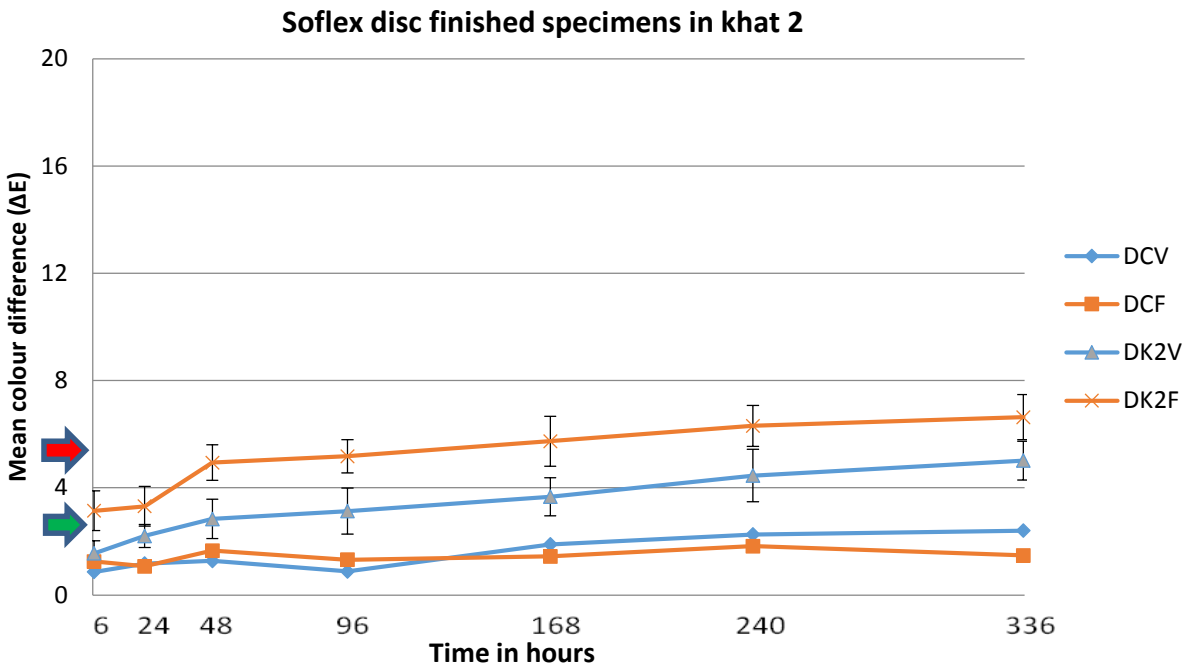

Figure 28: Mean colour difference (ΔE) for Soflex disc finished specimens in khat 2 showing the standard errors compared to the controls. (D-Soflex disc finish, C-control, K2-khat 2, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 16: Statistical results of the mean colour difference at the end of two weeks comparing Soflex disc finished Vit-l-escence and Filtek specimens in khat 2 using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*p*
				Lower Bound	Upper Bound			
DK2V	5	5.02	0.72	-2.76	-0.48	8	3.270*	0.011
DK2F	5	6.63	0.84					

*p<0.05

5.1.3 Comparison of white polishing stone finished specimens

In this section white polishing stone specimens of both materials were compared in the four different staining solutions, that is, tea, red wine, khat 1 and khat 2.

5.1.3.1 Effect of tea on white polishing stone finished specimens

The mean ΔE of polishing stone finished specimens for Vit-l-escence and Filtek in tea compared to the controls are presented with line graphs in Figure 29. Vit-l-escence specimens demonstrated less mean ΔE for the duration of the staining period. Results of statistical analysis for this group using independent t tests are shown in table 17. The mean ΔE between the two materials were statistically significant, p<0.001.

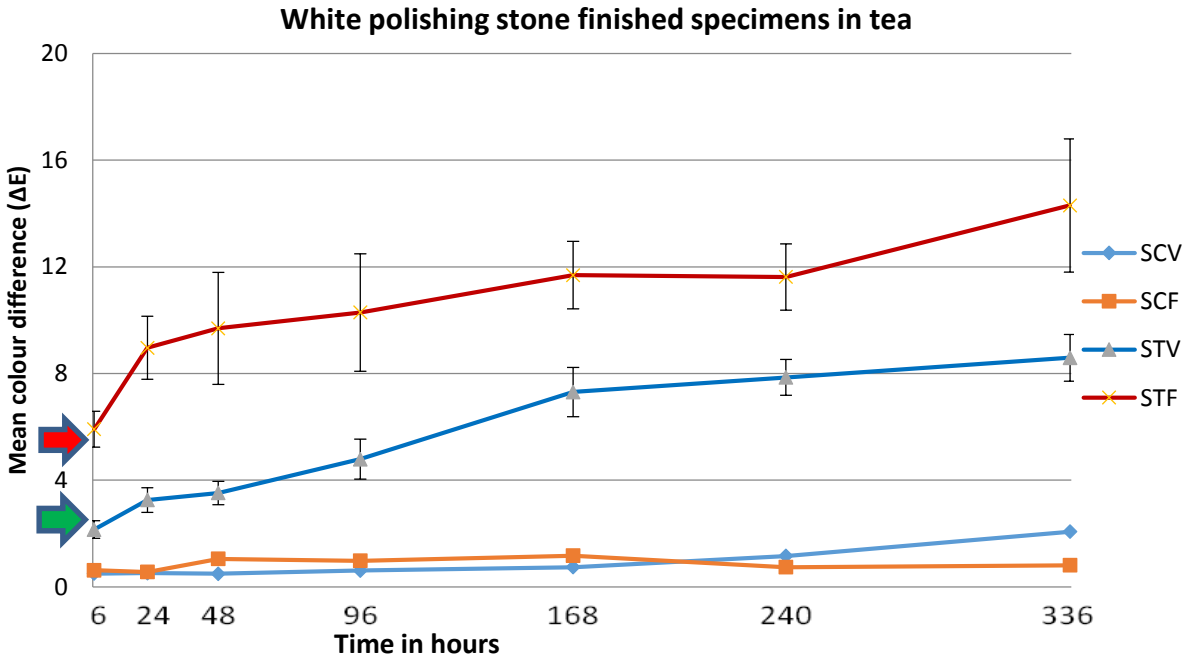

Figure 29: Mean colour difference (ΔE) for white polishing stone finished specimens in tea showing the standard errors compared to the controls (S-white polishing stone finish, C-control, T-tea, V-Vit-l-escence, F-Filtek, green arrow indicates the clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 17: Statistical results of the mean colour difference at the end of two weeks comparing white polishing stone finished Vit-l-escence and Filtek specimens in tea using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*p*
				Lower Bound	Upper Bound			
STV	5	8.59	0.88	-7.13	-4.29	8	9.275***	<0.001
STF	5	14.30	1.06					

***p<0.001

5.1.3.2 Effect of red wine on white polishing stone finished specimens

The mean ΔE of polishing stone finished specimens for Vit-l-escence and Filtek in tea and the controls are presented with line graphs in figure 30. Vit-l-escence specimens demonstrated less mean ΔE for the duration of the staining period. Results of statistical analysis for this group using independent t tests are shown in table 18. The mean ΔE between the two materials were statistically significant, p=0.001.

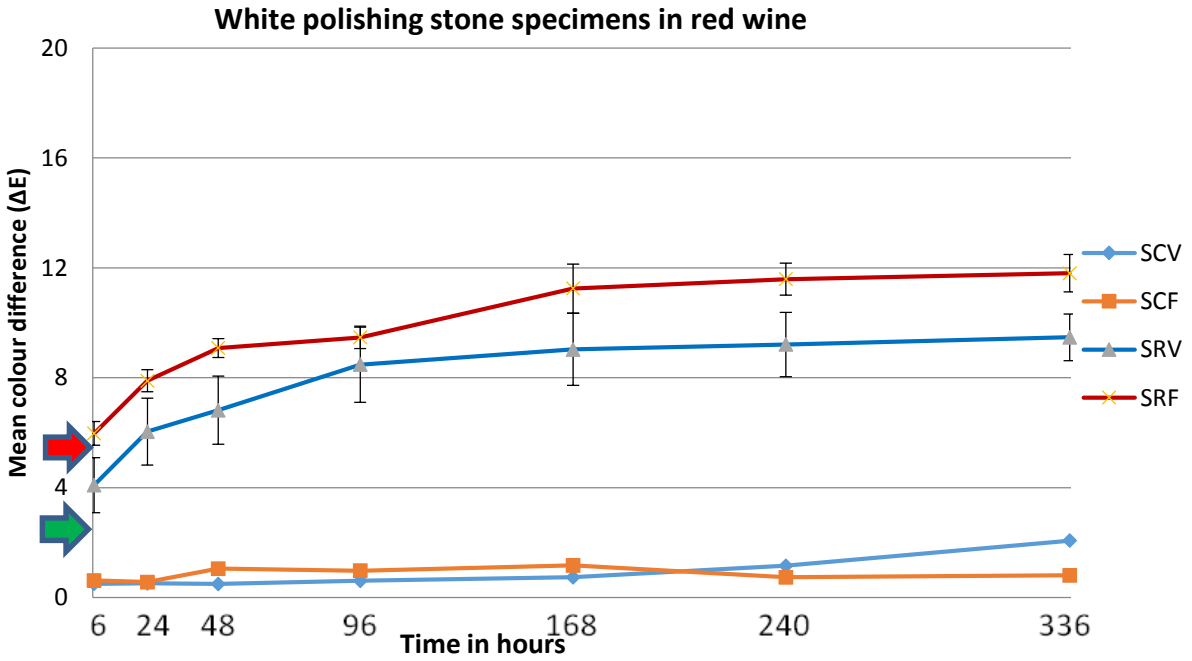

Figure 30: Mean colour difference (ΔE) for polishing stone finished specimens in red wine showing the standard errors compared to the controls. (S-polishing stone, C-control, R-red wine, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 18: Statistical results of the mean colour difference at the end of two weeks comparing White polishing stone finished Vit-l-escence and Filtek specimens in red wine using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*p*
				Lower Bound	Upper Bound			
SRV	5	9.47	0.85	-3.46	-1.21	8	4.785**	0.001
SRF	5	11.81	0.68					

**p<0.01

5.1.3.3 Effect of khat 1 on white polishing stone finished specimens

The ΔE of white polishing stone finished specimens for Vit-l-escence and Filtek in tea and the controls are presented with line graphs in Figure 31. Vit-l-escence specimens demonstrated less mean ΔE for the duration of the staining period. Results of statistical analysis for this group using independent t tests are shown in table 19. The mean ΔE between the two materials was statistically significant, p<0.001.

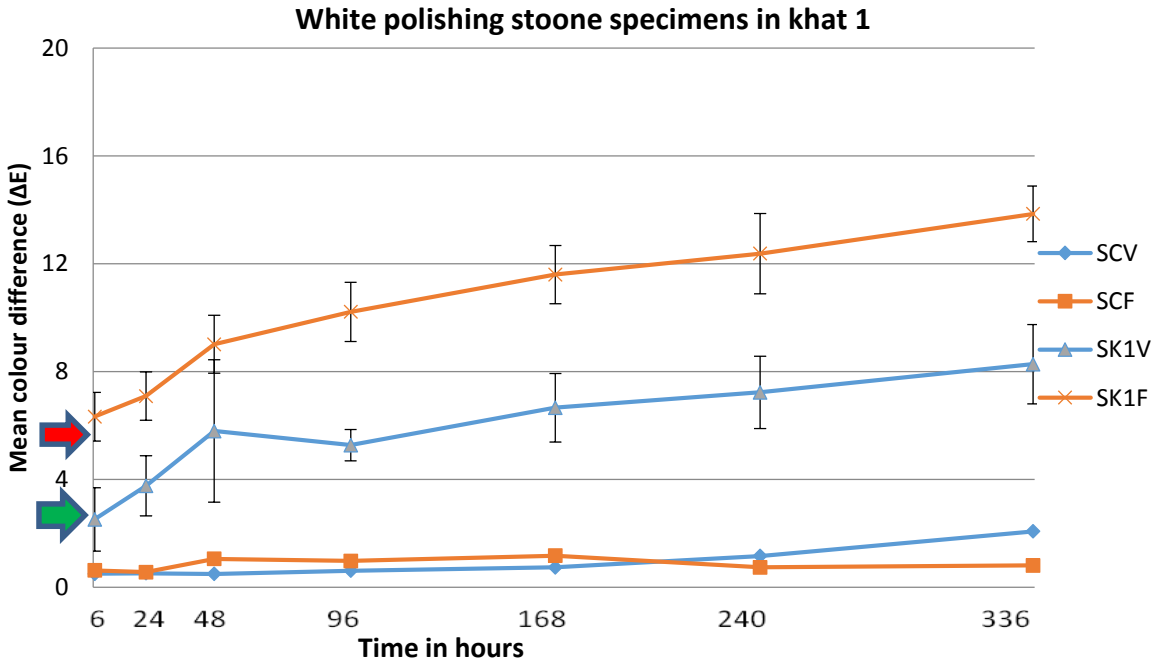

Figure 31: Mean colour difference (ΔE) for white polishing stone finished specimens in khat 1 showing the standard errors compared to the controls (S-white polishing stone, C-control, K-khat 1, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 19: Statistical results of the mean colour difference at the end of two weeks comparing white polishing stone finished Vit-l-escence and Filtek specimens in khat 1 using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*p*
				Lower Bound	Upper Bound			
SK1V	5	8.23	1.47	-7.43	-3.73	8	6.945***	<0.001
SK1F	5	13.85	1.03					

***p<0.001

5.1.3.4 Effect of khat 2 on white polishing stone finished specimens

The mean ΔE for polishing stone finished specimens for Vit-l-escence and Filtek in tea compared to the controls are presented with line graphs in Figure 32. Vit-l-escence specimens demonstrated less mean ΔE for the duration of the staining period. Results of statistical analysis for this group using independent t tests are shown in table 20. The mean ΔE between the two materials were statistically significant, p<0.001.

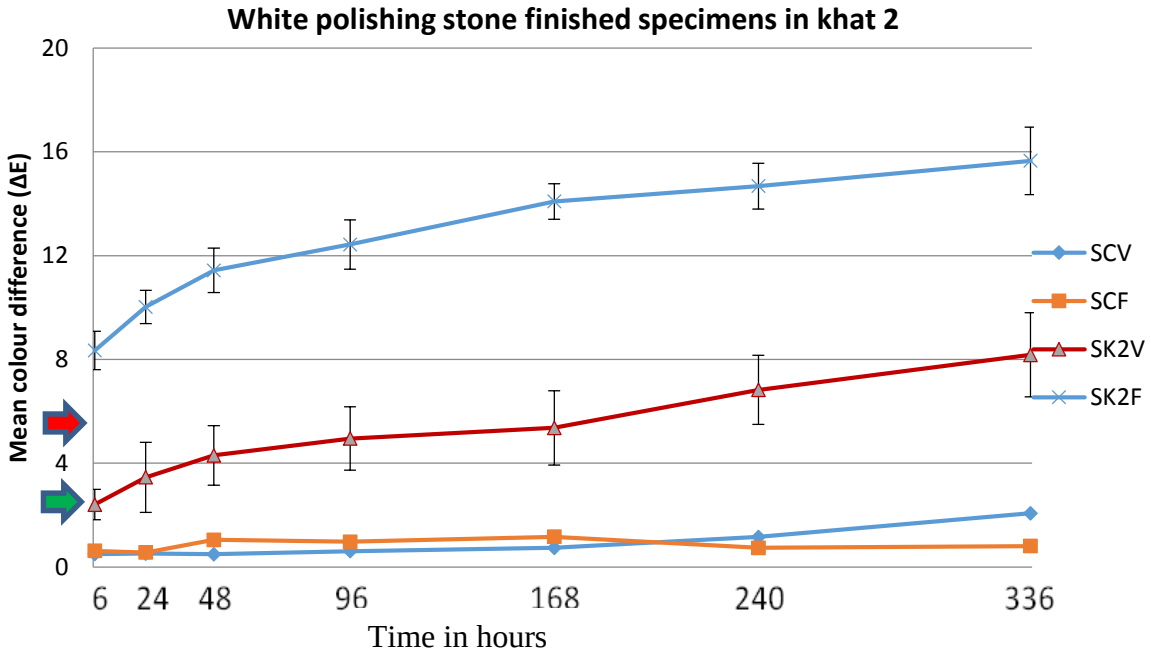

Figure 32: Mean colour difference (ΔE) for white polishing stone finished specimens in khat 2 showing the standard error compared to the controls (S-white polishing stone, C-control, K-khat 2, V-Vit-l-escence, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 20: Statistical results of the mean colour difference at the end of two weeks comparing white polishing stone finished Vit-l-escence and Filtek specimens in khat 2 using Independent-Sample t test

Specimen	*n*	*M*	*SD*	95% Confidence Interval		*df*	*t* test	*p*
				Lower Bound	Upper Bound			
SK2V	5	8.18	1.62	-7.48	0.93	8	8.069***	<0.001
SK2F	5	15.66	1.30					

***p<0.001

5.2 Comparison of effect of polishing protocol

In this section, the effect of polishing protocol on colour stability were considered under the staining solutions used in the study. Tukey HSD post hoc test were used to detect the differences within each group.

5.2.1 Filtek specimens in red wine

Figure 33 compares the effect of the three finishing protocols, mylar finish, Soflex disc and white polishing stone finish, on Filtek specimens in red wine. The samples finished with white polishing stone showed the highest mean ΔE while those with a mylar finish recorded the least difference throughout the staining period. Table 21. shows the statistical analysis for this group.

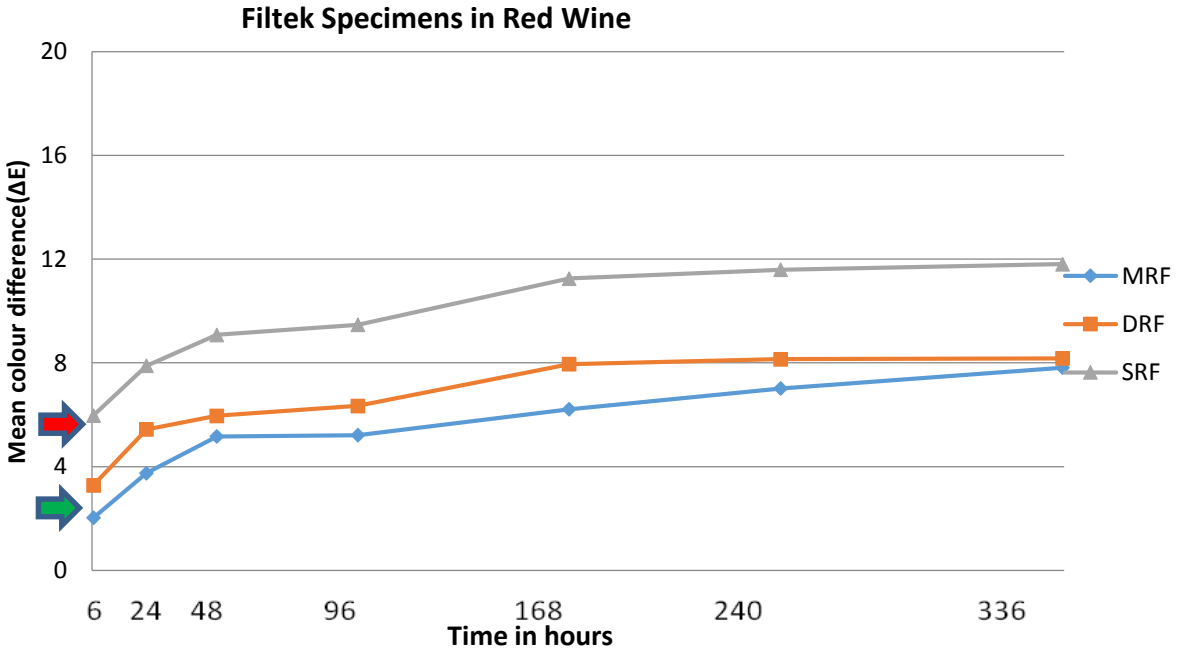

Figure 33: Comparison of mean colour difference (ΔE) for Filtek specimens in red wine finished with mylar, soflex dics and white polishing stone. (M-mylar, D-Soflex discs, S-white polishing stone, R- red wine, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 21: Results of statistical analysis of mean ΔE for different finishing protocols for Filtek specimens in red wine using Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
MRF^{case}	DRF^{case}	-0.35	-1.95	1.25	0.828
	SRF^{case}	-3.99***	-5.59	-2.39	<0.001
DRF^{case}	MRF^{case}	0.35	-1.25	1.95	0.828
	SRF^{case}	-3.63***	-5.23	-2.03	<0.001
SRF^{case}	MRF^{case}	3.99***	2.39	5.59	<0.001
	DRF^{case}	3.63***	2.03	5.23	<0.001

***. The mean difference is significant at the 0.001 level

5.2.2 Vit-l-escence specimens in red wine

Figure 34 compares the effect of the three finishing protocols, mylar finish, Soflex disc and polishing stone finish, on Vit-l-escence specimens in red wine. The samples finished with mylar showed the highest mean ΔE while those finished with Soflex discs recorded the least. Table 22 shows the statistical analysis results for this group.

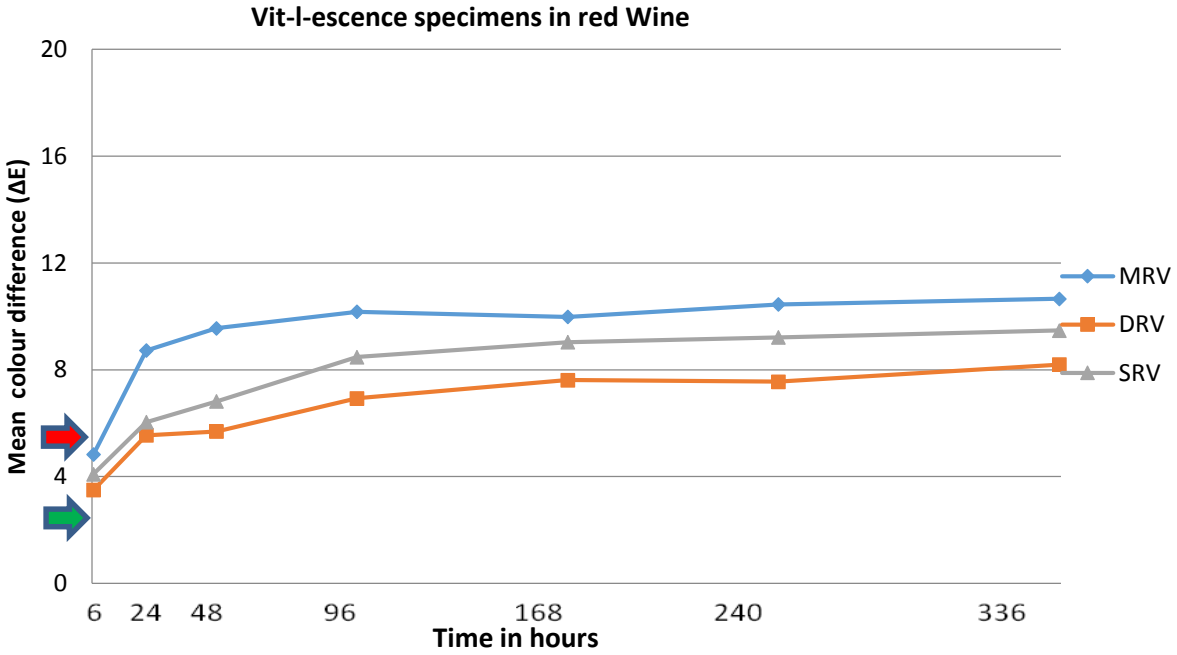

Figure 34: Comparison of mean colour difference (ΔE) for Vit-l-escence specimens in red wine finished with mylar, soflex dics and white polishing stone. (M-mylar, D-Soflex discs, S-white polishing stone, R- red wine, V-Vit-l-escence, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 22: Results of statistical analysis of mean ΔE for different finishing protocols for Vit-l-escence specimens in red wine using Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
MRV^{case}	DRV^{case}	2.46**	1.01	3.91	0.002
	SRV^{case}	1.18	-0.27	2.63	0.116
DRV^{case}	MRV^{case}	-2.46**	-3.91	-1.01	0.002
	SRV^{case}	-1.28	-2.72	0.17	0.087
SRV^{case}	MRV^{case}	-1.18	-2.63	0.27	0.116
	DRV^{case}	1.28	-0.17	2.72	0.087

**. The mean difference is significant at the 0.01 level.

5.2.3 Filtek specimens in tea

Figure 35 compares the effect of the three finishing protocols, mylar finish, Soflex disc and polishing stone finish, on Filtek specimens in tea. The samples finished with polishing stone showed the highest mean ΔE by the end of the staining period, while those finished with Soflex discs recorded the least colour difference. Table 23 shows the statistical analysis for this group.

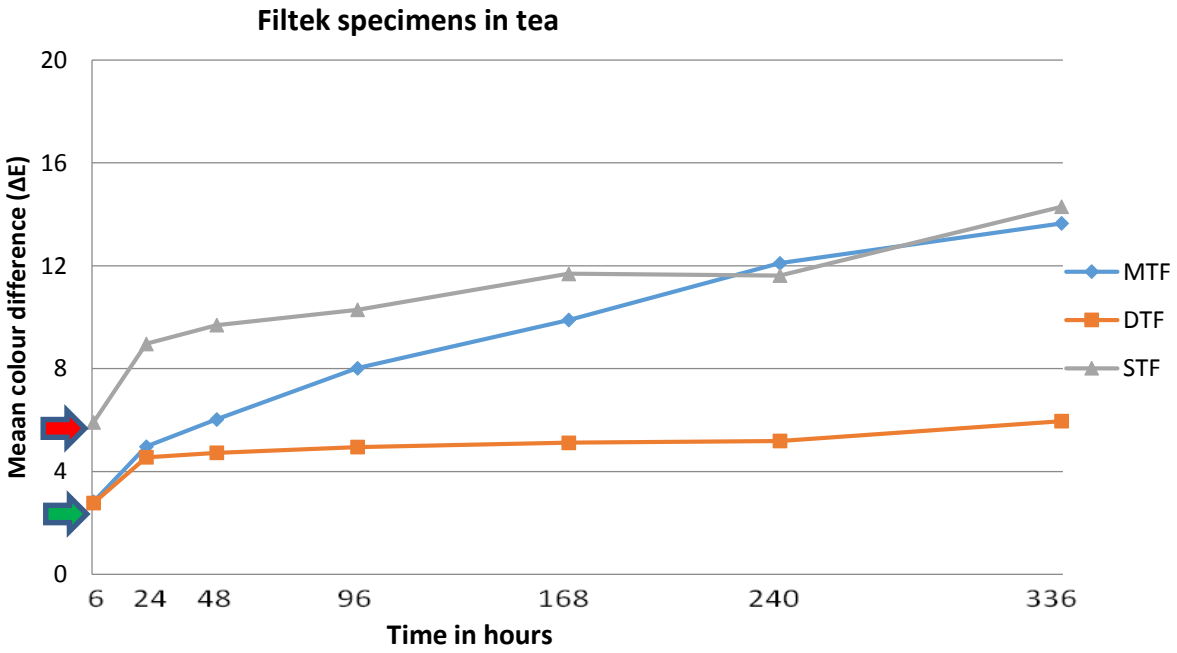

Figure 35: Comparison of mean colour difference (ΔE) for Filtek specimens in tea finished with mylar, soflex dics and white polishing stone. (M-mylar, D-Soflex discs, S-white polishing stone, T-tea, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 23: Results of statistical analysis of mean ΔE for different finishing protocols for Filtek specimens in tea using Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
DTF^{case}	MTF^{case}	7.69	-1.19	16.57	0.093
	STF^{case}	-4.57	-13.45	4.30	0.384
MTF^{case}	DTF^{case}	-7.69	-16.57	1.19	0.093
	STF^{case}	-12.26**	-21.14	-3.39	0.008
STF^{case}	DTF^{case}	4.57	-4.30	13.45	0.384
	MTF^{case}	12.26**	3.39	21.14	0.008

**. The mean difference is significant at the 0.01 level

5.2.4 Vit-l-escence specimens in tea

Figure 36 compares the effect of the three finishing protocols, mylar finish, Soflex disc and white polishing stone finish, on Vit-l-escence specimens in tea. The samples finished with mylar showed the highest mean ΔE while those finished with Soflex discs recorded the least mean ΔE. Table 24 shows the results of statistical analysis for this group.

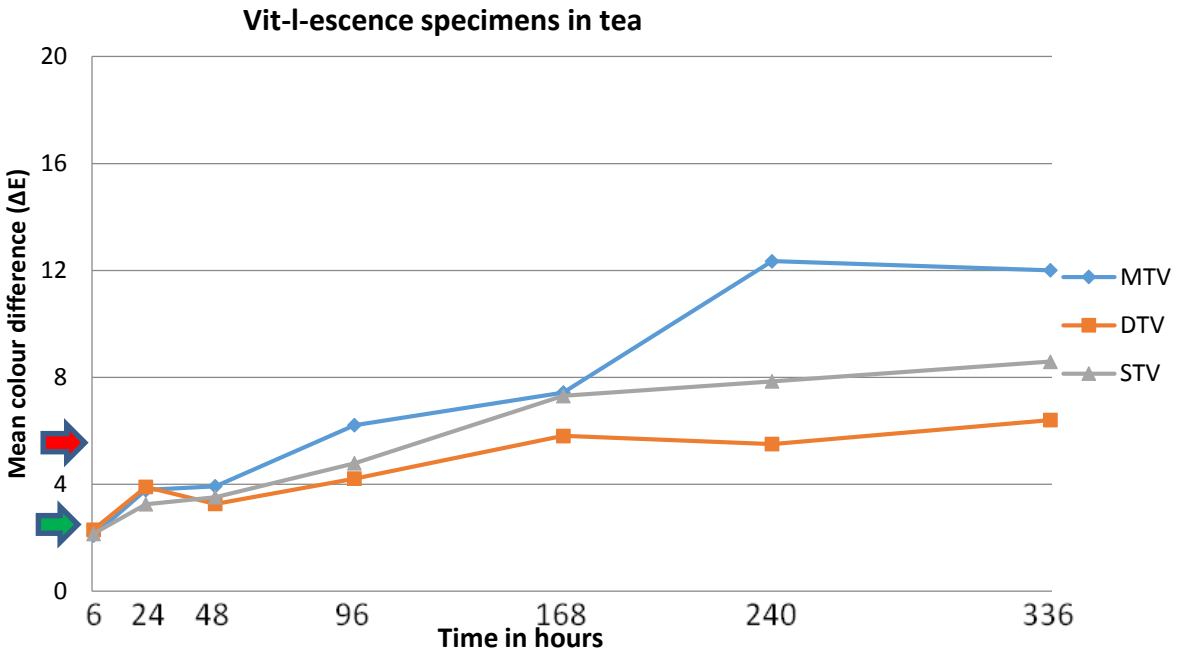

Figure 36: Comparison of mean colour difference (ΔE) for Vit-l-escence specimens in tea finished with mylar, soflex dics and white polishing stone. (M-mylar, D-Soflex discs, S-white polishing stone, T-tea, V-Vit-l-escence, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 24: Results of statistical analysis of mean ΔE for different finishing protocols for Vit-l-escence specimens in tea

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
DTV^{case}	MTV^{case}	5.60***	3.84	7.35	<0.001
	STV^{case}	3.41***	1.66	5.17	<0.001
MTV^{case}	DTV^{case}	-5.60***	-7.35	-3.84	<0.001
	STV^{case}	-2.18*	-3.94	-0.43	0.016
STV^{case}	DTV^{case}	-3.42***	-5.17	-1.66	<0.001
	MTV^{case}	2.18*	0.43	3.94	0.016

***. The mean difference is significant at the 0.001 level.
*. The mean difference is significant at the 0.05 level.

5.2.5 Filtek specimens in khat 1

Figure 37 compares the effect of the three finishing protocols, mylar finish, Soflex disc and white polishing stone finish, on Filtek specimens in khat 1. The samples finished with white polishing stone showed the highest mean ΔE while those finished with Soflex discs recorded the least mean ΔE. Table 25 shows the results of statistical analysis for this group.

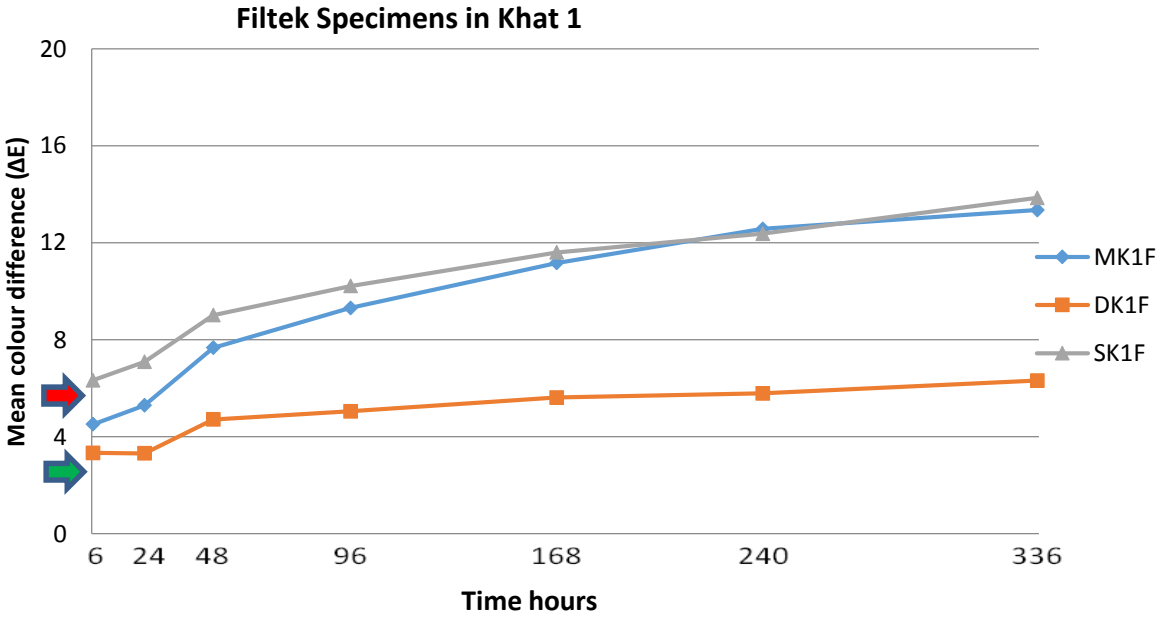

Figure 37: Comparison of mean colour difference (ΔE) for Filtek specimens in khat 1 finished with mylar, soflex dics and white polishing stone. (M-mylar, D-Soflex discs, S-white polishing stone, K1-khat 1, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 25: Results of statistical analysis of mean ΔE for different finishing protocols for Filtek specimens in khat 1

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$MK1F^{case}$	$DK1F^{case}$	7.03***	4.72	9.34	<0.001
	$SK1F^{case}$	-0.50	-2.81	1.80	0.833
$DK1F^{case}$	$MK1F^{case}$	-7.03***	-9.34	-4.72	<0.001
	$SK1F^{case}$	-7.53***	-9.84	-5.22	<0.001
$SK1F^{case}$	$MK1F^{case}$	0.50	-1.81	2.81	0.833
	$DK1F^{case}$	7.53***	5.22	9.85	<0.001

***. The mean difference is significant at the 0.001 level

5.2.6 Vit-l-escence specimens in khat 1

Figure 38 compares the effect of the three finishing protocols, mylar finish, Soflex disc and white polishing stone finish, on Vit-l-escence specimens in khat 1. The samples finished with mylar showed the highest mean ΔE while those finished with Soflex discs recorded the least mean ΔE. Table 26 shows the statistical analysis for this group.

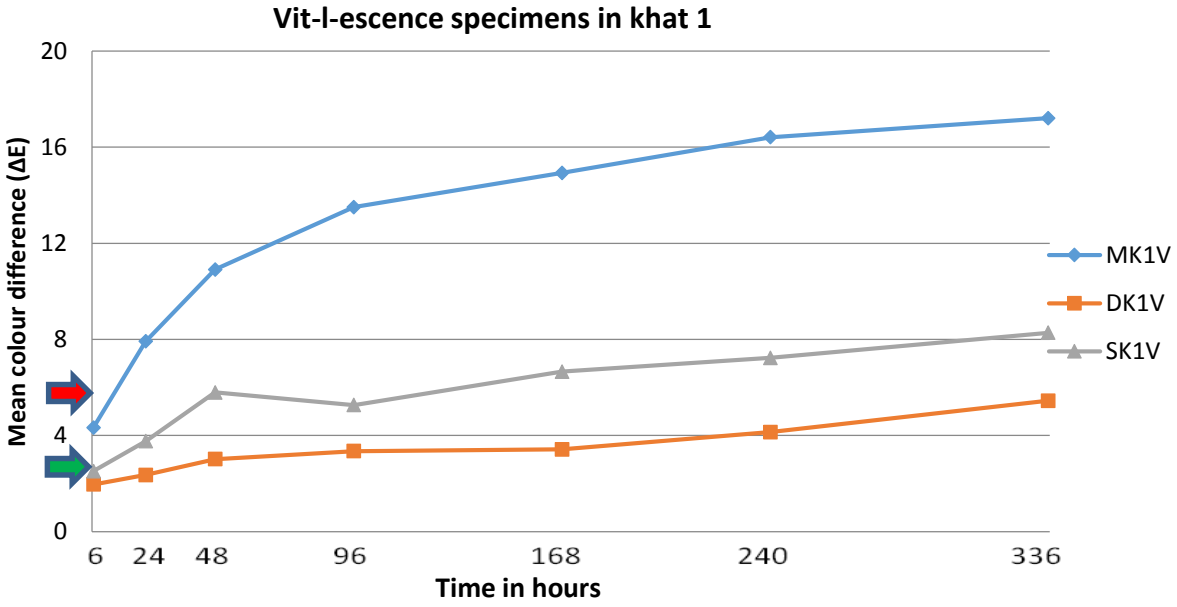

Figure 38: Comparison of mean colour difference (ΔE) for Vit-l-escence in khat 1 finished with mylar, soflex dics and white polishing stone. (M-mylar, D-Soflex discs, S-white polishing stone, K1-khat 1, V-Vit-l-escence green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 26: Results of statistical analysis of mean ΔE for different finishing protocols for Vit-l-escence specimens in khat 1 using Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
MK1V^{case}	DK1V^{case}	11.76***	9.17	14.35	<0.001
	SK1V^{case}	8.93***	6.34	11.52	<0.001
DK1V^{case}	MK1V^{case}	-11.76***	-14.35	-9.17	<0.001
	SK1V^{case}	-2.83*	-5.42	-0.24	0.032
SK1V^{case}	MK1V^{case}	-8.93***	-11.52	-6.34	<0.001
	DK1V^{case}	2.83*	0.24	5.42	0.032

***. The mean difference is significant at the 0.001 level.

*. The mean difference is significant at the 0.05 level

5.2.7 Filtek specimens in khat 2

Figure 39 compares the effect of the three finishing protocols, mylar finish, Soflex disc and white polishing stone finish, on Filtek specimens in khat 2. The samples finished with white polishing stone showed the highest mean ΔE while those finished with Soflex discs recorded the least colour difference. Table 27 shows the statistical analysis for this group.

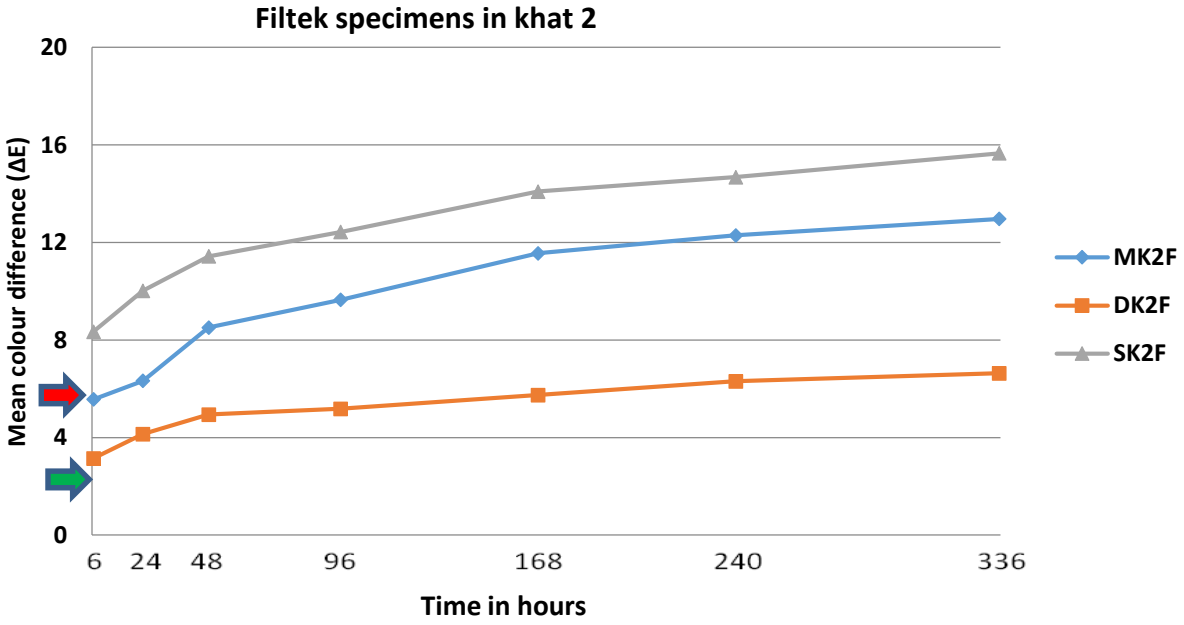

Figure 39: Comparison of mean colour difference (ΔE) for Filtek specimens in khat 2 finished with mylar, soflex dics and white polishing stone. (M-mylar, D-Soflex discs, S-white polishing stone, K2-khat 2, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 27: Results of statistical analysis of mean ΔE for different finishing protocols for Filtek specimens in khat 2 using Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$MK2F^{case}$	$DK2F^{case}$	6.34**	2.31	10.36	0.003
	$SK2F^{case}$	-2.69	-6.71	1.34	0.217
$DK2F^{case}$	$MK2F^{case}$	-6.34**	-10.36	-2.31	0.003
	$SK2F^{case}$	-9.02***	-13.05	-5.00	<0.001
$SK2F^{case}$	$MK2F^{case}$	2.69	-1.34	6.71	0.217
	$DK2F^{case}$	9.02***	5.00	13.05	<0.001

***. The mean difference is significant at the 0.001 level.

**. The mean difference is significant at the 0.01 level

5.2.8 Vit-l-escence specimens in khat 2

Figure 40 compares the effect of the three finishing protocols, mylar finish, Soflex disc and white polishing stone finish, on Vit-l-escence specimens in khat 2. The samples finished with mylar showed the highest mean ΔE while those finished with Soflex discs recorded the least mean ΔE. Table 28 shows the results of the statistical analysis for this group.

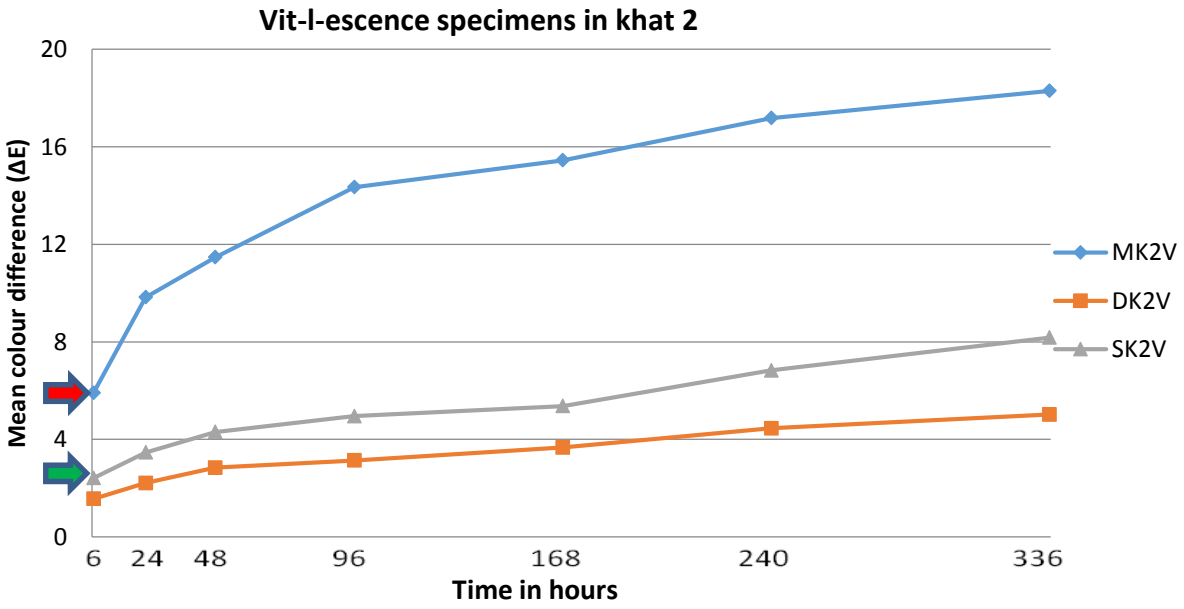

Figure 40: Comparison of mean colour difference (ΔE) for Vit-l-escence samples in khat 2 finished with mylar, soflex dics and white polishing stone. (M-mylar, D-Soflex discs, S-white polishing stone, K2-khat 2, V-Vit-l-escence, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 28: Results of statistical analysis of mean ΔE for different finishing protocols for Vit-l-escence specimens in khat 2

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$MK2V^{case}$	$DK2V^{case}$	13.28***	8.33	18.22	<0.001
	$SK2V^{case}$	10.12***	5.18	15.06	<0.001
$DK2V^{case}$	$MK2V^{case}$	-13.28***	-18.22	-8.33	<0.001
	$SK2V^{case}$	-3.16	-8.10	1.79	0.244
$SK2V^{case}$	$MK2V^{case}$	-10.12***	-15.06	-5.18	<0.001
	$DK2V^{case}$	3.16	-1.79	8.10	0.244

***. The mean difference is significant at the 0.001 level

5.3 The effect of the staining solutions after different polishing protocols

In this section, the effect of staining solutions on colour stability of the specimens was considered based on the finishing protocol. Tukey HSD post hoc test was used to detect the differences within each group.

5.3.1 Comparison of the effect of the staining solutions on Soflex disc finished specimens

Figure 41 and Figure 42 are line graphs illustrating the effect of staining solution on the colour stability of Soflex disc finished Filtek and Vit-l-escence specimens respectively. Table 29 and Table 30 show the statistical analysis of mean ΔE for these groups with Tukey-HSD post hoc test. Red wine is the only staining solution which produced a statistically significant mean colour difference for both materials.

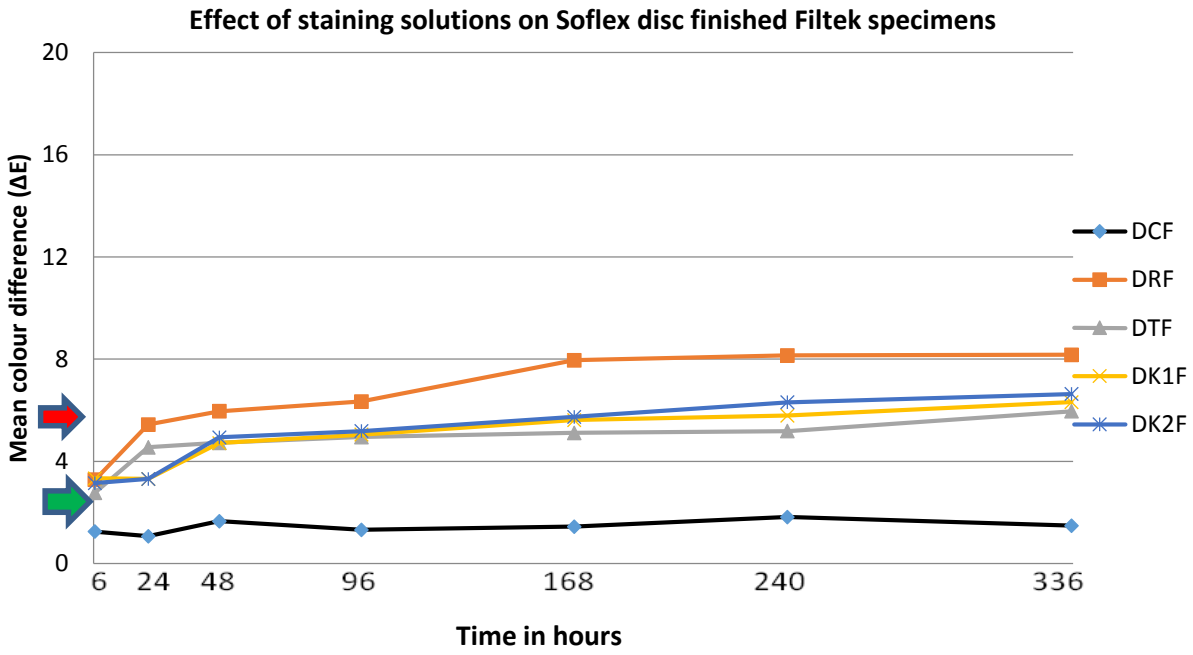

Figure 41: Comparison of the mean colour difference (ΔE) for Filtek-Soflex disc finished specimens (D-Soflex disc, R-red wine, T-tea, K1-khat 1, K2-Khat2, C-Control, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 29: Results of statistical analysis of mean ΔE for Soflex disc finished Filtek specimens with Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$DCF^{control}$	DRF^{case}	6.69**	1.25	12.14	0.002
	DTF^{case}	4.48	-0.97	9.92	0.294
	$DK1F^{case}$	4.84	-0.61	10.28	0.165
	$DK2F^{case}$	5.15	-0.29	10.60	0.091

**. The mean difference is significant at the 0.01 level

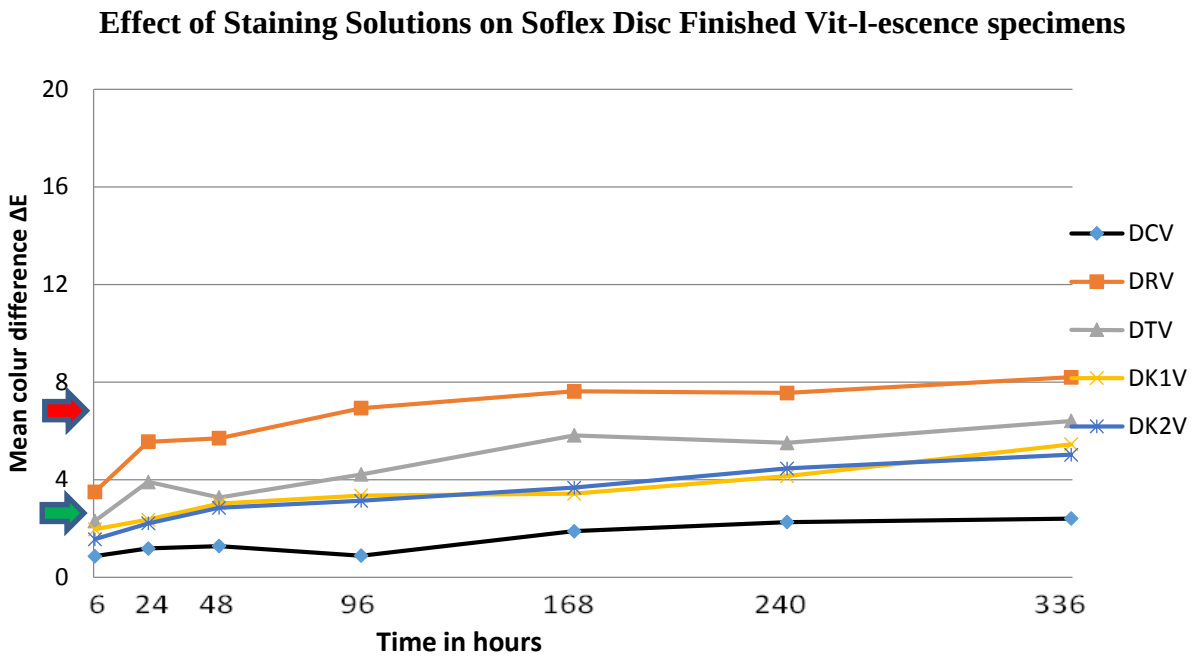

Figure 42: Comparison of the mean colour difference (ΔE) for Vit-l-escence- Soflex disc finished specimens(D-Soflex disc, R-red wine, T-tea, K1-khat 1, K2-Khat2, C-Control, V-Vit-l-escence, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 30: Results of statistical analysis of mean ΔE for Soflex disc finished Vit-l-escence specimems using Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$DCV^{control}$	DRV^{case}	5.80*	0.35	11.24	0.023
	DTV^{case}	4.00	-1.44	9.45	0.530
	$DK1V^{case}$	3.97	-1.48	9.41	0.551
	$DK2V^{case}$	2.62	-2.83	8.06	0.991

*. The mean difference is significant at the 0.05 level.

5.3.2 Comparison of the effect of staining solutions on mylar finished specimens

Figure 43 and 44 are line graphs illustrating the effect of staining solutions on mylar finished Vit-l-escence and Filtek specimens respectively. Table 31 and Table 32 show the results of the statistical analysis of ΔE for these groups with Tukey-HSD post hoc test. All staining solutions produced statistically significant mean colour differences ΔE.

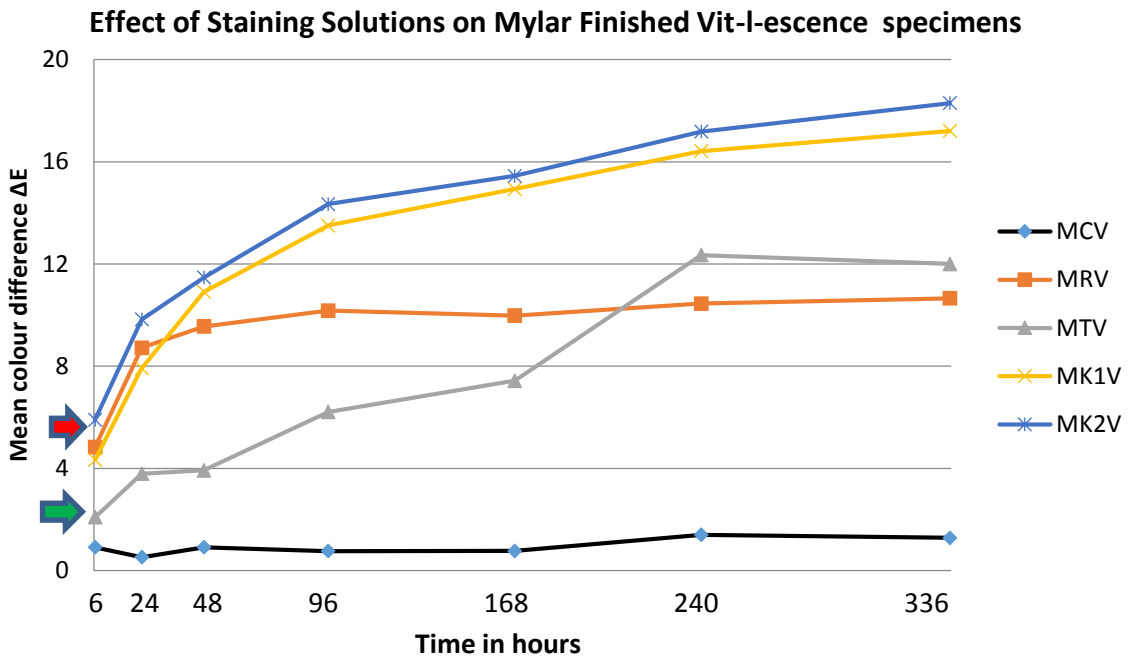

Figure 43: Comparisons of mean ΔE for Vit-l-escence mylar finished specimens (M-mylar, R-red wine, T-tea, K1-khat 1, K2-Khat2, C-Control, V-Vit-l-escence, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 31: Result of statistical analysis of ΔE for mylar finish Vit-l-escence specimens using Tukey-HSD post hoc test.

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$MCV^{control}$	MRV^{case}	9.38***	3.93	14.82	<0.001
	MTV^{case}	10.72***	5.28	16.17	<0.001
	$MK1V^{case}$	15.93***	10.48	21.37	<0.001
	$MK2V^{case}$	17.02***	11.57	22.46	<0.001

***. The mean difference is significant at the 0.001 level.

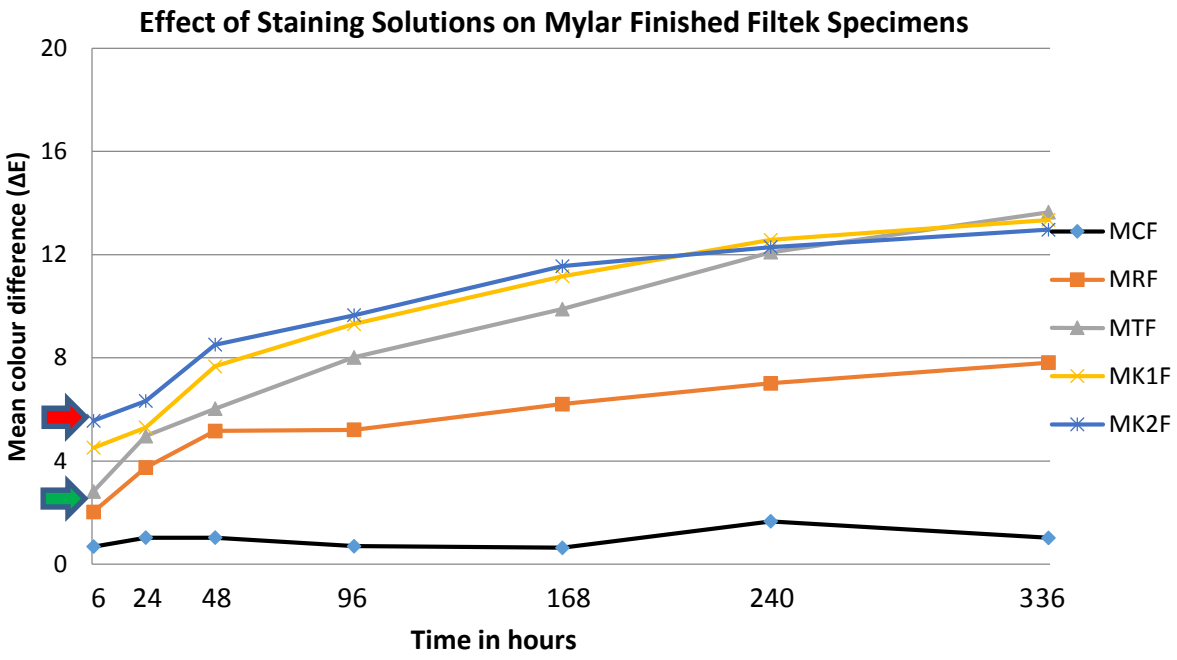

Figure 44: Comparison of mean colour difference (ΔE) for mylar finished Filtek specimens (M-mylar, R-red wine, T-tea, K1-khat 1, K2-Khat2, C-Control, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 32: Results of statistical analysis of mean ΔE for mylar finish Filtek specimens using Tukey HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$MCF^{control}$	MRF^{case}	6.79**	1.35	12.24	0.002
	MTF^{case}	12.62***	7.18	18.07	<0.001
	$MK1F^{case}$	12.32***	6.88	17.78	<0.001
	$MK2F^{case}$	11.94***	6.50	17.39	<0.001

***. The mean difference is significant at the 0.001 level.
**. The mean difference is significant at the 0.01 level.

5.3.3 Effect of staining solution on white polishing stone finished specimens

Figure 45 and 46 are line graphs illustrating the effect of staining solutions on white polishing stone finished Vit-l-escense and Filtek specimens respectively. Table 33 and Table 34 show the results of statistical analysis of mean ΔE for these groups with Tukey-HSD post hoc test. All staining solutions produced a statistically significant colour difference ΔE.

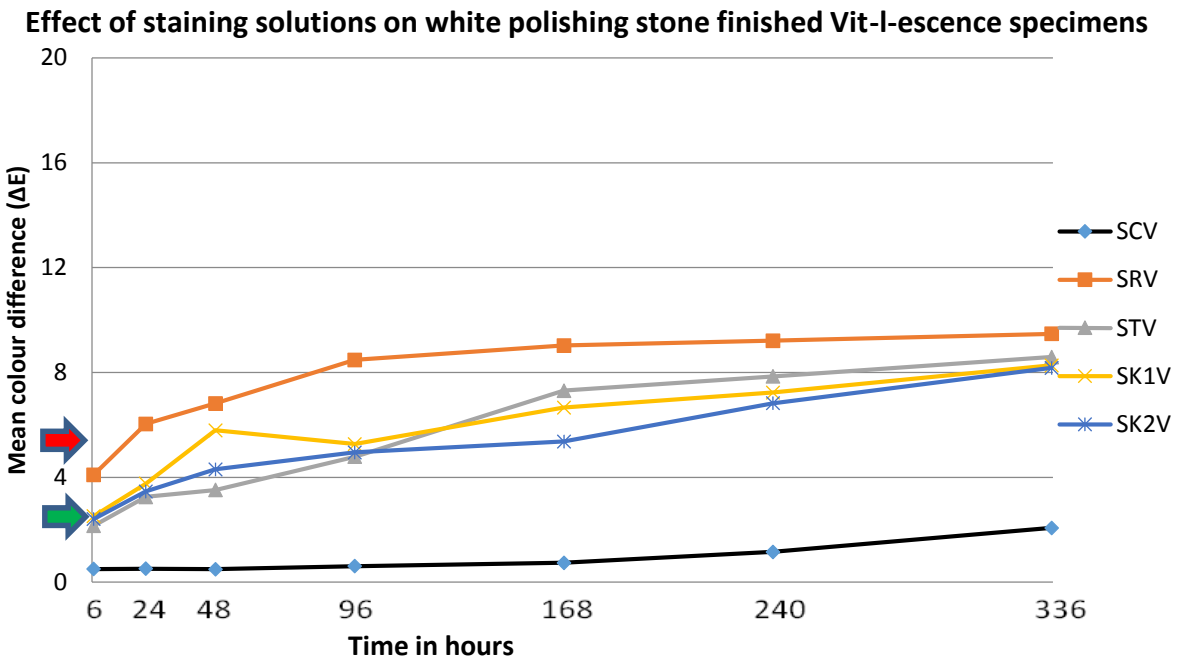

Figure 45: Comparison of the mean colour difference (ΔE) for Vit-l-escence specimens finished with white polishing stone burs. (S-white polishing stone, R-red wine, T-tea, K1-khat 1, K2-Khat2, C-Control, V-Vit-l-escence, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 33: Results of statistical analysis of mean ΔE for polishing stone specimens for Vit-l-essence using Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$SCV^{control}$	SRV^{case}	7.40***	1.95	12.84	<0.001
	STV^{case}	6.52**	1.07	11.96	0.004
	$SK1V^{case}$	6.20**	0.76	11.65	0.008
	$SK2V^{case}$	6.10*	0.66	11.54	0.011

***. The mean difference is significant at the 0.001 level.

**. The mean difference is significant at the 0.01 level.

*. The mean difference is significant at the 0.05 level.

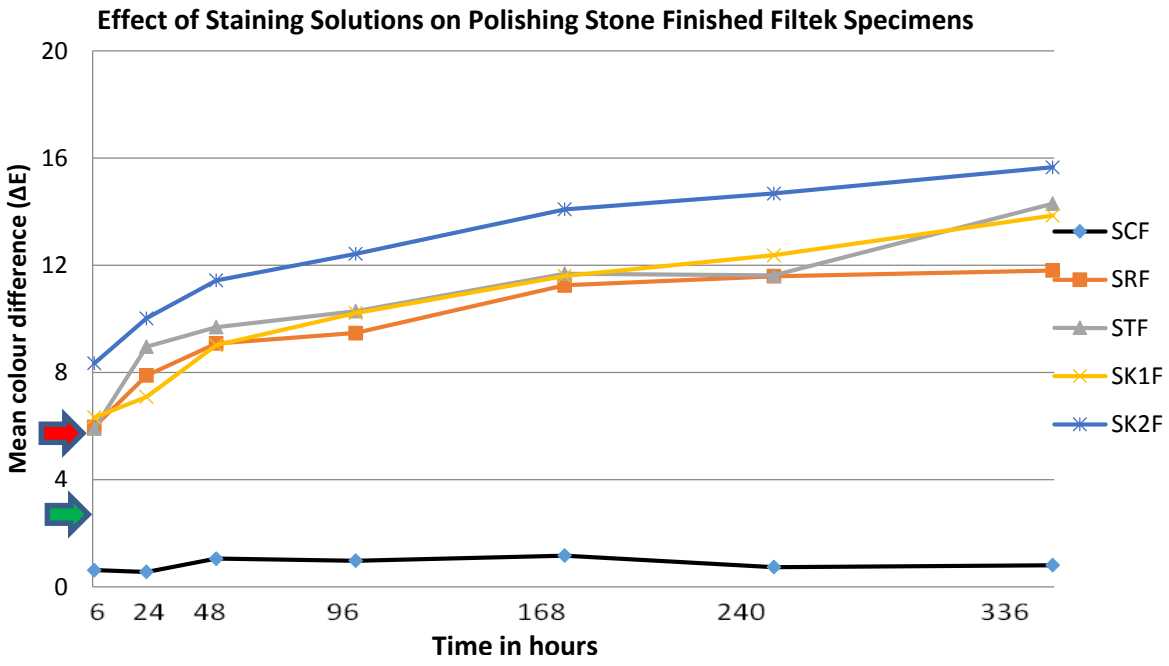

Figure 46: Comparison of the mean colour difference (ΔE) for Filtek - polishing stone finished specimens (S-white polishing stone, R-red wine, T-tea, K1-khat 1, K2-Khat2, C-Control, F-Filtek, green arrow indicates clinically perceptible ΔE level and the red arrow indicates the clinically unacceptable ΔE level)

Table 34: Results of statistical analysis of mean ΔE for polishing stone specimens for Filtek using Tukey-HSD post hoc test

Specimen		Mean Difference	95% Confidence Interval		*p*
			Lower Bound	Upper Bound	
$SCF^{control}$	SRF^{case}	10.99***	5.55	16.44	<0.001
	STF^{case}	17.41***	11.97	22.86	<0.001
	$SK1F^{case}$	13.04***	7.60	18.49	<0.001
	$SK2F^{case}$	14.85***	9.40	20.29	<0.001

***. The mean difference is significant at the 0.001 level.

5.4 Statistical analysis with ANOVA

An analysis of variance (ANOVA) test elicited a statistically significant difference in mean colour (ΔE) of the specimens $F = 42.658$, $p < 0.001$, Table 35.

Table 35: Results of statistical analysis with ANOVA test

Source	Sum of Squares	df	Mean Square	F	*p*
Between Groups	2008.678	29	69.265	42.658	<0.001
Within Groups	194.848	120	1.624		
Total	2203.526	149			

CHAPTER SIX: DISCUSSION

Resin composite restorations are exposed to diverse stains and other effects in the oral environment culminating in discolouration as demonstrated by the nano-fill and micro-hybrid composite materials evaluated in this study. The two materials specimens depicted unacceptable discolouration with a ΔE greater than 5.5 units at the end of two weeks with the exception of Soflex disc finished – microhybrid specimens (*Vit-l-escence*) in khat 1 and khat 2 staining solutions. For this study the clinically perceptible ΔE was considered to be 2.6 units and the clinically unacceptable ΔE was 5.5 units. These colour matching tolerances were determined by Douglas *et al.*[110] Distilled water was used as the control in this study and did not produce a clinically perceptible ΔE with any of the specimens, which is consistent with observations from several other studies, [43,85,90] indicating that adsorption of water did not cause significant colour change. The standard deviations for colour difference were high for mylar finished specimens and low for white polishing stone and Soflex disc finished specimens. This may be due to lack of uniformity on the surface layers after mylar finish compared to the other two finishing protocols.

Based on the results of this study, the null hypothesis is rejected and alternative hypothesis accepted. There is a significant difference in the colour stability of nanofill and micro-hybrid dental resin composite restorative materials with different surface finishing treatments, there is a significant difference in the colour stability of nanofill and micro-hybrid dental resin composite restorative materials when exposed to different staining solutions and there is a significant difference in the colour stability of micro-hybrid and nanofill dental resin composite materials following different finishing protocol when exposed to different staining solutions

6.1 Effect of finishing and polishing protocol on colour stability

The effect of finishing and polishing protocol on colour stability was slightly different for the two materials being investigated. In the microhybrid group (Vit-l-esence), Soflex disc finished specimens demonstrated the least ΔE while mylar finished specimens demonstrated the highest ΔE in all the staining solutions. On the other hand, for the nano-composite specimens (*Filtek Z350 XT*), the specimens finished with white polishing stone demonstrated the highest ΔE in all the staining solutions while Soflex discs demonstrated the least ΔE in all the staining solutions with the exception of the red wine staining group. In the red wine group, the mylar finish specimens

demonstrated a ΔE of 7.82 compared to the Soflex disc finish group which had a ΔE of 8.17, the difference between the two was however not statistically significant.

From these results, Soflex disc finish was generally associated with better colour stability for both materials. Red wine was the only staining solution which, with Soflex finish, produced a statistically significant ΔE with p=0.023 for the microhybrid and p=0.002 for the nanofill composite. These results are consistent with those from the following studies. Kumari *et al.* evaluated the effect of three finishing protocols (Soflex disc, diamond polishing paste and mylar finish) on the colour stability of a nanofilled composite and reported that Soflex discs finished specimens demonstrated the lowest ΔE.[25] Shmitt *et al*. evaluated the effect of polishing protocol on the colour stability of a microhybrid and nanohybrid composites and reported that specimens finished with a clear strip demonstrated the higher ΔE compared to Soflex disc finished specimens.

Although surface roughness of the samples were not analysed after the finishing treatments in this study, the mylar finished specimens were expected to produce the least surface roughness. This is attributed to the superficial layer being composed of more polymer matrix than fillers.[25] A smooth finish can improve aesthetics and longevity and mylar finish may be considered as the gold standard for finishing composite restorations.[18] On the other hand, the Soflex polishing discs were expected to produce a smoother finish compared to the white polishing stone. The Soflex polishing disc system incorporates a multistep polishing technique utilising sequential discs with decreasing abrasiveness, which results in a smoother final finish compared to one step materials like the white polishing stone. [61,119]

Mylar finished specimens demonstrated higher ΔE in particular with the microhybrid composites. The high ΔE with the mylar finish may also be attributed to the superficial layer being richer in resin than fillers, the same explanation provided for the smooth surface associated with the mylar finish. This resin rich layer is removed in the Soflex disc and polishing stone finished specimens. The resins have a high affinity for water and stains [61]. The results from this study are similar with those reported by of several other investigators which have also associated mylar finish with poor colour stability.[10,63,62]

In Kenya, it is thought that most dentists use white polishing stone perhaps due to cost and availability. No studies were available specifically evaluating the effect of polishing stone on colour stability of dental resin composites. The Soflex polishing discs were expected to produce a smoother finish compared to the white polishing stone and smoother finish is associated with better colour stability which was the case for both materials in the study.[10] Since polishing stone eliminates the resin rich superficial layer, it was expected to result in better colour stability compared to mylar finished specimens. This was the case with the microhybrid resin composite but not the nanofill resin composite. This perhaps may be attributed to the superficial layer in the nanofill having less resin compared to the microhybrid composite.

6.2 Effect of staining solution on colour stability

Both materials behaved similarly when immersed in the staining solutions. All polishing stone and mylar finished specimens demonstrated statistically significant ΔE which were also above the clinically unacceptable tolerance by the end of the staining period. With Soflex disc finished specimens, only red wine produced a statistically significant ΔE. Red wine has been known to cause significant ΔE in composites [13,43,45,46,90]. The red wine used for the study had seven and a half percent alcohol. A number of investigators have speculated that alcohol causes softening of the resin matrix of the composite facilitating staining.[45]

Several studies have also reported the tea can cause significant discolouration of dental composites [23,48,86]. Um and Ruyter [36] reported that the discolouration of dental composites due to tea was mainly due to adsorption of colourants in the form of tannins. In this study, tea produced statistically significant ΔE for both materials in all polishing groups except with Soflex discs.

Staining of teeth from khat chewing has been attributed to direct staining by chemicals (tannins) in the khat leaves and a change in the oral flora caused by the khat favouring chromogenic bacteria,[77,78] in which case the staining of specimens in the study would have been caused by the tannins. In khat staining solutions (khat 1 and khat 2), the nanofill composite demonstrated less ΔE for the mylar finished specimens compared to the microhybrid while the microhybrid demonstrated less ΔE with the Soflex disc and polishing stone specimens. Khat 2 demonstrated higher ΔE with white polishing stone finished-Filtek specimens and mylar finished Vit-l-escence

specimens compared to khat 1. This implies that in these groups, the diluted khat solution was more staining compared to the undiluted khat. This mean that as the khat is being chewed and becomes more dilute it may become more staining in certain situations. In all the other groups the ΔE from the 2 solutions was comparable.

Only one study was available on the effect of khat on dental composite material. The study investigated the effect of khat on the colour stability of a nanofilled and a microhybrid composite and reported that the nanofilled composite was more colour stable.[79] This is consistent with results from the mylar finished group but not the Soflex discs and white polishing stone finished groups from this study. From the abstract of the study, the investigator did not mention the finishing protocol used to prepare the specimens and may have used mylar finish which would be consistent with this study.

6.3 Comparison of type of resin composite and colour stability

Overall the nanofilled composite specimens where more colour stable compared to microhybrid when finished with mylar while the microhybrid was more colour stable for Soflex disc and white polishing stone finishing. Dental resin composites generally have a similar composition. They are all composed of silane coated inorganic filler particles with a blend of aromatic and/or aliphatic dimethahcrylate monomers such as Bis-GMA, UDMA and TEGDMA.[18] The resin matrix is an important determinant of colour stability.[36] In this study the volume of the composite materials resin component was not associated with increased staining in the Soflex disc and white polishing stone finished groups unlike in the mylar finish group. The filler component as stated in the product information was 63% for the nanofill composite (*Filtek Z350 XT*)[120] and 52% for the microhybrid (*Vit-l-escence*)[121] by volume. This is consistent with observations by Topcu[45] and Dietchi *et al*[40].

The filler particles do not absorb water but can contribute to water adsorption at the surface of the material. Dental resin composites with large filler discolour more in water compared to composites with small filler particles due to hydrolytic degradation of matrix filler interface.[23] However in a study by Ertas *et al*[43] in which colour stability of two nanohybrid (Filtek Supreme, 3M ESPE and Grandio, Voco) and two microhybid composite materials (Filtek P60 and Filtek Z250 both from 3M, ESPE) were evaluated, the two microhybrid composites and one of the nanohybrid

composites from the same manufacturer had nearly the same resin compositions and filler loading by volume. The microhybrids demonstrated better colour stability compared to the nanohybrid which is consistent with the results from this study. This is attributed to the high water sorption character of the agglomerated particles (nano-clusters).

Although surface roughness of the samples were not analysed after the finishing treatments in this study, the nanofill material specimens were expected to attain a lower surface roughness value compared to the microhybrid specimens. During finishing and polishing, filler particles might be plucked out leaving voids. The smaller particles in the nanofill composite would result in smaller voids compared to microhybrid composite.[54] Increased surface roughness is associated with decreased colour stability, however, in this study the microhybrid demonstrated better colour stability compared to the nano hybrid which can also be due to the high affinity for water by the agglomerated particles (nano-clusters).

The colour differences observed in this study may be a net effect from the combination of the above factors discussed which are, the effect of the polishing protocol used, the effect of the material's composition and the effect of the staining solutions.

6.4 Clinical significance of the study outcomes

The findings of this study may not be directly applied to the clinical situation because of the continuous staining procedure, lack of the effects of the complex oral environment and habitual effects but gives an indication of the effect of these staining solutions on the colour stability of dental resin composites. However within the limitations of this study, clinicians may execute best results if they finish microhybrid and nanofill resin composite restorations with Soflex polishing discs. All staining solutions caused clinically unacceptable discolouration to all specimens finished with mylar and white polishing stone. In the Soflex finish group only red wine produced clinically unacceptable colour difference. Patients should be advised to take potentially staining foods or beverages in moderation.

CHAPTER SEVEN: CONCLUSION AND RECOMMENDATIONS

7.1 Conclusions

1. Soflex polishing discs finish resulted in better colour stability compared to white polishing stone and mylar finish for both the nanofill and the microhybrid composites.

2. The effect of staining material is depended on the polishing protocol followed. There was no clear pattern as to which staining solution produced more discolouration though all the staining solutions can produce statistically significant ΔE which is above the clinically acceptable tolerance

3. The microhybrid composite material was more colour stable compared to the nanofill composite composite material when finished with Soflex discs or white polishing stone. The nanofill composite material was more stable when finished with mylar strips

4. The control specimens did not produce significant ΔE and remained below the clinically perceptible tolerance.

7.2 Recommendations

1. Patients with aesthetic resin composite restorations should be instructed to take potentially staining foods in moderation??
2. Dentists should be familiar with the composition of the resin composites they are using. This may further influence the effect of the finishing protocol hence should be considered during the manipulation of the composite materials.
3. A study on the effect of foods taken with khat chewing on the colour stability of dental resin composites.

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APPENDICES

APPENDIX I: DATA COLLECTION FORM USED FOR MYLAR FINISH VIT-L-ESCENCE SPECIMENS

Group	Time /hrs	ΔE from the 5 specimens					Average ΔE
		A	B	C	D	E	
MTV	6						
	24						
	48						
	96						
	168						
	240						
	336						
MRV	6						
	24						
	48						
	96						
	168						
	240						
	336						
MK1V	6						
	24						
	48						
	96						
	168						
	240						
	336						
MK2V	6						
	24						
	48						
	96						
	168						
	240						
	336						
MCV	6						
	24						
	48						
	96						
	168						
	240						
	336						

APPENDIX 2: DATA COLLECTION FORM USED FOR MYLAR FINISH FILTEK SPECIMENS

Group	Time /hrs	ΔE from the 5 specimens					Average ΔE
		A	B	C	D	E	
MTF	6						
	24						
	48						
	96						
	168						
	240						
	336						
MRF	6						
	24						
	48						
	96						
	168						
	240						
	336						
MK1F	6						
	24						
	48						
	96						
	168						
	240						
	336						
MK2F	6						
	24						
	48						
	96						
	168						
	240						
	336						
MCF	6						
	24						
	48						
	96						
	168						
	240						
	336						

APPENDIX 3: DATA COLLECTION FORM USED FOR SOFLEX DISC FINISH VIT-L-ESCENCE SPECIMENS

Group	Time /hrs	ΔE from the 5 specimens					Average ΔE
		A	B	C	D	E	
DTV	6						
	24						
	48						
	96						
	168						
	240						
	336						
DRV	6						
	24						
	48						
	96						
	168						
	240						
	336						
DK1V	6						
	24						
	48						
	96						
	168						
	240						
	336						
DK2V	6						
	24						
	48						
	96						
	168						
	240						
	336						
DCV	6						
	24						
	48						
	96						
	168						
	240						
	336						

APPENDIX 4: DATA COLLECTION FORM USED FOR SOFLEX DISC FINISH FILTEK SPECIMENS

Group	Time /hrs	ΔE from the 5 specimens					Average ΔE
		A	B	C	D	E	
DTF	6						
	24						
	48						
	96						
	168						
	240						
	336						
DRF	6						
	24						
	48						
	96						
	168						
	240						
	336						
DK1F	6						
	24						
	48						
	96						
	168						
	240						
	336						
DK2F	6						
	24						
	48						
	96						
	168						
	240						
	336						
DCF	6						
	24						
	48						
	96						
	168						
	240						
	336						

APPENDIX 5: DATA COLLECTION FORM USED FOR WHITE POLISHING STONE FINISH VIT-L-ESCENCE SPECIMENS

Group	Time /hrs	ΔE from the 5 specimens					Average ΔE
		A	B	C	D	E	
STV	6						
	24						
	48						
	96						
	168						
	240						
	336						
SRV	6						
	24						
	48						
	96						
	168						
	240						
	336						
SK1V	6						
	24						
	48						
	96						
	168						
	240						
	336						
SK2V	6						
	24						
	48						
	96						
	168						
	240						
	336						
SCV	6						
	24						
	48						
	96						
	168						
	240						
	336						

APPENDIX 6: DATA COLLECTION FORM USED FOR WHITE POLISHING STONE FINISH FILTEK SPECIMENS

Group	Time /hrs	ΔE from the 5 specimens					Average ΔE
		A	B	C	D	E	
STF	6						
	24						
	48						
	96						
	168						
	240						
	336						
SRF	6						
	24						
	48						
	96						
	168						
	240						
	336						
SK1F	6						
	24						
	48						
	96						
	168						
	240						
	336						
SK2F	6						
	24						
	48						
	96						
	168						
	240						
	336						
SCF	6						
	24						
	48						
	96						
	168						
	240						
	336						

APPENDIX 7: BUDGET

Activity	Number	Unit cost (Ksh)	Total cost (Ksh)
Proposal writing item			
Printing and binding proposal	15 copies	350	5,250
Institutional review board fees		5 000	5000
Data collection items			
Spectrophotometer	1	261 000	261 000
Filtek Z350 dental composite material	10	2 750	19 200
Vit-l-escence dental composite	10	1 385	9 695
Mylar strips	1	400	400
Soflex contouring and polishing disks	2	7000	1400
White polishing stone	5	5000	25000
Khat			10 000
Tea (ketepa)	1	500	500
Red wine (Robertsons)	1	2000	2000
Budget for data entry analysis and report writing			
Statistician			25000
Printing and binding of report			10 000
Sub total			362 000

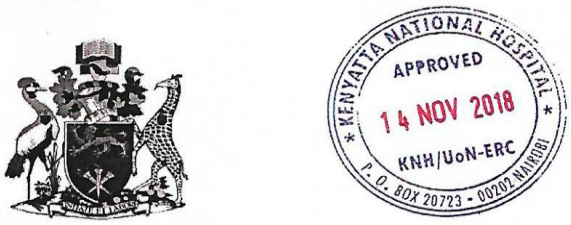
Budget summary

Total	362 000
Contingencies	55 000
Grand total	417 000
Source of funds	Self funded

APPENDIX 8: WORK PLAN

Activity	Timeline
Proposal writing	1st of August 2017 to 28th May 2018
Submission of proposal for departmental approval	1st of June 2018
Submission of proposal to faculty for approval	1st of July2018
Submission for ethical approval	21th of August 2018
Data collection	18th February to 13th of April 2019
Data analysis and report writing	15th of April 2019 to 1st of August 2019
Submission for supervisors approval	1st of August2019
Submission to external supervisor	30th of August 2019
Thesis defense	October 2019

APPENDIX 9: ETHICAL APPROVAL



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14th November 2018

Dr. Chamunorwa Marufu
Reg. No.V60/88169/2016
Dept. of Conservative and Prosthetic Dentistry
School of Dental Sciences,
College of Health Sciences
University of Nairobi

Dear Dr. Marufu

RESEARCH PROPOSAL – EFFECT OF POLISHING PROTOCOL AND EXPOSURE TO STAINING SOLUTIONS ON THE COLOUR STABILITY OF DENTAL COMPOSITE MATERIALS (P574/08/2018)

This is to inform you that the KNH- UoN Ethics & Research Committee (KNH- UoN ERC) has reviewed and **approved** your above research proposal. The approval period is 14th November 2018 – 13th November 2019.

This approval is subject to compliance with the following requirements:

a) Only approved documents (informed consents, study instruments, advertising materials etc) will be used.
b) All changes (amendments, deviations, violations etc.) are submitted for review and approval by KNH-UoN ERC before implementation.
c) Death and life threatening problems and serious adverse events (SAEs) or unexpected adverse events whether related or unrelated to the study must be reported to the KNH-UoN ERC within 72 hours of notification.
d) Any changes, anticipated or otherwise that may increase the risks or affect safety or welfare of study participants and others or affect the integrity of the research must be reported to KNH- UoN ERC within 72 hours.
e) Clearance for export of biological specimens must be obtained from KNH- UoN ERC for each batch of shipment.
f) Submission of a request for renewal of approval at least 60 days prior to expiry of the approval period. (*Attach a comprehensive progress report to support the renewal*).
g) Submission of an *executive summary* report within 90 days upon completion of the study. This information will form part of the data base that will be consulted in future when processing related research studies so as to minimize chances of study duplication and/ or plagiarism.

For more details consult the KNH- UoN ERC website **http://www.erc.uonbi.ac.ke**

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