

Original Article

Effect of energy drinks on microhardness of silorane and dimethacrylate-based composite resins

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Abstract

Introduction: Increased consumption of energy drinks has raised concerns about their effects on dental restorations. This study assessed the effects of two energy drinks on the surface microhardness of methacrylate and silorane-based composites after 1-week and 1-month periods.

Materials & Methods: In this in-vitro study, 90 cubic samples were prepared from Filtek P90, Filtek Z250 and Filtek Z350 XT composite resins. Vickers hardness test was performed to measure the baseline surface microhardness for each specimen. Ten randomly selected samples from each composite material were then immersed in one of the two sports drinks (Red Bull and Hype) or artificial saliva (control). Surface microhardness was re-evaluated after 1 week and 1 month of immersion. The data were evaluated using ANOVA via post-hoc Tukey tests and repeated measure test (α =0.05).

Results: Surface microhardness of all composites were significantly decreased in energy drinks in both evaluation periods (P<0.001). In artificial saliva, microhardness was significantly increased after 1 week and decreased after 1 month of immersion (P< 0.001). After 1 month, the lowest microhardness changes were observed in Filtek Z350 XT composite. (18% and 14% reduction in Hype and Redbull respectively). Differences between energy drinks were significant for Z350 XT composite only after 1 week (P=0.01) and for Z250 composite after 1 week and 1 month (P=0.020 and P< 0.001 respectively).

Conclusion: Hype and Red Bull energy drinks can affect the surface hardness of composite resins depending on their characteristics and exposure time.

Keywords: Composite resins, Energy drinks, Silorane composite resin

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اثر نوشیدنی های انرژی زا بر ریزسختی کامپوزیت رزین های سایلوران و بر پایه دی متاکریلات غزاله احمدی زنوز، بهناز اسماعیلی، سحر خورشیدی*، ثریا خفری

چکیده

مقدمه: افزایش مصرف نوشیدنی های انرژی زا نگرانی در مورد اثرات آن ها بر ترمیم های دندانی را بالا برده است.این مطالعه اثر دو نوشیدنی انرژی زا را بر ریزسختی سطحی کامپوزیت هایی بر پایه متاکریلات و سایلوران پس از یک هفته و یک ماه ارزیابی کرد. مواد و روش ها: در این مطالعه آزمایشگاهی، ۹۰ نمونه مکعبی از کامپوزیت های Z250،P90 و Z350XT تهیه شد. سختی سنجی ویکرز برای اندازه گیری سختی اولیه هر نمونه انجام شد. ۱۰ نمونه ی تصادفی انتخاب شده از هر کامپوزیت ،در دو نوشیدنی انرژی زاای اندازه گیری سختی اولیه هر نمونه انجام شد. ۱۰ نمونه ی تصادفی انتخاب شده از یک هفته و یک ماه دوباره انرژی زاای و بزاق مصنوعی (کنترل) غوطه ور شدند. سختی سطحی بعد از یک هفته و یک ماه دوباره ارزیابی داده ها با استفاده از آزمون های آماری Post-hoc Tukey، ANOVA و Post-hoc Tukey، شد. (α=0.05)

یافته ها: سختی سطحی همه کامپوزیت ها بطور معناداری در نوشیدنی های انرژی زا در هر دو زمان کاهش یافت(P<0.001). بعد از در از اور معناداری بعد از یک هفته غوطه ور سازی افزایش و بعد از یک ماه کاهش یافت(P<0.001). بعد از یک ماه کاهش یافت(P=0.001). بعد از یک ماه کمترین تغییرات در کامپوزیت (P=0.001) دیده شد(به ترتیب (P=0.001) و برای کامپوزیت (P=0.001) بعد از یک هفته و (P=0.001) و برای کامپوزیت (P=0.001) بعد از یک هفته و یک ماه معنادار بود (به ترتیب (P=0.001) و (P=0.001).

نتیجه گیری: نوشیدنی های انرژی زا Hype و Red Bull می توانند بر سختی سطحی کامپوزیت ها اثر گذارند که این اثرات وابسته به ویژگی های آن ها و زمان غوطه ور سازی است.

واژگان کلیدی: کامپوزیت رزین ها، نوشیدنی های انرژی زا، کامپوزیت رزین سایلوران

Introduction

In recent years, due to the better esthetic, improved formulation and bonding methods, the use of resinbased restorative materials has considerably increased in dentistry. [1-4] The physical properties of composite resins are important factors in determining the lifespan of the restorations. Of these, surface hardness is an imperative aspect, which is related to compressive strength, resistance to intraoral softening and degree of conversion. [5] Reduction in surface hardness increases the possibility of wear and fatigue in the dental materials, and can lead to failure of restorations. [5, 6] Surface hardness is influenced by the composition of the material, the environment to which they are exposed and the time of exposure. [7, 8] Previous studies have shown that the consumption of some chemically acidic foods and drinks can cause surface degradation of the restorative materials and changes in the surface hardness of the glass ionomer cements, composite resins

and componers. [2, 7-9] In recent years, energy drinks have become popular with a growing trend, especially among adults aged 18 to 35 years. [3, 6, 10] Alarmingly, these beverages can cause dental erosion and affect dental restorations after long-term consumption. Although different products are presented in the market, most energy drinks have similar ingredients, including simple sugars, caffeine, taurine, taurynie, inositol, B vitamins, glucuronolactone and herbal extracts. Most energy drinks contain about 30-35mg of caffeine per 100ml. Some of the positive effects of using energy drinks include the increased body function, better concentration, decreased fatigue and overcoming stress, but they also have side effects in the body, some of which occur in the oral cavity. [4, 11-15] The chemicals present in these drinks can lead to fatigue and surface degradation of composite restorations. [7, 8] The low pH and acidity of these drinks on one side lead to erosion of



the surface of the enamel and restorative materials. On the other hand, their sugar content is metabolized through microorganisms in the plaque to produce organic acids which can cause demineralization and consequently dental caries. ^[16]

The advancements in nanotechnology have led to the production of nanofilled composites with lower filler size (approximately 25 nm and nanoaggregates of approximately 75 nm) and improvement of their physical properties due to their higher filler content (up to 79.5%). [7, 17, 18] Low shrinkage silorane-based composite is another type of composite. Silorane resin is an alternative to the methacrylate resin matrix, thereby creating lower polymerization shrinkage and better hydraulic stability. Silorane is synthesized as a result of the reaction of oxirane and siloxane molecules. [19] Siloxane determines the nature of the highly hydrophobic silorane, and the oxirane is responsible for the lower polymerization shrinkage of silorane compared to methacrylate-based composites. Cationic ring-opening in silorane-based composite is the mechanism that reduces shrinkage compared to free radical polymerization in methacrylates. [20, 21] These composites have shown promising physical properties in previous studies in comparison to conventional methacrylate-based composites. [7]

Since limited studies were conducted on the effect of energy drinks on the properties of restorative materials, the purpose of the present in-vitro study was to compare the surface hardness variations of microhybrid, nanofilled and silorane-based composite resins under the influence of energy drinks.

The null hypotheses tested were:

- 1- Energy drinks do not reduce the surface hardness of composite resins.
- 2-There are no differences in the hardness value variations among nanofilled, silorane-based and microhybrid composites in energy drinks.

Materials & Methods

The current invitro research was conducted on the three composite resins provided in the A3 shade including Filtek Z350 XT nanofilled composite resin, Filtek Z250 microhybrid composite resin and Filtek P90 silorane-based composite resin. The characteristics, manufacturers and constituents of the composites used in this study are presented in table 1. In total, 90 cubic samples (30 samples from each composite) were

prepared in a length of 5 mm, a width of 5 mm and a thickness of 2 mm using a polyvinyl chloride (PVC) mold. Two 1-mm-thick composite resins were incrementally placed with a plastic instrument in the mold and pressed by a piece of transparent polyester matrix tape (Mylar Strip, SS White Co., Philadelphia, PA, USA) and a glass slide to prevent air retention and create a smooth surface. Each layer was then light cured with a LED light-curing unit at a light intensity over 800 mW/cm2 (Valo, Ultradent Product Inc. South Jordan, the USA) for 20 seconds in accordance with the manufacturer's instructions. Prior to onset of the radiometer (Demetron LED polymerization, a Radiometer, Kerr, Orange, the USA) was utilized to ensure the power of emitting light. The head of the light-curing unit was held in contact with the glass slide of 1 mm for standardizing the distance between the light source and the sample surfaces. A scalpel was used to mark the bottom surface of each sample. For achieving complete polymerization, all the samples were then immersed in distilled water at the temperature of 37°C for 24 hours.

With the intention of simulating the clinical condition, the upper surfaces of samples were polished with 600 to 1200-grit silicon carbide abrasive papers consecutively for 30 seconds. The samples were washed carefully after each polishing stage under running water for 10 seconds to eliminate the debris.

According to the immersion solutions (artificial saliva as a control, Red Bull energy drink and MPF Hype energy drink), their type, composition and manufacturers are presented in table2, samples from each composite resin were randomly divided into three subgroups of 10. Samples were placed in 30 mL of Red Bull energy drink and MPF Hype energy drink at the lab temperature for 5 min/day in a sealed container. Samples were then washed with distilled water and kept in artificial saliva at 37°C for the rest of the day. The controls were left in sealed containers in the presence of 30 mL of artificial saliva (Hypozalix, Biocodex, France) at 37°C for 24 hours. In each subgroup, containers were refilled with fresh solutions once daily.

Assessment of surface microhardness: The hardness of the specimens was measured at baseline, after 7 days and after one month using Vickers microhardness indenter (MH1.6 Microhardness Tester, KOOPA, Mashhad, Iran). After each storage period, the samples were washed under running water, and then additional water on the surfaces was gently dried using tissue



paper. Three indentations were made and measurements were obtained at different points on each specimen, with a 0.5 kg load for a 10 s dwell time.

The hardness number was automatically measured using the software of the device and the average value of three indentations was recorded as the Vickers Hardness Number (VHN) for each sample expressed in kg/mm2. After a week and then after a month, the same procedure was repeated on the samples. In order to compare the changes in surface microhardness of different composites in two time intervals, the hardness

variation percentage with respect to the baseline was calculated for each group.

Statistical analysis: Data obtained from the present study were analyzed using SPSS22. The normal distribution of data was examined by Kolmogorov-Smirnov test. ANOVA test was used to compare different groups of composites and drinks, and Post-hoc Tukey for paired test between the two groups. RM (repeated measure) test was employed to check the trend of time variation. P-value <0.05 was statistically considered as significant level.

Table 1. The characteristics, manufacturers and constituents of the composites used in this study

Product	Manufacturer	Shade	Type	Content					Lot
(code)				Organic matrix	Fillers	Particle	Filler	Filler	number
						size	weight	volume	
Filtek	3M Espe, St.	A3	Silorane	Siloranes	Quartz, yttrium	0.47 µm	76%	55%	N468933
P90	Paul,USA				fluoride				
Filtek	3M Espe, St.	A3	Microhybrid	BisGMA, UDMA,	Zirconia/silica	0.01-3.5	84.5%	60%	N528844
Z250	Paul,USA			Bis-EMA		μm			
Filtek	2M Eana St	A3	Nanofilled	BisGMA, UDMA.	Zirconia/nanosilica	5-20 nm	82%	59.5%	N495372
	3M Espe, St.	AS	Nanonned	DISGWIA, UDIWIA,	Zircoma/nanosmca	3-20 IIII	82%	39.3%	N493572
Z350	Paul,USA			TEGDMA, Bis-					
				EMA					

Bis-GMA: Bisphenol A glycol dimethacrylate; UDMA: Urethane dimethacrylate; Bis-EMA: Ethoxylated bisphenol A glycol dimethacrylate; TEGDMA: Triethylene glycol dimethacrylate; Bis-EMA: ethoxylated bisphenol A glycidyl methacrylate

Table 2. Immersions solution, Composition and Manufactures

Staining Solutions	Composition	Manufacturer
Red Bull	Water, Sucrose, Glucose, Acidity Regulators (Sodium Citrates, Magnesium Carbonate), Carbon Dioxide, Acidifier citric acid, Taurine 0.4 %, Caffeine 0.03%, Inositol, Vitamins (Niacin, Pantothenic Acid, B6, B12), Flavouring, Colours (Caramel, Riboflavin)	
Нуре	Carbonated Water , Sugar , Acidifier Citric Acid E330, Acidity Regulator Sodium Citrate E331, Taurine , Caramel Sugar Syrup , Caffeine 0.032% , Flavouring , Glucuronolactone 0.024%, Vitamins (Niacin , Pantothenic Acid , B6, B2, B12)	Warsaw, Poland, pH=3.42
Artificial saliva	Sodium chloride $86.550~mg/100mL$, calcium chloride $16.625~mg/100mL$, dipotassium phosphate $32.600~mg/100mL$, potassium chloride $62.450~mg/100mL$, magnesium chloride $5.875~mg/100mL$, sorbitol, sodium carboxymethyl-celulose, purified water Preservative: methyle parahydroxybenzoate(E218)	Hypozalix,Biocodex,France

Results

Mean values and standard deviations of surface hardness of different composites in the immersion solutions at base line, after one week and after one month are presented in table 3. Significant differences were observed in baseline surface microhardness among composite resins (P<0.001). The baseline surface microhardness of P90 composite was significantly lower



than Z250 and Z350XT composites .Statistically, significant differences were revealed in the hardness of each composite resin in various immersion periods in different solutions (P<0.001). Mean surface hardness values of all three composite resins before immersion in energy drinks were higher than those after 1-week storage. However, in artificial saliva, the mean surface hardness values of all three materials were increased after 1-week immersion compared to the baseline. After one month, all three composite resins showed significantly lower surface hardness in comparison to baseline for both the energy drinks and control surface microhardness variation The percentages in composite groups immersed in different solutions after a week and a month are shown in table 4. Differences in surface microhardness variations for each composite in different solutions and both time periods were significant (p<0.001). Accordingly, after one-week immersion in the Hype energy drink, significant differences were observed between the surface microhardness variations of the composite resins (P<0.001). Changes in the microhardness value of Filtek Z350 XT composite resin were significantly lower than those of Filtek P90 and Filtek Z250 composites (P=0.008 and P<0.001, respectively). Differences between P90 and Z250 were not significant (P=0.08).

In the Red Bull energy drink after a week, the changes in all three composites were close and no significant difference was observed (P=0.8). After a week, in artificial saliva, there were also no significant differences between the surface microhardness variations of composites (P=0.4, but unlike reducing the composite microhardness in the presence of energy

drinks, increased surface microhardness was observed in artificial saliva in all three composites. After 1-month immersion in Hype energy drink, significant differences were found among surface microhardness variations of composite resins (P=0.007). Among three composite resins tested, Z250 and Z350 XT showed the highest and lowest surface microhardness reduction, respectively. Only differences between Z250 and Z350 XT composites were significant (P=0.005).

After one month, in Red Bull energy drink, differences between surface microhardness variations of composite resins were significant (P=0.01). the highest surface microhardness reduction was observed in P90 composite, which had a significant difference with the Z350 XT composite with the least changes (P=0.012). In the artificial saliva, after one month immersion, differences between surface microhardness variations of composite resins were significant (P=0.001).

Z350 XT compared to Z250 and P90 composite resins had significantly lower reduction in surface microhardness was observed (P=0.001 and P=0.01, respectively). Z250 composite showed the highest reduction of surface microhardness, however the microhardness variation percentage did not differ significantly fromP90 composite (P=0.5). Pairwise comparison of energy drinks in each time period for each composite indicated no significant differences except in Z350XT composite after 1 week (P=0.01) and in Z250 composite after 1 week and 1 month (P=0.02 and P<0.001, respectively). In addition, specimens immersed in artificial saliva demonstrated lower mean surface microhardness reduction compared to the ones stored in energy drinks after a 1 month.

Table 3. Mean values and standard deviation of surface microhardness of tested composites before and after immersion in different solutions

Beverages comp	Нуре			Red Bull Time			Artificial saliva		
comp	Baseline	after a week	after a month	Baseline	after a	after a month	Baseline	after a week	after a month
Filtek P90	78.15+4.63 ^{A a}	72.51+5.24 ^{A b}	61.31+5.2 ^{A c}	75.9+6.05 ^{A a}	70.37+5.9 ^{A b}	60.27+2.93 ^A	77.7+4.27 ^{A a}	78.21+4.24 ^{A b}	73.73+4.6 ^{Ac}
Filtek Z250	109.14+3.9 ^B a	97.8+2.5 ^{B b}	79.72+5.38 ^{Bc}	112.28+4.7 ^B a	105.01+2.93 ^{Bb}	93.83+3.42 ^{B c}	108.17+4.49 ^{Ba}	108.96+2.81 ^{Ba}	101.32+3.22 ^{Bb}
Filtek Z350XT	103.72+3.62 ^{Ca}	100.78+1.63 ^{Bb}	84.75+8.44 ^{Bc}	105.16+2.88 ^{Ca}	97.72+5.1 ^{C b}	90.19+6.07 ^{B c}	102.93+1.7 ^{C a}	104.4+1.65 ^{Съ}	101.08+1.66 ^{Bc}

A/B/C: Within each immersion period, different capital letters in each column indicate significant differences between the composites. a/b/c: Within each composite resin, different small letters in each row indicate significant differences between the immersion periods.



Table 4. Mean surface microhardness variation percentages and standard deviation of tested composites before and after immersion in solutions

Beverages Comp	Hype Percentage of surface hardness variations		Percentag hardness	d Bull ge of surface s variations ime	Artificial saliva Percentage of surface hardness variations		
	after a week	after a month	after a week	after a month	after a week	after a month	
Filtek P90	7.26+2.32 ^A	21.60+3.62 ^a	7.28+2.22 ^A	20.34+4.43 ^a	66+.63 ^B	5.13+1.75 ^b	
Filtek Z250	10.30+3.72 ^A	26.92+4.85 ^a	6.39+2.88 ^B	16.35+3.53 ^b	80+2.31 ^C	6.24+3.60 °	
Filtek Z350XT	2.75+2.98 ^A	18.29+7.57 ^a	7.07+4.24 ^B	14.24+5.14 ^a	-1.43+1.16 ^C	1.79+.86 ^b	

A/B/C: Within each composite, the same capital letters in each row indicate no significant differences between solutions after a week. a/b/c: Within each composite, the same small letters in each row indicate no significant differences between solutions after a month.

Discussion

The aim of the present study was to evaluate the changes in surface microhardness of three types of resin composites from the same manufacturer after exposure to two popular energy drinks (Hype and Red Bull). Our results suggested a significant reduction in surface microhardness of all tested composite resins after oneweek and one-month immersion in Hype and Red Bull energy drinks. Surface microhardness variations were significantly higher after one month compared to one week of immersion. This finding is in accordance with the results of Fatima and Hussain [6] who evaluated the effect of two commonly available energy drinks on surface microhardness of tooth color restorative materials. They observed that the surface microhardness of the composite resin materials was significantly decreased, and nano composite exhibited less reduction than other composites.

In another study by Erdemir et al, [5] surface hardness values of the composite resin materials were significantly decreased, either immersed in distilled water or immersed in sports and energy drinks after 1month evaluation period. In this study, Filtek Silorane showed significantly lower initial surface microhardness compared to the tested methacrylatebased composites. In a study by Yesilyurt et al, [21] similar results were reported. In our research, siloranebased Filtek P90 with silorane-based was selected due to having new monomeric system and comparing with Z250 and Z350XT, two widely used methacrylate based composites with two different structure (microhybrid and nanofilled). Filtek Silorane is based on the silorane chemistry and does not contain methacrylates. The name silorane derives from its constituting molecules, oxirane and siloxane. The organic matrix of Filtek P90 is mainly composed of silorane resin and its inorganic particles include quartz and yttrium fluoride. Desirable abrasion resistance of Filtek P90 can be attributed to the small size of its filler particles and its stable chemical structure due to conjugation with silicon atoms. [19, 22] Microhardness of a composite is a function of several factors including the composition of organic matrix, type and size of filler particles and degree of conversion (DC). According to Yesilyurt et al, [21] the difference in surface hardness of restorative materials can be attributed to the difference in their filler or monomer ratio. The low initial surface microhardness of siloranebased composite can be due to its lower filler content (55 %vol) compared to methacrylate-based Z250 and Z350 XT composite resins (60 %vol and 59.5 %vol respectively).

For all restorative materials, the surface microhardness is varied with the immersion solution and immersion period. For each composite in both immersion periods, significant differences were found in surface microhardness variations between energy drinks and artificial saliva. Therefore, the first null hypothesis, which stated that energy drinks do not reduce the surface microhardness of composite resins, was rejected. [2]

Deterioration of resin materials is likely because of the water absorption. The presence of water can soften the resin by swelling the polymer network and reducing the frictional forces between the polymer chains. ^[2, 6] In



addition, composite resins are highly soluble in low pH solutions, and this can lead to matrix softening, surface abrasion and loss of structural ions. The acid in the energy drinks can penetrate into the resin matrix and accelerate the release of unreacted monomers via reducing the surface hardness. The energy drinks used in this study contained citric acid, which is known to have a damaging effect on hardness of dental surfaces and resin-based restorative materials. Of course, these degradation effects depend on the solubility of the resin restorative materials, which differ in the composite resins. Therefore, the increase of the interaction and reaction between solution and resin materials such as water absorption and erosion due to the acidic condition leads to the decrease in the surface hardness of resin composites. [23, 24]

Variuos erosive potentials of different energy drinks can also be explained by other factors such as buffering capacity of saliva and acid type and non-reducing sugar contents of energy drink. ^[12] In contrast to the results of immersion in energy drinks, a week-long immersion in artificial saliva increased the surface microhardness of all three composite resins. Similar results have been reported in previous studies. ^[2, 5, 25] This finding can be attributed to the post-curing cross-linking reactions in the resin matrix which increases the monomer conversion and allows chemical bonds to continue to be made.

The results of this study illustrated a reduction in surface microhardness of the composites after one-month immersion in test and control groups. In the oral environment, resin materials are susceptible to degradation and quality reduction owing to water absorption and, as stated, the presence of water can damage the resin hardness and lead to its softening. ^[2] According to Awliya et al, the amount of water absorption depends on the resin content of the composite material and the quality of the bond between the resin and filler particles. It has been reported that excessive absorption of water may reduce the lifespan of composite resins by expanding and plasticizing the resin components. ^[8]

The results also ruled out our second null hypothesis, stating that the changes in surface microhardness after immersion in energy drink solutions were the same in nanofilled, silorane, and microhybrid composites. Based on the immersion solution, different results were obtained from the comparison of surface microhardness variations of these composites.

Nevertheless, energy drinks had a lower impact on the surface microhardness of Filtek Z350 than that of other composites.

The nanofilled Z350 XT composite used in our study contains silica fillers of 20 nm in size and zirconia / silica particles in sizes ranging from 0.6 to $1.4\mu m$. It seems that the small filler size of Filtek Z350 compared to two other composites tested allows a smoother surface to emerge after polishing, and this perhaps results in more stability against surface alteration including alteration in surface hardness. [26-28]

After 1 month for all three composites, surface microhardness reduction of samples immersed in Hype energy drink was higher than those immersed in Red Bull solution. However, this finding was statistically significant only in Z250 composite resin. This result may be attributed to the slight difference in the pH of two drinks and higher acidity of Hype, which have a greater softening effect on the resin matrix and has led to dislodgment of filler particles and reduced load resistance of the composite resins. It is worth to mention that since the aforementioned energy drinks have a largely similar chemical composition, the sample size and test duration of this study are not broad enough to allow a definitive judgment to be made on the differences of these two drinks, and such conclusion requires further research on this subject. In addition to PH, temperature can be an important factor in the abrasive effects of energy drinks. If these drinks are used at higher temperatures, these effects will be exacerbated. [2]

In the present study, composite specimens were immersed, for 5 minutes per day, in the energy drink solutions stored at room temperature ($23 \pm 1^{\circ}$ C). Since energy drinks are typically stored and consumed at low temperature, future studies are recommended to also examine the effect of temperature.

In addition, this study made no direct evaluation on the effect of pH of energy drinks on the surface hardness of restorative materials and the arguments were based on the results and hypotheses of recent studies. Hence, there is still a need for further work in this venue of research.

Conclusion

Within the limitations of this invitro study, it can be concluded that Hype and Red Bull energy drinks have a significant damaging effect on the surface hardness of composite resins, and these effects are increased with



duration of exposure so the patients who have a regular diet of such drinks should consider this issue.

The composition of composite resins had a noticeable effect on the surface microhardness changes. Variation in the surface microhardness of Z350 XT composite was lower than Z250 and Filtek P90 composites.

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Authors' Contributions

Ghazaleh Ahmadizenouz developed the original concept and design and supervised the in vitro procedure and preparation of manuscript. Sahar Khorshidi carried out the in vitro procedures, acquisition of data and writing the manuscript. Behnaz Esmaeili supervised the procedure and edition of manuscript. Soraya Khafri developed the interpretation of data and statistical analysis.

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