





Research Article

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Effect of Energy Drinks on Surface Roughness and Weight Loss of Custom-Made Mouthguard Material

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Article Info	ABSTRACT
Article History	Aim: To investigate the effect of different drinks on the surface roughness and weight loss of custom-made mouthguard material prepared in two different thicknesses.
Received: 20.06.2024 Accepted: 23.09.2024 Published: 15.10.2024	Materials and Methods: The custom-made mouthguard material Ethylene vinyl acetate (EVA) (4.0x125 mm) was used in the current research. 102 rectangular prism specimens (8 × 8 mm) from two different thickness (3 and 4 mm) were prepared. They were randomly divided into four different groups (n=13), numbered, and initial weight and roughness measurements were recorded. All specimens were then stored in 4 different drinks, including 2 different energy drinks, an isotonic sports drink, and water. At the end of
Keywords: Energy drinks, Etilen Vinil Asetat, Mouthguard, Roughness, Weight.	the 7-day and 28-day periods, weight and roughness measurements of the specimens were again performed and recorded. The data were analyzed the repeated measures ANOVA, one-way ANOVA, and the paired sample t tests (α =.05). Results: There were no statistically significant differences in the weight × beverage and roughness × beverage interactions (p>0.05) both weight and roughness changes of specimens at 0, 7, and 28 days. Conclusion: Energy drinks and sports drinks have no effect on the surface liquid and weight of the specially produced oral material.

Enerji İçeceklerinin Kişiye Özel Üretilen Mouthguard Materyalinin Yüzey Pürüzlülüğü ve Ağırlık Kaybı Üzerindeki Etkisi

Makale Bilgisi	ÖZET					
Makale Geçmişi	Amaç: Bu çalışmanın amacı, iki farklı kalınlıkta hazırlanan kişiye özel mouthguard malzemesinin yüzey pürüzlülüğü ve ağırlık kaybı üzerine farklı içeceklerin etkisini araştırmaktır.					
Geliş Tarihi: 20.06.2024 Kabul Tarihi: 23.09.2024	Gereç ve Yöntemler: Bu çalışmada özel yapım ağızlık malzemesi Etilen vinil asetat (EVA) (4,0x125 mm) kullanılmıştır. İki farklı kalınlıkta (3 ve 4 mm) 102 dikdörtgen prizma numunesi (8 × 8 mm) hazırlanmıştır. Numuneler daha sonra rastgele dört gruba ayrılmış (n=13), numaralandırılmış ve ilk ağırlık ve pürüzlülük					
Yayın Tarihi: 15.10.2024	ölçümleri kaydedilmiştir. Tüm numuneler daha sonra 2 farklı enerji içeceği, bir izotonik spor içeceği ve su olmak üzere 4 farklı içecekte saklanmıştır. 7 günlük ve 28 günlük sürelerin sonunda numunelerin ağırlık ve pürüzlülük ölçümleri tekrar yapılmış ve kaydedilmiştir. Veriler tekrarlanan ölçümler ANOVA, tek yönlü					
Anahtar Kelimeler:	ANOVA ve eşleştirilmiş örneklem t testleri ile analiz edilmiştir (α =,05).					
Enerji içeçeği,	Bulgular: Ağırlık \times içecek ve pürüzlülük \times içecek etkileşimlerinde (p>0,05) 0, 7 ve 28. günlerde					
Etilen Vinil Asetat,	numunelerin hem ağırlık hem de pürüzlülük değişimlerinde istatistiksel olarak anlamlı bir fark					
Ağız koruyucu,	bulunmamıştır.					
Pürüzlülük,	Sonuç: Enerji içecekleri ve spor içeceklerinin özel olarak üretilen ağız materyalinin yüzey sıvısı ve ağırlığı üzerinde herhangi bir etkisi yoktur.					
Ağırlık.						
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INTRODUCTION

One of the etiologic factors for traumatic dental injuries is sports accidents. Mouthguards are widely used in many sports and can significantly reduce the risk of orofacial injuries.^{1,2} Mouthguards are a highly protective and low-cost device designed to absorb the impacts that cause orofacial injuries.³ Many sports use it as effective personal protective equipment. The production, materials used, and designs of mouth guards have changed over time according to their effectiveness. A properly manufactured mouthguard absorbs the impact inside the mouth, restricting the force to the hard and soft tissues in the mouth and distributing it evenly throughout the orofacial complex.³ Mouthguards must be of sufficient thickness labially, palatially, lingually, and occlusally to be protective. The proper distribution of force that the athlete will experience during the impact will enable them to endure the stress and strain.

The mouthguard's thickness needs to fall between the minimum required values for this function.⁴ In no place should the mouthguard's average thickness be less than 3 mm. In general, the thickness of the mouthguard on different surfaces should be between 3-4 mm.⁵ If it is even thinner, it will not provide appropriate protection against potential hits and will also have a negative influence on the athlete's airway and speech.⁶ Following shaping with a vacuum shapping equipment, a thickness reduction of 47% on the surface and 54% on the buccal surface may be observed, with 38% on the incisal edge and 38% on the labial edge.^{7,8} Due to the model's varying height and width, thickness variations may also happen when making the mouthguard in the vacuum forming device, depending on the athlete's shaped and deepened palate dome as well as the axes of their anterior teeth.⁴ According to the American National Standards Institute / American Dental

Association (ADA), the types of mouthguards are classified as type I stock, type II mouth (Class 1. Thermoplastic, Class 2. Chemical Hardening), type III special (Class 1. Vacuum, Class 2. Model).⁹ Mouthguards that are custommade fall under type III. Custom mouthguards are currently thought of as the standard of the best option,^{1,10,11} because it can be completely altered to fit the patient's preferences for style and anatomy. EVA is a commonly used material for custom-made mouthguards.^{12,13} EVA is a biocompatible, highly elastic, aesthetic, heatformable, and non-allergenic material.¹⁴

The use of energy drinks is particularly popular among athletes as they have a performance-enhancing effect. Energy drinks are non-carbonated, sugar-sweetened beverages that are believed to enhance cognitive and physical performance.¹⁵ Energy drinks help to replace the fluid electrolytes lost during and after exercise, which contributes to improving the athlete's performance.¹⁶ Energy drinks are commonly eaten by athletes to enhance their physical performance and endurance, as well as to prevent dehydration.¹⁷ Athletes have a dangerous consumption pattern during training and competition when they take "small sips from a bottle" of sports drinks and gels for carbohydrates.^{18,19} In addition, consumption of energy drinks may pose a risk to oral hygiene and may cause dental erosion.²⁰ There are studies^{21,22} in the literature that demonstrate that energy drinks can promote dental erosion, but none have looked at how they may affect the material of mouthguards worn by athletes.

This study set out to look into the effects of several energy drinks and sport drinks on the surface roughness and weight change of custom-made mouthguard material prepared in two different thicknesses. The null hypothesis of the study is that different drinks will affect the weight and roughness values of the mouthguard material, regardless of the thickness difference.

MATERIALS AND METHODS

A power analysis was conducted utilizing the G*Power software application. (v3.0.10; Heinrich Heine University Düsseldorf) to obtain the highest power level with the smallest specimen size. The examination indicated that a minimum of thirteen specimens were necessary to achieve the highest level of power, with a total of 102 specimens utilized in the study (power=80, α =0.05).

The mouthguard material in the study was a 4.0x125 mm clear Bioplast Xtreme (Scheu-Dental GmbH, Iserlohn, Germany). Before the mouthguard is applied to the mouth, the material is adapted to the model using a special machine (Biostar; Scheu-Dental GmbH, Iserlohn, Germany) in accordance with the heat and pressure technique. The Bioplast Xtreme disks used in the study were heat treated in line with the manufacturer's instructions and let to cool naturally without being molded onto any models to simulate the mouthguard forming processes of the material. Differences that could have resulted from heat treatments were removed in this way. The thickness of the lamination surface of the mouthguard material, which protects against impact, is slightly higher than the remainder of the material. Both surfaces were considered as parameters in determining the study groups. Specimens were prepared by cutting a rectangular prism of $8 \times 8 \times 4$ mm for thicker (T4) and $8 \times 8 \times 3$ mm for thinner (T3) under water cooling with a diamond bur. The accuracy of the specimen dimensions after each cut the measurement was taken using a digital caliper (Absolute AOS; Mitutoyoto Corp., Kanagawa, Japan) to ensure standardization. Every specimen was given a number, and a computerized analytical balance Mettler (XS105, Toledo. Greifensee. Switzerland) was used to take the initial measurements of the weigh. Each specimen was weighed on analytic balance with a precision of 0.1 mg. The initial surface roughness measuerements of the specimens were recorded by two-dimensional profilometer (Surtronic 25; Taylor Hobson, Leicester, U.K.) (cutoff value (λc) of 0.80 mm, an evaluation length of 2.4 mm). The initial weight values (w0) and the surface roughness values (Ra, r0) values were recorded. The specimens were then randomly divided into 4 groups (n=13) and placed in boxes (70×20 mm) with transparent plastic lids. The specimens were stored in 4 different beverages, including three commercial energy drinks and water. Information about the energy drinks used is given in Table 1. The storage of the specimens in beverages was determined as follows:

Table 1. Ingredients of energy drinks

Energy drink Manufacturer		pН	Ingredient			
Red Bull	Red Bull Company; Austria	3.81	Water, sucrose, glucose, acidifier sodium citrates, carbon dioxide, taurine (0.4%), glucuronolactone (0.24%), caffeine (0.03%), inositol, vitamins (niacin, pantothenic acid, B6, B12), flavourings, and colours (caramel, riboflavin). Sugar content: 27 g (per 8.46 oz)			
Burn	The Coca-Cola Company; Atlanta, Georgia, USA	3.03	Carbonated water, sucrose, citric acid, taurine (0.4%), acidity regulator: sodium citrate, coloring agents: E163, E150d, preservatives: potassium sorbate, sodium benzoate, flavor, caffeine (0.03%), inositol, vitamins [nicotinamide (B3), d calcium pantothenate, pyridoxine hydrochloride (B6), cyanocobalamin (B12)], seed extract of guarana (0.005%), antioxidants (ascorbic acid)			
Powerade	The Coca-Cola Company; Atlanta, Georgia, USA	3.79	Water, Glucose, Acid (Citric Acid), Acidity Regulators (Sodium Citrates, Potassium Citrates), Fructose, Stabilisers (Acacia Gum, Glycerol Esters Of Wood Rosins), Sweeteners (Aspartame, Acesulfame-K), Flavourings, Colour (Brilliant Blue), Vitamin B6			

The bottle containing each specimen was filled with 10 mL of the beverage to exceed the surface of the specimen and placed in an oven (Nükleon NIN-30; Nükleon, Ankara, Turkey) at 37 °C for 2 minutes.17 Then, the beverages in each specimen were removed from the oven, and 10 mL of distilled water was added to exceed the specimen surface and stored at room temperature until the next beverage application. The purpose of storing the specimens in distilled water was to prevent weight loss by storing them in a dry environment, and the reason for closing the mouths of the containers was to prevent the water from evaporating and reducing the level. This procedure was performed every day at the same time and by the same researcher in order to complete each 24hour period. At the conclusion of the 7-day period, weight and roughness measurements of the specimens were again performed and recorded according to the initial measurement procedures (w7, r7). The storage of the specimens in the beverages was repeated for 28 days, and at the end of the 28th day, weight and roughness measurements were performed again, and the data were recorded (w28, r28). The color changes caused by different drinks on the specimens were also visually examined.

The data was statistically analyzed using a statistical software tool (IBM SPSS Statistics, v20; IBM Corp). The analyses employed the paired sample t test, repeated measures ANOVA, one-way ANOVA, and the Kolmogorov-Smirnov test for normalcy (α =.05).

RESULTS

There were no statistically significant differences in the weight×beverage and roughness×beverage interactions (p>0.05), according to the two-way ANOVA test results used to assess the weight and roughness changes of both T3 and T4 specimens at 0, 7, and 28 days. Based on the results of the weight measurements of both T3 and T4 specimens;

There were no statistically significant differences specimens (T3, T4)xbeverages interactions (p=0.58), there but were statistically significant differences between specimens (p=0.001), and among the beverages (p=0.004) at 0 day. T4 (0.258930±0.0365999) weight than had less T3 (0.283988 ±0.0414878). Burn and Powerade had lower weight values than water, while the weight differences between RedBull and water and other drinks were not significant.

There were no statistically significant differences specimens (T3, T4) x beverages interactions (p=0.65), but there were statistically significant differences between specimens (p=0.004), and among the beverages (p=0.016) at 7 days. T4 (0.263168 \pm 0.0353625) had less weight than T3 (0.284944 \pm 0.041556). Burn had lower weight values than water, while the weight differences between the others were not significant.

There were no statistically significant differences specimens (T3,T4)xbeverages (p=0.59), interactions but there were statistically significant differences between specimens (p=0.001), and among the beverages (p=0.01) at 28 days. T4 (0.26358±0.0353647) had less weight than T3 (0.288475±0.041924). Burn had lower weight values than water, while the weight differences between the others were not significant.

Based on the results of the surface roughness measurements of both T3 and T4 specimens;

There were no statistically significant differences specimens (T3,T4)xbeverages interactions (p=0.43), but there were no statistically significant differences between specimens (p=0.91), and among the beverages (p=0.3) at 0 day.

There were statistically significant differences specimens (T3,T4)xbeverages interactions (p=0.01), but there were no statistically significant differences between specimens (p=0.65), and among the beverages (p=0.88) at 7 days.

There were statistically significant differences specimens (T3, T4)xbeverages interactions (p=0.03), but there were no statistically significant differences between specimens (p=0.53), and among the beverages (p=0.15) at 28 days. According to the one-way

ANOVA results for the comparison of T3 and T4 group specimens, there were no significant differences (p>0.05) in terms of both weight and roughness changes at 7 and 28 days. Tables 2 and 3 display the mean and standard deviation values.

		w0		w7		w28		Ν
		Mean	Standard Deviation	Mean	Standard Deviation	Mean	Standard Deviation	
Т3	Water	.302354	.0436971	.302923	.0439729	.307554	.0436894	13
	Red Bull	.285469	.0422565	.288115	.0411814	.291838	.0411432	13
	Powerade	.284446	.0318508	.284092	.0336150	.288292	.0334585	13
	Burn	.263685	.0423218	.264646	.0420909	.266215	.0424908	13
	Total	.283988	.0414878	.284944	.0415560	.288475	.0419240	52
T4	Water	.282831	.0259529	.283062	.0259560	.283638	.0256130	13
	Red Bull	.262915	.0376436	.263038	.0375385	.264046	.0374451	13
	Powerade	.241462	.0408700	.249162	.0420741	.249162	.0420741	13
	Burn	.246618	.0269066	.256364	.0261400	.256364	.0261400	13
	Total	.258930	.0365999	.263168	.0353625	.263580	.0353647	52

Table 2. Least square means and standard deviation (SD) for weight loss

(T3: 3 mm thick specimens, T4: 4 mm thick specimens; w0: Weight measurement on day 0, w7: Weight measurement on day 7, w28: Weight measurement on day 28)

		r0			r7	r28		Ν
		Mean	Standard Deviation	Mean	Standard Deviation	Mean	Standard Deviation	
Т3	Water	.54	.33	.76	.24	.44	.13	13
	Red Bull	.41	.11	.65	.66	.68	.51	13
	Powerade	.51	.21	.41	.07	.52	.21	13
	Burn	.69	.52	.68	.57	.72	.52	13
	Total	.54	.34	.63	.46	.59	.39	52
T4	Water	.52	.15	.61	.28	.53	.16	13
	Red Bull	.5	.13	.64	.2	.6	.32	13
	Powerade	.56	.32	.88	.39	.88	.39	13
	Burn	.53	.22	.51	.11	.52	.11	13
	Total	.53	.21	.67	.29	.64	.3	52

 Table 3. Least square means and standard deviation (SD) for surface roughness

(T3: 3 mm thick specimens, T4: 4 mm thick specimens; r0: Roughness measurement on day 0, r7: Roughness measurement on day 7, r28: Roughness measurement on day 28)

increases in weight or roughness (p>0.05).

The paired sample t test's results, which analyzed the weight and roughness changes of the beverages individually for the specimens from the T3 group: w0-w7 (p<0.05), w0-w28 (p<0.001), and w7-w28 (p<0.001) for water, Only the change from w0-w28 (p<0.05) for Red Bull, Only the change w7-w28 (p<0.05) for Powerade showed significance.

Burn experienced no significant

The paired sample t test's results, which analyzed the weight and roughness changes of the beverages individually for the specimens from the T4 group:

Changes in w0-w7 and w0-w28 for water (p<0.05), w0-w28 (p<0.05), w7-w28 (p<0.05), and r0-r7 (p<0.05) adjustments for Red Bull, Changes in r0-r7 and r0-r28 for Powerade were significant (p<0.05).

Burn experienced no significant increases in weight or roughness (p>0.05).

DISCUSSION

The purpose of the study was to investigate the roughness and weight changes of custom-made mouthguard materials made in two different thicknesses after storage in various energy drinks. The null hypothesis was partially rejected since sample beverage interactions and weight measurements did not show statistical differences, and although there was no difference in roughness results on day 0, there was a difference on days 7 and 28.

Mouthguards may have different thicknesses or different thicknesses in different parts of the apparatus depending on the type of sport. A thicker mouthguard has higher protection than a thinner mouthguard.²³ EVA reduces impact energy to act as a form of shock absorber for hard forces.²⁴ Another aspect of EVA that should be considered for athletes is its great capacity for cushioning, given that it can soak up extra moisture from the oral area.²⁵ Water absorption and material thickness are the two most crucial characteristics required of the materials used in the manufacture of mouth guards.²⁶ Additionally, because microorganisms stick to surfaces more readily, the surface roughness of materials used in the mouth can have a direct impact on bacterial colonization. A higher bacterial colonization rate is a sign of developing gum and tooth disease.²⁷ One of the many diverse characteristics of dental materials is surface roughness, which is crucial because it has to do with microbial adhesion. Borro et all. assessed the surface roughness of EVA both before and after various cleaning techniques in their investigation into the significance of surface roughness's impact on bacterial adherence and bio-film formation.²⁸ There is research in the literature on how EVA layer thickness and color affect surface roughness.29 It has been demonstrated in the literature that mechanical biofilm cleansing (tooth brushing) and friction and abrasion from use may have contributed to

the increase in surface hardness of sports mouthguards manufactured of EVA plates.³⁰ The hardness and surface roughness of acrylic resin samples with alloys fixed on top that had been soaked in artificial saliva for five minutes before being submerged in sodium bicarbonate, hydrogen peroxide, and water (control) were examined in a study conducted by Garcia et all., which supported our findings.³¹ Similar to the increase in roughness values in the days that followed our investigation, the data demonstrated that the surface roughness of the other samples increased in comparison to those submerged in water. In a different investigation, it was demonstrated that the immersion method in sodium perborate, which was used to disinfect the mouthguard material, greatly raised the surface roughness values.32 This demonstrates that the energy drink effect in our investigation is equally significant to the dipping and holding strategy.

Energy drinks have been reported to have deleterious effects on dental tissues and restorative materials because of their low pH levels and large quantities of non-reducing carbohydrates, even though there are remarkably few studies in the literature looking at how they affect mouthguard materials. Energy drinks like Red Bull and Burn have been reported to significant surface degradation of restorative materials,³³ while Red Bull was found to have the highest total titratable acidity and a strong propensity to erode enamel. Tanthanuch et all.³⁴ evaluated the effect of repeated exposure to sports and energy beverages on the surface characteristics of glass ionomer, nanohybrid, and bulk-fill resin composites. Surface hardness decreased significantly, whereas surface roughness and color increased significantly. Investigations on the effects of sports and energy drinks on the surface characteristics of dental materials have been published in the literature; however, investigations on the impact of mouthguards on surface roughness have not. Contrary to evidence in the literature, energy drinks had partially effect on the surface roughness and weight change of EVA material, according to the results of the current study. This might be attributed to alterations in the chemical composition of the EVA material employed, as well as a difference in retention durations. Although the time interval measurements of Red Bull and Powerade brand energy drinks differed, there were no discrepancies in the time interval measurements of Burn brand energy drink. These discrepancies are assumed to be caused by the ingredients in the energy drinks.

The present study has some limitations, including the use of just two distinct thicknesses, the lack of a surface hardness evaluation, and a maximum time interval of 28 days for the specimens. Future studies may yield different findings when using specimens with various thicknesses, storage times, and time intervals.

CONCLUSIONS

Within the limitations of this in vitro study, the following conclusions were drawn:

- 1. Weight measures are not significantly affected by interactions with beverages.
- 2. Energy drinks have effect on the surface roughness of EVA-based custom-made mouthguards.

Ethical Approval

This in-vitro study does not require ethics committee approval.

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The authors declare that this study received no financial support.

Conflict of Interest

The authors deny any conflicts of interest related to this study.

Author Contributions

Design: TS, AO, TK, Data collection and processing: TS, TK, Analysis and interpretation: TS, AO, Literature review: TS, AO, TK, Writing: TS, AO.

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