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## EFEKAT RASTVORA EKSTRAKTA BILJNE VRSTE *FOENICULUM VULGARE MILL.* NA MEHANIČKA I SVOJSTVA KVAŠENJA TOPLOPOLIMERIZUJUĆEG AKRILATA ZA BAZU PLOČASTE PROTEZE

### THE EFFECT OF HERBAL EXTRACT *FOENICULUM VULGARE MILL.* SOLUTION ON THE MECHANICAL AND WETTING PROPERTIES OF HEAT POLYMERIZED DENTURE BASE RESIN

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#### Sažetak

**Uvod.** Komercijalno dostupna hemijska sredstva koja se upotrebljavaju za čišćenje proteza imaju negativan uticaj na mehanička svojstva proteze. Stoga, postoji potreba za pronalaženjem alternative koja neće menjati površinska svojstva materijala.

**Cilj.** U ovom radu je ispitivan uticaj ekstrakta biljne vrste *Foeniculum vulgare Mill.* na kvašenje i mehanička svojstva topopolimerizujućeg akrilata za bazu pločaste zubne proteze u poređenju sa istim svojstvima iste smole u prirodnom stanju (na vazduhu) i izložene samo vodenj sredini.

**Maaterijali i metode.** Uzorci akrilata u obliku parvougaoika (15 mm x 15 mm x 3 mm) su pripremljeni i podeljeni na: (i) uzorak koji je koninuirano bio izložen vazduhu – AIR; (ii) uzorak koji je koninuirano bio izložen destilovanoj vodi - DW; i (iii) uzorak koji je bio izložen 10% rastvoru lista morača – PES. Svojstva kvašenja su merena metodom sesilovije kapi, pri čemu su mehanička svojstva analizirana metodom nanoindentacije.

**Rezultati.** Rezultati su pokazali da su uzorci AIR imali najnižu vrednost ΘC vrednost 64.5±2.3 °, dok su drugi uzorci DW i PES imali sličnu ΘC vrednost (75.4±3.3 °, 75.7±4.7 °, redom). Štaviše, nije bilo statistički značajne razlike u koeficijentu tvrdoće, dok je Jungov modul elastičnosti pokazao značajnu razliku (p < 0,005) samo između uzoraka AIR i DW.

**Zaključak.** Na osnovu dobijenih vrednosti potvrđeno je da ekstrakt biljne vrste morač može biti uspešno korišćen i efikasan kao sredstvo za čišćenje, bez degradacije površine proteze.

**ključne reči:** baza ploče proteze, ekstrakt morača, kvašenje, tvrdoća, Jungov modul elastičnosti

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#### Abstract

**Introduction.** Commercially used chemical dental cleansers adversely affect mechanical properties of denture. Therefore, there is a need for finding an alternative which will not change the surface properties of material.

**Aim.** This paper studied the effect of the herbal extract (*Foeniculum vulgare Mill.*) solution on the wetting and mechanical properties of heat-polymerized denture base resin compared to the same properties of the same resin in the native state (in air) and exposed only to aqueous environment.

**Material and methods.** Rectangular shape of heat polymerized acrylic resin (15 mm x 15 mm x 3 mm) were prepared and divided into: (i) sample continuously exposed to air – AIR; (ii) sample continuously exposed to distilled water - DW; and (iii) sample exposed to 10% solution of the fennel leaves extract – PES. Wetting properties were measured by sessile drop method, whereas mechanical properties were analyzed by nanoindentation method.

**Results.** Results showed that samples AIR had the lowest ΘC value 64.5±2.3 °, while the other two samples DW and PES had similar ΘC values (75.4±3.3 °, 75.7±4.7 °, respectively). Furthermore, there was no statistically significant difference in hardness coefficient, while Young's modulus showed a significant difference (p < 0.005) only between the samples AIR and DW.

**Conclusion.** The obtained values confirmed that fennel extract can be used as successful and efficient cleanser, without degradation of denture surface.

**Key words:** denture base resin, fennel extract, wetting hardness, Young's modulus

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## Uvod

Polimetil-metakrilat (PMMA) je polimer koji ima široku primenu u biomedicinskim aplikacijama, posebno u stomatologiji, kao glavna komponenta za izradu baze pločaste zubne proteze<sup>1</sup>. Tokom upotrebe mobilne proteze se nalaze unutar usne duplje, a kada nisu u upotrebi, drže se u vodi ili u tečnom sredstvu za čišćenje proteza<sup>2</sup>. Na ovaj način, površina zubne proteze je redovno u direktnom kontaktu sa oralnom mukozom. Stoga je od izuzetne važnosti održavanje fizičke, hemijske i biološke čistoće baze zubne proteze<sup>2,3</sup>. Fizička čistoća se postiže mehaničkim metodama čišćenja, od kojih je četkanje jedan od najefikasnijih i najčešće korišćenih metoda<sup>3,4</sup>. Hemijsko čišćenje proteza uobičajno podrazumeva korišćenje hemijski agresivnih jedinjenja koja često sadrže hlor ili razne peroksidge<sup>5</sup>. Oba sastojka utiču na degradaciju PMMA (čak pojedina jedinjenja hlora, kao što je hloroform ili hloretan, mogu potpuno da rastvore PMMA)<sup>5,6</sup>. Kao posledica toga, površina baze pločaste zubne proteze gubi svoja inherentna mehanička svojstva (kao što su relativno visoka vrednost modula elastičnosti i tvrdoće za plastiku) i postaje mekana. Štaviše, interakcije gornjeg sloja baze proteze i hemijskih sredstava za čišćenje utiče na površinsku morfologiju<sup>2</sup>. Usled navedenog, svojstva kvašenja baze pločaste zubne proteze su takođe modifikovana. Zapravo, najveći problem u održavanju baze zubne proteze čistom tiče se biološkog aspekta čistoće, koji podrazumeva uklanjanje i prevenciju adherencije bakterija i gljivica<sup>8</sup>. U toku standardnog korišćenja zubne proteze u usnoj duplji, lako može doći do kolonizacije oralnih mikroorganizama. Naime, različiti mikroorganizmi gljivice i bakterije, kao što su *Candida spp*, *Streptococci* i drugi respiratorni patogeni, mogu da kolonizuju unutrašnju površinu proteze i da formiraju biofilm<sup>3,9</sup>. Na taj način, proteza se ponaša kao rezervoar infekcije, koja često može da dovede do razvoja proteznog stomatitisa. Prema tome, protezni stomatitis se javlja kod više od 65% pacijenata koji nose pločaste proteze<sup>10</sup>. Dodatno, zubne proteze mogu da budu kontaminirane u nekom od koraka same izrade kao i u usnoj duplji. Takođe, unakrsna kontaminacija može da nastupi između zubotehničke laboratorije i stomatološke ordinacije, kao i između pacijenta i stomatologa.

## Introduction

Polymethyl methacrylate (PMMA) is acrylic polymer widely used in biomedical applications, especially in dentistry as a main component of denture base<sup>1</sup>. Dentures are on the daily base placed in the oral cavity and in-between the daily use in tap-water or some denture cleanser<sup>2</sup>. In this way, surface of denture base is regularly in direct contact with oral mucosa. Therefore, maintaining physical, chemical and biological cleanness of denture base is essential<sup>2,3</sup>. Physical cleanness is provided by mechanical methods among which brushing is the most common and effective one<sup>3,4</sup>. Chemical denture cleaning typically assumes the use of chemically aggressive compounds which often contain chlorine or form peroxides<sup>5</sup>. Both are known to degrade PMMA (even some compounds of chlorine, such as chloroform or some chlorethanes, can completely dissolve PMMA)<sup>5,6</sup>. Consequently, the surface of denture base loses its inherent mechanical properties (such as for-plastic relatively high modulus and hardness) and becomes soft. Moreover, interactions of the top-most layer of the denture base surface and the chemical cleansers also affect the surface morphology<sup>2</sup>. Thus, wetting properties of denture base are also modified. In fact, the biggest issue in maintaining a denture base clean poses the biological aspect of cleanness which involves the removal and prevention of bacterial and fungal attachment<sup>8</sup>. During the standard use of denture in the oral cavity, it can be easily colonized by oral microorganism. Thus, different fungal and bacterial microorganism, such as *Candida spp*, *Streptococci*, and other respiratory pathogens can colonize the inner surface of denture and form a biofilm<sup>3,9</sup>. In that way, denture acts as a reservoir of infection which often causes denture stomatitis<sup>9</sup>. Indeed, more than 65% of denture wearers are affected by denture stomatitis<sup>10</sup>. In addition, denture can be contaminated already at some steps of its fabrication and insertion in the oral cavity. Moreover, cross-contamination can occur between the prosthodontic laboratory and dental office as well as between patients and dental personnel. It has been reported that even denture laboratory pumice and polishing wheel can be contaminated<sup>2,11</sup>.

U radovima se navode slučajevi kontaminacije bimštajna i četki za poliranje u zubotehničkim laboratorijama<sup>2,11</sup>.

Iz navedenih razloga, adekvatna higijena zubnih proteza je veoma važna za prevenciju kontaminacije proteza sa stanovišta bioloških aspekata. Zapravo, smatra se da idealni proizvod za čišćenje proteza treba da ima baktericidna i fungicidna svojstva, a takođe treba da bude jeftin i bez negativnih efekata na fizička i mehanička svojstva proteze nakon čišćenja<sup>2,8</sup>.

Imajući u vidu sve gore navedene aspekte, postoji potreba za sredstvom za čišćenje pločastih proteza koji ne poseduje hemijski agresivne komponente (kao sredstva koje sadrže hlor, perokside itd.), koji je efikasan u čišćenju i koji takođe poseduje antimikrobne efekte, ali istovremeno ne utiče na mehanička svojstva i svojstva kvašenja zubnih proteza. Usled navedenog, brojna istraživanja su usmerena ka primeni prirodnih proizvoda i pronalaženju novih terapijskih alternativa u etarskim uljima i ekstraktima biljaka<sup>12,13</sup>. Ekstrakt morača (*Foeniculum vulgare Mill.*) se na primer intenzivno proučava za različite primene u medicini zbog svojih antibakterijskih, antigljivičnih, antioksidansnih i antiinflamatornih svojstava. Povrh toga, pokazano je da su i eterska ulja i ekstrakti ploda morača efikasni u borbi protiv gljiva roda *Candida* koje su najčešći uzrok proteznog stomatitisa<sup>14,15</sup>. Ekstrakti lista morača ispoljavaju antibakterijska, antigljivična, antioksidativna i antiinflamatorna svojstva usled visokog sadržaja polifenola kao što su hlorogena kiselina, kafeoil, heterozidi kvercetina, kemferola i izoramnetina<sup>14</sup>.

Imajući u vidu antiinflamatorne i antimikrobne efekte morača (naročito na gljive roda *Candida*), kao i njegovu nisku cenu, ekstrakti njegovih listova mogu se smatrati obećavajućim sredstvom za čišćenje proteze. Međutim, nije poznato da li ovaj ekstrakt može da utiče na mehaničke osobine i osobine kvašenja baze pločaste zubne proteze.

### Cilj

Cilj ovog rada bio je da se prouči uticaj rastvora ekstrakta morača na svojstva kvašenja (merenjem kontaktnog ugla) i osnovna mehanička svojstva (Jungov modul elastičnosti i tvrdoća) površine baze pločaste zubne proteze korišćenjem nano indentacije i poređenje tih svojstava sa svojstvima netretiranih uzoraka.

Therefore, adequate denture hygiene is very important to prevent denture contamination, especially from a biological aspect. Actually, it has been proposed that an ideal denture care product should have bactericidal and fungicidal properties and should be cost-effective without an adverse effect on physical and mechanical properties of denture after the cleaning process<sup>2,8</sup>.

Having in mind all these, there is a need for a denture cleanser which does not contain chemically aggressive components (such as ones having chlorine, peroxides, etc.), which is effective in cleaning and which also has antimicrobial effect, but which simultaneously does not affect the mechanical and wetting properties of denture. For this purpose, numerous research efforts are directed towards natural products and a search of new therapeutic alternatives among plants and their essential oils and extracts<sup>12,13</sup>. For instance, an herbal extract of fennel (*Foeniculum vulgare Mill.*) has been extensively studied for various health applications because of its antibacterial, antifungal, antioxidant and anti-inflammatory effects. Moreover, it has been shown that both an essential oil and extracts of seeds and fruits of fennel are effective against *Candida* which is the most often cause of denture stomatitis<sup>14,15</sup>. Fennel leaves' extracts have antibacterial, antifungal, antioxidant and anti-inflammatory effects due to high content of polyphenols, such as quinic acid, caffeoyl quinic acids, heterosides of quercetin, kaempferol and isoramnetin<sup>14</sup>.

Bearing in mind the anti-inflammatory and antimicrobial effect of fennel (especially on *Candida*), and its low cost, leaves extract of fennel can be considered as the promising denture cleanser. However, it is not known if this extract could affect the mechanical and wetting properties of denture base acrylic resin.

### Aim

Therefore, the aim of this work was to study the effect of fennel's extract solution on the wetting properties (by measuring contact angle) and on the essential mechanical properties, by measuring Young's modulus and hardness of the surface of denture base resin (by nanoindentation method) and comparing these properties with those on non-treated samples.

## ***Materijali i metode***

### ***Priprema uzoraka***

U ovom istraživanju pripremljena su tri uzorka od akrilata standardnom procedurom. Ukratko, prvo su izrađeni gipsani kalupovi, neophodni za izradu uzoraka. Ovi gipsani kalupovi su napravljeni na sledeći način: gips tipa IV je uliven u metalnu kivetu; voštani uzorci pravougaonog oblika dimenzija koje odgovaraju dimenzijama uzoraka (15x15x3 mm) su uloženi u gips kako bi se dobili otisci ovih pravougaonika u gipsu; zatim je nakon stvrdnjavanja gipsa, vosak uklonjen i tako dobijeni gipsani kalup je očišćen. Na ovaj način dobijeni gipsani kalupovi korišćeni su za izradu uzoraka na sledeći način: toplo polimerizujući akrilat (Probase, Ivoclar Vivadent, Lichtenstein) pripremljen je prema uputstvu proizvođača mešanjem PMMA i MMA u preporučenom odnosu i nakon toga unešen u prethodno pripremljen gipsani kalup. Toplotna polimerizacija akrilata je vršena u vremenskom periodu od 45 minuta stavljanjem kivete sa gipsanim kalupovima u ključalu vodu. Nakon polimerizacionog postupka, kivete su stavljene na sobnu temperaturu da se ohlade pre nego što su otvorene, izvađen je gipsani kalup i oslobođeni su uzorci od akrilata. Tako dobijeni uzorcima su polirani standardnom procedurom (uključujući prah, bimsstaj i kožnu četku). Nakon poliranja, svi uzorci su potopljeni u destilovanu vodu na sobnoj temperaturi 24h, kako bi se eliminisao rezidualni monomer.

### ***Priprema ekstrakta***

Listovi morača su sakupljeni na teritoriji Niša u oktobru na početku sazrevanja ploda. Materijal je osušen na suvom promajnom mestu, bez direktnog delovanja sunčevog zračenja. Osušeni listovi su čuvani u papirnoj kesi do ekstrakcije. Ekstrakcija je izvedena sa biljnim materijalom usitnjenim do nivoa praška i vodom (1:10), u erlenmajeru sa brušenim čepom na ultrazvučnom kupatilu u trajanju od 20 minuta. Nakon ekstrakcije, smeša je filtrirana, a voda je uklonjena rotacionim vakuum uparivačem. Prinos ekstrakta iznosio je 25,3%. Dobijeni suvi ekstrakt je prebačen u staklenu bočicu i čuvan na 4°C. Za ovu studiju ekstrakt je rekonstituisan u vodi kako bi se dobilo 10% rastvora ekstrakta (ovde imenovan kao rastvor biljnog ekstrakta, PES), koji je dalje korišćen u eksperimentu.

## ***Materials and methods***

### ***Sample fabrication***

In this study, three samples of denture-based acrylic resin were prepared by the standard dental procedure. Briefly, we first made dental-stone molds which are needed for sample fabrication. These dental-stone molds were prepared as follows: type IV dental stone in still liquid state was placed in a metal flask; wax patterns which have rectangular shape with dimensions which correspond to dimensions of samples (15 mm x 15 mm x 3 mm) were inserted into dental stone to obtain imprints of these rectangles in stone; upon solidification of stone, wax was removed and the obtained stone mold was cleaned. Thus-obtained stone molds were used for fabrication of samples as follows: heat polymerized acrylic resin (Probase, Ivoclar Vivadent, Lichtenstein) was prepared according to manufactures instruction by mixing PMMA and MMA in the recommended ratio and subsequently deposited into a previously prepared dental stone molds. Heat polymerization of resin was performed during the period of 45 min by placing the stone mold, which was in the metal flask, into the boiling water. Upon the polymerization process, metal flasks was left to cool down at room temperature before it was open in order to take out stone mold, open it and recover resin samples. The thus-obtained samples were polished by the standard dental procedure (involving powder, polishing stone and leather buff). In order to eliminate residual monomers, after the polishing step, all samples were immersed in distilled water at room temperature for 24h.

### ***Preparation of plant extract***

Fennel leaves were collected in Nis in October at the beginning of fruiting. Material was air-dried in a dry place without direct sunlight. Dried leaves were kept in a paper bag before extraction. Extraction was carried out with pulverized plant material and water (1:10) in Erlenmeyer flask with ground stopper placed in ultrasound bath for 20 min. After extraction, the mixture was filtered and water residue was removed by rotary vacuum evaporator. The yield of the extract was 25.3%. The obtained dried extract was transferred in a glass bottle and stored at 4 °C. For this study, the extract was dissolved in water to obtain 10% solution of the extract

### **Tretiranje akrilatnih uzoraka**

Akrilatni uzorci su nasumično podeljeni i izloženi na tri različita načina (uzorci dobijeni nakon ovih izlaganja su imenovani u odnosu na odgovarajuće stanje).

1. Uzorak izložen vazduhu (ime uzorka A) – ovaj uzorak je stalno držan na vazduhu;
2. Uzorak izložen destilovanoj vodi (ime uzorka DW) – ovaj uzorak je stalno držan u destilovanoj vodi;
3. Uzorak izložen biljnom ekstraktu (ime uzorka PES) – ovaj uzorak je potopljen u 7 ml PES-a 5 minuta, zatim izvađen iz PES-a, detaljno ispran tekućom vodom i osušen apsorbujućim papirom. Ova procedura je ponavljena 45 puta dnevno, u periodu od četiri dana. Stoga, naša eksperimentalna procedura simulira 180 dana čišćenja proteze od strane pacijenata, uzimajući u obzir učestalost čišćenja jednom dnevno. Uzorci su držani na sobnoj temperaturi u destilovanoj vodi između procedura potapanja.

### **Karakterizacija svojstva kvašenja**

Kontaktni ugao se merio pomoću metode sesilove kapi. Tri kapi vode 10  $\mu$ l su dodate pipetom na površinu svakog uzorka. Tokom ove metode karakterizacije, uzorci su postavljeni na ravnu površinu, pri temperaturi od  $22 \pm 2$  °C. Slike su napravljene fotoaparatom (Nikon Coolpix L840), koji je bio fiksiran na držaču kamere. Tri merenja su urađena za svaki uzorak. Kontaktni ugao je meren pomoću ImageJ softera, a dobijene vrednosti su predstavljene kao srednja vrednost ( $\pm$  standard devijacija).

### **Karakterizacija mehaničkih karakteristika**

Mehanička karakterizacija uzoraka je izvršena korišćenjem Agilent Nano Indenter G200, sa Berkovich vrstom dijamantskog vrha (tropovršinska piramida sa velikim uglom od  $142.3^\circ$ ). Uzorak je montiran na držaču uzorka korišćenjem dvostruko lepljive trake. Merenje je izvršeno sa sledećim parametarima: maksimalna sila prilikom utiskivanja 10mN, vreme primene sile 10 s, vreme zadržavanja utiskivača 15 s kada se dostigne maksimalna sila i jedno utiskivanje po poziciji. Tri indentacije su vršene za svaki uzorak i izračunata je srednja vrednost.

### **Treatment of resin samples**

The samples of denture base resin were randomly divided and exposed to the three different conditions (the samples obtained after these exposures were named according to the corresponding study condition):

1. Sample exposed to air (sample name: AIR) – this sample was continuously kept in air;
2. Sample exposed to distilled water (sample name DW) – this sample was continuously kept in distilled water;
3. Sample exposed to PES (sample name: PES) – this sample was immersed into 7 ml of PES for 5 min, subsequently removed from PES, thoroughly washed in running water, and dried with absorbing paper. This procedure was repeated 45 times per day over a period of 4 days. Thus, our experimental procedure simulates 180 days of denture cleaning by the patient by taking into account the cleaning rate to be one cleaning per day. The samples were kept in distilled water at room temperature between the immersion procedures.

### **Characterization of wetting properties**

Contact angles were measured by sessile drop method. Three water drops of 10  $\mu$ l were pipetted on the surface of each sample. During this characterization, samples were placed on the flat surface at room temperature,  $22 \pm 2$  °C. Images were taken with the camera (Nikon Coolpix L840) which was fixed on a camera holder. Three measurements were done for each sample. Contact angle were measurements by ImageJ softer and the obtained values were given as mean ( $\pm$  standard deviation).

### **Characterization of mechanical properties**

Mechanical properties of the above-mentioned samples were performed by nanoindentation method. Mechanical characterization was conducted by the Agilent Nano Indenter G200, with Berkovich type diamond tip (three-sided pyramid with large included angle of  $142.3^\circ$ ). Measurements included the following parameters: the maximum indentation force was 10 mN, peak hold time was 10 s, time to load 15 s, and single load per indent. Three indentations were performed for each specimen, and consequently the mean value was calculated.

### **Statistička analiza**

Dobijeni rezultati za svaku karakterizaciju su analizirani pomoću jednofaktorske analize varijanse (ANOVA), a razlike između srednjih vrednosti su poređene korišćenjem Dankanovog testa ( $p < 0,005$ ). Statistička analiza je izvršena pomoću SPSS 17 (SPSS, Inc., Chicago, IL).

### **Resultati**

#### **Kontaktni ugao**

Rezultati testa sesilove kapi na našim uzorcima su prikazani na Slici 1. Dobijene srednje vrednosti izmerenih kontaktnih uglova ( $\Theta_C$ ) su prikazane u Tabeli 1 kao srednja vrednost i standardna devijacija. Uzorci čuvani na vazduhu (AIR) su imali najnižu vrednost  $\Theta_C$   $64,5 \pm 2,3^\circ$ , dok druga dva uzorka (DW i PES uzorci) imala sličnu  $\Theta_C$  vrednost ( $75,4 \pm 3,3^\circ$ ,  $75,7 \pm 4,7^\circ$ , redom). Dobijene vrednosti  $\Theta_C$  između ispitivanih uzoraka nisu bile statistički značajne ( $p < 0,005$ ).

#### **Jungov modul elastičnosti**

Na osnovu rezultata nanoindentacije dobijene su vrednosti za Jungov modul elastičnosti ( $E_Y$ ). Ove vrednosti su prikazane u Tabeli 1. Ove vrednosti su prikazane na grafiku na Slici 2, a statistička analiza dobijenih vrednosti pokazuje da nije bilo značajne razlike ( $p < 0,005$ ) samo između uzoraka AIR i DW. Razlika između uzoraka AIR and PES, kao i DW i PES nije bila statistički značajna.

#### **Tvrdoća**

Rezultati dobijenih vrednosti tvrdoće (H) analiziranih uzoraka su prikazani u Tabeli 1. Na Slici 3 su prikazane vrednosti tvrdoće koje su veoma slične. Vrednosti tvrdoće svih uzoraka nisu bile statistički značajne ( $p < 0,005$ ).

### **Statistical analysis**

The obtained sets of results of each characterization were analysed by one-way analysis of variance (ANOVA) and differences among means were compared using the Duncan test ( $p < 0.005$ ). Statistical analyses were performed by the use of SPSS 17 (SPSS, Inc., Chicago, IL).

### **Results**

#### **Contact angle**

Results of the sessile drop test performed on our samples are showed in Figure 1. The obtained values of measured contact angles ( $\Theta_C$ ) are given in Table 1 as the mean values and standard deviations. Samples kept on the air (AIR) had the lowest  $\Theta_C$  value  $64.5 \pm 2.3^\circ$ , while the other two samples (DW and PES samples) had similar  $\Theta_C$  values ( $75.4 \pm 3.3^\circ$ ,  $75.7 \pm 4.7^\circ$ , respectively). There is no statistical significant difference in the obtained  $\Theta_C$  values among studied samples ( $p < 0.005$ ).

#### **Young's modulus**

Based on the results of nanoindentation, the values of Young's modulus ( $E_Y$ ) were obtained. These values are presented in Table 1. Figure 2 depicts the graph of these values and statistical analysis revealed that there was a significant difference ( $p < 0.005$ ) only between the samples AIR and DW. The difference between samples AIR and PES was not significant, as well as between samples DW and PES.

#### **Hardness**

The obtained results for hardness (H) for all analyzed samples are presented in Table 1. Figure 3 shows the graphic representation of these values which are similar. Indeed, there was no statistically significant difference ( $p < 0.005$ ) in values for hardness among the studied samples.

**Tabela 1.** Dobijene vrednosti kontaktnog ugla, Jungovog modula elastičnosti i tvrdoće svih ispitivanih uzoraka. Sve vrednosti su predstavljene kao srednja vrednost i standardna devijacija

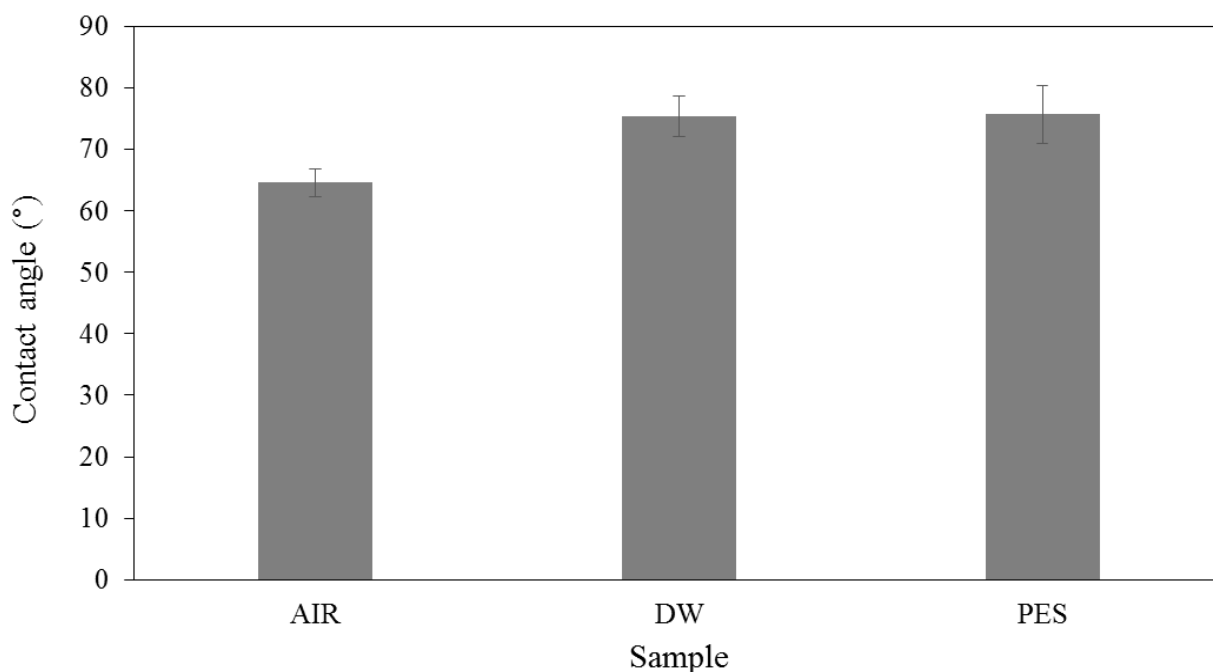
**Table 1.** The obtained values of contact angle, Young's modulus and hardness for studied samples. All values are given as mean and standard deviation

	$\Theta_c (^{\circ})$	$E_Y$ (GPa)	H (GPa)
AIR	$64,5 \pm 2,3^a$	$2,125 \pm 0,090^a$	$0,060 \pm 0,002^a$
DW	$75,4 \pm 3,3^a$	$2,513 \pm 0,118^b$	$0,066 \pm 0,004^a$
PES	$75,7 \pm 4,7^a$	$2,238 \pm 0,018^{a,b}$	$0,067 \pm 0,001^a$

Ista slova u kolonama pokazuju statistički neznačajnu razliku između varijeteta (Duncan test,  $p < 0,005$ ).

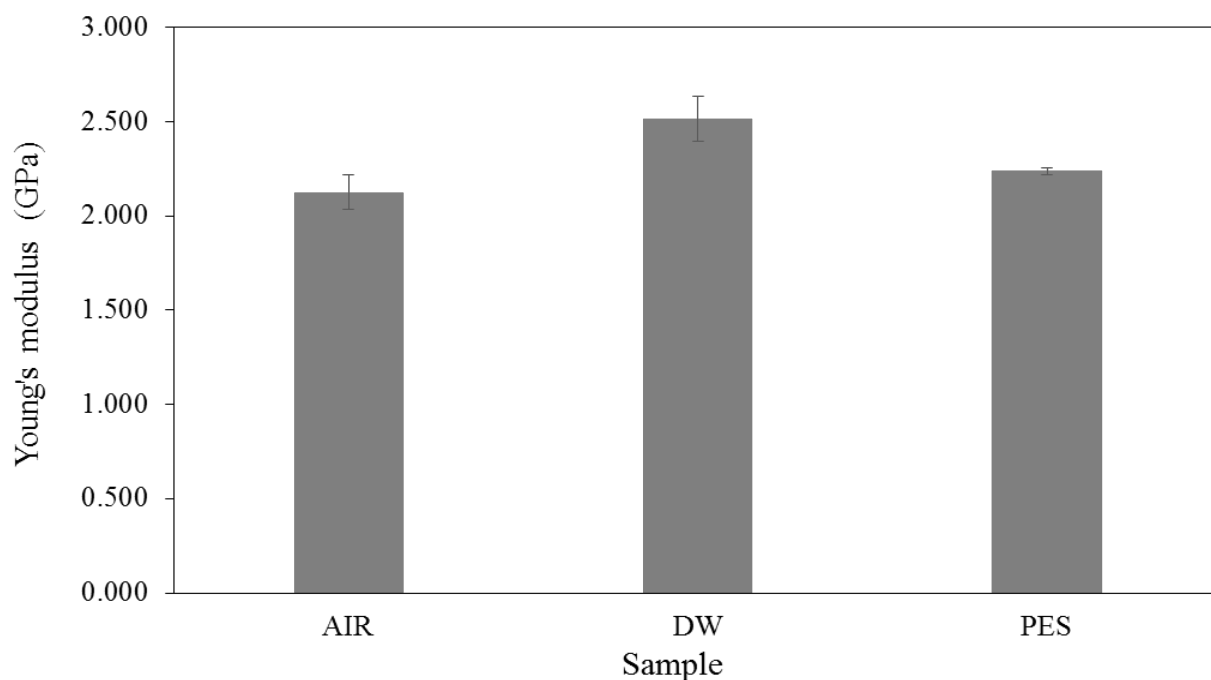
Različita slova u kolonama pokazuju statistički značajnu razliku između varijeteta (Duncan test,  $p < 0,005$ ).

The same letters in columns indicate insignificant difference among varieties (Duncan test,  $p < 0.005$ ). Different letters in columns indicate significant difference among varieties (Duncan test,  $p < 0.005$ ).



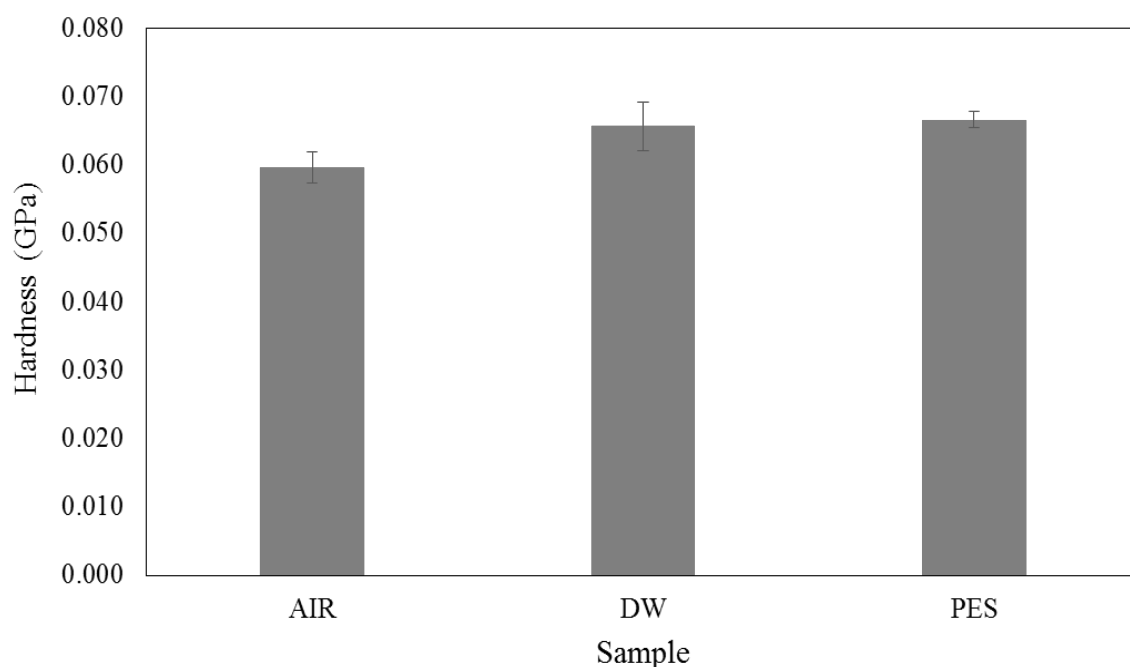
**Slika 1.** Kontaktni ugao vode na ispitivanom uzorku za bazu proteze izloženom vazduhu (AIR), destilovanoj vodi (DW) i rastvoru ekstrakta lista morača (PES)

**Figure 1.** Contact angle of water on the studied denture base resin samples exposed to air (AIR), distilled water (DW) and solution of Fennel leaves extract (PES)



**Slika2.** Jungov modul elastičnosti dobijen nanoindentaciom ispitivanih uzorka za bazu proteze izloženih vazduhu (AIR), destilovanoj vodi (DW) i rastvoru ekstrakta lista morača (PES)

**Figure 2.** Young's modulus obtained by nanoindentation of studied denture base resin samples exposed to air (AIR), distilled water (DW) and solution of Fennel leaves extract (PES)



**Slika3.** Tvrdća dobijena nanoindentaciom ispitivanih uzorka za bazu proteze izloženih vazduhu (AIR), destilovanoj vodi (DW) i rastvoru ekstrakta lista morača (PES)

**Figure 3.** Hardness obtained by nanoindentation of studied denture base resin samples exposed to air (AIR), distilled water (DW) and solution of Fennel leaves extract (PES)



## Diskusija

U cilju ispitivanja efekta morača na kvašenje i mehanička svojstva toplopolimerizujućih akrilata, mereni su  $\Theta C$ , EY i H. Ove vrednosti su takođe urađene za uzorke akrilata koji su: bili izloženi samo vazduhu (sa ciljem da se upoređi uticaj PES na akrilat u odnosu na akrilat bez ikakve modifikacije), i samo destilovanoj vodi (kako bi se razlikovao uticaj biljnog ekstrakta na akrilat u odnosu na vodu koja je glavna komponenta PES-a).

Osobine kvašenja materijala za bazu zubne proteze su u jakoj korelaciji sa afinitetom vezivanja biomolekula na površinu. U usnoj duplji, proteza je u kontaktu sa oralnom mukozom i izložena je takođe različitim česticama i mikroorganizmima iz oralne sredine. Tako, komponente pljuvačke mogu da se talože na površini, zatim mikroorganizmi koji su deo oralne flore mogu da se zalepe na površini proteze i formiraju biofilm<sup>16</sup>. Međutim, različiti mehanički i hemijski pristupi koji se koriste za održavanje higijene proteze, mogu da promene površinska svojstva proteze. Zbog toga je veoma bitno čuvati površinu materijala nepromenjenom.

Kvašenje je jedna od fizičkih osobina površine koja označava mogućnost tečnosti da održava kontakt sa čvrstom površinom<sup>17</sup>. Step en kvašenja se može definisati merenjem kontaktnog ugla formiranog između kapljice vode i čvrste površine. Kontaktni ugao kapljice vode na površini akrilata treba da bude oko 75°. Ovo je veoma važno zbog toga pljuvačke preko površine proteze<sup>16,17</sup>. Naši rezultati su pokazali da je kontaktni ugao bio nepromenjen nakon potapanja u ekstrakt morača 75,4 ± 3,3, u poređenju sa onim uzorcima koji su čuvani u vodi na vazduhu (75,7 ± 4,7; 64,5 ± 2,3), redom, i u rangu zahtevanog kontaktnog ugla za akrilat.

Među brojnim mehaničkim svojstvima, jedan od esencijalnih je Jungov modul elastičnosti (EY), koji predstavlja meru krutosti (pokazuje u kojoj meri neki objekat odoleva deformaciji kao odgovor na neku primenjenu silu). Jungov modul može biti izveden merenjem noindenterom, što se zasniva se na uvlačenju vrha indentera u testirani materijal, gde se događaju elastična i neelastična deformacija, elastična deformacija se vraća u prvobitno stanje kada se indenter povuče iz materijala.

## Discussion

In order to study the effect of fennel extract on the wetting and mechanical properties of heat polymerized acrylic resin, we measured  $\Theta C$ , EY and H. These measurements were also done for acrylic resin samples which were: exposed to air only (in order to compare the influence of PES on resin with respect to native resin without any modifications), and exposed to distilled water only (in order to distinguish the influence of plant extract on acrylic resin with respect to water which is the major component of PES).

Wetting properties of the surface of denture base material are strongly correlated to bonding affinity of biomolecules to that surface. In the oral cavity, denture is in the contact with oral mucosa and also exposed to the different particles and microorganism from the oral environment. Thus, saliva ingredients can precipitate on the surface, while microorganisms from the oral flora can attach and form biofilm on denture<sup>16</sup>. However, the use of different mechanical and chemical approach in maintaining the denture hygiene could change the surface properties. Therefore, it is important to keep the surface of material unchanged.

Wettability is one of the physical properties of the surface, which means the ability of liquid to maintain contact with a solid surface<sup>17</sup>. The degree of wetting could be determined through the measurement of contact angle formed between a water drop with solid surface. The contact angle of the water drop over the acrylic resin should be around 75°. It is very important for saliva flow over the denture surface<sup>16,17</sup>. Our results showed that contact angle was unchanged after immersion in fennel extract 75.4 ± 3.3, compared to those kept in water and on air (75.7 ± 4.7; 64.5 ± 2.3), respectively, and in the range of the required contact angle for acrylic resin.

Among numerous mechanical properties, one of the essential is Young's modulus ( $E_Y$ ) which is a measure of stiffness (shows to which extent some object resists to deformation as a response to some applied force). Young's modulus can be deduced from nanoindentation measurements which are based on pushing an indenter tip into the tested material where both elastic and plastic deformations occur; the elastic deformation

Na osnovu ove procedure, dobijene su vrednosti za sve testirane uzorke. Za uzorak AIR, koji odgovara standardnoj bazi pločaste proteze koja se koristi u stomatologiji, dobili smo vrednosti koje se slažu sa prethodno objavljenim vrednostima za čist PMAA (1,8-3,6 GPa)<sup>18</sup>. Ovo su očekivani rezultati, budući da je PMMA glavna komponenta baze proteze.

Međutim, maksimalna vrednost  $E_Y$  za PMMA (3,6 GPa) se ne može dobiti za bazu zubne proteze, jer su za samu protezu dodate brojne druge komponente (kao što su aditivi, pigmenti, katalizatori, stabilizatori itd.), što modifikuje njegova mehanička svojstva u poređenju sa čistim PMMA-om. Pokazalo se da je dugoročno potapanje u vodi uticalo na mehanička svojstva pločaste proteze, pri čemu se modul elastičnosti povećao nakon 180 dana potapanja u vodi<sup>19</sup>. Tvrdoća je takođe važno svojstvo materijala koje meri mogućnost materijala da se odupre plastičnoj deformaciji nakon primenjene sile. Primenjenom procedurom nanoindentacije na svim uzorcima, dobijene su takođe vrednosti koeficijenta tvrdoće. Tvrdoća uzorka AIR (0,06 GPa) nije bila u opsegu objavljenih vrednosti za čist PMMA (0,16 0,21 GPa). Na tvrdoću materijala može uticati vreme procesa polimerizacije što utiče na sadržaj rezidualnog monomera. Rezidualni monomer može delovati kao plastifikator smanjujući interlančane sile, omogućavajući da se deformacija lako dogodi pod opterećenjem tokom testa<sup>19,2</sup>.

Do sada su ispitivana antimikrobna svojstva nekih biljaka (kao što su *Ricinus communis*, *Salvia officinalis*), ali ne i njihov uticaj na fizička svojstva zubne proteze<sup>20,21</sup>. Stoga, u prezentovanoj studiji je pokazano da potapanje u ekstrakt lista morača nakon simuliranog perioda čišćenja od 180 dana nije značajno uticalo na mehaničke osobine i kvašenje uzoraka akrilata.

is recovered when the indenter is withdrawn from the material. Based on this procedure, the values for  $E_Y$  were obtained for all studied samples. For the sample AIR, which corresponds to standard denture base resin as it is routinely used in dentistry, we obtained the value which is in agreement with previously reported values for pure PMMA (1.8-3.6 GPa)<sup>18</sup>. That is an expected result, since PMMA is the main component of denture base resin.

However, the maximal value of  $E_Y$  for PMMA (3.6 GPa) cannot be obtained in denture base resin because the later contains numerous additional components (such as various additives, pigments, catalysts, stabilizers etc.) which modify its mechanical properties compared to the pure PMMA. It has been shown that long-term water immersion affect the mechanical properties of acrylic resin denture base whereby the elastic moduli significantly increased after 180 days of water immersion<sup>19</sup>. Hardness is also an important mechanical property of material which is a measure of the ability of a material to resist plastic deformation when force is applied. Applying nanoindentation procedure on all samples, the values of hardness coefficient (H) were obtained as well. The hardness of sample AIR (0.06 GPa) was not in the range of reported various for pure PMMA (0.16 0.21 GPa). Hardness of material could be affected by the time of polymerization process which affects the content of residual monomer. Residual monomer can act as a plasticizer, reducing the interchain forces enabling that deformation easily occur under load during the test.<sup>19,2</sup>

Antimicrobial and antifungal properties of some herbs (such as *Ricinus communis*, *Salvia officinalis*) have been studied, but not their influence on the physical properties of acrylic resins, especially denture base resin<sup>20,21</sup>. Therefore, the present study showed that fennel leaves extract after simulated 180-days long period of use in cleaning purpose do not significantly affect mechanical and wetting properties of denture base acrylic resins.

## **Zaključak**

Na osnovu dobijenih rezultata da ekstrakt lista morača nije imao negativan uticaj na kvašenje i mehaničke osobine površine akrilata za bazu proteze, može se zaključiti da ekstrakt ne sadrži neke komponente koje bi mogle da penetriraju i oštete strukturu polimernog matriksa, za razliku od uobičajno korišćenih hemijskih sredstava za čišćenje. Izmerene vrednosti za kontaktni ugao, tvrdoću i Jungov modul elastičnosti su pokazale da nema značajne razlike u ovim parametrima za tri analizirane grupe uzoraka, AIR, DW and PES. To znači da bi se ekstrakt morača mogao koristiti kao efikasno sredstvo za dezinfekciju proteza umesto hemijski agresivnih supstanci. Međutim, dalje studije su potrebne da ispituju njegove efekte na fizička svojstva proteze kao i na različite mikroorganizme.

## **Zahvalnica**

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Autori izjavljuju da ne postoji sukob interesa.

## **Conclusion**

Based on our results that fennel extract did not adversely affect wetting and mechanical properties of the surface of denture base resin, it can be concluded that it does not contain any component that could penetrate and damage the structure of polymer matrix, unlike usually used chemical cleanser. The obtained results for contact angle, hardness coefficient and Young's modulus revealed that there is no significant difference in these parameters for three analyzed group of samples, AIR, DW and PES. It means that fennel herbal extract can be used as an efficient cleanser instead of chemically aggressive substances. However, further studies needed to investigate the effect on its physical properties of denture base resin as well as on different microorganisms.

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The authors declare that there is no conflict of interest.

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