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IZVORNI ZNANSTVENI RAD
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Analiza topografije i hrapavosti površine Co-Cr uzoraka uz pomoć mikroskopa atomskih sila

Analysis of Surface Topography and Surface Roughnes of CoCr Alloy Samples by Atomic Force Microscopy

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Sažetak
Svrha rada bila je dobiti trodimenzionalne kvalitativne i kvantitativne podatke o topografiji elektropoliranih uzoraka Co-Cr dentalne legure na nano-ljestvici, točnije podatke o dvjema različitim čvrstim fazama opisanima u literaturi kao dendriti i interdendritski prostori. **Materijal i metode:** Napravljeno je, izliveno i elektropolirano deset uzoraka Co-Cr legure (Wironit® extra-hard, Bego, Njemačka). Njihova površina snimljena je i analizirana mikroskopijom atomskih sila (AFM-om) te optičkim mikroskopom. AFM-snimke i analiza omogućili su trodimenzionalno promatranje površine Co-Cr legure. Vidljivo je da se njezina dendritska struktura sastoji od nakupina nanokristalita oblikovanih u otoke (odgovaraju tamnom interdendritskom području na optičkom mikroskopu), a izdižu se iz glatke undulirajuće površine koja odgovara svijetloj površini, odnosno dendritima na snimkama optičkog mikroskopa. **Rezultati:** Devedeset i pet postotni interval pouzdanosti ($p<0,05$) za maksimalne z-vrijednosti kreće se od 819,98 do 1020,99 nm, za vrijednosti RMS-a od 66,93 do 108,54 nm i za Ra (srednja hrapavost) od 51,53 do 82,48 nm. **Zaključak:** Površina ima vrlo dobra svojstva što se tiče hrapavosti i može se smatrati doista glatkom, jer nema elemente koji bi mogli retinirati i zarobiti mikroorganizme.

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Uvod

Kobaltno-kromne (Co-Cr) legure ubrajaju se u skupinu baznih legura. Otkako su uvedene na stomatološko tržište godine 1929. (1) koriste se u protetici za metalne skelete djelomičnih proteza (DP) i fiksno-protetskih konstrukcija. Bazne legure imaju manju gustoću od zlatnih, a modus elastičnosti gotovo im je dva puta veći. Zato Co-Cr legure imaju prednost pred zlatnima jer su čvrste i imaju manji

Introduction

Cobalt-chromium (CoCr), a base metal alloy, has been widely used in the fabrication of removable (RPD) and fixed partial denture (FPD) frameworks since being introduced to dentistry in 1929 (1). The base metal alloys have a lower density than gold alloys and a modulus of elasticity nearly double than that of gold alloys. Therefore, the CoCr RPDs have the advantage of maintaining rigidity with less bulk obujam (2). One su uglavnom nadomjestile zlatne legure, posebice zbog poboljšanih mehaničkih svojstava i znatno manje cijene (3,4). No, Co-Cr legure imaju i tehničke nedostatke – teže i kompliciranije se lijevaju, bruse i poliraju (5).

Kvaliteta površine (hrapavost i morfologija) bilo koje legure koja se koristi u stomatologiji vrlo je važna jer određuje površinu kontakta s oralnom tekućinom (slinom). Kvaliteta površine bilo kojeg stomatološkoga metalnog rada utječe na korozivnost, biokompatibilnost, otpuštanje iona metala i interakciju s mikroorganizmima usne šupljine. Adherencija zubnog plaka znatno ovisi o hrapavosti površine (6).

Biomedicinski prihvatljive Co-Cr legure imaju površinu dendritičke mikrostrukture ako ih se polako hladi tijekom stvrdnjavanja. Kako bi se postigla dobra struktura površine, prije mikroskopiranja uzorci legura se jetkaju. (7). Smatra se da je potrebno oko 30% kroma da Co-Cr legura dobije dendritičku strukturu (8,9). Kad se njezina površina gleda optičkim ili elektroničkim mikroskopom [Scanning electron microscope (SEM)], vide se svjetlija područja ili dendriti i tamnija područja ili interdendritske zone (7-9). Prema najnovijim spoznajama misli se da tamne interdendritske površine i svijetle dendritske površine nastaju tijekom sporog stvrdnjavanja te predstavljaju dvije različite vrste krute otopine različite prema strukturi, a samo neznatno prema kemijskom sastavu (8,9). Pritom svjetlija površina (dendriti) predstavlja kristale plošno centrirane kubične rešetke (fcc), a tamna (interdendritska površina) heksagonalnu kristalnu rešetku (hcp). Također se smatra da je mikrotvrdoća svjetlijeg područja veća od tamnijeg područja kod Co-Cr legura (8,9).

Promatrajući slike optičkim mikroskopom ili SEM-om, ne možemo dobro procijeniti kakva je topografija površine. I SEM i optički mikroskop omogućuju promatranje samo u dvjema dimenzijama. Premda je nedostatak SEM-a u kompliciranosti pripreme uzorka, jer je za mikroskopiranje potreban vakuum, ipak je on mnogo bolji od optičkog mikroskopa jer znatno povećava površinu, tako da je na nanometraskoj ljestvici moguće promatrati detalje. No, ipak daje samo dvodimenzionalni prikaz promatrane površine (10-15).

Tijekom prošlog desetljeća izumljena je nova tehnika mikroskopiranja – tzv. mikroskopiranje atomskih sila (AFM) (10-15). Ta tehnika rekonstruira površinu u svim trima dimenzijama i skenira topografiju zadane površine kao skup z (vertikalna os), x (horizontalna os) i y (transverzalna os) vrijednosti (10-13). Na taj način AFM bilježi «istinit» trodimenzionalni profil. AFM ima i bolju rezoluciju od SEM-

(2). Base alloys have increasingly replaced gold alloys owing to the improved mechanical properties and decreased cost (3,4). However, technical shortcomings include the increased difficulty of casting, grinding and polishing procedures (5).

The surface quality (the surface roughness and the surface morphology) of any dental alloy is very important, as it determines the area of the contact surface with oral fluids. The quality of a surface of a dental restoration influences the corrosion behaviour, the biocompatibility, the release of metal ions, and microorganism interactions with dental alloys. Plaque adherence greatly depends on surface roughness (6).

Biomedically acceptable CoCr alloys have been reported to have surfaces with dendritic microstructure when cooled slowly through a solidification range. To reveal the surface structure the alloy samples have to be etched prior to viewing (7). Moreover, approximately 30% of chromium is required to achieve the dendritic structure (8,9). When viewed with optical microscope or scanning electron microscope (SEM), the surface of a CoCr dental alloy presents light regions, referred to as dendrites, and dark regions, referred to as interdendritic regions. It was reported that the dark interdendritic regions and the light dentritic regions, originating from a slow solidification, actually correspond to the two types of CoCr solid solutions, which differed in structure, but only slightly in composition. It has been reported that the dendrites (light region) represent cubic close packed crystal lattice structure, actually the face centered cubic lattice (fcc), while the dark regions (interdendritic region) represented hexagonal close packed lattice structure (hcp). It has also been reported that the microhardness of the light zones is greater than that of the dark zones in CoCr alloy electropolished samples (8,9).

From the optical micrographs or SEM images it is difficult to assess the topographic characteristics of the surfaces. They allow only a two-dimensional view. The SEM, although operating under vaccum, is superior to optical microscope as it resolves a structure down to the nanometer scale, but still gives a two-dimensional 'photographic' image of the samples (10-15).

During the last decade a new microscope-imaging technique has been developed – the atomic force microscopy (AFM). The AFM technique reconstructs the three-dimensional image of the sample topography and records the experimental data in digital form as sets of z (vertical axis), x (horizontal axis), and y (transversal axis) values (10-13). The AFM provides a true three-dimensional surface proa (14,15) te može mikroskopirati i u sobnim uvjetima (zrak), ali i u tekućinama. Prikazuje strukturu čak i na subnanometarskoj ljestvici (vertikalna rezolucija 0,1 nm). S AFM-om se može raditi na nekoliko načina, ovisno o primjeni. Općenito, načini rada dijele se na statički modus (nazvan i kontaktni) i nekoliko dinamičkih. Različiti kompjutorski programi (softwari) mogu se priključiti AFM-u, a omogućuju računalnu (off-line) statističku analizu parametara koji definiraju hrapavost površine, preciznu analizu bilo kojeg presjeka te mnoga druga ispitivanja i pretraživanja.

Svrha ovog rada bila je AFM-analizom dobiti trodimenzionalne kvalitativne i kvantitativne prikaze topografije površine Co-Cr dentalne legure, tj. prikazati u tri dimenzije dvije krute otopine (faze) karakteristične za tu leguru, a opisane su kao dendriti i interdendritsko područje (svijetla i tamna područja) na snimkama optičkog mikroskopa i SEM-a.

Materijal i metode rada

Lijevanje uzoraka

Bilo je izliveno deset uzoraka Co-Cr legure (Wironit® extra-hard, Bego, Njemačka), tvorničkog sastava: 63% Co, 30% Cr, 5% Mo te Si, Mn i C < 2%. Kvadratni predlošci bili su pripremljeni od voska za modeliranje u obliku kvadrata, dimenzija 8x8x2 mm (Modelling wax standard, DeguDent GmbH, Njemačka) te prema uputama proizvođača uloženi u Castorit super C, uložnu masu za neplemenite legure (Dentaurum, Ispringen, Njemačka). Predgrijavanje je obavljeno na temperaturi od 1000^0C, a uzorci su izliveni uz pomoć vakuumskog ljevača (Nautilus, Bego, Njemačka) na temperaturi od 1420^0C. Nakon lijevanja pažljivo su očišćeni, a ostaci materijala za ulaganje uklonjeni su u pjeskari (Austenal Dentastrahl VE2, Njemačka) uz pomoć čestica aluminijskog oksida veličine 250 μm.

Pripremanje uzoraka prije AFM-a

Nakon pjeskarenja uzorci su bili 15 minuta podvrgnuti postupku elektropoliranja (Eltropol SL electropolishing machine, Bego, Njemačka), a koristila se otopina za elektropoliranje za Co-Cr legure (Megalyt Megadental GmbH, Njemačka).

Čišćenje uzoraka prije AFM-analize

Prije mikroskopiranja uzorci su bili pažljivo očišćeni alkoholom te isprani u potpuno čistoj destiliranoj vodi, a nakon toga su dodatno očišćeni ultrazvučno u trajanju od 10 minuta, također u potpuno

file. In principle, AFM can provide higher resolution than SEM (14,15). Moreover, the AFM can be operated in ambient conditions and in fluids. It resolves the structure down to subnanometer scale (vertical resolution of 0.1 nm). The AFM can be operated in a number of modes, depending on the application. In general, possible imaging modes are divided into static (also called contact) mode and a variety of dynamic modes. Different software programs can be included in the AFM, allowing the off-line statistical analysis of surface roughness parameters, precise section analysis of any regions of interest, and it also allows many other tests and searches.

The aim of this study was to acquire a three-dimensional qualitative and quantitative nano-scale data of surface topology of CoCr dental alloy, actually of the two different solid solutions described as dendrites and interdendritic regions, using the AFM technique.

Materials and methods

Sample casting

Ten CoCr alloy samples (Wironit® extra-hard, Bego, Germany), consisting of 63% Co, 30% Cr, 5% Mo, and Si, Mn and C < 2% were prepared and cast. Square shaped wax patterns 8x8x2 mm were prepared from a modelling pink wax (Modelling wax standard, DeguDent GmbH, Germany) and invested in the Castorit super C investment material for non-precious alloys (Dentaurum, Ispringen, Germany) according to the manufacturer's recommendation. Preheating was made at a temperature of 1000 °C and afterwards specimens were cast using a vacuum casting machine (Nautilus, Bego, Germany) at a temperature of 1420 °C according to the manufacturer's recommendation. The cast specimens were carefully cleaned from the particles of the investment material using a sandblasting machine (Austenal Dentastrahl VE2, Germany) with airborne-particles of aluminium oxide (250 μm) to remove investment residues.

Sample preparation prior to AFM

After sandblasting all the prepared samples were electropolished (Eltropol SL electropolishing machine, Bego, Germany) using an electrolyte polishing solution for CoCr alloys (Megalyt Megadental GmbH, Germany) for 15 minutes.

Sample cleaning prior to AFM analysis

Prior to analysis, surfaces were carefully cleaned with alcohol, then rinsed with ultrapure water and finally sonicated for 10 min in ultrapure water. The samples were then dried. Excess water was removed čistoj destiliranoj vodi. Nakon toga su osušeni, višak vode upijen je u staničevinu te su spremljeni u plastične Petrijeve zdjelice.

AFM-snimanje i analiza

Slike uzoraka površine Co-Cr legure snimljene su kontaktnim načinom rada multimodalnog AFM-uređaja s Nanoscopom III-a kontrolnim programom (Veeco Instruments, Santa Barbara, SAD) u sobnim uvjetima. Optička kamera (mikroskop) (Sony high-resolution CCD camera, Japan) omogućila je preciznu lokaciju područja interesa, a i snimanje površina kod manjih povećanja. Pod kontrolom kamere optičkog mikroskopa vršak sonde (poluge) AFM-a doveden je u bliski ("atomski") odnos s površinom snimanog uzorka te je zatim obavljeno skeniranje područja piezoelektričnim skenerom. Laserska zraka fokusirana na kraj sonde (poluge) zajedno s fotodetektorom osjetljivim na "položaj", mjerila je mala izvijanja vrška sonde. Njezini pokreti (poluge) snimljeni su kao funkcija položaja vrška. Uporabljene su sonde (poluge) u obliku slova V, s vrškom od silikonskih nitrida u obliku piramide (NP-20, Veeco), a nominalna sila tijekom skeniranja iznosila je 0,32 N/m. Slike su snimljene u najvećoj mogućoj rezoluciji od 512 x 512 piksela po snimci. Skener (JV) je imao maksimalni raspon od 125 μm u x- i y- smjeru, a 5 μm u z-smjeru (visina). Dimenzije skeniranih površina iznosile su: 12 x 12 μm, 30 x 30 μm i 100 x 100 μm. Po tri različite površine (svaka je uključila tri dimenzije skeniranja: 12 x 12 μm, 30 x 30 μm i 100 x 100 μm) snimljene su za svaki uzorak, a napravljena je i po jedna snimka kamerom optičkog mikroskopa. Sve AFM-snimke bile su sirovi podaci, osim dvodimenzijskog poravnavanja prvog reda. Pregledavanje snimaka, analiza hrapavosti površine tj. dobivanje vrijednosti RMS-a (korijen srednjeg kvadrata visinskih devijacija od prosječne ravnine), R_a-vrijednosti (aritmetička sredina apsolutnih vrijednosti devijacija visine od prosječne ravnine) i vrijednosti Z raspona (maksimalna vertikalna udaljenost između najviše i najniže točke na snimci) te analiza presjeka snimke na područjima interesa, obavljene su računalno (off-line) uz pomoć programa AFM NanoScope Control, verzija 5,12r5 (Veeco Instruments, Santa Barbara, SAD). Za statističku obradu uzete su aritmetičke sredine triju skeniranih površina dimenzija 100 x 100 μm za svaki uzorak.

Statistička analiza

Statistička obrada podataka obavljena je statističkim paketom SPSS 12 for Windows (Chicago, SAD). Deskriptivna statistika napravljena je za pa-

by gentle blotting with absorbent paper, and stored in a plastic Petry dish.

AFM imaging and analysis

The images of the CoCr samples were recorded using the contact mode operation on the Multimode AFM with Nanoscope IIIa controller (Veeco Instruments, Santa Barbara, USA) under ambient conditions in air. The optical camera (Sony high-resolution CCD camera, Japan) enabled to locate the region of interest in an easy way and to record optical micrographs as well. Under the optical camera control, the probe associated with a cantilever was brought into light (atomic) contact with the sample surface and then rasterscanned across the surface by the piezoelectric scanner. A laser beam focused on the back of the cantilever together with a position-sensitive photodetector measured minute deflection of the sensor probe tip. The motion of the sensor tip was recorded as a function of tip position. The V shaped cantilevers with pyramidal silicon-nitride tips, NP-20, Veeco, with nominal spring constant of 0.32 N/m were used to record images. The images were recorded at the highest possible resolution of 512 x 512 pixels per image. The scanner (JV) had maximum ranges of 125 μm in x- and y- direction and 5 μm in z- direction (height). The dimensions of the scanned surface areas were: 10 x 10 μm^2, 30 x 30 μm^2 and 100 x 100 μm^2. Three different surfaces including all dimensions (10 x 10 μm^2, 30 x 30 μm^2 and 100 x 100 μm^2) were obtained from each sample, as well as one optical micrograph. All images were raw data except for the first order two-dimensional flattening. Image browsing, the roughness analysis of the RMS values (root mean square average of height deviations taken from the mean plane); the R_a values (arithmetic average of the absolute values of the surface height deviations measured from the mean plane) and the Z-range values (maximum vertical distance between the highest and the lowest data points in the image), as well as section analysis of the profiles of the regions of interest were made off-line using the AFM NanoScope Control software, version 5.12r5 (Veeco Instruments, Santa Barbara, SAD). Mean values of the three scanned regions of 100 x 100 μm^2 of each sample were taken for statistical analysis.

Statistical analysis

Statistical analysis was performed using the SPSS 12 for Windows (Chicago, USA). Descriptive statistics was made for the roughness paramerametre koji opisuju hrapavost površine (RMS, R_a i Z-raspon vrijednosti), uključujući i intervale pouzdanosti kod 95% vjerojatnosti. Od podataka dobivenih analizom presjeka područja interesa, također su izračunane srednje vrijednosti i rasponi.

Rezultati

Mikrograf jednog od elektropoliranih uzoraka (312,5 x 312,5 μm) snimljen kamerom optičkog mikroskopa prikazan je na Slici 1A. Vidi se tipična dendritička struktura sa svijetlim područjima koja predstavljaju dendrite i tamnim područjima koja odgovaraju interdendritskim zonama. Površina istog uzorka (100 x 100 μm) snimljena AFM-om i predstavljena u trima dimenzijama prikazana je na Slici 1B. Promatrajući trodimenzionalno, jasno se vide elevacije: nakupine nanokristalita u obliku otoka, a izlaze iz glatke undulirajuće površine. Poneki su otoci (nakupine mikrokristalita) međusobno povezani. Nakupine mikrokristalita (otoci) odgovaraju tamnim područjima (interdendritsko područje) na snimkama optičkog mikroskopa, a undulirajuća površina svijetlim područjima (dendriti). Analiza presjeka otkriva nazubljenost nanokristalita koji odozgo izgledaju poput zubaca pile (Slika 1C).

Slika 2A predstavlja također 3-D površinu istog uzorka, ali povećanu na 30 x 30 μm, a Slika 2B analizu presjeka iste površine. Slika 2. mnogo bolje prikazuje detalje koji se odnose na izgled otoka (nakupine nanokristalita). Na snimci je jedan otok nalik na čizmu, a drugi sliči leptiru. Otoci koji se sastoje od nanokristalita imaju tipičan duguljasti oblik, a površina ima raspon od 4 do 80 μm^2. Visina otoka kreće se od 90 do160 nm, a može se točno izmjeriti. Nakupine nanokristalita u otoku poredane su paralelno i međusobno su udaljene 160 ili 320 μm. Nazubljena površina otoka mnogo se bolje uočava kod takvoga povećanja.

Slika 3. prikazuje površinu istog uzorka, ali još više povećanu - na 12 x 12 μm. Trodimenzionalni prikaz (3A) predstavlja nakupinu nanokristalita (jedan otok koji sliči leptiru), a uzdiže se iz glatke površine. Slika 3B prikazuje analizu presjeka iste površine.

Tablica 1. prikazuje deskriptivnu statistiku podataka dobivenih analizom hrapavosti površine deset elektropoliranih Co-Cr uzoraka. Devedeset pet postotni interval pouzdanosti ($p<0,05$) za maksimalni z-raspon iznosi od 819,98 do 1020,99 nm, za RMS vrijednosti od 66,93 do 108,54 nm, a za R_a (srednja hrapavost) od 51,53 do 82,48 nm.

ters acquired through the roughness analysis (RMS, R_a and Z-range values), including a 95% confidence intervals of mean. The data from the section analysis considering the dimensions of the newly revealed shapes were also obtained to calculate means and ranges.

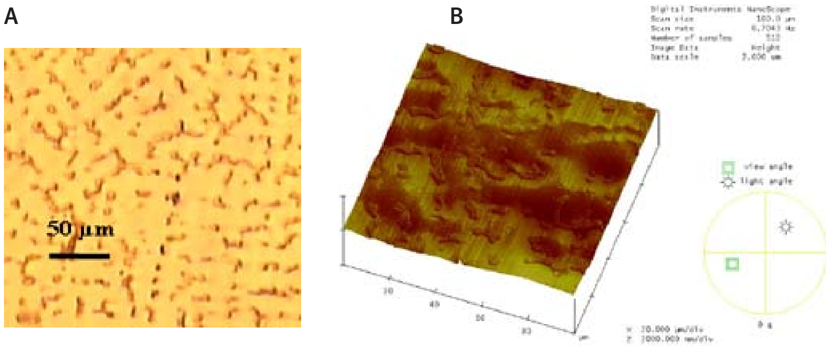
Results

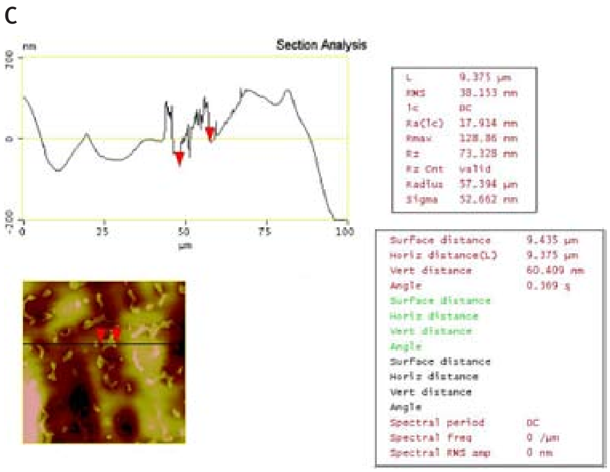
The optical camera micrograph of a surface of one electropolished sample (312.5 x 312.5 μm) is presented in Figure 1A. Typical dendritic feature can be observed with light areas representing dendrites and dark area representing interdendritic regions. A surface (100 x 100 μm) of the same sample obtained by AFM and presented as a 3-D image is shown in Fig. 1B. When viewed three-dimensionally it can be clearly seen that elevations (clusters of nanocrystallites forming islands) are emerging from a smooth undulating surface. The islands are sometimes connected. The clusters of nanocrystallites (islands) actually correspond to the darker areas (interdendritic region) on the optical-camera micrograph, while undulating surface corresponds to the light (dendritic) areas. Section analysis reveals saw-tooth character of the nanocrystallites when viewed from the top (Fig. 1C).

Figure 2A shows the 3-D view of the surface of the same sample enlarged to a 30 x 30 μm and Fig. 2B shows section analysis of the same feature. Fig. 2 reveals more details considering islands (clusters of nanocrystallites). One island resembles a boot and the other one a butterfly. The islands of nanocrystallites have characteristic oblong shapes and the surface areas ranging from 4 to 80 μm^2. The islands' height ranges from 90 to160 nm and can be determined precisely. Clusters of nanocrystallites within the island are ordered in parallel rows 160 or 320 μm apart. A saw-tooth top view pattern can be seen better at this enlargement.

Figure 3 shows the surface of the same sample, but at the enlargement of only 12 x 12 μm. The 3-D image (Fig. 3A) shows one cluster of nanocrystallites (island resembling a butterfly) emerging from a smooth surface and Fig. 3B shows the section analysis of the same surface.

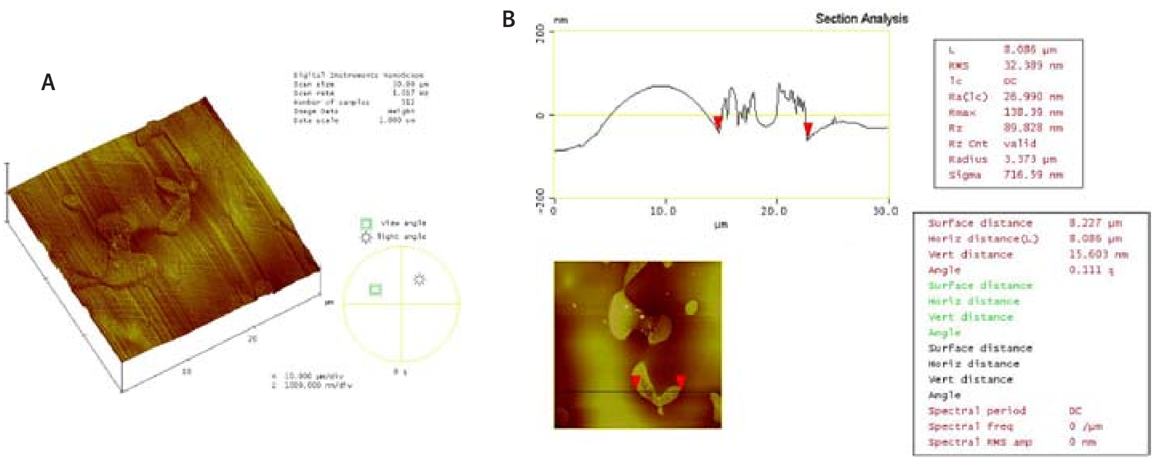
Table 1 presents descriptive statistics obtained from roughness analysis of 10 electropolished CoCr samples investigated in this study. The 95% confidence interval ($p<0.05$) reveals that the maximum z-value is from 819,98 to 1020,99 nm, RMS is from 66,93 to 108,54 nm and R_a (mean roughness) is from 51,53 to 82,48 nm.





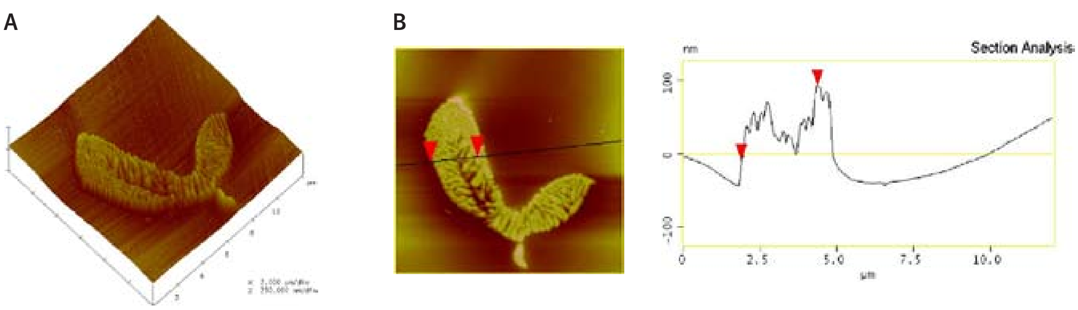
Slika 1. A) Optički mikrograf prikazuje tipičnu dendritsku strukturu površine CoCr legure (dendriti-svijetle zone i interdendritska područja-tamne zone) (325x325 µm); B) 3-D AFM snimka (100x100 µm): nakupine nanokristalita formiraju otoke (odgovaraju tamnoj zoni kod A) i glatka undulirajuća površina (odgovara svijetloj zoni kod A); C) Analiza presjeka pomoću AFM

Figure 1 A) Optical micrograph revealing typical dendritic structure (dendrites-light zone and interdendritic region-dark zone) of a surface of the CoCr dental alloy (325x325 µm); B) The 3-D AFM image (100x100 µm): clusters of nanocrystallites forming islands correspond to the dark zone in A and a smooth undulating surface corresponds to the light zone in A; C) Section analysis by AFM



Slika 2. A) 3-D AFM snimka (30x30 µm): Dva otoka nanokristalita; B) Analiza presjeka iste snimke pomoću AFMa

Figure 2 A) The 3-D AFM image (30x30 µm): Two islands of nanocrystallites; B) Section analysis of the same image by AFM



Slika 3. 3-D AFM snimka (12x12 µm): A) Jedan otok nanokristalita; B) Analiza presjeka

Figure 3 The 3-D AFM image (12x12 µm): A) A single island of nanocrystalites; B) section analysis

Tablica 1. Deskriptivna statistika podataka dobivenih analizom hrapavosti površine 10 elektropoliranih Co-Cr uzoraka
Table 1 Descriptive statistics obtained from roughness analysis of 10 electropolished CoCr samples

	x (nm)	SD	SE	hrvatski • 95% Confidence Interval for Mean	Minimum	Maximum
Z-range	930,10	97,29	39,91	819,98 - 1020,99	827,33	1099,00
RMS	88,29	20,57	8,03	66,93 - 108,54	64,00	117,42
R_a	66,67	15,19	6,74	51,53 - 82,48	46,39	88,89

Legenda • Legend:
x = srednja vrijednost • mean, SD = standardna devijacija • standard deviation, SE = standardna pogreška • standard error;
RMS = korijen srednjeg kvadrata • root mean square, R_a = srednja hrapavost • mean roughness

Rasprava

Vrlo je važno postići što glatkiji izgled površine zubnih legura, jer upravo ona određuje područje kontakta s intraoralnom tekućinom (slinom) i mikroorganizmima usne šupljine.

Nedavno razvijena tehnika mikroskopiranja – AFM (mikroskopiranje atomskim silama) omogućila je trodimenzionalni prikaz povećane promatrane površine na subnanometarskoj ljestvici (10-15). Optički i elektronički mikroskop (SEM) prikazuju povećanu površinu jedino u dvjema dimenzijama.

Co-Cr legure imaju površine dendritičke mikrostrukture, ako se polagano hlade tijekom stvrdnjavanja (prelazak iz tekućeg rastaljenog u kruto stanje) (7-9). Ako se njihova površina gleda pod svjetlosnim mikroskopom ili SEM-om, vide se svjetlija područja poznata kao dendriti i tamnija – interdendritska područja. U jednom istraživanju objašnjeno je da tamna interdendritička područja i svijetla dendritička zapravo predstavljaju dvije vrste krutih otopina različitih prema mikrostrukturi, a samo neznatno u kemijskom sastavu. Smatra se da dendriti (svijetlo područje) predstavljaju fcc (kubičnu plošnu kristalnu rešetku), a tamna područja predstavljaju hcp (heksagonalnu kristalnu rešetku). Također se smatra da je mikrotvrdoća fcc-a veća od hcp-a (8,9).

Prvi put su u ovom istraživanju prikazane i analizirane površine Co-Cr legura pomoću AFM-a. Na taj način jasno je vidljivo da su tamna područja na optičkom mikroskopu nakupine nanokristalita koji formiraju otoke, a uzdižu se iz undulirajuće glatke površine. AFM-analizom izmjereni su visine i opseg otoka. Tamna boja te strukture na optičkom mikroskopu i SEM-u vjerojatno nastaje zbog loma svjetla, jer su otoci (sastavljeni od nanokristalita) nazubljeni na vrhu, pa dvodimenzionalno daju dojam tamne boje. Svijetla područja (dendriti) na snimkama SEM-a i optičkog mikroskopa odgovaraju glatkoj undulirajućoj površini na trodimenzionalnim snimkama AFM-a.

Discussion

It is very important that the surfaces of dental alloys are smooth as they determine the area of the contact surface with oral fluids and microorganisms.

A new recently developed microscope-imaging technique – the AFM - enabled the three-dimensional view and enlargement of the observed surface to the subnanometer scale (10-15). The optical micrographs or SEM images present only a two-dimensional view.

The CoCr alloys have surfaces with dendritic microstructure when cooled slowly through a solidification range (7-9). The surface of a CoCr dental alloy viewed by optical microscope or SEM presents light regions, referred to as dendrites, and dark regions, referred to as interdendritic regions. It was reported that the dark interdendritic regions and the light dentritic regions, correspond to the two types of CoCr solid solutions, which differed in structure, but only slightly in composition. It has been reported that the dendrites (light region) represent fcc, while the dark regions hcp. It has also been reported that the microhardness of the fcc is greater than that of the hcp (8,9).

For the first time, this study observed three-dimensionally and analysed in details the surface of a CoCr dental alloy by AFM. Now, it is clearly demonstrated that the “dark” regions on optical micrographs and SEM images are actually clusters of nanocrystallites forming islands, emerging from a smooth undulating surface. The height and the perimeters of the islands were measured by the AFM section analysis. The dark colour in SEM images and optical micrographs is probably due to the sawtooth pattern of islands, i.e. to the peaks of nanocrystallites, which reflect the light in the way that the surface appears dark when viewed two-dimensionally. The light regions (dendrites) on SEM images and optical micrographs correspond to the smooth undulating surface on AFM images.

Maksimalne z-vrijednosti predstavljaju najveću vertikalnu udaljenost između najviše i najniže točke na snimci i variraju od 819,98 do 1020,99 nm ($p<0,05$). Prosječna visina otoka je 100-120 nm ($p<0,05$). Ako se uzme u obzir veličina najmanjih mikroorganizama poput bakterija (1μ) ili gljivica (4μ) (16), možemo biti zadovoljni izgledom površine Co-Cr legure. Prosječne vrijednosti z-raspona od 930,10 nm ($0,93\mu$), prosječne RMS-vrijednosti 88,29 nm ($0,088\mu$) i srednja hrapavost 66,67 nm ($0,0667\mu$) pokazuju da se čak i najmanje bakterije (1μ) ne mogu mehanički zarobiti na površini elektropoliranih uzoraka.

No, bilo bi zanimljivo promatrati učinak daljnjeg mehaničkog poliranja i efekt biokorozije na svaku od dviju čvrstih faza (otoci nanokristalita i undulirajuća glatka površina), koje su detaljno prikazane pomoću mikroskopiranja atomskih sila. No, to će biti područje budućeg istraživanja.

Zaključci

Mikroskopiranje atomskim silama (AFM) i analiza promatranih površina omogućili su trodimenzionalni prikaz Co-Cr legure. Otkriveno je da se njezina dendritička struktura sastoji od elevacija - nakupina nanokristalita koji oblikuju otoke (odgovaraju tamnim interdendritskim prostorima na snimkama optičkog mikroskopa), a izlaze iz glatke undulirajuće površine (odgovara svijetlim područjima – dendritima na snimkama optičkog mikroskopa). Površina elektropolirane legure ima dobra svojstva te se može smatrati doista glatkom sa stajališta mehaničkog zarobljavanja mikroorganizama.

The maximum z-value represents maximum vertical distance between the highest and the lowest data points in the image, and varied from 819.98 to 1020.99 nm ($p<0.05$). The average height of the island was 100-120 nm. Considering the size of small microorganisms, such as bacteria (1μ) and yeasts (4μ) (16), we can be satisfied with the surface characteristics of the CoCr alloy. The average value of the maximum z range was 930.10 nm (0.93μ), and the average root mean square roughness was 88.29 nm (0.088μ), and mean roughness was 66.67 nm (0.0667μ), revealing that even small bacteria (1μ) cannot be captured mechanically on the surface of electropolished samples.

However, it will be interesting to observe the effect of further mechanical polishing and the effect of biocorrosion on each of the two solidification phases (islands of nanocrystallites and the undulating smooth surface) identified by AFM and it will be the subject of the further research.

Conclusion

The AFM imaging and analysis made it possible to observe a surface of the CoCr dental alloy three-dimensionally. It has been understood that the dendritic surface structure of the CoCr alloy consists of clusters of nanocrystallites forming islands (corresponding to the dark interdendritic regions on optical micrographs) which emerge from a smooth undulating surface (corresponding to the light dendritic regions on optical micrographs). The surface reveals favourable characteristics regarding roughness, moreover it can be considered really "smooth" without any mechanical retentive features for microorganisms' trapping.

Abstract
The aim of this study was to acquire a three-dimensional qualitative and quantitative nano-scale data of surface topology of electropolished CoCr dental alloy, actually of the two different solid solutions previously described as dendrites and interdendritic regions. **Material and Methods:** Ten CoCr alloy samples (Wironit® extra-hard, Bego, Germany) were prepared, cast and electropolished. The surface of the samples was recorded and analysed by atomic force microscopy (AFM), as well as with optical microscope. The AFM imaging and analysis made it possible to observe a surface of the CoCr dental alloy three-dimensionally. It has been understood that the dendritic surface structure of the CoCr alloy consists of clusters of nanocrystallites forming islands (corresponding the dark interdendritic regions on optical micrographs), which emerge from a smooth undulating surface (corresponding the light dendritic regions on optical micrographs). **Results:** The 95% confidence interval ($p<0.05$) for the maximum z-value was from 819,98 to 1020,99 nm, for RMS values from 66,93 to 108,54 nm and for Ra (mean roughness) values from 51,53 to 82,48 nm. **Conclusion:** The surface revealed favourable characteristics regarding roughness, moreover the surface of the alloy can be considered really "smooth", without any mechanical retentive features for microorganisms' trapping.

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References

1. Taylor DF, Leibfritz WA, Adler AG. Physical properties of chromium-cobalt dental alloys. J Am Dent Assoc. 1958;56:343-51.
2. Cunningham DM. Comparison of base metal alloys and type IV gold alloys for removable partial denture frameworks. Dent Clin North Am. 1973;17:719-22.
3. Lane JR. A survey of dental alloys. J Am Dent Assoc. 1949;39:414-37.
4. Taylor DF, Sweeney WT. Proposed specification for dental chromium-cobalt casting alloys. J Am Dent Assoc. 1957;54:44-8.
5. Bezzon OL, Pedrazzi H, Zaniquelli O, da Silva TB. Effect of casting technique on surface roughness and consequent mass loss after polishing of NiCr and CoCr base metal alloys: A comparative study with titanium. J Prosthet Dent. 2004;92(3):274-7.
6. Gough JE, Clupper DC, Hench LL. Osteoblast responses to tape-cast and sintered bioactive glass ceramics. J Biomed Mater Res A. 2004;69(4):621-8.
7. Živko J, Jerolimov V. Metali u stomatološkoj protetici. Zagreb: Školska knjiga; 2005.
8. Matkovic T, Matkovic P, Malina J. Effects of Ni and Mo on the microstructure and some other properties of Co-Cr dental alloys. J Alloy Comp. 2004;366(1):293-7.
9. Matkovic T, Slokar Lj, Matkovic P. Structure and properties of biomedical Co-Cr-Ti alloys. J Alloy Comp. 2006;407(1-2):294-8.
10. Mendez-Vilas A, Donoso MG, Gonzalez-Carrasco JL, Gonzalez-Martin ML. Looking at the micro-topography of polished and blasted Ti-based biomaterials using atomic force microscopy and contact angle goniometry. Colloids Surf B Biointerfaces. 2006;52(2):157-66.
11. Jandt KD. Atomic force microscopy of biomaterials surfaces and interfaces. Surf Sci. 2001;491(3):303-2.
12. Radmacher M, Tillamnn RW, Fritz M, Gaub HE. From molecules to cells: imaging soft samples with the atomic force microscope. Science. 1992;257(5078):1900-5.
13. Valois CR, Silva LP, Azevedo RB, Costa ED Jr. Atomic force microscopy study of gutta-percha cone topography. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2004;98(2):250-5.
14. Giessibl F. Advances in Atomic Force Microscopy. Rev Mod Phys. 2003;75 (3):949-83.
15. Lapshin RV. Feature-oriented scanning methodology for probe microscopy and nanotechnology. Nanotechnology 2004;15 (9):1135-51.
16. Alcamo E. The Microbiology Coloring Book. New York: AddisonWesley; 1995.