단일, 이중 산처리 임플란트의 회전제거력 비교

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Comparison of removal torque of dual-acid etched and single-acid etched implants in rabbit tibias

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Purpose: Chemically strong-acids (HF and HCl/H₂SO₄) dual etching implant surfaces have higher strengths of osseointegration than machined implant surfaces. However, the dual acid treatment deteriorates the physical properties of the titanium by weakening the fatigue resistance of the implant and causing microcracks. The removal torque comparison between the dual-acid etched (hydrochloric acid, sulfuric acid, HS) and single-acid etched implants (hydrochloric acid, H) could reveal the efficiency of implant surface acid treatment. **Materials and methods:** Nine 3.75×4 mm dual-acid etched SLA implants and nine single-acid etched SLA implants were inserted into New Zealand rabbit tibias. After 10 days, removal torque, roughness, and wetting angle were measured. **Results:** Mean removal torque values were as follows: Mean removal torque were 9.94 Ncm for HS group and 9.96 Ncm for H group (*P*=.995). Mean surface roughness value were 0.93 µm for HS group and 0.84 µm for H group (*P*=.170). Root mean square roughness (RSq) values were 1.21 µm for HS group and 1.08 µm for H group (*P*=.294), and mean wetting angle values were 99° for HS group and 98° for H group (*P*=.829). Statistical analysis showed no significant difference between the removal torques, roughness, or wetting angles of the two groups. **Conclusion:** In this experiment, we found no significant difference in removal torque, roughness, or wetting angle between dual-acid etched and single-acid etched implants. (*J Korean Acad Prosthodont 2019;57:335-41*)

Keywords: Osseointegration; Biocompatibility; Implant surface treatment; Biomechanical evaluation

Introduction

The relationship between dental implants and bone was developed based on the concept of osseointegration of titanium, as claimed by Brånemark.¹ The high success rate of implants and good long-term prognosis are influenced by patient factors and the condition of the implant.^{2,3} Patient factors were not the topic of this study; the study focused instead on the conditions of the implants.

Implant conditions include physical properties, surface treatments, corrosion resistance, abrasion resistance, and others. The condition of the implant surface that directly integrates with the bone is an essential factor.^{4,5} This implant condition is actively studied to enlarge

the contact area between the bone and the implant and to increase the removal torque using surface treatments. $^{6\cdot22}$

Acid treatment methods using hydrochloric, sulfuric, and hydrofluoric acids have been introduced, which produce micropits with a diameter of 0.5 - 2.0 μ m on the implant surface.^{15,16} These acidtreated surfaces increase the apposition of bone on the implant surface by enhancing the osteogenic mechanism through the attachment of fibrin and osteoblasts.²³ Studies have determined that the implant surface treated with blasting and acid etching (SLA) results in a high BIC, high osseointegration strength, and high clinical success rates.^{6,16-22}

The acid treatment used for Straumann SLA implants is the dual-

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acid etching method, which mixes high-concentration hydrochloric and sulfuric acids at a temperature above 100°C, and Osseotite is in implants made using hydrofluoric, hydrochloric, and sulfuric acids. Other acid etching methods used depend upon the manufacturer. The treatment coats the implant surface with 20 - 40% titanium hydride (TiH); Straumann SLA implants consist of 37% TiH and 63% TiH.

However, the acid treatment deteriorates the mechanical properties of the titanium by weakening the fatigue resistance of the implant. Titanium hydride needles that were known as hydrogen embrittlement of titanium may be triggered by TiH. Stronger acid treatments increase surface roughness, but also increase the concentration of TiH resulting in weaker mechanical properties and increased possibility of fracture.^{24,25}

Surface wettability and roughness are also important physical properties of surface-treated implants that affect implant osseointegration. The adhesion and spread of cells on the bioconjugate material depends on the wettability of the material. Surface roughness affects wettability.²⁶⁻²⁸ At the early stage of implantation, cell attachment is affected by physicochemical properties including wettability. Therefore, the degree of wettability affects the initial stage of osseointegration of implants.

The dual-acid etching method is preferentially used for currently existing products. The treatment increases surface roughness at a microscopic level. It also causes various problems compared to the single-acid method, such as decreased mechanical properties of the implant and environmental pollution.

In this study, we compared the removal torque, roughness and wettability of two groups of integrating implants manufactured using dual-acid etching (hydrochloric acid, sulfuric acid) as the control group and single-acid etching (hydrochloric acid) as the experimental group.

Materials and methods

A total of 9 SLA implants (CSM Implant Co., Ltd., Daegu, Korea) manufactured using dual-acid etching with hydrochloric and sulfuric acids (HS group) and 9 SLA implants (CSM Implant Co., Ltd., Korea) manufactured using single-acid etching with hydrochloric acid (H group) were studied. The implants were 3.75 mm in diameter and 4 mm in length.

This experiment was approved by the Animal Care and Use Committee of Kyung-Pook National University. A total of 9 New Zealand rabbits with an average weight of 3.5 kg were included in the study. The rabbits were anesthetized intramuscularly with 0.2 mL/kg of tiletamine/zolazepam (Zolet, Virbac Laboratories, France) and 0.25 mL/kg of xylazine (Rompun, Bayer Korea, Korea).

After removing the hair from the intended surgical site, the site was pre-treated with iodine and a 75% ethanol hybrid solution before incision. After the tibia was exposed, the implants were placed according to the manufacturer's surgical guide. 9 implants surfacetreated with hydrochloric acid and sulfuric acid was inserted in the tibia under constant irrigation of physiological saline solution, and 9 implants surface-treated with hydrochloric acid was placed in the tibia. After implantation, the incision was closed with 4-0 Vicryl and 3-0 silk.

After the surgical procedure, antibiotics and medications were administered and appropriate management was performed. After 10 days, this period may be regarded as the early stage of the healing period, presuming that 10 days in a rabbit is equivalent to approximately 25 days in a human.²⁹ anesthesia was performed as described above and the surgical site was re-opened. After the implant was exposed, the removal torque (MGT-12 digital torque gauge, Mark-10 Corp, New York, NY, USA) was measured and the rabbits were sacrificed.

Roughness was measured in average surface roughness (RSa) and mean root square of the surface (RSq) using confocal laser scanning microscopy (Carl Zeiss, LSM 700, Oberkochen, Germany). A total of 3 specimens were measured at 3 middle point of implant.

Plates 10 mm square were made using the same material as the implants and received the same acid treatment. The wetting angle was measured by CA goniometer (SmartDrop, Femtobiomed Inc., Korea) using distilled water.

The two group were compared through t-test in statistical analysis.

Results

The average removal torque was 9.94 Ncm in HS group and 9.96 Ncm in H group (Table 1). The surface roughness of HS group was 0.93 μ m in RSa, 1.21 μ m in RSq and H group was 0.84 μ m in RSa, 1.08 μ m in RSq (Table 2). The wetting angle was 99° in HS group

Table 1. Removal torque of HS group and H group. The average removal torque was 9.94 Ncm in HS group and 9.96 Ncm in H group (Unit: Ncm)

Group	1	2	3	4	5	6	7	8	9
HS	8.2	13.7	8.6	10.0	11.4	10.1	9.5	10.8	7.2
Н	22.2	8.9	7.4	9.1	6.8	11.9	9.6	6.4	7.3

There is no statistically difference between dual acid treated group (HS Group) and single acid treated group (H group).

Group		1	2	3	4	5	6	7	8	9
HS	*Rsa	0.817	0.901	0.884	1.049	1.097	1.083	0.799	0.836	0.925
	†RSq	1.032	1.137	1.182	1.391	1.444	1.415	1.008	1.063	1.179
Н	*Rsa	0.672	0.762	0.696	0.750	0.739	0.842	1.073	0.982	1.045
	RSq	0.853	0.967	0.879	0.944	0.922	1.093	1.405	1.275	1.378

Table 2. Surface roughness of HS group and H group. The surface roughness of HS group was 0.93 μm in RSa, 1.21 μm in RSq and H group was 0.84 μm in RSa, 1.08 μm in RSq (Unit: μm)

* average surface roughness; † roughness mean root square of the surface

There is no statistically difference between dual acid treated group (HS Group) and single acid treated group (H group).

Table 3. Wetting angle of HS group and H group. The wetting angle was 99° in HS group and 98° in H group (Measurements are given as the wetting angle in degrees) (Unit: µm)

Group	1	2	3	4	5	6	7	8	9
HS	94.5°	98°	91.8°	81°	87.7°	103.9°	112.9°	104.5°	116.1°
Н	99°	98.6°	106.6°	101.3°	103.5°	94.1°	92.9°	94.1°	91.9°

There is no statistically difference between dual acid treated group (HS Group) and single acid treated group (H group).

Table 4. Statistical analysis between HS group and H group

	Group	Ν	Mean	SD	t	Р	
Denne (NI-m)	HS	9	9.9444	1.9223	0.006	005	
Kemoval torque (Ncm)	Н	9	9.9556	4.8972	0.000	.995	
$\mathbf{D}_{1} = (\mathbf{D}_{1}^{T} \mathbf{D}_{2}^{T} \mathbf{D}_{2}^{T})$	HS	9	0.9323	0.1157	1 426	170	
Koughness (KSa, µm)	Н	9	0.8401	0.1540	1.430	.170	
Roughness (RSq, µm)	HS	9	1.2057	0.1697	1 176	204	
	Н	9	1.0806	0.2197	1.170	.294	
Wetting angle (°)	HS	9	98.9333	11.5359	0.222	820	
	Н	9	98.0000	5.1247	0.222	.829	

Statistical analysis revealed no difference in removal torque, roughness, and wetting angle between the two groups.

and 98° in H group (Table 3). Statistical analysis revealed no difference in removal torque, roughness, or wetting angle between the two groups (Table 4).

Discussion

The removal torque test is used to measure the extent of osseointegration of the implant. The first use was in 1988 by Carlsson *et al.*, and measured the sheer force of the machined-screw implant required to cause a fracture between un-integrated and integrated bone.³⁰ Removal torque is not obtained by a break between the bone and the implant, but by a rupture between un-integrated and integrated bone. Thus, the measurement of removal torque detects the degree of osseointegration of the implant, and the removal torque varies depending on the surface roughness and the time of measurement. The timing at which the surface roughness of the test material affects osseointegration changes during osseointegration, so the timing of the measurement of the removal torque should be adjusted according to surface roughness.

Rapid bone attachment after implant placement is an important factor in the success of the implant. In clinical practice, the removal torque value (RTV) was approximately 140 Ncm in 4 weeks after insertion of acid-treated implants.³¹ Based on this, there have been studies of the 99% success rate of implants over 3 years in a clinical trial of prostheses 6 weeks after implantation.^{22,32} The results of these studies suggested that the initial bone attachment after implant place-

ment had a significant impact on the implant success rate.

In a rabbit, the removal torque changes significantly until 1 month after placement of the implant, but there is little or no change after 1 month.¹⁶ The removal torque after 1 month of implantation ranges from 20 to 30 Ncm.^{16,30} In addition, a study by Albrektsson et al. compared the healing time of rabbits with that of humans and concluded that 6 weeks of rabbit healing was equivalent to 3 - 4 months of human healing.²⁹ This period may be regarded as the early stage of the healing period, presuming that 10 days in a rabbit is equivalent to approximately 25 days in a human. The average removal torque after 10 days was approximately 10 Ncm in this experiment. In comparison with the removal torque after 1 month, this degree shows a fairly high removal torque during a normal healing process. Therefore, the surface of the implant with a micro-texture due to acid treatments seems to affect osseointegration at the early stage of implantation. As there was no difference between the mean values of the experimental and control groups, the results of the dual-acid etching method using hydrochloric acid and sulfuric acid and the single-acid etching method using only hydrochloric acid were not significantly different.

The surface roughness of implants in this experiment ranged from 0.5 to 2 μ m, which generally results from acid surface treatments. Implants with this range of roughness promote early osseointegration.¹⁶ Thus, the roughness of an implant affects the relationship between surface energy and cell proliferation.³³

According to Wennerberg *et al.*,³⁴ a histological experiment using 5 μ m, 75 μ m, and 250 μ m particle blasted implants showed that 250 μ m implants have the highest BIC. Another study reported that plasma spray methods or coating methods lead to increased roughness but decreased BIC, removal torque, and osseointegration strength compared to implants treated with blasting or acid.⁶

Webb *et al.*³⁵ demonstrated that cell differentiation is affected by surface roughness. Cells grown on rough surfaces produce more osteocalcin and alkaline phosphatase, suggesting that these mechanisms are the result of the differentiation and proliferation of osteoblasts.

During the healing process after implantation, cells migrate toward and adhere to the surface of the implant to formulate bone, which results in osseointegration of the implant into the bone. Cells require a certain roughness to move rapidly and adhere to the implant surface. Thus, not just high but suitable roughness is important. The acid treatment is an essential factor for the preparation of implants with rough surfaces, but there was no difference in roughness between the control and the experimental groups in this experiment. Thus, the dual-acid and single-acid etching methods were not different. Dualacid etching cannot be considered superior to single-acid etching, as there was no difference between the experimental and control groups and the ideal roughness has not yet been determined.

Physical properties of the surface and surface energy can be mea-

sured using the wettability and contact angle between the surface and the liquid. These values indicate whether a surface is hydrophilic or hydrophobic. Surfaces with contact angle close to 0° are hydrophilic, while those with contact angle close to 140° are hydrophobic.

The average contact angles of the experimental and control groups were 98° and 99°, respectively, suggesting no difference. Therefore, the contact angle, or wettability, did not appear to differ according to single-acid or dual-acid etching. These results suggest that both experimental and control implants are more hydrophobic than hydrophilic.

The higher hydrophilicity encourages cell and tissue interaction.³⁶ Proteins on the surface of implants are highly influenced by surface properties in their absorption and adhesion. Therefore, the degree of hydrophilicity of the implant surface affects initial osseointegration.

Surface properties such as roughness, wettability, and surface energy affect the cells involved in early osseointegration. Studies by Boyan *et al.*³⁷ have shown that higher wettability increases cell differentiation, and that more fibroblasts attach to a hydrophilic surface than to a hydrophobic surface. These hydrophilic implant surfaces enhance adhesion of fibrin to the surface, and the attached fibrin induces the attachment of osteoblasts. The attachment mechanism of cells to the implant surface affects the differentiation of cells and the formation of surrounding tissues.

Chemical acid treatments weaken the physical properties of titanium. These treatments produce microcracks that weaken the fatigue resistance of the implant. The effect is due to hydrogen ions (H⁺) generated during the acid treatment of the implant surface that rapidly penetrate the surface and accumulate underneath. These ions produce the titanium hydride layer.^{38,39} Titanium hydride can be up to 40% of the implant. This high concentration of titanium hydrides triggers a hydride needle, which weakens fatigue resistance. This is known as hydrogen embrittlement of titanium.^{25,40} SLAactive is manufactured with dual-acid etching and includes 37% titanium hydride.²⁴ This titanium hydride weakens the implant, which decreases the prognosis of the implant over time.

However, according to a study of sandblasted and acid-etched implants by Zhang *et al.*,⁴¹ the experimental group containing TiH showed increased alkaline phosphatase and osteocalcin. The study determined that TiH is eliminated by heat treatment. In addition, cell differentiation and cell adhesion were significantly higher in the TiH group, suggesting that TiH improves osseointegration by affecting cells and surrounding tissues during the early stage of osseointegration.

As there are a number of methods that do not use blasting or acidetching surface treatments, and implants treated with those methods also show good osseointegration,^{7,9,11} the use of blasting and strong, dual, triple etching should be reconsidered. TiH, which is produced at high concentrations by these acid treatments, ultimately weakens the implant. In addition, more acid treatments increase the industrial pollution generated by the treatments, and damage the environment. In this experiment using implants treated by the dual-acid and single-acid etching methods in rabbits, we found that there is no significant difference in the removal torques, roughness, or contact angles between the two types of treated implants. So we expect that implant surface treatment should be not by stronger acid etching and more surface roughness, but by making ideal surface roughness through study and researches.

Conclusion

The results of this study show no significant difference in removal torque, roughness, or wetting angle between dual-acid etched and single-acid etched implants in rabbit tibias.

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단일, 이중 산처리 임플란트의 회전제거력 비교

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목적:불산과 염산/ 황산을 두 번 사용하는 임플란트 표면처리 방식은 선반가공한 표면에 비하여 강한 결합력을 보이지만 두 번 산처리하는 방식은 티 타늄의 피로도를 야기시켜 미소균열을 야기시킬 수 있으므로, 염산으로 한번 산처리하는 방식이 비슷한 결합력을 가진다면 단순하고 효율적인 산처리 방식이 될 수 있다.

재료 및 방법: 9개의 지름 3.75, 길이 4 mm의 두 번 처리 방식의 임플란트와 같은 크기의 한 번 처리 방식의 임플란트를 토끼의 경골에 심어서, 10일 후 에 회전제거력, 거칠기, 젖음각도를 측정하였다

결과: 두 번 처리 방식과 한 번 처리 방식의 회전제거력은 차이가 없었고 (*P* = .995). 두 번 처리군의 거칠기는 0.93 μm, 한 번 처리는 0.84 μm이었다 (*P* = .170). 평균젖음각은 두 번 처리는 99°, 한 번 처리는 98° 이었다 (*P* = .829). 각 변수 모두 두 그룹의 차이는 없었다.

결론:산으로 한번 처리하거나 두 번 처리 하거나 두 그룹의 회전제거력, 거칠기, 평균젖음각의 차이는 없었기 때문에 종래의 dual acid treatment보다 한 번 처리하여도 유용한 것으로 보여진다. (대한치과보철학회지 2019;57:335-41)

주요단어: 골유착; 생물학적 적합성; 임플란트 표면처리; 생체역학적 평가