



Basic Research for New Tooth Whitening Applied the Powder Jet Deposition

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Received: July 22, 2019; **Published:** August 14, 2019

Abstract

Objectives: The powder jet deposition (PJD) process is used for creation of hydroxyapatite (HA) layers on human teeth. The fabricated HA layers showed excellent microstructural and mechanical properties *in vitro* and *in vivo*. Titanium dioxide (TiO₂) changed the whiteness of the layers because of the selective reflection of light. This raises the possibility that TiO₂-HA layer deposition may improve new whitening treatment for discolored teeth. For the development of new tooth whitening by PJD, the microstructural and mechanical properties of the TiO₂-HA layers, particularly the effects of thermal stress, the color change obtained, and the color stability of the layers after thermal cycling were evaluated.

Materials and Methods: The microstructural properties of the TiO₂-HA layers, namely surface thickness and roughness were evaluated from scanning electron microscopy (SEM) images and three-dimensional profiles. The mechanical properties were evaluated by micro-Vickers hardness and bonding strength tests. For color differences and stability, the CIE L*a*b* color system was used, and ΔE* values were calculated before and after the fabrication of the TiO₂-HA layers. For visual color evaluation, images of specimens were taken with a digital camera. Furthermore, these material properties and color differences were evaluated before and after 500 thermal cycles (5–55 °C).

Results and Discussion: The TiO₂-HA particles were packed densely in the layers, the maximum thickness of layers was about 60 μm. There were no significant differences in surface roughness, hardness, or bonding strength before and after the thermal cycling. Moreover, these properties were equivalent to those of HA layers fabricated by PJD. The TiO₂-HA layers had increased L* and decreased b* parameter compared with those of the enamel substrates, and the color difference ΔE* was about 6.7 units, indicating that TiO₂-HA layers whitened the color of the treated tooth. Furthermore, it was confirmed from digital camera images that TiO₂-HA layers showed visually perceptible level of whiteness. This color was maintained even after thermal cycling.

Conclusion: The fabrication of TiO₂-HA layers by PJD may be a suitable new whitening treatment for discolored teeth.

Keywords: Powder Jet Deposition; Hydroxyapatite; Titanium Dioxide; Whitening; Discolored Teeth

Abbreviations

PJD: Powder Jet Deposition; HA: Hydroxyapatite; TiO₂: Titanium dioxide; SEM: Scanning electron microscopy; TiO₂-HA layers: HA layers containing TiO₂ particles

Introduction

The self-esteem and professional relationships were influenced by the discolored teeth, white and light-colored teeth are regarded as important [1,2]. Tooth discoloration is classified by intrinsic or extrinsic factors (or a combination of both) according to etiology, and varies based on location, appearance, affinity to the tooth structure, and severity [3,4]. For removing the extrinsic discoloration, including chromogens with coffee, tea, cigarettes, or dental plaque, professional tooth polishing using abrasive compounds is generally used [1-5]. On the other hand, for treatment of teeth with intrinsic discoloration, that may be drug-related or caused by pulp tissue remnants after endodontic therapy or pulp necrosis, the bleaching with using radical agents or tooth restoration with conventional dental materials such as composite resin and porcelain are generally taken [2,6]. However, several studies showed side-effects of tooth bleaching agents, such as irritation of dental pulp, degreased enamel hardness, and tooth hypersensitivity [7,8]. Conversely, conventional restoration methods are well-established or developed, especially adhesive restoration [2], but require invasive excavation of tooth substance. Furthermore, the restorative materials used have quite different chemical and mechanical properties from those of tooth substance, enamel, and dentin, leading to clinical problems in the long run such as the loss of restorative materials from the tooth or secondary caries [9]. These observations suggest that the development of a novel method that provides a biomaterial interface layer with the tooth surface is necessary for the whitening treatment of tooth discoloration without invasion of the tooth substance and dental pulp.

In previous studies, the possibility of applying HA layer as a new restorative material with compositional, chemical and mechanical properties corresponding to those of tooth substance was presented [10-13]. The PJD is a precise mechanical coating technique with using ultrafine particles which accelerated to several hundred meters per second by the jet flow of carrier gas to fabricate functional surfaces with high efficiency. PJD is used at room temperature and atmospheric pressure, which enables

it to be used with HA particles for the fabrication of HA layers on human teeth [10-13]. In *in vitro* studies showed that PJD technique can be used to fabricate thick HA layers on human enamel. These HA layers showed excellent microstructural and mechanical properties, including a micro-Vickers hardness equivalent to that of human enamel and a bonding strength almost equivalent to that of composite resin-enamel. These properties were not influenced by thermal stress [12]. Furthermore, PJD technique and fabricated HA layers showed high biocompatibility and safety in a preclinical trial [13].

TiO₂ is a common additive in many pigments, personal care products, and other consumer products used in daily life. TiO₂-based ingredients are mostly used as coloring agents in enamels, plastics, glazes, paper, foods, pharmaceuticals, cosmetics, and toothpastes [14]. TiO₂ is also an important additive for dental composites, which must match the color of the natural tooth. TiO₂ particles are non-toxic and stable in the oral environment and show high color stability over a long period [15]. According to a study carried out to determine the influence of TiO₂ particles on the color, translucency, fluorescence, and opalescence of light curing composite resin, a concentration of 0.10–0.25% TiO₂ particles induced a whiter tooth color and simulated opalescence of human enamel [16]. In a study assessing the effect in terms of color stability of adding TiO₂ nanoparticles to acrylic resin intended for the artificial sclera of ocular prostheses, the incorporation of 2.5% TiO₂ helped to maintain color stability after irradiation with ultraviolet light, simulating the aging of human eyes [17]. Furthermore, in this previous study, X-ray diffraction analyses confirmed that no new chemical compounds were generated from the TiO₂ and HA particles. Therefore, the application of the HA layers containing TiO₂ particles (TiO₂-HA layers) to the human tooth surface by PJD was proposed as a new whitening treatment for discolored teeth. The previous studies have led to an understanding of the excellent properties of HA layers fabricated by PJD. However, the properties of TiO₂-HA layers have not yet been clarified. Furthermore, the degree to which TiO₂-HA layers influence whitening and color stability have not been well documented. For the development of new whitening treatment system, the microstructural and mechanical properties of TiO₂-HA layers before and after thermal cycling, and the color difference and color stability of the TiO₂-HA layers were evaluated as a basic research. The null hypotheses of

the present study were: (1) TiO_2 -HA layers could not be created by PJD process and would not demonstrate excellent microstructural and mechanical properties, (2) TiO_2 -HA layers would not whiten the tooth color, and (3) the material properties and white color of the TiO_2 -HA layers would not be maintained after thermal cycling.

Materials and Methods

Preparation of specimens and PJD process

Caries-free human molars extracted during orthodontic treatment were used in the present study, with the informed consent of the patients. All protocol and consent forms were approved by the Ethics Committee (approval number 25-10) of the Tohoku University Graduate School of Dentistry. The absence of caries was determined according to clinical parameters using a sharp explorer and visual inspection. The human molars were also checked by a diagnostic laser (DIAGNO dent, GC Co., Ltd. Tokyo, Japan) to make sure that no caries was present. The crown of each human molar was severed into lingual and buccal parts with using a diamond blade cutter. The buccal piece of the tooth was fixed onto the aluminum SEM stage using the epoxy resin-based adhesive agent. For present study, a flat enamel surface perpendicular to the enamel rods was prepared. To obtain an identical surface, the specimens were polished successively with 320-, 600-, 1000-, 1200-, 2000-, and 4000-grit silicon carbide grinding wheels with diamond paste, and the surface roughness was adjusted to about $0.4 \mu\text{m Ra}$. Before the experiment, the specimens were cleaned in an ultrasonic cleaning bath for 30 min and then dried in an air flow. The specimens were treated with PJD to fabricate TiO_2 -HA layers on their surfaces using HA- TiO_2 particles synthesized by Sangi Co., Ltd. (Tokyo, Japan), to which TiO_2 was added as a color regulator [13].

In the present study, the dental PJD device developed by Kuriyagawa and Sangi Co., Ltd. was used for fabrication of the TiO_2 -HA layers on the enamel substrate specimens (Figure 1). The blasting nozzle of the device was of a similar size to that of normal dental hand-pieces (Figure 2). The HA and TiO_2 particles were mixed in the chamber of device with continuous flow of accelerated gas (air) and blasted from the nozzle onto the enamel substrate at room temperature (25°C) and atmospheric pressure (1 atm). Table 1 shows the details of the experimental conditions of the blasting.



Figure 1: Dental hand-piece type PJD device.



Figure 2: Blasting nozzle of PJD device.

Substrate	Enamel
Size of particles (containing TiO_2) (μm)	3.0 ± 1.0
Accelerated gas pressure (MPa)	0.5
Feed gas pressure (MPa)	0.5
Blasting angle ($^\circ$)	90
Blasting time (s)	30
Nozzle scan rate (mm/s)	5
Gap between nozzle and substrate (mm)	1.0

Table 1: The details of the experimental conditions of the PJD blasting.

Specimens were fixed on the stage, with their enamel surface perpendicular to the blasting nozzle. Each specimen was blasted using a fixed time and nozzle scan rate, overlapping about 15 times, to fabricate TiO₂-HA layers with an area of about 3.5 mm × 3.5 mm. After the deposition, the layers were polished by diamond polishing paste (Dia Polisher Paste, GC Co., Ltd., Tokyo, Japan) and a felt wheel for 30 s with a pressure of 5000 g [12].

Thermal cycling

Two-temperature computer-controlled thermal cycler (Thermal cycling K-179; Tokyo-Giken Co., Ltd., Tokyo, Japan) was used. The two water baths of the thermal cycler were maintained at 5°C and 55°C. Each cycle is consisted of 20 s immersion in each water bath and a travel time of 10 s. The water baths were constantly stirred by stirrers, and the variation in the temperature of each water bath remained within 1°C of the set temperature. The specimens were immersed in distilled water at 37°C for 1 day. Following this immersion, the specimens were subjected to 500 thermal cycles [12,18].

Evaluation of microstructural and mechanical properties

The cross-section of the TiO₂-HA layers was observed by SEM (JSM-6500F, JEOL Co., Ltd., Tokyo, Japan) for evaluation of their microstructure. The three-dimensional profiles, including surface thickness and roughness, of the layers were evaluated by three-dimensional non-contact measurement system (NH-3, Mitaka Kohki Co., Ltd., Tokyo, Japan). For evaluation of the mechanical properties, micro-Vickers hardness was measured by dynamic micro-hardness tester (FM-ARS 9000, Future-Tech Co., Ltd. Kawasaki, Japan). A load of 100 gf was applied for 5 s with a pyramid-shaped die; the depth of the resulting impression was used to calculate the hardness of the specimens [12,19]. Furthermore, the bonding strength of the layers to the enamel substrates was evaluated by micro-tensile test (Romulus, Quad Group, Spokane, WA, USA) [12,20]. An epoxy pre-coated alumina stud was prepared perpendicularly onto the surface of the specimen. The area of the surface coated with the epoxy glue was about 2.7 mm in diameter. The specimen was set into the machine and gripped after curing at 150°C for 1 h. The stud was pulled until destruction of the specimen occurred, and the bonding strength was determined to be the maximum load recorded [12]. SEM images, three-dimensional profiles, surface thickness and roughness for microstructural properties, and micro-Vickers hardness and bonding strength for mechanical properties were evaluated before and after the above-mentioned thermal cycling.

Evaluation of color difference and stability

The color of the specimens was measured according to the CIE L*a*b* color scale relative to the standard illuminant D65 and using a 10° observer on a spectrophotometer (CMS-35FS/C, Murakami Color Research Laboratory Co., Ltd., Tokyo, Japan) at wavelengths of 390–730 nm in reflectance mode [13]. The CIE L*a*b* system is composed of three axes or coordinates: L* (lightness, from 0 = black to 100 = white), a* (from -a = green to +a = red) and b* (from -b = blue to +b = yellow) [21,22]. The area of irradiation and color measurement was φ6 mm and φ3 mm, respectively. The areas of TiO₂-HA layers and the control HA layers were set to be wider than the spectrophotometer measurement area. Each specimen was chromatically measured three times under different conditions – (1) before and (2) after the fabrication of the layers, and (3) after the thermal cycling – and the average values were calculated. Total color difference between the above-mentioned three conditions were calculated by the following equation:

$$\Delta E^* = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2}$$

For evaluation of the degree of whitening, the color difference between conditions (1) and (2) was calculated as ΔE^*_1 . For evaluation of the color stability, the difference between conditions (2) and (3) was calculated as ΔE^*_2 . Furthermore, images of the TiO₂-HA layers at the above-mentioned three conditions were obtained by digital camera (Nikon D70S, Nikon Co., Ltd., Tokyo, Japan) for visual evaluation of the color difference and color stability.

Statistical analysis

The Mann-Whitney U-test was used to detect significant differences in surface roughness, micro-Vickers hardness and bonding strength before and after thermal cycling. For evaluation of color difference and stability, the Turkey-Kramer test was used for the among-group comparison. The IBM SPSS Statistics software version 25 (Japan IBM Co., Ltd, Tokyo, Japan) was used for statistical analysis, and the significance level was set at 0.05.

Results and Discussion

Microstructural and mechanical properties

Figure 3 shows cross-sectional SEM images of the TiO₂-HA layers and the enamel substrate before and after the thermal cycling. There were no cracks or pores in the layers, indicating the deposition of fully dense TiO₂-HA layers. Furthermore, there were no observable gaps between the layer and the enamel substrate. In a

previous study on the microstructure of a plasma-sprayed 50 μm -thick HA coating on titanium alloy, cross-sectional SEMs revealed the presence of cracks in the titanium alloy and along the HA coating-substrate interface [23]. Such cracks may directly influence the bonding strength and may affect the long-term stability of such HA coatings [23,24]. Although the substrate was different, it could be considered HA layers with excellent material properties are more effectively fabricated using PJD process than the above-mentioned plasma spray coating technique.

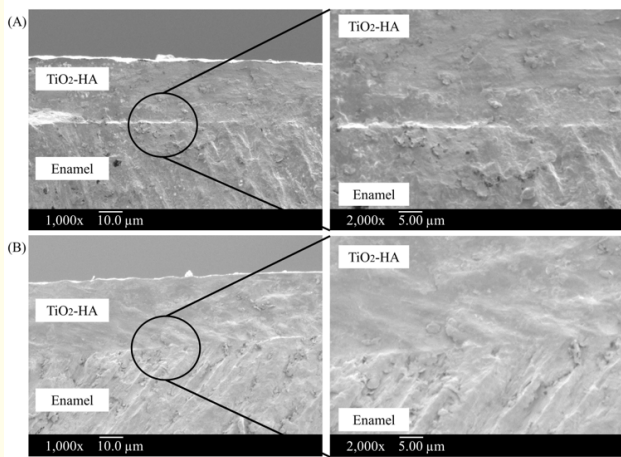


Figure 3: SEM images of the cross-section of TiO_2 -HA layer before (A) and after (B) thermal-cycling.

Figure 4 shows three-dimensional view of the fabricated TiO_2 -HA layers. Under the present conditions, the maximum and average thickness were about 60 μm and 50 μm , respectively. In a previous study, the maximum and average thicknesses of HA layers fabricated on enamel substrates by PJD on the same experimental conditions were also about 60 μm and 50 μm , respectively [12]. It was expected that the present TiO_2 -HA layers would show almost the same microstructural properties and high stability as those of the previous HA layers.

Table 2 shows the average surface roughness, the micro-Vickers hardness, and the bonding strength of the layers. The surface of TiO_2 -HA layers polished smoothly by diamond polishing paste, and their surface roughness was almost equal to that of the HA layers in the previous study [12]. It is well known that smoothly

polished dental restorations show high aesthetic quality and compatibility compared with rougher surface restorations in oral cavities [25,26]. Studies investigating the surface roughness of dental composite resin have reported that Ra values above 0.2 μm may lead to increased periodontal inflammation, plaque accumulation, and greater stain absorption than relatively smooth surfaces [27-29]. According to a review of the literature, the roughness of enamel surfaces in the oral cavity influences initial bacterial adhesion, and plaque tends to accumulate more on surface irregularities such as cracks, grooves, or abrasion defects on enamel [30]. Therefore, the present TiO_2 -HA layers were expected to show low bacterial adhesion, low plaque accumulation, and low stain absorption after polishing because they had a much lower Ra than those reported in other similar studies.

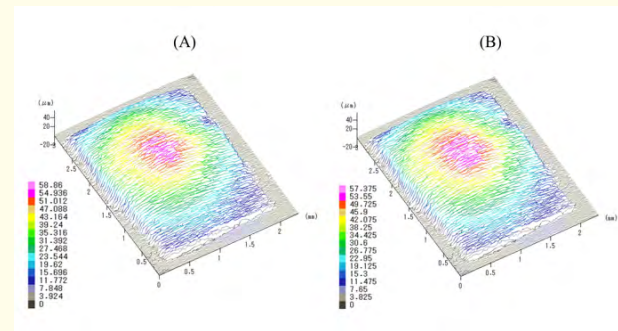


Figure 4: Three-dimensional view of TiO_2 -HA layer fabricated on enamel substrate before (A) and after (B) thermal cycling.

Evaluation items	Before thermal cycling	After thermal cycling	P-value
Surface roughness (μm) ^a	0.0774 (0.025)	0.0793 (0.030)	0.946 ^b
Micro-Vickers hardness (Hv) ^a	371.38 (20.68)	365.6 (27.53)	0.496 ^b
Bonding strength (MPa) ^a	15.71 (1.48)	15.49 (3.82)	0.791 ^b

^a Data are shown as mean (standard deviation).

^b By Mann-Whitney *U*-test.

Table 2: Surface roughness, micro-Vickers hardness and bonding strength of TiO_2 -HA layers before and after thermal cycling. (n=10).

With regard to the micro-Vickers hardness and bonding strength, these results are also almost the same as those obtained for the HA layers fabricated on enamel under the same PJD conditions in the previous study [12]. Furthermore, several studies have showed composite resin to enamel substrate bonding strengths of 5.9 to 22.2 MPa; these values are similar to the 15 MPa bonding strength measured for the present TiO₂-HA layers [31-33]. Thus, it was confirmed that the PJD process could fabricate TiO₂-HA layers with excellent material properties on enamel substrates. Furthermore, the above results indicate that included TiO₂ particles did not influence the layer fabrication process or the material properties of the HA. Therefore, the first null hypothesis was rejected.

Color difference

The color difference results of the TiO₂-HA layers and HA layers are shown in Table 3 and Table 4, and digital camera images are shown in Figure 5. The TiO₂-HA layers, between conditions (1) and (2), and (1) and (3), the L* parameter significantly increased, the b* parameter significantly decreased, and the a* parameter did not change. In contrast, none of the parameters significantly changed between any of the conditions for the HA layers. These results indicated that the fabricated TiO₂-HA layer whitened the color of the specimens; their color became slightly bluer. Furthermore, the results suggested that the HA layers were whitened by the addition of TiO₂ particles. Previous studies that have evaluated whitening effects *in vitro* have reported that whitening occurs mainly by increasing the L* parameter (higher L*), reducing yellowness (lower b*) and, to a lesser extent, reducing redness (lower a*) [2,34]. According to a review of the literature¹, the subjective responses to whiteness improvement and satisfaction are significantly correlated with changes in the b* parameter and not the L* or a* parameters, and the yellow-blue shift is perceptual importance in tooth whitening [1]. The reason for the color change observed

in the present study might be related to selective reflection and absorption of incident light by the TiO₂ particles. The total color differences ΔE*₁ of the TiO₂-HA layers and HA layers, which evaluate the degree of whitening, were about 6.7 units and 0.5 units, respectively. Several reports showed the threshold levels of total color difference which can be visually perceived [35,36]. In a study of intraoral determination of perceptibility and acceptability tolerances for shade mismatch, the color difference of the test denture which 50% of dentists could perceive was 2.6 units, while the value at which 50% of dentists would remake the denture owing to color mismatch was 5.5 units [36]. The digital camera images (Figure 5) clearly confirmed that the TiO₂-HA layers showed a visually perceptible level of whiteness. This indicated that the TiO₂ particles contributed to the whitening of the HA layers. Therefore, the second null hypothesis was also rejected. However, the present TiO₂-HA layers significantly whitened the enamel substrates beyond the perceptible threshold level, their color was too whitening and mismatched the optical properties of natural teeth.

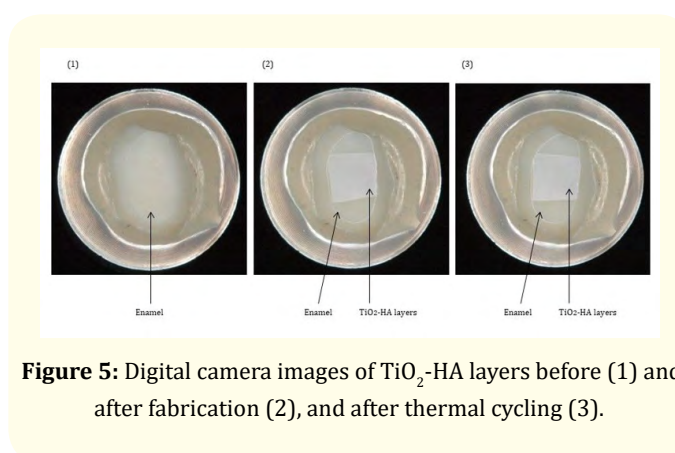


Figure 5: Digital camera images of TiO₂-HA layers before (1) and after fabrication (2), and after thermal cycling (3).

	Color parameter	(1) Before fabrication	(2) After fabrication	(3) After thermal cycling	P-value
TiO ₂ -HA ^a	L*	71.02 (1.81) ^{c,d}	77.27 (1.36) ^c	76.73 (1.43) ^d	<0.001 ^b
	a*	-2.25 (0.28)	-2.33 (0.35)	-2.25 (0.31)	0.537 ^b
	b*	-1.54 (0.23) ^{c,d}	-3.73 (0.46) ^c	-3.72 (0.42) ^d	<0.001 ^b
HA ^a	L*	70.11 (0.91)	70.60 (1.00)	70.33 (0.89)	0.121 ^b
	a*	-2.38 (0.33)	-2.44 (0.35)	-2.41 (0.37)	0.835 ^b
	b*	-1.43 (0.20)	-1.44 (0.20)	-1.45 (0.22)	0.932 ^b

^a Data are shown as mean (standard deviation).

^b By Turkey-Kramer test.

^c Significant within-group difference between “(1) Before fabrication” and “(2) After fabrication” by Turkey-Kramer test.

^d Significant within-group difference between “(1) Before fabrication” and “(3) After thermal cycling” by Turkey-Kramer test.

Table 3: The results of color parameters of TiO₂-HA and HA layers before (1) and after fabrication (2), and after thermal cycling. (n=10)

	ΔE^*_1	ΔE^*_2
TiO ₂ -HA	6.67 (1.48)	0.75 (0.65)
HA	0.54 (0.23)	0.34 (0.23)

Table 4: Total color differences of the TiO₂-HA and HA layers. (n=10)

Material and color stability

Thermal cycling simulated the frequent changes in intra-oral temperature induced by drinking, eating, and breathing in the present study. Thermal stresses are related to the mechanical stresses; differential thermal changes can induce crack propagation through bonded interfaces. The changes of gap dimensions result in volume changes which pump pathogenic oral fluids in and out of the gaps [37-40]. In the present study, thermal cycling between 5°C and 55 °C for 500 cycles based on the International Organization for Standardization specifications for evaluation of dental materials durability was adopted to evaluate the TiO₂-HA layers under the same conditions as those used for the HA layers in the previous study [12].

Figure 3 and 4 shows the TiO₂-HA layers maintained their three-dimensional morphology even after thermal cycling. Table 2 shows that there were no significant differences in the surface roughness, micro-Vickers hardness, and bonding strength of the layers before and after thermal cycling. The results of color difference of the TiO₂-HA layers (Table 3 and 4) confirmed that no significant changes in any of the color parameters occurred after thermal cycling; although a color difference ΔE^*_1 was observed, while ΔE^*_2 was negligible in the present study. Similarly, for the HA layers, there were no significant differences in any of the color parameters, and ΔE^*_2 was also negligible. Visual evaluation by digital camera images (Figure 5) revealed no differences in the color of the layers before and after the thermal cycling. In a previous investigation on the color stability of three light-polymerized veneer materials exposed to xenon light to simulate several years of clinical use, the obtained ΔE^* values of lower than approximately 3.3 units were considered to indicate high color stability [35]. Thus, the TiO₂-HA layers not only whitened the enamel substrates but also maintained their whiteness after thermal cycling. As a result, the third null hypothesis was also rejected.

In conclusion, the PJD process was used to fabricate a thick TiO₂-HA layers on human enamel substrates. The layers demonstrated

and maintained excellent microstructural and mechanical properties comparable to those of the HA layers in the previous study even after thermal cycling simulated the oral environment. Furthermore, it was confirmed that the TiO₂-HA layers whitened the color of the tooth surface and showed high color stability even after thermal cycling. However, the whiteness of the layers mismatched the natural tooth color to too great an extent. One of the major goals in esthetic dentistry is to produce restorations that match the properties of natural teeth [1]. It has been reported that adjusting the concentration of TiO₂ particles to be added to the composite resin makes it possible to control the dental shade [16]. Thus, adjusting the ratio of TiO₂ particles to HA particles used in the present PJD process could be used to fabricate layers matching natural tooth shades, as patients desire. Whether layers containing adjusted concentrations of TiO₂ will demonstrate the same excellent material properties is unclear but warrants examination. Furthermore, the present PJD process could be applicable to particularly discolored teeth because the TiO₂-HA layers showed a perceptible white color. However, it is unknown whether this process could be applied to tooth substrate with quantitative enamel deficiencies such as amelogenesis imperfecta, because the present study is based on the results of layers fabricated on sound enamel substrate. It is necessary to verify the layer formability on amelogenesis imperfecta affected tooth substrates, for example, on which most of the enamel has been chipped away or worn off, exposing large areas of dentin with yellowish-brown discoloration [41]. Achieving this would increase the potential of the PJD process for clinical use as a new whitening treatment for intrinsic tooth discoloration.

Conclusion

Within the limitations of the present study, it was concluded that a PJD process can be used to fabricate thick TiO₂-HA layers on enamel substrates. The TiO₂-HA layers demonstrated excellent microstructural and mechanical properties comparable to those of PJD fabricated HA layers, even after a thermal cycling. Furthermore, the TiO₂-HA layers whitened the tooth color and exhibited high color stability. The fabrication of TiO₂-HA layers by PJD may be a valuable new whitening treatment for discolored teeth.

Acknowledgements

The present work was supported by Sangi Co., Ltd. (Tokyo, Japan) for expert technical assistance. The present study received no external funding to conduct this research through any of the authors involved.

Conflict of Interest

The authors have no conflicts of interest.

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Volume 3 Issue 9 September 2019

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