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The effect of blocking the oxygen in the air during curing on the polymerization of sealant

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광중합 시 공기 중 산소의 차단이
치면열구전색제의 중합에 미치는 영향

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국문초록

광중합 시 공기 중 산소의 차단이 치면열구전색제의 중합에 미치는 영향

오 유 향

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광중합 레진의 중합과정에서 공기 중의 산소가 모노머와 경쟁적으로 라디칼과 결합하여 중합반응의 진행을 억제하므로써 레진의 표층에 산소 억제층이라 불리우는 10-15㎛ 두께의 미중합층을 형성한다. 미중합된 모노머들은 수복물의 물리적 성질을 저하시킴은 물론 구강내로 유출되어, 세포독성 및 알러지 반응을 일으킬 수 있다. 따라서 이러한 미반응 모노머를 다량 함유하고 있는 산소 억제층의 형성을 방지하거나 제거하여 레진의 중합률을 증가시키므로써 내구성의 향상과 함께 변색과 마모에 대한 저항성을 증가시켜 줄 수 있으며 이와함께 미반응 모노머에 의한 생체 유해성을 감소시킬 수 있다.

본 연구에서는 할로겐 광중합기, 플라즈마 광중합기, 그리고 2세대 고강도 LED 광중합기를 이용하여 치면열구전색제를 중합하는 과정에서 산소 차단 용액인 glycerin gel (DeOx®)의 도포, 질소 가스와 탄산 가스를 분사 시켜 공기 중 산소와의 접촉을 차단시킴으로써 산소억제층의 감소 여부를 규명하고자 하였다. 이에 광중합 광원의 종류와 공기 중 산소의 차단 방법에 따른 산소억제층의 두께, 치면열구전색제의 중합률, 그리고 표

면경도를 측정, 평가하였다. 산소 차단 방법을 시행하며 광중합 하여 제작된 시편을 HPLC에서 역상크로마토그래피를 이용하여 미반응 모노머 TEGDMA의 용출 양을 측정하여 중합률을 평가하였고, Vickers hardness tester를 이용하여 표면미세경도를 측정, 광학현미경을 이용하여 산소억제층의 두께를 측정하여 다음과 같은 결과를 얻었다.

1. 질소 및 탄산 가스를 분사하면서 중합한 군, DeOx®를 도포한 후 중합한 군 모두 공기 중에서 중합한 군보다 TEGDMA 용출량이 감소되었다 ($p<0.05$).
2. 할로겐 광으로 20초간 중합한 경우 DeOx®를 도포한 군과 질소 및 탄산 가스를 분사한 군의 TEGDMA 용출량은 유사하였지만($p>0.05$), 40초로 중합한 경우 탄산가스 분사군이 질소 가스 분사군 보다 TEGDMA 용출량이 적었다($p<0.05$).
3. 플라즈마 광으로 10초간 중합한 경우 DeOx®를 도포한 군의 TEGDMA 용출량이 가장 적었고($p<0.05$), 탄산 가스 분사군이 질소 가스 분사군 보다 용출량이 적었다($p<0.05$).
4. LED 광원에서는 탄산 가스 분사군이 질소 가스 분사군보다 TEGDMA의 용출량이 적었으나 유의할만 한 차이는 없었다($p>0.05$).
5. 세 광원 공히 공기 중에서 중합한 군보다 산소를 차단한 상태에서 중합한 군에서 미세경도가 크게 나타났다($p<0.05$).
6. DeOx®로 처리했을 때 플라즈마 광 10초와 LED 광 20초 중합군이 할로겐 광 40초 중합군보다 미세경도 값이 높았고, 질소 가스와 탄산 가스 분사 하에서 플라즈마 광으로 10초간 중합한 경우와 LED 광으로 20초간 중합 한 경우가 할로겐 광으로 40초간 중합한 경우보다 높은 미세경도 값을 보였다($P<0.05$).
7. 세 광원 공히 공기 중 중합한 군에 비해 질소 및 탄산 가스 분사를

분사하면서 중합한 군, DeOx®를 도포한 후 중합한 군이 산소억제층의 두께가 평균 49%의 감소되었으며($p<0.05$), 이들 산소를 차단한 군 간의 유의차는 없었다.

이상과 같은 결과를 종합해보면 치면열구전색제를 광중합할 때 DeOx®의 도포, 질소가스 및 탄산가스를 분사가 수복물의 최외층에서 산소와의 접촉을 차단하여 산소억제층을 감소시키는데 효과적이며 이러한 산소억제층의 감소가 미세경도 증가와 미반응 모노머의 유출 감소에 직접적인 영향을 미침을 알 수 있었다. 그러나 DeOx®의 도포는 시술의 단계를 증가시키며 유동성 있는 재료의 상층에 도포하는데는 어려움이 있어 임상적 적용이 쉽지 않다는 단점이 있다. 따라서 산소억제층 감소에 DeOx® 도포와 유사한 결과를 보인 질소가스와 탄산가스의 분사가 임상적 적용이 보다 유리 할 것으로 사료된다.

I. Introduction

Resin-based dental restorative materials are becoming the primary choice of clinicians. These products are used in a wide variety of ways: in Class I through Class Ⅵ preparations, as the cementing agents, the core buildup materials and the pit and fissure sealants. The advantages of these products are their ability to bond to tooth structure and the rapid rate at which they are set, especially if they are photo-activated.

Typically, commercial dental composites are random copolymers filled by various types of inorganic particles consisting of 2,2-bis[4-(2-hydroxy-3-methacryloxypropoxy)phenyl]-propane(Bis-GMA) and triethyleneglycol dim -ethacrylate(TEGDMA) as major reactors.

Bis-GMA and TEGDMA are bi-functional methacrylate monomers that harden by the free-radical-induced polymerization reaction. Multifunctional monomers used for dental restorations exhibit final double-bond conversion of 55 to 75%[1-3]. Furthermore, researchers have found that up to 6% unreacted monomer remained in the BisGMA/TEGDMA resin system after curing[4].

The pit and fissure sealants have the greatest potential for leaving uncured resin components in a restorative material. Because these materials are cured without an occlusal matrix, oxygen at the restoration/air interface inhibits the setting reaction on the outer layer of the sealant[5]. This mechanism is related to a free radical's preference to react with oxygen and

form a peroxy radical, which is very stable and quenches the radical's polymer-forming potential[6,7].

As a result, a uncured monomer layer covers the outer surface of the sealants. This layer is commonly referred to an oxygen-inhibited layer because of its origin, and it is primarily composed of unreacted monomers[5]. This unreacted monomer could be extractable and leach into the body, where various fates are possible. The previous researches indicate that components of resin-based restorative materials[8,9]and unreacted components leaching from cured restorations demonstrate *in vitro* cytotoxicity[10,11]. In addition, release of TEGDMA from resin composites was found to stimulate the growth of bacteria around the restoration[2]. Furthermore, unreacted functional groups can act as plasticizers, reducing the mechanical strength of the material and increasing the swelling. Therefore, elimination of uncured residual, unreacted monomers and removal of the oxygen-inhibited layer have been found to reduce cytotoxicity in cell cultures and improve the mechanical properties[12.13].

Various methods for reducing oxygen inhibition and unreacted monomer have been demonstrated[14-16]. One of them, oxygen inhibition can further be reduced by using high intensity of curing light. Halogen bulb based light curing units(halogen light curing units) have been as the most popular method of curing dental composites in the clinical setting. Recently, many different types of units have been developed, with newer types of light curing units such as laser, xenon arc plasma and light-emitting diode(LED)-based

technologies. Laser and xenon arc units have the high power intensity and the advantage of the reduced curing time, however, these light-curing units have a larger and more complicated construction, and are also more expensive than halogen light-curing units and LED-units. Recently, a second-generation LED-units were introduced to the market. The second-generation light delivers a different spectral distribution with a greater power output than the first-generation light and may therefore offer better performance and shorter curing times.

Another methods involves that oxygen impermeable barriers have been used to block the resin/atmosphere interface[17] and composite resin was cured under in the argon or nitrogen atmosphere in an attempt to eliminate oxygen inhibition[18,19].

It has not been not reported that carbon dioxide is applied in the dental materials to remove oxygen inhibited layer.

The present research were performed for following purposes: (1) to measure the amount of leachable monomer, (2) to determine the microhardness of upper surface, (3) to measure the thickness of oxygen inhibited layer and (4) to compare the efficacy of reducing the oxygen inhibited layer of photoactivated sealant. All curing were performed with various light curing units under the application of oxygen gel barrier, stream of nitrogen and carbon dioxide gas for inhibition of oxygen diffusion into sealant surface.

Ⅱ. Materials and methods

Materials

The visible light-curing pit and fissure sealant (Ultraseal XT plus™, Ultradent, USA) was used in this study. This material was based on a bisphenol glycidyl methacrylate (BisGMA), triethylene glycol dimethacrylate resin matrix, camphoroquinone as photoinitiator, and 58% (v/v) inorganic filler.

Light-curing units

The quart tungsten halogen (QTH) unit (XL3000™, 3M ESPE, USA), the plasma-arc curing (PAC) unit (Flipo™, LOKKI, France) and the second generation light emitted diode(LED) units (Elipar FreeLight II™, 3M/ESPE, Germany) were used for polymerization of pit and fissure sealant in standard mode. Light intensities, given by the light unit manufacturers, energy densities(light intensity x curing time) and curing times, recommended by the restorative manufacturer, are listed in Table 1.

Specimens preparation

The Teflon mold (PELCO, Microwell Staining Mold, TedPella Inc, USA) with the diameter of 4.8 mm and the depth of 1.8 mm was carefully filled with the pit and fissure sealant, attempting to avoid air bubbles entrapment. The specimens were then cured with three different light sources under room-air atmosphere (Control) and also assembled in the room-air atmosphere while under the stream of nitrogen (Air/N_2) and the carbon dioxide (Air/CO_2). Stream of N_2 and CO_2 has been blown at 1 MPa on the distance of 1 cm from the sealant surface.

An oxygen gel barrier (DeOx®, Ultradent, USA) was placed on the surface of sealant to eliminate the formation of an oxygen inhibited layer and then, the sealants was cured with three different light sources for different curing time.

Table 1. The light-curing units and irradiation conditions used in this study

Light-curing unit	Light source	Irradiation time (sec)	Light intensity (㎽/㎠)	Energy density (J/㎠)
XL 3000™	QTH	10	450	4500
		20		9000
		40		18000
Flipo™	PAC	3	1900	5700
		5		9500
		10		19000

		5		4000
Elipar-Freelight 2™	LED	10	800	8000
		20		16000

Detection of residual TEGDMA on high performance liquid chromatography(HPLC)

A cured specimen of each condition was pulverized into granules, and the residual monomers were extracted with 1 ml of PBS at 37 ℃ for 24 h. The eluted amount of TEGDMA was determined by HPLC (LC-10AD, SHIMADZU Co., Kyoto, Japan) using the standard curve established from known concentrations of monomer (Fig. 1). The experimental condition for HPLC are shown in Table 2. The values of eluted monomer were calculated as an average of independent experiments performed in triplicate.

Table 2. Experimental conditions for HPLC

Column	C_{18} reverse phase column
Mobile phase	A: Water with 0.1% TFA B: Acetonitrile with 0.1% TFA Gradient : 20% B to 60% B over 25 min
Flow rate	1 ㎖/min
Detector	UV 205㎚

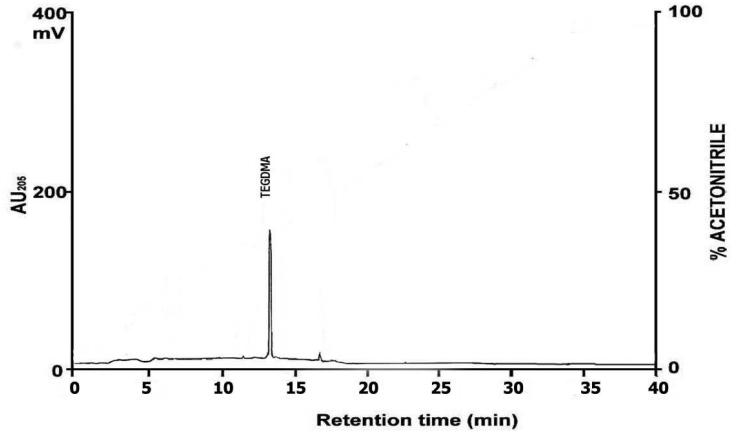

Fig. 1. Chromatogram of the standard mixture used for identification of the different substance contained in the composite.

Measurement of microhardness

Ten specimens were cured with desired curing time under treated atmosphere. The upper surface of these were measured with a Vicker's hardness-measuring instrument (HM-112, AKASHI CO., JAPAN). The indenter point was kept on the surface for 10 sec with 50 g load.

Measurement of thickness on oxygen inhibited layer (OIL)

The method for measuring the thickness of OIL was similar to that of Ruyter(1981) with little modification. A small drop of pit and fissure sealant was placed on a clean glass microscope slide. A coverslip was laid over this drop, and the fluid was permitted to flow under the weight of the coverslip. The pit and fissure sealant was then cured with a halogen light curing unit for 40 sec, with plasma arc light curing unit for 10 sec, and with a second generation LED light curing unit for 20 sec. These specimens were made as mentioned above.

A light microscope was used for measuring of thickness of the OIL at five locations around the periphery of the cured drop. The mean thickness of the oxygen inhibited layer of each drop was then determined, and this value represented the inhibited layer thickness was determined, and this value represented the thickness of OIL for that specimen.

Statistical analysis

The data were analyzed by means of ANOVA and Tukey post hoc test. The values of $p<0.05$ were considered statistically significant.

Ⅲ. Results

Amount of residual monomer (TEGDMA)

The eluted TEGDMA was decreased by increasing curing time in all experimental conditions. The eluted TEGDMA form the specimens cured with QTH light under the Air/ N_2 and Air/CO_2 gas and application of Oxygen gel barrier (DeOx®) were significantly lower than that cured under the room-air atmosphere(Control). The amount of eluted TEGDMA from the specimens cured with QTH light for 40 sec in Air/CO_2 conditions showed the most lowest, but there was no significant difference with DeOx® treated group (Fig. 2).

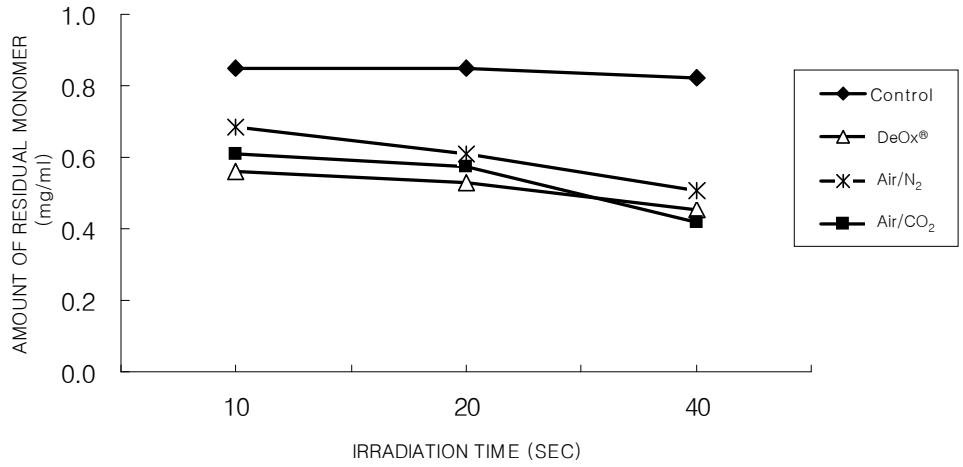

Fig. 2. Amount of TEGDMA released from specimens cured with QTH light.

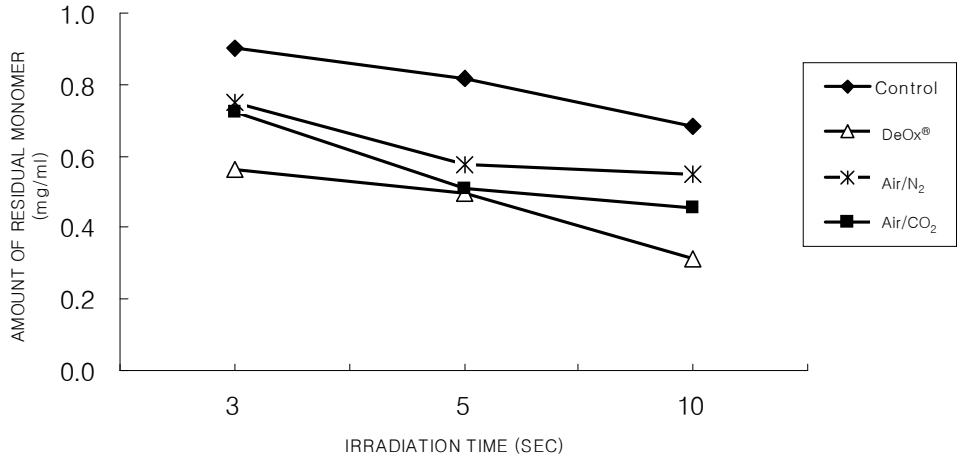

Fig. 3. Amount of TEGDMA released from specimens cured with PAC light.

In the room-air atmosphere the specimens cured with PAC light for 10 sec were lower than that cured for 5 sec ($p<0.05$). As shown to Fig. 3., when the specimens were cured with PAC light for 5 or 10 sec in stream of N_2 and CO_2 gas, the eluted TEGDMA was lower than that cured under room-air atmosphere($p<0.05$). In the Air/N_2 and Air/CO_2 atmospheric conditions, there was no statistically significant released TEGDMA between the curing for 5 sec and 10 sec. In the DeOx® application, the specimen cured with PAC light for 10 seconds was less than that cured in the Air/N_2 and Air/CO_2 atmospheric conditions (Fig. 3).

As shown Fig. 4, the results corresponding to 10 sec and 20 sec were similar ($p>0.05$) in LED light under room-air atmosphere. In the LED using

10 or 20 sec irradiation times under the stream of N_2 and CO_2, the unreacted TEGDMA showed no significant difference ($p>0.05$), whereas, the specimens applied with DeOx® showed the lowest release of TEGDMA in the all experimented group ($p<0.05$).

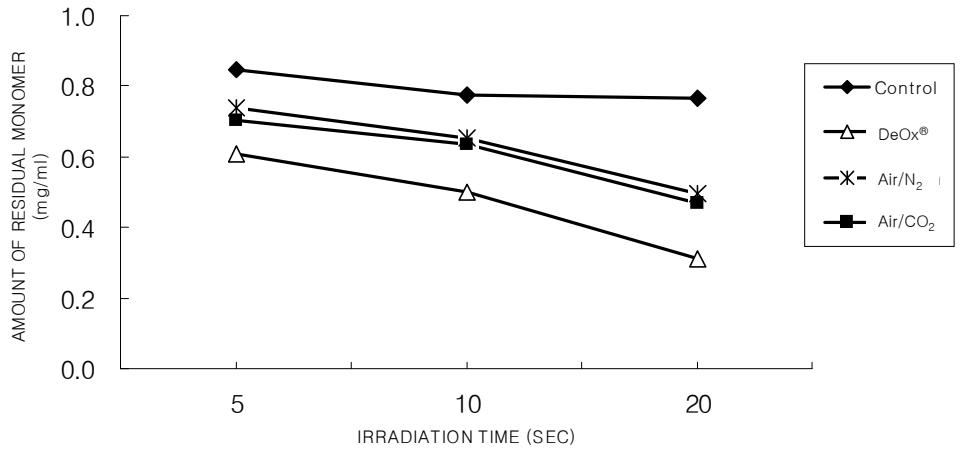

Fig. 4. Amount of TEGDMA released from specimens cured with LED curing light.

Table 3. Statistical analysis of quantities of TEGDMA released from the curing time with different light sources under various condition

Intensity energy	Light source	Curing time	Methods of oxygen blocking			
			Control (in Air)	DeOx® application	Air/N_2 blow	Air/CO_2 blow
Small	QTH	10	$0.85\pm0.023^{a\alpha}$	$0.56\pm0.188^{ab\beta}$	$0.69\pm0.176^{ab\alpha\beta}$	$0.61\pm0.075^{abc\alpha\beta}$
	PAC	3	$0.90\pm0.024^{b\alpha}$	$0.56\pm0.103^{ab\beta}$	$0.75\pm0.042^{a\gamma}$	$0.72\pm0.007^{a\gamma}$
	LED	5	$0.85\pm0.023^{a\alpha}$	$0.61\pm0.012^{a\beta}$	$0.74\pm0.005^{a\gamma}$	$0.70\pm0.061^{ab\gamma}$
Medium	QTH	20	$0.85\pm0.003^{a\alpha}$	$0.53\pm0.076^{abc\beta}$	$0.61\pm0.075^{abc\beta}$	$0.57\pm0.776^{bcd\beta}$
	PAC	5	$0.82\pm0.023^{ac\alpha}$	$0.50\pm0.014^{bc\beta}$	$0.58\pm0.044^{abc\gamma}$	$0.51\pm0.027^{cde\beta\gamma}$
	LED	10	$0.77\pm0.013^{c\alpha}$	$0.50\pm0.011^{bc\beta}$	$0.65\pm0.006^{abc\gamma}$	$0.63\pm0.038^{abc\gamma}$
Large	QTH	40	$0.82\pm0.007^{c\alpha}$	$0.45\pm0.885^{c\beta\gamma}$	$0.51\pm0.024^{bc\beta}$	$0.42\pm0.040^{e\gamma}$
	PAC	10	$0.68\pm0.001^{d\alpha}$	$0.31\pm0.014^{d\beta}$	$0.55\pm0.040^{bc\gamma}$	$0.45\pm0.043^{de\delta}$
	LED	20	$0.77\pm0.017^{c\alpha}$	$0.31\pm0.021^{d\beta}$	$0.49\pm0.008^{c\gamma}$	$0.47\pm0.020^{de\gamma}$

a,b:different letters show the significant different among groups of different various light sources and intensity

α,β: different letters show the significant different among groups of different tested conditions in each light sources and intensity

Microhardness of top surface

The mean Vickers hardness number (VHN) and the SDs of the groups after polymerization with different light-curing units under appropriated conditions were performed and the results of the one-way ANOVA are shown in Figure 5 and Table 4. With QTH light curing, microhardness of the specimens irradiated for 40 sec in the stream of CO_2 was higher than that in the room-air atmosphere. With PAC light curing for 10 sec, there was no statistical difference among tested groups. With LED light curing, values for 20 sec in the stream of CO_2 and application of DeOx® were higher than that in the room-air atmospere.

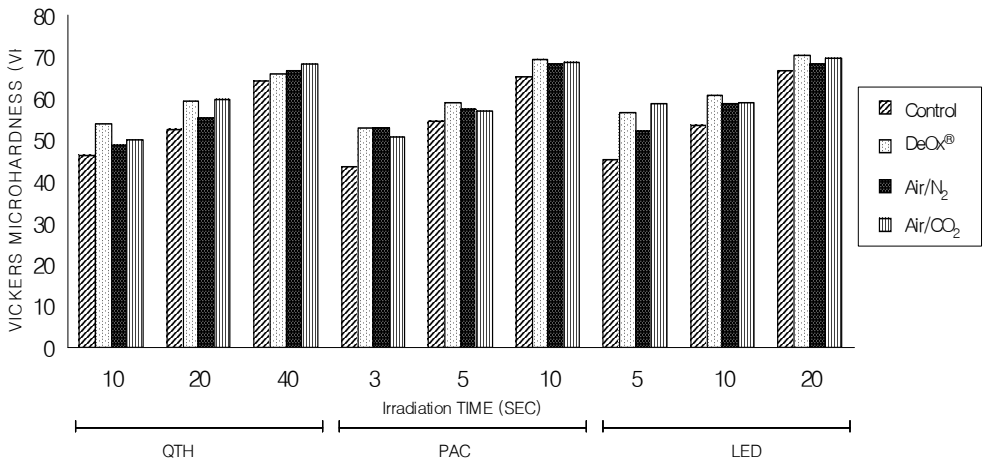

Fig. 5. The mean microhardness of the specimens cured by different type of light-curing units with different times.

Table 4. Statistical analysis of microhardness from the curing time with different light sources under various condition

Intensity energy	Light sources	Curing time	Methods of oxygen blocking			
			Control (in Air)	DeOx® application	Air/N_2 blow	Air/CO_2 blow
Small	QTH	10	46.2±1.66$^{a\alpha}$	53.7±1.70$^{a\beta}$	48.6±1.55$^{a\gamma}$	49.9±1.71$^{a\gamma}$
	PAC	3	43.4±1.64$^{a\alpha}$	52.8±2.00$^{a\beta}$	52.9±1.81$^{bc\beta}$	50.8±1.24$^{a\gamma}$
	LED	5	45.1±2.83$^{a\alpha}$	56.4±2.31$^{b\beta}$	51.9±2.16$^{b\gamma}$	58.5±1.03$^{bc\beta}$
Medium	QTH	20	52.3±1.57^{ba}	59.3±0.84$^{c\beta}$	55.3±1.96$^{cd\gamma}$	59.5±1.27$^{c\beta}$
	PAC	5	54.5±1.40^{ba}	59.1±1.20$^{c\beta}$	57.1±1.79$^{de\beta}$	56.9±2.32$^{b\gamma}$
	LED	10	53.3±3.10^{ba}	60.6±0.83$^{c\beta}$	58.6±1.57$^{de\beta}$	58.8±0.95$^{bc\beta}$
Large	QTH	40	64.3±2.43$^{c\alpha}$	66.0±1.74$^{d\alpha\beta}$	66.7±1.65$^{f\alpha\beta}$	68.2±2.91$^{d\beta}$
	PAC	10	65.2±1.63$^{c\alpha}$	69.2±1.71$^{e\beta}$	68.4±2.10$^{f\beta}$	68.6±1.14$^{d\beta}$
	LED	20	66.6±1.38$^{c\alpha}$	70.2±1.37$^{e\beta}$	68.3±1.59$^{f\gamma}$	69.5±0.89$^{d\beta\gamma}$

a,b:different letters show the significant different among groups of different various light sources and intensity

α,β: different letters show the significant different among groups of different tested conditions in each light sources and intensity

The microhardnesses of the specimens cured for 40 sec with QTH light, 10 sec with PAC light and 20 sec with LED curing light under each atmosphere conditions were similar and no statistical difference. On the other hands, in the DeOx® treated group, the curing for 40 sec with QTH light was lower than that cured for 10 sec with PAC light and 20 sec with LED curing light ($P<0.05$).

Thickness of oxygen inhibited layer (OIL)

The light microscopy was used to evaluate the thickness of OIL on photo-activated sealant, (Fig 6-8). Table 5 shows the values of the thickness of OIL with different light sources in appropriated condition.

The mean thickness of OIL in room-air atmosphere was approximately 2 times higher than that of cured in treated groups ($p<0.05$). As comparing to each surface treated method, the values have no statistically difference ($p>0.05$).

When the specimens cured by LED light for 20 sec in room-air atmosphere, the thickness of OIL was significantly less than that cured by QTH light for 40 sec and PAC light for 10 sec in the same condition. On the

other hands, in the curing under the stream of CO_2 and the surface treatment of Deox®, the thickness of OIL with specimens cured by LED light for 20 sec and PAC light for 10 sec was significantly less than that cured by QTH light for 40 seconds.

The specimens cured by three light sources under the stream of N_2 were showed to be no statistically significant difference ($p>0.05$).

Table 5. Statistical analysis of thickness of oxygen inhibited layer(㎛) from the curing time with different light sources under various condition

Light sources	Curing Time	Methods of oxygen blocking			
		Control (in Air)	DeOx® application	Air/N_2 blow	Air/CO_2 blow
QTH	**40**	**21.2±1.6$^{a\alpha}$**	**10.3 ±0.4$^{a\beta}$**	**10.2 ±1.0$^{a\beta}$**	**9.9 ±1.1$^{a\beta}$**
PAC	**10**	**20.0±1.0$^{a\alpha}$**	**8.4 ±0.5$^{b\beta}$**	**9.2 ±1.1$^{a\beta}$**	**8.8 ±0.9$^{b\beta}$**
LED	**20**	**15.4±1.4$^{b\alpha}$**	**8.2 ±0.2$^{b\beta}$**	**9.2 ±0.9$^{a\gamma}$**	**8.6 ±0.3$^{b\beta\gamma}$**

a,b:different letters show the significant different among groups of different various light sources and intensity

α,β: different letters show the significant different among groups of different tested conditions in each light sources and intensity

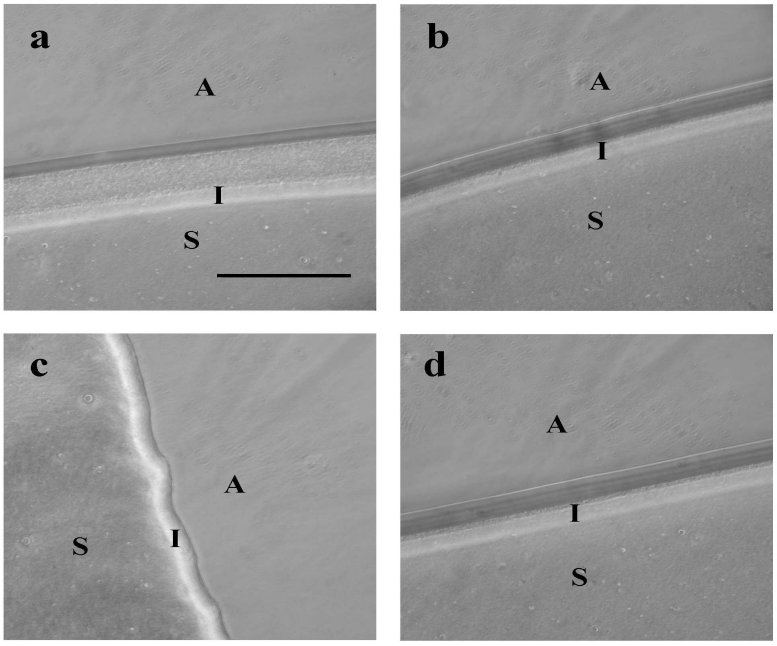

Fig. 6. Micrographs of oxygen inhibited layer on sealant cured with QTH light (a) in air atmosphere, (b) the stream of CO_2, (c) application of DeOx® and (d) the stream of N_2. Original magnification 400x, bur = 100㎛, S: sealant, I: oxygen inhibited layer, A: Air

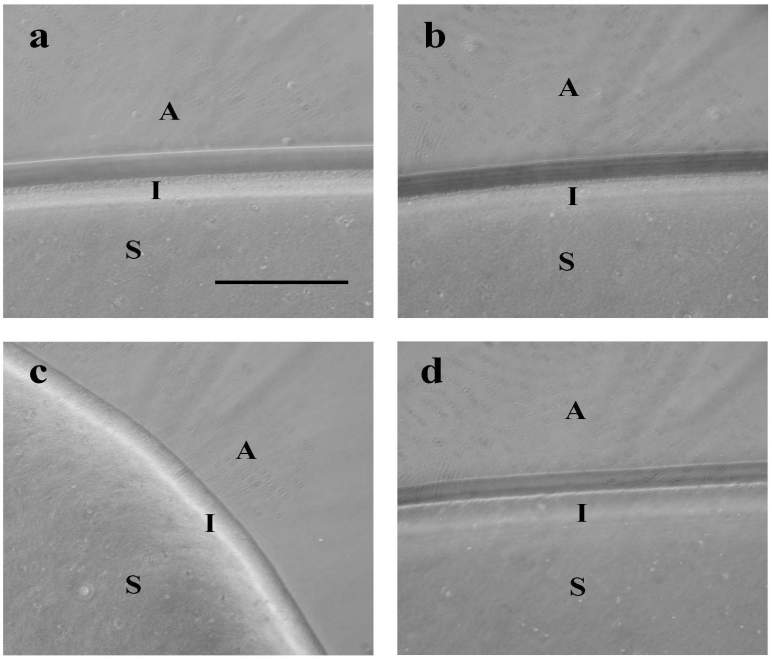

Fig. 7. Micrographs of oxygen inhibited layer on sealant cured with PAC light (a) in air atmosphere, (b) the stream of CO_2, (c) application of DeOx® and (d) the stream of N_2. Original magnification 400x, bur = 100㎛, S: sealant, I: oxygen inhibited layer, A: Air

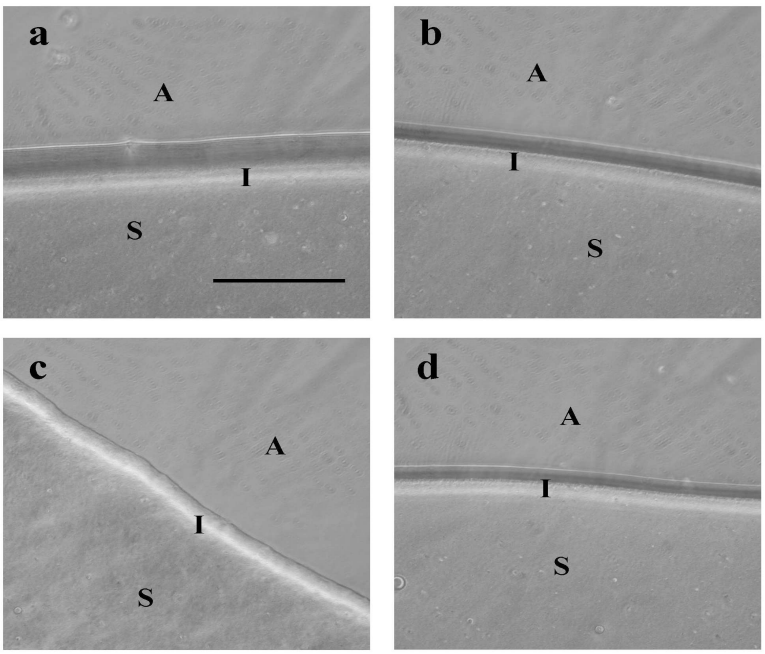

Fig. 8. Micrographs of oxygen inhibited layer on sealant cured with LED light (a) in air atmosphere, (b) the stream of CO_2, (c) application of DeOx® and (d) the stream of N_2. Original magnification 400x, bur = 100㎛, S: sealant, I: oxygen inhibited layer, A: Air

Ⅳ. Discussion

Oxygen is know to inhibit the polymerization of the surface layer of composite resins[5]. The reason is the ability of oxygen, compared with that of a monomer molecule, to react with growing radical[6,7]. It was found that the effects of oxygen were responsible for the formation of an inhibited layer on the surface of the resin in contact with room air. This layer is commonly referred to an oxygen inhibited layer due to its origin, and it is primarily composed of unreacted monomers and oligomers. The thickness of this layer is highly dependent on resin viscosity: the less viscous the resin, the greater the potential for oxygen diffusion and the thicker the unpolymerized layer[5].

The presence of this uncured monomer has demonstrated cytotoxic reactions in tissue culture[13] and allergic response[20,21]. Especially, Gercina and Hume[22] reported TEGDMA as a major cytotoxic component eluting from uncured resin composite. The TEGDMA has been identified as the main compound release from polymerized resin composites into aqueous media and wetting environment such as the intra oral cavity. It is related to the TEGDMA's relative hydrophilicity in comparison with the more hydrophobic Bis-GMA and Bis-DMA. Hence, we determined the respective amount of residual TEGDMA in the cured specimens by quantitative HPLC. The extraction and determination of unreacted free monomers using HPLC is the only convenient method to measure the residual amount of each monomer separately, and we consider that it gives useful information.

Fig. 2-4 show the amount of TEGDMA release from specimens cured by three light sources; QTH light, PAC light and second generation LED light units. For the specimens cured for 40 seconds with QTH light in Air/CO_2 conditions, the eluted amount of TEGDMA was lower than that the specimens cured in Air/N_2 condition($p<0.05$), and was similar results corresponding with the DeOx® treated group($p>0.05$). In the specimens cured with PAC light for 10 seconds under the Air/CO_2 atmospheric condition, the amount of eluted TEGDMA was lower than that cured under the Air/N_2 atmospheric condition ($p<0.05$). Whereas, when the specimens were photopolymerized with the LED curing light using 10 or 20 seconds irradiation times under the stream of N_2 and CO_2, the amount of unreacted TEGDMA showed to be no statistically significant difference ($p>0.05$). The specimens included application of DeOx® showed the lowest release of eluted TEGDMA among all groups ($p<0.05$).

In previous studies, oxygen impermeable barriers (Mylar strip, glass slide) have been used to block the resin/atmosphere interface due to eliminate oxygen inhibition[23,24]. Recently, the oxygen barrier gel (DeOx®) were introduced to the dental clinic and it was place on restorative materials to eliminate the formation of an oxygen inhibited layer[25]. It was composed the glycerine and polyethylene glycol. In this study, surface treatment with DeOx® was more effective than other treated groups. However, application of DeOx® will extend step of treatment and spreading of this on teeth surface is difficult to clinical application due to mobile character of

material. Therefore, this paper propose that the stream of N_2 and CO_2 gas may be beneficial effect in clinical application due to similar results in reduction of oxygen inhibited layer.

Rueggeberg and Margeson[19] reported that the degree of conversion was the highest when the resin was cured in an argon atmosphere. When the composite resin was cured under air and nitrogen atmosphere, exhibited no significant difference. However there are few reports on the effect of reducing the oxygen inhibited layer by CO_2. Therefore, in this research, to reduce the oxygen inhibited layer, the curing methods of sealant under a continuous stream of CO_2 and N_2 approached because this methods supplies can be available in dental clinics.

In this study, it is shows that the stream of CO_2 is more effective in the reduction of unreacted TEGDMA and the increase of surface microhardness than that of N_2 and air atmospheric conditions. The reason might that CO_2 is heavier than air and can be easily maintained in a surface of resin , without much loss. The beneficial effects are that CO_2 is more available and has more low cost than nitrogen. Therefore, the visible photo-activated light curing under the stream of CO_2 atmosphere will have its greatest potential for inhibition of oxygen diffusion to reduced the formation of oxygen inhibited layer.

In comparison of three light units, the release of TEGDMA in curing with PAC light for 10 sec under air atmosphere was lower than that with QTH light for 40 sec and with LED light for 20 sec. These with QTH for 40

sec and with LED light for 20 sec were showed to be no statistically significant difference. When the DeOx® was applied, these for 10 sec with PAC light and 20 sec with LED light was significantly lower than that cured for 40 sec with QTH light. When the N_2 and CO_2 was blow, the specimens cured for 10 seconds with PAC light, for 20 seconds with LED light and for 40 seconds with QTH light were showed to be no statistically significant difference. These study consistent with previous results that the high power curing light, PAC light and second-generation LED light could be obtained the optimal polymerization of resin restoration[26-29]. In addition to, the surface treatment by DeOx®, N_2 or CO_2 atmospheric conditions could perform the reduction of difference about the amount of eluted TEGDMA among all light units.

As comparing to the surface microhardness values of the specimens cured with three lights under each treated conditions, that cured with all three different light sources in application of DeOx® and the stream of N_2 and CO_2 are higher than that cured in the room-air atmosphere. The thickness of oxygen inhibited layer on photoactivated sealant was readily detected by light microscopy[5,30,31]. The surface treatment by DeOx®, N_2 and CO_2 reduces the thickness of oxygen inhibited layer by approximately 49% of the untreated control value. The microhardness of the specimens cured for 40 seconds with QTH light in application of DeOx® and the stream of CO2 was higher than that cured in the room-air atmosphere. In the same condition mentioned as above, the thickness of oxygen inhibited layer was lower than that cured in

the room-air atmosphere. With PAC light curing for 10 sec, the microhardness and the thickness of the oxygen inhibited layer were no statistical difference among of tested groups. With LED light curing, microhardenss of the specimens irradiated for 20 sec in application of DeOx® and the stream of N_2 and CO_2 was significantly higher than that under the room-air atmosphere. The thickness of oxygen inhibited layer in all treated conditions was significantly lower than that in the room-air atmosphere. We indicated that the negative correlation between the microhardness and thickness of oxygen inhibited layer was demonstrated. This indicate that the reduction of oxygen inhibited layer contributes directly to increase microhrdness of surface.

This study will give help to develop clinical approach in the reduction of the the oxygen inhibited layer and the increase of microhardness, but the pressure and amount of N_2 and CO_2 gas will be adjusted in clinical application.

Ⅴ. Conclusion

The aim of this study was to investigate the effect of oxygne inhibition on the polymerization of sealant cured with different light-curing units. The amount of TEGDMA of each specimens were analyzed by HPLC and the surface microhardness was measured by Vicker's hardness tester Data were analyzed by means of ANOVA.

The result of present study can be summarized as follows:

1. The amount of eluted TEGDMA form the specimens cured with all the tree different light units in the stream of N_2 and CO_2 gas and application of Oxygen gel barrier (DeOx®) were significantly lower than in the room-air atmosphere(Control)($p<0.05$).
2. The amount of eluted TEGDMA form specimens cured with QTH light for 40 seconds in Air/CO_2 conditions was most lowest, but there was no statistically significant difference comparing to DeOx® treated group.
3. In the DeOx® application, the amount of eluted TEGDMA the specimen cured with PAC light for 10 seconds was less than that cured in the stream of N_2 and CO_2 atmospheric conditions($p<0.05$).
4. In the LED using 10 or 20 sec irradiation times under the stream of N_2 and CO_2, the unreacted TEGDMA showed to be no statistically significant difference ($p>0.05$), whereas, the specimens applied with DeOx® showed the lowest release of TEGDMA in the all test group

($p<0.05$).

5. With QTH light curing, microhardness of the specimens irradiated for 40 sec in the stream of CO_2 was higher than that in the room-air atmosphere ($p<0.05$). Otherwise, with PAC light curing for 10 sec, there was no statistical difference among tested groups.
6. With LED light curing, values for 20 sec in the stream of CO_2 and application of DeOx® were higher than that in the room-air atmospere ($p<0.05$).
7. The microhardnesses of the specimens cured for 40 sec with QTH light, 10 sec with PAC light and 20 sec with LED curing light under each atmosphere conditions were similar and no statistical difference.
8. In the DeOx® treated group, the microhardness of the specimens cured with QTH for 40 sec light was lower than that cured for 10 sec with PAC light and 20 sec with LED curing light ($P<0.05$).
9. The surface treatment by DeOx®, N_2 and CO_2 reduces the thickness of oxygen inhibited layer by approximately 49% of the untreated control value.

On the basis of the results, all curing were performed with various light curing units under the application of oxygen gel barrier, stream of nitrogen and carbon dioxide gas for inhibition of oxygen diffusion. However, application of DeOx® will extend step of treatment and spreading of this on teeth surface is difficult to clinical application due to mobile

character of material. Therefore, this research will give help to develop clinical approach in the reduction of the oxygen inhibited layer and the increase of microhardness, but the pressure and amount of N_2 and CO_2 gas will be adjusted in clinical application.

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