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Original Article

Calcium Phosphates as Fissure Sealant Materials Fused to Enamel by CO_2 Laser. I. Monocalcium Phosphate Monohydrate and Dicalcium Phosphate Dihydrate as Fissure Sealant Materials

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Monocalcium phosphate monohydrate (MCPM) and dicalcium phosphate dihydrate (DCPD) were evaluated as pit and fissure sealants, which melt with low energy density laser irradiation and obsturate pits and fissures of enamel surfaces without accompanying surface destruction. The melting points of MCPM and DCPD, estimated by differential thermal analysis, are 971℃ and 1348℃, respectively; thus the melting point of MCPM was lower than that of DCPD. When heated, MCPM decomposed to γ-metacalcium phosphate, β-metacalcium phosphate and δ-metacalcium phosphate. On the other hand, DCPD changed to DCPA, γ-calcium pyrophosphate, β-calcium pyrophosphate and α-calcium pyrophosphate. Solubility in 200 mM acetic acid buffer was higher with heated MCPM than with heated DCPD samples. This leads us to conclude that DCPD would be more effective as a sealant material than MCPM. Comparison of lased disks made of MCPM and DCPD at various energy densities showed that with DCPD disks, fewer craters were formed, and no craters were formed at an energy density of 45.6 J/cm^2. All these findings suggest that DCPD would have advantages over MCPM as a sealant that can be fused at a lower energy density with no damage on the enamel surface.

Key words: CO_2 laser, Calcium phosphate, Fissure sealants

INTRODUCTION

The risk of caries on occlusal surfaces of molars just after eruption is very high, due to long-term coverage with gingiva and the time necessary to reach the occlusal plane after eruption, and the bottom of pits and fissures is hard to clean due to their complex shape. Therefore, the caries risk is higher within 2 years of permanent molar tooth eruption, and 2 or 3 years after eruption of post-natal molars.

In pedodontics, pit and fissure sealants function as a preventative measure against caries. As pit and fissure sealants, resin materials such as Bis-GMA or fluoride sustained-release polymer, and cement sealants such as glassionomer cement, resin-additional glassionomer cement or polycarboxylate cement[1)] are employed. The aim of fissure sealants is to physically seal the complex pits and fissures. For both resin and cement sealants, numerous studies[1~6)] have reported their efficacy against caries, but also demonstrated that there are many cases in which resealing is needed due to the removal of materials or fracture of material edges.

Another preservation method uses irradiation from a dental laser to prevent dental caries and at the same time to provide acid resistance to enamel[6~8)]. For example, Sognnaes et al.[9)] and Stern et al.[10)] each reported the enhancement of acid resistance by irradiating enamel surface with a dental laser. Walsh et al.[11)] also studied the possibility of melting and blocking pits and fissures with a CO_2 laser. Practically, however, it is very difficult to evenly irradiate the pit and fissure region of occlusal surfaces because of their complexity. In addition, if the level of laser irradiation is not lower than a certain threshold,[12,13)] there is the possibility of voids being formed due to evaporation of enamel, cracking, and heat damage to dental pulp. In vivo studies by Brugnera et al.[14)] have thus suggested that CO_2 laser irradiation of enamel alone is ineffective.

Of the many studies investigating the fusion of various calcium phosphates[15,16)], bioglass (Na_2O-CaO-SiO_2-P_2O_5)[17,18)], calcium phosphate-glass, or low-melting porcelain[19)] to enamel using CO_2 or Nd-YAG lasers, Stewart et al.[15)] tried to fuse fluoride-containing hydroxyapatite to enamel using a CO_2 laser and suggested that fusion of calcium phosphate by laser irradiation at less than 24 J/cm^2 will not allow the approach of ink into the junction between enamel and fused materials. Shimizu et al.[16)] fused α-tricalcium phosphate to enamel with a Nd-YAG laser and investigated the thermal transformation of fused materials and enamel by micro X-ray analysis. The Nd-YAG laser, however, is not suitable for pit and fissure sealing, because the laser wave needs black carbon ink or something black to effectively melt and fuse α-tricalcium phosphate to enamel, resulting in colored pits and fissures that are less esthetically pleasing. On the other hand, Lin et al.[17,18)] attempted to fuse bioglass to enamel and Nihei et al.[19)] investigated calcium phosphate-glass and low-melting porcelain. However, these studies only showed the pos-

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sibility of fusing the materials to enamel with laser irradiation and did not investigate the condition of the fused materials, sealing method or laser irradiation conditions for pit and fissure caries prevention.

In this study, we used commercial monocalcium phosphate monohydrate and dicalcium phosphate dihydrate as pit and fissure sealants, as these materials melt with low energy density laser irradiation and obsturate pits and fissures of enamel surfaces without accompanying surface destruction.

MATERIALS AND METHODS

Monocalcium phosphate monohydrate ($Ca(H_2PO_4)_2$: MCPM) (SIGMA-ALDRICH Co., Ltd. St. Louis, MO, USA) and dicalcium phosphate dihydrate monohydrate ($CaHPO_4$: DCPD) (NAKARAI-TESC Co., Ltd., Kyoto, Japan) were used as laser-fused sealants without further purification.

Thermal transformation of MCPM and DCPD

Thermal transformation of MCPM and DCPD was investigated by thermogravimetric analysis (TG) and differential thermal analysis (DTA) from room temperature to 1500°C at a heating rate of 10°C/min (Thermo Plus TG 8120 RIGAKU Electronics Co., Ltd., Tokyo, Japan). To further investigate the thermal transformation of MCPM and DCPD at a specified temperature, MCPM was heated at 110, 180, 300, 500, 1000 and 1500°C and DCPD was heated at 110, 160, 250, 450, 1200 and 1500°C in a platinum melting pot for 1 h, followed by cooling to room temperature. Heated samples were examined by X-ray diffraction (RINT 2000, RIGAKU Electronics Co., Ltd., Tokyo, Japan) operating at a scanning speed of 2.0 θ/min, and at 56 kV and 200 mA.

Solubility of heated MCPM and DCPD disks in acetic acid buffer

In order to mimic the acid resistance of lased MCPM and DCPD in the oral cavity, MCPM and DCPD samples that were heated at 1500°C for 1 h and then quenched to room temperature were placed in acetic acid-acetic sodium buffer (5 g in 50 m*l*, 200 mM, pH 5.0). Aliquots of 0.1 m*l* were withdrawn at 1, 3, 5, 8, 12 and 24 h, and were then filtered through 0.2-μm Millipore filters for calcium measurement. Calcium concentrations were determined by ion-chromatography (ION Chromatograph HIC-6 A Shimazu Co, Ltd., Kyoto, Japan) using 4 mM tartaric acid and 2 mM ethyrendiamin solution (3.0 *l*, 40°C) as a mobile phase.

Disks of MCPM and DCPD for laser irradiation

MCPM or DCPD powders were placed in a cylindric die and one-way pressed (50 MPa) to make disks (diameter: 12 mm; thickness: 1 mm; density, MCPM: 1.83 g/cm^3; density, DCPD: 1.83 g/cm^3). We used a CO_2 laser (NANO LASER GL-1, GC Co., Ltd.) and F 120 hand piece for laser irradiation. The distance between the hand piece and tablet was 10 mm (just in focus) and irradiation power was fixed at 1.0 W. Irradiation time was 0.1, 0.2, 0.4 or 0.8 s. After laser irradiation, the shape and size of laser-irradiated spots on the disks were examined using a light microscope (VHX Digital microscope Keyence CO., Ltd.). Disks were then mounted in plastic (B.P.S. set Q Kyoto Chemical Co., Ltd.), and after 24 h at room temperature, each specimen was cut with a low-speed diamond saw (ISOMET, Buehler Co., Ltd, Lake Bluff, IL, USA) cooled with water. The section obtained was further ground with emery paper No. 800 and 2000 to a thickness of about 20 μm. The cut plane of each section was investigated with a deflected light microscope (BH-2, Olympus Co., Ltd.) and the depth of the laser irradiation spot was measured.

RESULTS

Figure 1 shows the TG and DTA curves for MCPM, and Fig.2 shows the corresponding curves for DCPD. As the DTA curves

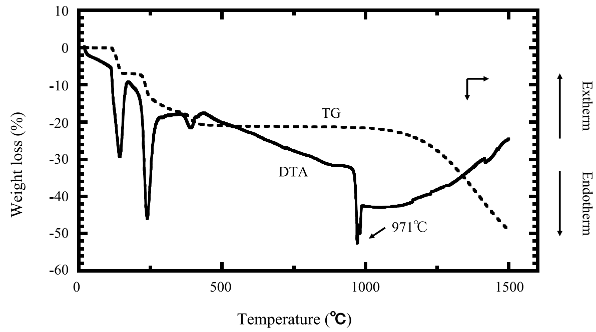

Fig. 1 Thermogravimetric and differential thermal analyses of MCPM

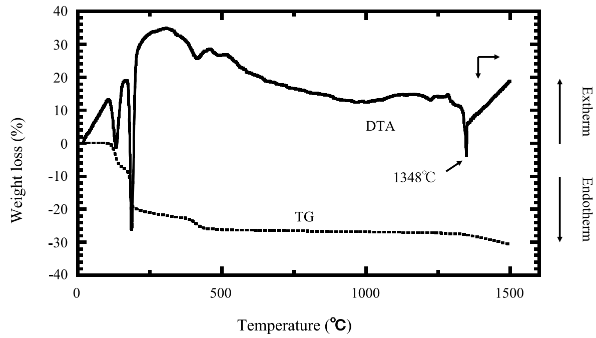

Fig. 2 Thermogravimetric and differential thermal analyses of DCPD

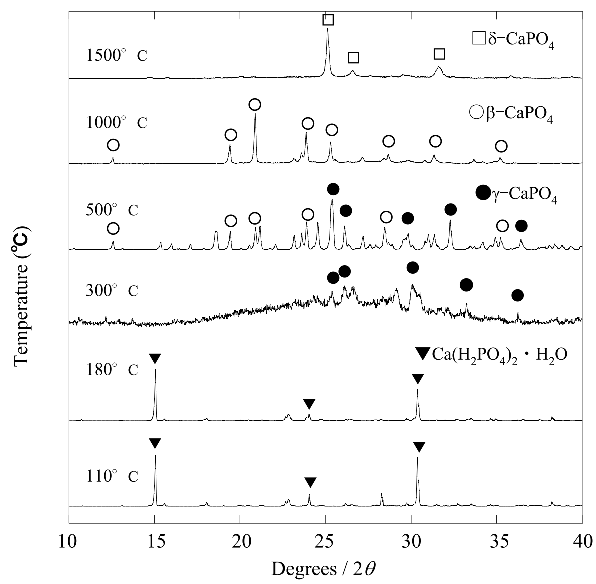

Fig. 3 X-ray diffraction patterns of MCPM heated at various temperatures

show, the melting points of MCPM and DCPD are 971℃ and 1348℃, respectively, indicating that the melting point of MCPM is lower than that of DCPD.

Figures 3 and 4 show the X-ray diffraction patterns of MCPM and DCPD after heating from room temperature up to 1500℃, and being held for 1 h at the specified temperature. MCPM changed to γ-metacalcium phosphate, β-metacalcium phosphate and δ-metacalcium phosphate with heating. On the other hand, DCPD changed to DCPA, γ-calcium pyrophosphate, β-calcium pyrophosphate and α-calcium pyrophosphate.

Figure 5 shows the dissolution behavior of MCPM and DCPD, which were heated at 1500℃ and then either quenched to or slowly cooled down to room temperature, by plotting calcium concentrations in 200 mM acetic buffer as a function of time. As compared with heated DCPD, heated MCPM dissolved more and quenched MCPM specimens dissolved the most.

Figures 6 and 7 show photomicrographs of CO_2-lased disks of MCPM and DCPD, respectively. As the energy density increased, lased lesions become larger, and with lased MCPM at an energy density of 365 J/cm^2, a crater formed at the center of a circular spot, while no craters were formed in DCPD at the same energy density.

Figure 8 shows a polarized light micrograph of a cross section of a DCPD disk that was CO^2-lased at an energy density of 91.2 J/cm^2. As the figure shows, the boundary between the lased and unlased areas is distinct, making it possible to estimate the penetration depth of laser beams.

Figures 9 and 10 show the diameter and depth of lased specimens, respectively. As Fig.9 shows, no crater was formed in DCPD at an energy density of 45.6 J/cm^2, although craters became evident as energy density increased. In contrast to DCPD, MCPM showed craters, even at the lowest energy density employed in the present study. With regard to the diameter of lased

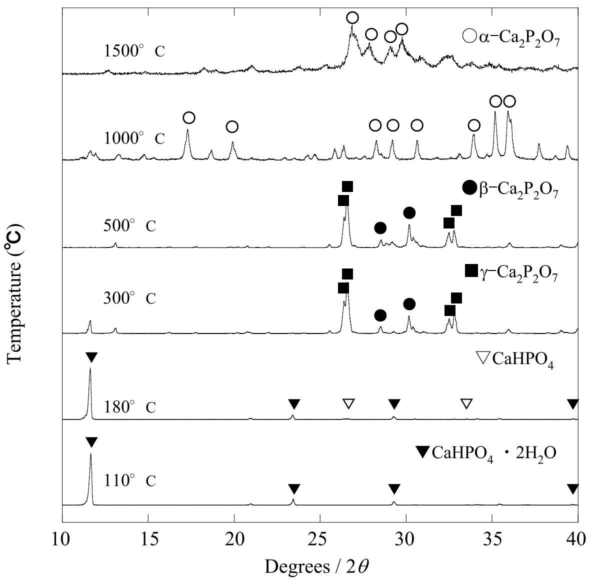

Fig. 4 X-ray diffraction patterns of DCPD heated at various temperatures

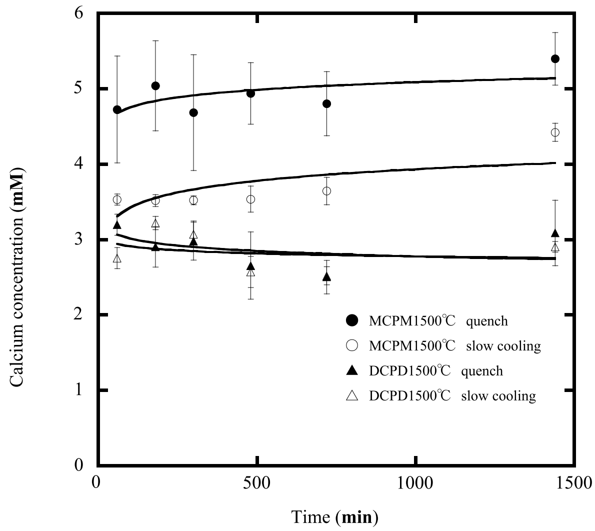

Fig. 5 Time dependence of concentration of Ca^{2+} eluted from heated MCPM and DCPD in 200 mM acetic acid buffer solutions
For both MCPM and DCPD samples, two cooling conditions were employed, i.e., under one condition samples were slowly cooled down to the room temperature after heating at 1500℃ and under the other condition samples were quenched.

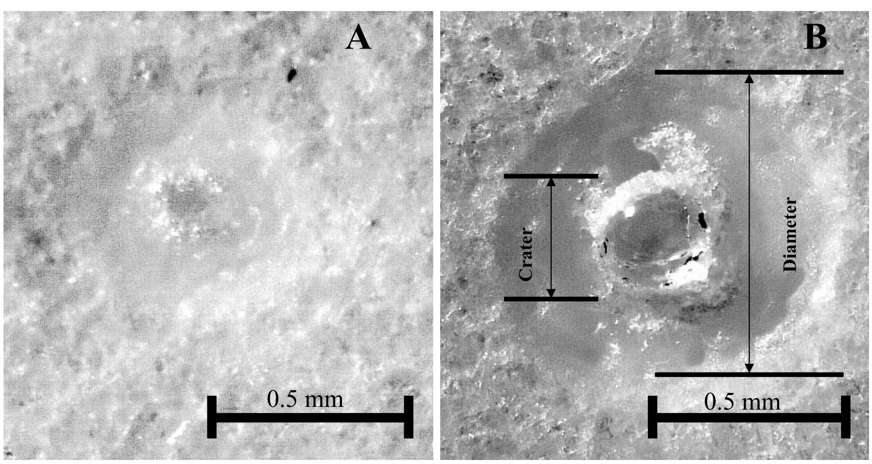

Fig. 6 Photo micrographs of lased spots formed on MCPM disks.
Disks were laser-irradiated at an energy density of 45.6 J/cm^2 (1.0 W for 0.1 s) (A) and at an energy density of 365 J/cm^2 (8.0 W for 0.1 s) (B)

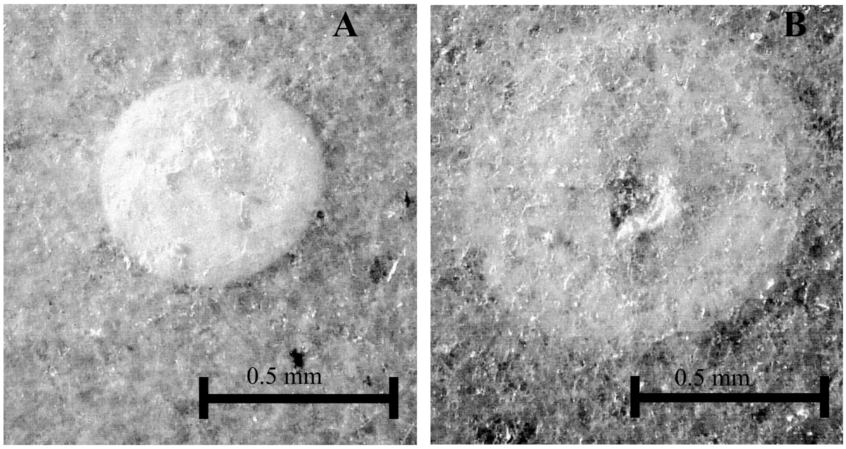

Fig. 7 Photo micrographs of lased spots formed on DCPD disks.
Disks were laser-irradiated at an energy density of 45.6 J/cm^2 (1.0 W for 0.1 s) (A) and at an energy density of 365 J/cm^2 (8.0 W for 0.1 s) (B)

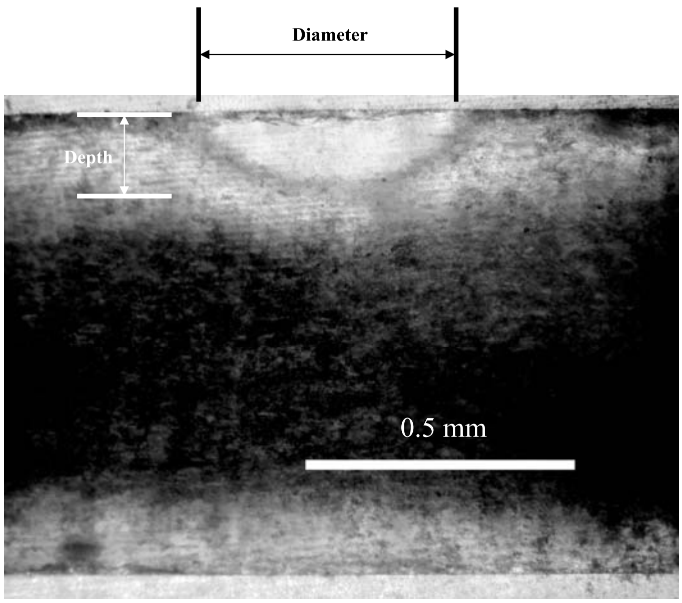

Fig. 8 A polarized light micrograph of a cross section of the spot of a DCPD disk that was CO_2-lased at the energy density of 91.2 J/cm^2 (1.0 W for 0.2 s)

lesions including crater lesions, which increased as the energy density increased, both DCPD and MCPM behaved in essentially the same manner as a function of energy density. The depth of lased lesions, however, strongly depended on the type of calcium phosphate, MCPM or DCPD. As Fig.10 shows, the depth of the lased lesion was significantly greater in MCPM than DCPD.

DISCUSSION

Sealant materials that can be melted and fused using dental lasers must satisfy several requirements. First, sealant materials need to melt with laser radiation at an energy density low enough to cause no damage to the enamel surface. Second, considering the possible elimination of phosphorus due to rapid evaporation by laser irradiation, the Ca/P ratios of sealant materials need to be lower than that of enamel or hydroxyapatite. In the present study, DCPD and MCPM were selected as sealant materials because they satisfy these two requirements, i.e., they melt at moderately low temperatures (971℃ for MCPM and 1348℃ for DCPD) and they have Ca/P molar ratios of 0.5 for MCPM and 1.0 for DCPD, which is smaller than the 1.67 in stoichiometric hydroxyapatite.

The potential applications of various lasers, such as neodymium-doped YAG (Nd: YAG) lasers, erbium-doped YAG (Er: YAG) lasers and carbon dioxide (CO_2) lasers have been demonstrated. Among these, the Nd: YAG laser has the shortest wavelength of 1.064 μm[20] and has high light permeability for enamel and dentin, primarily because the wavelength does not correspond to the absorption bands of the hard and/or soft tissues in enamel and dentine. For efficient adsorption, color mediators, typically substances primarily consisting of carbon, are employed for Nd: YAG lasers. Without color mediators, colored lesions adsorb

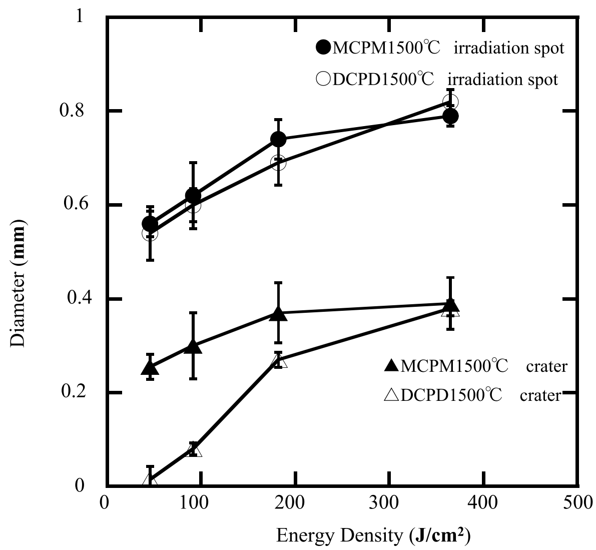

Fig. 9 Diameters of lased spots and craters of MCPM and DCPD disks at different energy densities

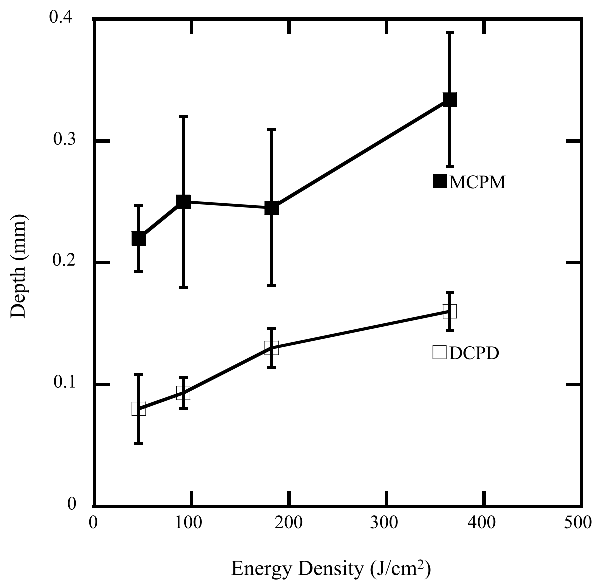

Fig. 10 Depth of lased spots of MCPM and DCPD disks at different energy densities.

Nd: YAG laser more efficiently, which is sometimes known as the carious focal perspiration effect[20~22]. Er: YAG lasers have a wave length of 2.94 μm[23]. The absorption efficiency of water is extremely high, and thus vapor produced by laser energy can result in internal pressure until a microexplosion takes place, ejecting substrate in the form of microsopic particles[24,25]. Thus, the majority of incident energy is consumed in the ablative process and teeth can be cut with this laser[26,27]. On the other hand, the CO_2 laser has the longest wave length, 10.6 μm[28,29] among the three lasers, and this coincides closely with some of the absorption bands of apatite. As the infrared absorption bands of apatite comes from phosphate, hydroxyl and carbonate vibration modes, they also coincide closely with those of MCPM and DCPD. This suggests that with a CO_2 laser, MCPM, DCPD and enamel can be efficiently fused together with minimal heating effects on the organism[28].

The TG and DTA data shown in Figs.1 and 2 clearly suggest that MCPM melts at a lower temperature than DCPD. MCPM decomposed to γ-metacalcium phosphate, β-metacalcium phosphate and δ-metacalcium phosphate upon heating. On the other hand, DCPD changed to DCPA, γ-calcium pyrophosphate, β-calcium pyrophosphate and α-calcium pyrophosphate. Solubility in 200 mM acetic acid buffer was higher for heated MCPM than for heated DCPD samples. This finding can be explained by the fact that methaclaium phosphates are more soluble than calcium pyrophosphates. If the transfer of laser energy to these samples is the same, lased MCPM may be more soluble than lased DCPD. This evidence leads us to conclude that DCPD would be more effective as a sealant material than MCPM.

The laser irradiation spot became lager and deeper as the energy density increased (Figs.9 and 10). No significant differences were observed between DCPD and MCPM in spot size at any energy density. With MCPM, however, craters formed even at the lowest energy density used in this study (Fig.9), while no craters were formed on the surface of DCPD. This finding suggests that DCPD has advantages over MCPM, as it can be fused at a lower energy density with no damage to the enamel surface.

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CONCLUSIONS

Monocalcium phosphate monohydrate (MCMP) and dicalcium phosphate hydrate (DCPD), which both have a calcium/phosphate molar ratio less than that of hydroxyapatite, were investigated as sealant materials. DCPD was found to be more suitable as a pit and fissure sealant, as it melts and is fused using a CO_2 laser at relatively low energy densities. The fact that MCPM melts at a lower temperature than DCPD may suggest that the former calcium phosphate is more suitable as a pit and fissure sealant, as a lower energy density would be needed for fusion. However, dissolution experiments using heated samples of MCMP and DCPD clearly suggested that the heat-induced decomposition of DCPD give less soluble products. Analysis of lased spots also suggested that MCPM formed craters, even at the lowest energy density, thus indicating that damage cannot be averted on the enamel surface, on which the materials would be placed before CO_2 irradiation.

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CO_2レーザー融着による小窩裂溝予防填塞としてのリン酸カルシウム
１．小窩裂溝予防填塞としてのリン酸一カルシウム・一水和物および，リン酸二カルシウム・二水和物

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低エネルギー密度の炭酸ガスレーザー照射で溶融し，エナメル質表面を破壊することなく小窩裂溝を塞ぐことのできる材料として，リン酸一カルシウム・一水和物（MCPM）および，リン酸二カルシウム・二水和物（DCPD）の可能性を検討した．示唆熱分析によりMCPMとDCPDの融点はそれぞれ，971℃と1348℃であることが分かり，MCPMの融点がDCPDより低いことが分かった．加熱により，MCPMはγ-メタリン酸カルシウム，β-メタリン酸カルシウム，δ-メタリン酸カルシウムに変化し，DCPDはDCPA，γ-ピロリン酸カルシウム，β-ピロリン酸カルシウム，α-ピロリン酸カルシウムに変化した．加熱試料の0.2モル酢酸緩衝液中の溶解性はDCPD試料より，MCPM試料の方が高く，シーラント材として，DCPDの方がMCPMよりも，より効果的であることがわかった．

MCPMとDCPDの圧粉体について様々の出力レーザーを照射すると，DCPDではクレーターの形成が小さく，45.6 J/cm^{2}の照射では，クレーターは見られなかった．これらのことより，DCPDはエナメル質表面に悪影響を及ぼすことなく，低いエネルギー密度で融着することができ，シーラント材として，MCPMに比べ優れていることが示唆された．

キーワード：炭酸ガスレーザー，リン酸カルシウム，フィッシャーシーラント

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