研究者:印思瑜 (所属:大阪歯科大学大学院歯学研究科小児歯科学専攻)

研究題目(Study title):

新規アパタイトアイオノマーセメントの細菌学的特性に関する研究 Study on microbiological properties of novel apatite-ionomer cement

目 的 (Objective):

ICCMSTM (International Caries Classification and Management System) is the new concept of caries management based on ICDAS (International Caries Detection and Assessment System) which is a detection and assessment system classifying stage of the caries process, suggests that we should only remove the soft dentin in deep caries cavity (ICDAS Code 5-6). Glass ionomer cement (GIC) is one of the recommended materials for the deep caries region in ICCMS. We found that the addition of hydroxyapatite (HAp) strengthened conventional GIC, named apatite-ionomer-cement (AIC). However, there were many uncertain properties. This study was focused on discussing the multi-mineral ion release and the antibacterial activity of novel AIC.

材料および方法(Materials and Methods):

1) Preparation of specimens

Conventional GICs for restoration (Fuji IX GP, GC, Tokyo, Japan) were used as the control and fundamental materials. AIC powder which 8 wt% of GIC powder was replaced with spherical-shaped HAp was used for restoration. GIC and AIC powder were mixed with the IX-GP-liquid at powder/liquid ratio of 3.6 as recommended by the manufacturer. In addition, GIOMERTM (Beautifil II, SHOFU, Kyoto, Japan) was used as positive control, and cured by a dental light-cure device for 20 seconds on the each side.

2) Multi-mineral release test

All the specimens were prepared to 10mm in diameter and 2mm in thickness disc, then respectively suspended by cotton rope and immersed into 18ml deionized water at 37°C for seven days. For the measuring, each disc was removed from the water, washed by 2ml deionized water over the immersed water. The concentration of aluminum (Al), silicon (Si), phosphorus (P), Calcium (Ca), and strontium (Sr) ion were analyzed by using Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES, Shimadzu Co., Kyoto, Japan) and F ion were analyzed by a fluoride selected electrode (6561-10C, HORIBA Ltd., Kyoto, Japan) connected to an ion analyzer (D-53, HORIBA Ltd.).

3) Antibacterial activity test

Antibacterial activity test was using cariogenic bacteria, *Streptococccus mutans* ATCC25175 (*S. mutans*). *S. mutans* were preliminary incubated in 20ml Luria-Bertani broth medium at 37° C for 24 hours, then washed twice with phosphate buffered saline solution by centrifuging

at 4°C, 4000 rpm for 10 minute. S. *mutans* suspension was spectrophotometrically standardized to a concentration of 8 X 10⁶ CFU/ml with brain-heart infusion medium. Next, every specimen was immersed into the S. *mutans* suspension and incubated at 37°C for 4 hours. The No Sample group was used as negative control, which incubated S. *mutans* suspension without any specimen. After incubated, the luminescence intensity of the suspension of each group was evaluated by the ATP luminescence method, which was performed by luciferin-luciferase ATP assay reagent kit (Lucifer HS Set, Kikkoman Co., Chiba, Japan) and luminometer (Lumitester C-110, Kikkoman Co.).

4) Statistical analysis

In this study, data were analyzed by one-way ANOVA and Tukey's test. The p-values less than 0.05 were considered that significantly different.

結果(Results):

1) Multi-mineral release properties

Doses of ions released from GIC, AIC and Giomer are shown in Figure 1. The doses of F, Al, Si, and P of the AIC specimens were significantly higher than those of the GIC and the Giomer specimens. However, there were no significant differences in the release doses of Ca and Sr between three groups.



Figure 1. Multi-mineral release doses from GIC, AIC and Giomer

2) Antibacterial properties

The results of antibacterial activity of GIC, AIC and Giomer are shown in Figure 2. The ATP luminescence method performed in this study was using the reaction between ATP and an enzyme called luciferase which can generate light. The presence of ATP can be considered proof of the presence of a living bacteria. The antibacterial test was evaluated by measuring



Figure 2. Antibacterial activity of GIC, AIC and Giomer

the luminescence intensity with the ATP-luciferase reaction. However, there were no specimens immersed in the No Sample group, which can represent the original growth ability of *S. mutans*. Based on the luminescence intensity of Giomer has no significant differences with that of No Sample group, Giomer has no effective influence on inhibit the growth of bacterial. However, compared to the Giomer, the intensity of AIC and GIC were significantly decreased especially the group of AIC.

考察(Discussion):

In this study, novel material, AIC was compared on the multi-mineral ion release property and antibacterial activity to conventional GIC and Giomer. Fuji IX GP, which was used as a conventional GIC in this study, is one of the strongest commercially available restorative GICs. Therefore, it has been widely used for atraumatic restorative treatment (ART). Furthermore, Giomer is famous as a functional material. It contains the surface pre-reacted glass ionomer (S-PRG) filler, and has several anti-cariogenic properties, such as multi-ion (F, boron (B) and Sr, and so on) release and recharge. On both multi-mineral ion release property and antibacterial property, AIC was superior to GIC and Giomer. It was suggested that the increased of multi-mineral release was related to the increased ability of inhibit the growth of *S. mutans.* ICDAS suggests that we should remove the minimum of soft dentin in deep caries cavity which has high risk on pulp exposure. However, there is one concern that part of bacteria could be sealed in the lesions which may cause secondary caries. Excellent multi-mineral ion release property of AIC is expected not only the anti-bacterial property but also the re-mineralization of soft dentin. Moreover, it was expected that these advantages could decrease the chances on secondary caries which caused by bacteria remain in lesions.

結論 (Conclusion):

Addition of the HAp improved multi-mineral ion release and antibacterial properties of GIC.

成果発表(Research Achievement):

- 1) <u>邱 思瑜</u>, 邱 秀慧, 西村貴子, 篠永ゆかり, 阿部洋子, 有田憲司: アパタイトアイオノ マーセメントの電子顕微鏡的研究. 第53回日本小児歯科学会大会, 広島市, 2015年5月21 日 (Poster Presentation)
- 2) <u>邱 思瑜</u>, 邱 秀慧, 篠永ゆかり, 西村貴子, 阿部洋子, 有田憲司:充填用アパタイトアイ オノマーセメントの抗菌性に関する研究. 第34回日本小児歯科学会近畿地方会大会, 大阪 市, 2015年10月25日 (Poster Presentation)
- <u>Szu-Yu Chiu</u>, Yukari Shinonaga, Yoko Abe, Kyoko Harada, Kenji Arita: Effects of adding hydroxyapatite on functional properties of GICs. 10th Biennial Conference of the Pediatric Dentistry Association of Asia, Tokyo, 2016.5.26-28 (Poster Presentation 予定)