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Giga-Enhancement of Hydroxyapatite Effective Range in Bone Marrow by Electrical Polarization

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Electrically polarized hydroxyapatite (HAp) ceramics were implanted in the medullary cavities of canines. The effects of the polarized HAp on bone tissue reactions were investigated *in vivo* by histological evaluations. The HAp ceramics were electrically polarized at 300°C in a DC field of 1.0 $kVcm^{-1}$. The polarized and non-polarized HAp specimens were implanted in the medullary cavities of femoral and tibial bones of canines. Although at 7 days, no newly formed bone was observed in the medullary cavities in the vicinity of the non-polarized HAp surfaces, the newly calcified layer contacted with the negative charge induced surfaces. The osteoid tissues surrounded with connective tissues were formed approximately 0.5 mm above the positive charge induced surfaces. It was demonstrated that the effective range of the polarized HAp on bone formation was extended across the connective tissues.
Key words: hydroxyapatite, remanent charge, histological evaluation, electrical polarization

1. INTRODUCTION

Hydroxyapatite ($Ca_{10}(PO_4)_6(OH)_2$; HAp) ceramics have superior biocompatibility to bone tissues and are appropriate for artificial grafts in orthopedic and dental fields. The surface charge of HAp induced by electrical polarization altered the overgrowth rates of bone-like hydroxyapatite in simulated body fluid (SBF)[1-4].

In the present study, the electrically polarized HAp ceramics were implanted in the medullary cavities of rabbits. The effects of the polarized HAp on bone tissue reactions were investigated *in vivo* by histological evaluations. It was demonstrated that the effective range of the polarized HAp on bone formation was extended across the connective tissues.

2. EXPERIMENTAL PROCEDURE

2.1 Preparation of HAp ceramics

HAp powder calcined at 850°C was pressed in a mold at 200 MPa. The HAp pellets were sintered at 1200°C for 2 h in saturated water vapor pressure atmosphere. The samples, 5×8×0.5 mm^3, were cut from disc-shaped sintered ceramics with a low-speed diamond saw. The ceramic samples were characterized by X-ray powder diffractometry (XRD) and infrared absorption spectroscopy(IR).

2.2 Electrical polarization treatments

The HAp ceramics were electrically polarized at 300°C in a DC field of 1.0 $kVcm^{-1}$ in air as shown in Fig. 1. The negatively charged surface, the positively charged surface, and the non-polarized surface are abbreviated as n-surface, p-surface and 0-surface, respectively. The polarized HAp specimens were cooled to room temperature in the electric field. The remanent charge of the polarized HAp ceramics was confirmed by thermally stimulated current measurements.

2.3 *In vivo* evaluations

The polarized and non-polarized HAp samples sterilized with ethylene oxide gas were implanted in the medullary cavities of femoral and tibial bones of canines. After anesthetization, the femoral and tibial bones were exposed by lateral luxation. The HAp samples were placed in the slots obtained in the bones with a dental fisher bur of 0.8 mm in diameter. The bones containing the HAp blocks were extracted under the anesthesia 3 and 7 days after the implantation. The histological evaluations were examined with the decalcified sections stained by hematoxylin eosin (H-E) and toluidine blue methods as well as the non-decalcified sections stained by H-E and Villanueva methods.

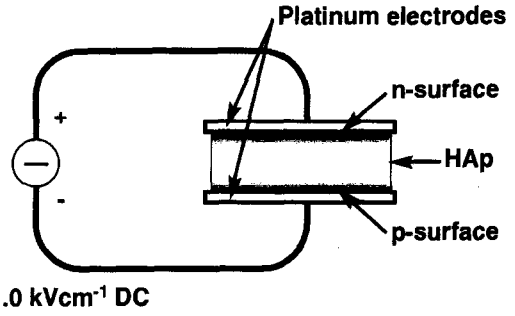


Fig. 1 Schematic illustration of electrical polarization treatment.

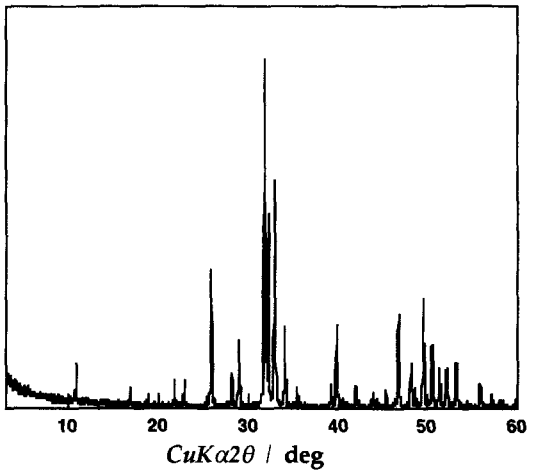


Fig. 2 XRD pattern of HAp ceramics sinterd at 1200°C for 1h.

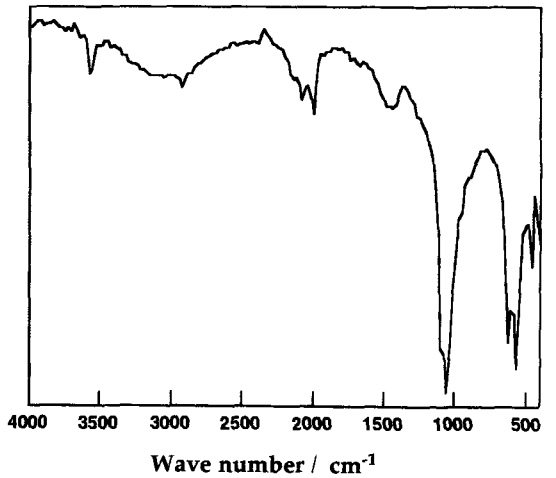


Fig. 3 IR spectrum of HAp ceramics sinterd at 1200°C for 1h.

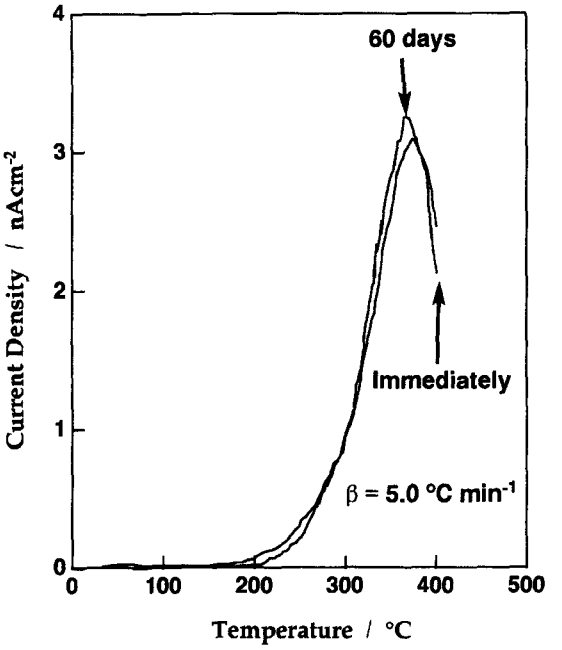


Fig. 4 TSC curve of HAp ceramics immediately after electrically polarization at 300°C in DC field of 1.0 kVcm^{-1}, compared with that 60 days after polarization.

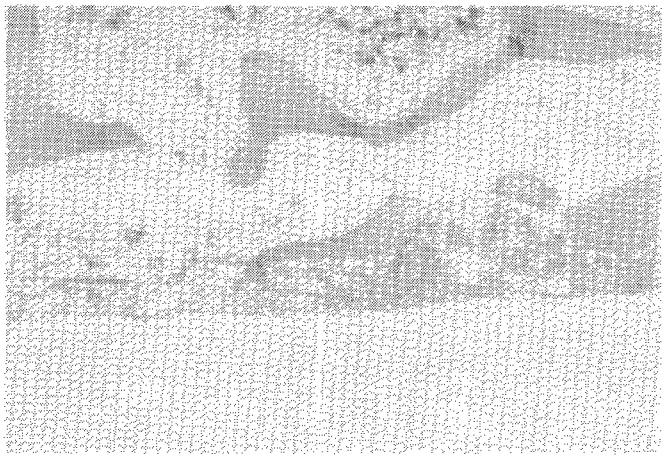
3. RESULTS AND DISCUSSION

3.1 Characterization of HAp ceramics

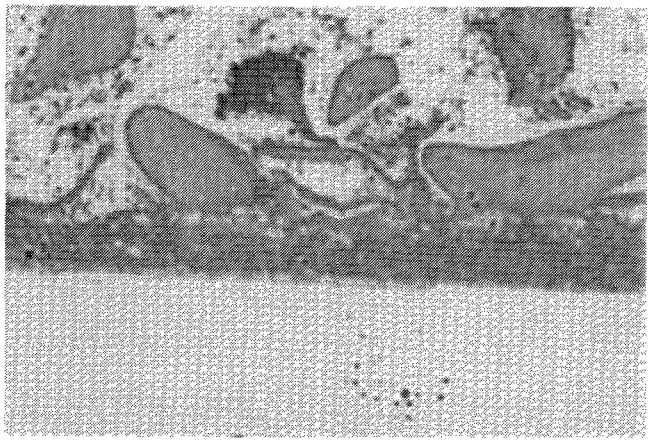
The sintered ceramics were identified as a HAp polycrystalline single phase by XRD as shown in Fig. 2. No other calcium phosphate phase was detected. The IR spectrum of the HAp ceramics is shown in Fig. 3. The absorption peaks at 568, 962 and 1035 cm^{-1} indicated the existence of tetrahedral PO_4^{3-}. The other absorption peaks at 425, 605, 1055 and 1095 cm^{-1} were assigned to the combined mode vibrations of PO_4^{3-} tetrahedron. The OH vibrations at 630 and 3550 cm^{-1} was attributed to the existence of OH^- ions in apatitic structure.

3.2 TSC measurements

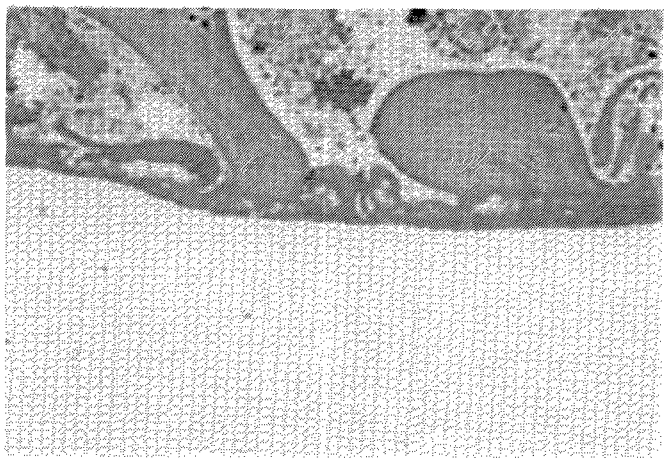
The TSC curves of the HAp ceramics immediately after the electrical polarization treatment and 60 days after the polarization are shown in Fig. 4. No significant difference was detected in the total charge values and the profiles. The TSC results suggested that the remanent charge was almost completely preserved for a long time.



(a) 0-surface



(b) n-surface



(c) p-surface

Fig. 5 Histological sections of bone marrow 7 days after implantation of electrically polarized and non-polarized HAp ceramic; (a) 0-surface, (b) n-surface and (c) p-surface.

3.3 *In vivo* evaluations

The layers consisting of blood cells and fibroblasts on the n- and p-surfaces were observed in the H-E stained sections obtained from both parts of the cortical bone and bone marrow neighborhood at 3 days after the implantation. Although no newly formed bone was observed in the medullary cavities near by the 0-surfaces, the newly calcified layer contacted with the n-surfaces 7 days after the implantation. The osteoid tissues surrounded with connective tissues were formed approximately 0.5 mm above the p-surfaces.

4 CONCLUSION

The significantly accelerated new bone formations were observed on the n-surface. On the contrary, the osteoid tissues surrounded with connective tissues were formed approximately 0.5 mm above the p-surfaces. It was demonstrated that the effective range of the polarized HAp on bone formation was extended across the connective tissues

ACKNOWLEDGEMENTS

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REFERENCES

1. M.Ohgaki, K.Yamashita, *Phos. Res. Bull.* **8**, 37-42 (1998).
2. M.Ohgaki, K.Yamashita, S.Nakamura *et al.*, *Am. Ceram. Soc. 100th Annual Meeting Abstract Book* 7, (1998).
3. M.Ohgaki, J.Hamagami, T.Umegaki, K.Yamashita, *Bioceramics*, **11**, 493-496 (1998).
4. K.Yamashita, K.Kitagakiq, T.Umegaki, *J. Am. Ceram. Soc.*, **78**, 1191-1195 (1995).

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