EFFECTIVE CELLULAR RESPONSES TO POLARIZED HYDROXYAPATITE VECTOR CERAMICS IN BODY FLUID

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Abstract The polarized Hydroxyapatite (HA) and non polarized HA pellets were intraperitoneally implanted into rats and reacted with the body fluid. The samples were extracted at 5 min, 30 min, 1 and 3 day after the implantation. Clear differences can be seen between on the polarized and non polarized surfaces of HA.

INTRODUCTION

Hydroxyapatite ($\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$; HA) has an excellent biocompatibility and valuable electric properties.^{1, 2)} HA can be polarized under a dc field at an elevated temperature.^{3, 4)} The polarized HA has a strong biological effect in an in vivo interaction with bones.^{5, 6, 7)} We have studied the electrovector effects of the polarized HA with the nature of cell-biomaterial surface interaction corresponding to survival, growth and differentiation in body fluid.

EXPERIMENTAL

Preparation of polarized HA ceramics

HA powder was pressed in a mold at 200 MPa. The HA pellets were sintered in the saturated water at 1250°C for 2h.

Polarization

The samples were sandwiched between a pair of platinum electrode plates, heated to 300°C in air, and electrically polarized in a dc field of 1kV/cm for 1h. The surface negatively and positively charged by the polarization are abbreviated as N surface and P surface, respectively. Non polarized surface is denoted by 0

surface. The electrical storage of the polarized HA pellets was confirmed by thermally stimulated depolarized current (TSDC) measurement.

Animals

The biological reaction of the polarized HA was investigated by implanting into the abdominal cavity of 6 weeks Wistar rats. The samples were extracted at 5, 30 min, 1, 3 days after the implantation and soaked in 2.5% glutaraldehyde in 0.1 M PBS. The surfaces of the polarized HA and the non polarized HA were examined by scanning electron microscopy (SEM).

RESULTS AND DISCUSSION

The electrical surface charge induced by the space-charge polarization was the order of $1.5\mu\text{Ccm}^{-2}$ from TSDC data (Fig.1).

Fig.1 TSDC spectra of HA polarized at 300°C for 1h in a dc field of 1.0 kV cm^{-1} . Heating rate was 5°Cmin^{-1} .

At the first 5 min, there was not much difference between the surfaces. All of the surfaces were partially covered with some fibrin formation and red blood cells. However, all surfaces also involved some macrophages. The cells were more attached and the fibrin was more developed on the P surface.

After 30min, on the 0 surface, the fibrin network was still very weakly developed. In some areas, apparently associated with the fibrin, the number of cells increased considerably. A strong fibrin network was formed on the N surface. Red blood cells were imbedded in the network but also some

macrophages and cells with fibroblastic morphology. The fibrin network on the P surface was not so strong and stabilized as on the N surface case. Cells could be seen mostly in the fibrin coated area. They appeared to grow between the HA and the fibrin network.

At 1 day, the fibrin network on the 0 surface did not exist. There was hardly any fibrin to be seen, however, the number of cells had increased. The majority of the cells had fibroblastic morphology and appeared to be well attached to the surface suggesting rapid spreading and proliferation. The fibrin network on the N surface appeared to have stabilized further. Macrophages were trapped between the fibrin layer and the HA surface.

At 3 days, a strong fibrin network was also seen on the N and the P surfaces. However, it was not stabilized as on the 0 surface (Fig.2).

Fig. 2 SEM photographs for 0, N, P surfaces of HA at 3 days after the implantation.

In summary, clear differences can be seen on the 0, N and P surfaces of the HA. The cell adhesions and proliferation were dominated on the 0 surface. In the contrast, fibrin network formation and stabilization were dominated on the N and the P surfaces.

A stabilized fibrin network was associated with growth and differentiation factors that support for example bone formation. The fact that the cells grew better on the 0 surface did not necessarily mean that the cells differentiated into specific tissues such as bone and capillaries. It suggested that the electrical

polarized HA induced effective effects on cell adhesion and growth in body fluid.

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