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Rapid communication

Adsorption and irreversible binding of adriamycin incorporated into hydroxyapatite beads

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Summary

Adriamycin-loaded porous hydroxyapatite (HAP) beads were investigated in terms of drug adsorption and drug release from solution-loaded wet beads and the corresponding dry beads. The wet beads resulted in almost 100% release of the drug into water while the dry beads released only about 60%. A possible explanation for this observation is that direct contact between the drug and the surface of HAP in the dry state induced strong irreversible binding through electrostatic forces and hydrogen bond and/or complex formation with surface calcium.

Hydroxyapatite (HAP) is the major mineral component of bone and teeth. Synthetic HAP is known to be similar to naturally occurring HAP on the basis of chemical and crystallographic studies (Posner, 1985; Driessens, 1988). Because of its biocompatible nature, HAP has been used in various forms in orthopedics as an aid in bone grafts following surgery of bone tumors and osteoarthritis (Bucholz et al., 1987; Koeneman et al., 1990).

Drug delivery applications of drug-loaded HAP implants are also considered to be advantageous for local adjuvant chemotherapy, where the implant is expected to release active agents at a predetermined rate for a specific duration (Lemons et al., 1988). Therapeutic agents to be incorporated include antibiotics, antineoplastics, and hormones such as bone growth factor.

The present study is concerned with adriamycin (ADR) loading of model HAP beads, and the behavior of the adsorption and release of drug from wet and dry beads containing the drug.

ADR-HCl was a gift from Kyowa Hakko, Tokyo and was used as received. Porous HAP beads were donated by NGK Spark Plug Co., Nagoya, Japan. The physical data of the beads (XVC-56) were given as follows: sintering temperature, 1100°C; weight per bead, 531 ± 0.7 mg (mean ± SD, $n = 5$); diameter, 8.48 mm; bulk density, 1.66; true density of the material, 2.97; and Ca/P, 1.67. HAP beads were washed once

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daily for 1 week with freshly distilled water to remove unknown water-soluble substances and ions, and then dried in a desiccator with a vacuum pump. To determine the water-accessible volume of the internal pore spaces, the beads were allowed to stand overnight in distilled water at 25°C. The volume was estimated from the weight difference between the empty and water-filled beads where the density of distilled water was assumed to be 1.0 at 25°C. Mean full displacement of the pore spaces with water was 0.140 ± 0.001 ml (mean $\pm$ SD, $n = 5$). Assuming that the water-accessible volume in the bead was equivalent to the pore volume, the bulk porosity was calculated to be 0.439.

Examination of the adsorption isotherm of ADR was carried out as follows: two beads were placed in various drug solutions containing up to 120 μg/ml (distilled water or pH 5.0–7.4, 0.067 M phosphate buffers, each 4 ml) and allowed to stand overnight at 5°C. The concentration of drug in the bulk solution was determined by spectrophotometric assay at 480 nm and pH 6.0 after appropriate dilution. Drug release behavior was investigated using two types of beads differing in the extent of drug loading: (i) wet beads in which the drug solution remains loaded (335 μg/bead, estimated from the pore volume); (ii) dry beads in which the drug was deposited to dryness at room temperature in a desiccator with a vacuum pump. Beads of either the wet or dry type were placed in distilled water (40 ml) at 37°C and the amount of drug released was assayed as a function of time.

Fig. 1 shows the adsorption isotherm for adsorption of ADR onto HAP beads in various aqueous media. It is clearly demonstrated that drug adsorption reached a saturation maximum in distilled water while no such tendency was observed in phosphate buffers (pH 5.0, 6.0 and 7.4) within the range of drug concentrations examined, however, their linear behavior was pH dependent. In the pH region studied and in distilled water, the exclusively existent species of ADR should be in the monocationic form with a protonated amino sugar moiety, since the microscopic dissociation constants are known to fall within the range of pK 8–10 (Sturgeon and Shulman, 1977). Therefore, the drug is likely to be bound primarily by electrostatic forces between the protonated form and negatively charged groups such as HAP-PO_4^- and HAP-OH^- (Gorbunoff and Timasheff, 1984). However, it is of interest that the magnitude of adsorption increased with increasing concentration of alkali metal (from pH 5 to 7.4) originating from buffer species, particularly Na^+ which has a relatively high affinity for HAP (Shimabayashi et al., 1981).

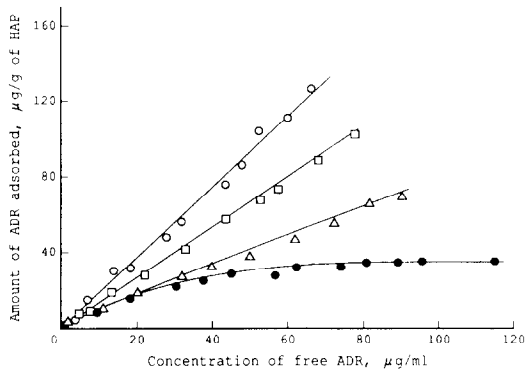

Fig. 1. Adsorption isotherm of adriamycin onto HAP beads in various aqueous media. (●) Distilled water, (△) pH 5.0 phosphate buffer, (□) pH 6.0, (○) pH 7.4 (temperature, 5°C). Symbols are means of 2 separate runs. Bead weight: 531 mg/bead.

Fig. 2 depicts plots of the rates of release ADR from drug-loaded wet and dried HAP beads. It should be noted that the wet beads gave rise to about 98% release of the drug while the dried beads released only about 60% despite the lack of drastic processing of beads containing drug solution except for drying at room temperature. Thus, the process of drying led to considerable immobilization of drug on the surface of the HAP material. It should again be emphasized that the major difference between the two conditions is only whether the drug is accommodated in the beads as an aqueous solution or deposited in a dry state on the HAP pore walls. A possible explanation is as follows: due to possessing ample phosphate oxygens, the HAP surface strongly adsorbs one or two full layers of water molecules through hydrogen bonding in water (Posner, 1985). Accordingly, the water layers are likely to prevent binding of ADR on the HAP surface.

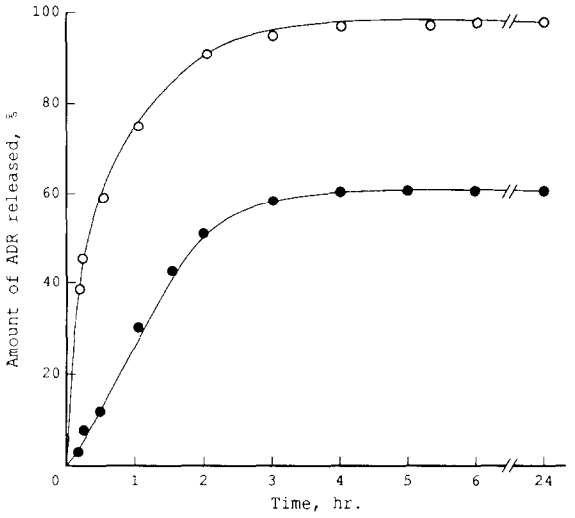

Fig. 2. Release behavior of adriamycin from wet and dried HAP beads. (○) Wet beads, (●) dried beads. The bulk porosity of HAP beads was 0.439. Symbols are means of 3 separate runs and the absence of a bar indicates that the SD is smaller than the symbol. Amount of drug loaded, 0.335 mg/bead; temperature, 37°C.

During the process of drying, the drug is concentrated in solution followed by precipitation on the pore walls. It is therefore probable that the direct contact contributes to strong irreversible binding between the drug and the surface of HAP through electrostatic forces and hydrogen bonds. Another possible mechanism is that of complex formation with surface calcium during drying, resulting in the immobilization of ADR.

Dry drug-loaded HAP beads are more advantageous than the corresponding wet beads in terms of drug stability and practical use. The present results suggest that carefully controlled further studies on the loading, adsorption and release of drugs are required in order for the rational design of drug delivery HAP beads to be achieved, especially drugs with a high affinity for the surface of HAP.

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