Intra-Arterial Administration of Heated Albumin Microspheres Containing Mitomycin C to Rabbits with VX-2 Tumor

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ABSTRACT: In an attempt to enhance antitumor effects, we prepared heated albumin microspheres containing mitomycin C (MMC). These MMC microspheres have an average diameter of 45 ± 8 μm and contain about 5 per cent of MMC. The intra-arterial MMC microsphere treatment, for albino rabbits with implanted VX-2 tumor, increased remarkably the tissue MMC levels, compared to that with conventional MMC, and resulted in conspicuous antitumor efficacy. This approach to antitumor chemotherapy should be effective for selected patients with malignant tumor receiving a blood supply from an end-artery.

KEY WORDS: mitomycin microsphere, intra-arterial chemotherapy

INTRODUCTION

Intra-arterial infusion chemotherapy is prescribed to attain high localized levels of antitumor drugs, especially in case of unresectable tumors. With conventional infusion treatment, however, the infusates are eliminated rapidly from the drainage veins. For accumulation of the antitumor drugs in the target tumor area, we devised biodegradable albumin microspheres containing MMC. In a foregoing paper,1 we reported the in vitro and in vivo drug release from these microspheres. We now report the in vivo antitumor efficacy and pharmacokinetics of the biodegradable microspheres.

MATERIALS AND METHODS

Bovine serum albumin microspheres containing MMC were prepared by the modified method of Scheffel et al.,2 as reported previously.1 In short, a mixed aqueous solution of bovine serum albumin and MMC is emulsified in cotton-seed oil containing 10 per cent Span 85, a surface active agent (Fig. 1). Thereafter, it is gradually heated and solid-
ified at 170°C. After cooling of these solidified microspheres, the spheres are separated by centrifugation and are washed with ethyl ether to remove adhering oil. The average diameter of these microspheres is $45 \pm 8 \mu m$ and the drug content is about 5 per cent (Fig. 2).

Male albino rabbits and VX-2 carcinoma were used. When the VX-2 tumor implanted into the right hind leg reached about 2 cm in diameter, 1.2 mg/kg of MMC microspheres (1.2 mg/kg as MMC) or conventional MMC was directly given into the right femoral artery, following pentobarbital-Na anesthesia. Peripheral blood and that from the right femoral vein were obtained to measure the amounts of MMC leaked from the MMC microspheres, at a determined time. The VX-2 tumor and the muscle tissue in the hind legs were excised to measure MMC tissue levels, at a determined time. VX-2 tumors and the muscle tissues, which were removed aseptically as soon as possible, were homogenized with a phosphate buffered solution in an ice-cold homogenizer. The homogenate was centrifuged at 1,000×g for 10 minutes at 2–4°C, and the resultant supernatant was used for a bioassay method for MMC concentration determination. MMC concentration in sera and these tissues were bioassayed using Escherichia coli B strain.³

VX-2 tumors were excised 2 weeks after these treatments and histologic examinations, including the entrapped microspheres, were done microscopically, under conditions of conventional hematoxylin-eosin staining.

**RESULTS**

Time course of MMC levels in the peripheral blood is shown in Fig. 3. With conventional MMC, the peripheral levels dropped with a rapid half-reduction time of 7–10 minutes, while in the MMC microsphere group, it decreased with two types of half-reduction time. The drug concentrations in the venous blood were markedly high, that is, 0.1 μg/ml within 120 minutes after the infusion.

Figure 4 illustrates the time course of muscle tissue drug levels in the case of MMC microsphere and conventional MMC. As for drug concentration in the MMC microsphere group, high drug levels continued for 240 minutes, while in the case of conventional MMC, the level decreased 60 minutes later, below the limitation of bioassay.