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Sustained Release of Sulfamethizole, 5-Fluorouracil, and Doxorubicin from Ethylcellulose-Polylactic Acid Microcapsules

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The preparation of ethylcellulose-polylactic acid microcapsules with long release half-lives of chemotherapeutic agents was investigated. The microcapsules were prepared by means of coacervation-phase separation of ethylcellulose from ethyl acetate solution by nonsolvent addition in the presence of polylactic acid.

In the case of sulfamethizole microcapsules, longer release half-life was obtained with larger amounts of polylactic acid in the preparation. The release rate of sulfamethizole was not very dependent on the pH of the release media.

5-Fluorouracil and doxorubicin hydrochloride were similarly microencapsulated. 5-Fluorouracil microcapsules were sieved prior to release studies. A release half life of 2.6 hr was obtained with the 5-fluorouracil microcapsules. In the case of doxorubicin microcapsules, the drug was released with a half-life of 84 min, whereas uncoated doxorubicin dissolved completely within 15 min.

Keywords——sulfamethizole; 5-fluorouracil; doxorubicin; ethylcellulose; polylactic acid; microencapsulation; coacervation-phase separation; microcapsules; sustained release; photomicrograph

Microencapsulation, a technique of encapsulating drug powders and beads, has been developed to achieve sustained release of drugs. Various microencapsulation processes such as air suspension, coacervation-phase separation, spray drying and congealing, and pan coating techniques have been adapted to pharmaceutical use.[2)]

Since microencapsulation by coacervation-phase separation does not require any elaborate manufacturing equipment, it is often employed in small laboratories. The authors have demonstrated the usefulness of enteric coated microcapsules in sustaining the release of an antibacterial agent, sulfamethizole, from microcapsules coated with carboxymethylethylcellulose, a new enteric coating material.[3)]

Water-insoluble polymers such as ethylcellulose have also been used to prepare sustained release microcapsules.[4—7)] In the preparation of microcapsules employing ethylcellulose as a coating material, temperature change,[4,6,8,9)] incompatible polymer addition,[10)] and nonsolvent addition,[7,11)] have been used to effect coacervation-phase separation.

In this work, we examined the effect of an added complementary polymer on the release characteristics of drugs from ethylcellulose microcapsules prepared by the nonsolvent addition technique.

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Chemotherapeutic agents were employed in the present study since sustained release preparations are often required for the control of infections and neoplastic diseases.

Experimental

Materials——Ethylcellulose standard 100 (EC) with an ethoxy content of 48.0—49.5% (lot AIK4978) was purchased from Wako Pure Chemical Ind., Osaka. Polylactic acid was synthesized following the reported procedure.[12] The average molecular weights of polylactic acid samples measured with a vapor pressure osmometer (Knauer, Germany) were 2000 and 1600. Sulfamethizole of JP IX grade (lot XC14GG) was a product of Eisai Co., Tokyo, and 5-fluorouracil (lot 27098) and doxorubicin hydrochloride (adriamycin, lot 64498) were generously supplied by Kyowa Hakko Kogyo Co., Tokyo. All other chemicals were of reagent grade.

Preparation of Microcapsules——Polylactic acid corresponding to 20 or 50% (w/w) of the amount of EC was completely dissolved in 5% EC solution in ethyl acetate. Sulfamethizole corresponding to 20 or 40% (w/w) of the amount of EC was then suspended in the solution and the suspension was stirred for several hours at a rate of 300 rpm. The drug powders were then dispersed finely by ultrasonification (model UR-200, Tomy Seiko Co., Tokyo; 100W, 1—2 min) and *n*-pentane was added dropwise to the suspension at a rate of 0.5—0.8 ml/min until the phase separation was completed. The microcapsules thus obtained were washed twice with *n*-pentane by mixing and decantation and then collected on a filter paper. The microcapsules were dried *in vacuo* for 24—48 hr at ambient temperature.

In the microencapsulation of 5-fluorouracil and doxorubicin hydrochloride powders, the microencapsulation procedures used for sulfamethizole were followed except that the amounts of polylactic acid and the drugs used were set at 50 and 20 (and also 8 for doxorubicin) % (w/w) respectively. Standard sieves were employed for the separation of microcapsule particles of 5-fluorouracil into various size ranges depending on the sizes of microcapsular clusters. Sieves with numbers 150 (sieve opening 105 μm), 100 (149 μm), and 80 (177 μm) were used for this purpose. Sulfamethizole and doxorubicin microcapsules were not sieved.

Scanning Electron Microscopic Observation——The dried microcapsules were observed with a scanning electron microscope (model S-430, Hitachi Manufacturing Co., Tokyo) to examine their shapes, surface characteristics, and particle sizes.

Release Studies——Release profiles of the drugs from the microcapsules in JP IX disintegration medium No. 1 at pH 1.2 and in medium No. 2 at pH 7.5 (sulfamethizole) or in physiological saline (5-fluorouracil and doxorubicin) were obtained by a modification of the beaker method. An Erlenmeyer flask containing 200 ml of the release medium was placed in a constant-temperature water bath maintained at $37.0 \pm 0.2°$ which was placed on top of a constant torque magnetic stirrer (High Magmix, Mitamura Riken Kogyo Inc., Tokyo). A weighed quantity (200 mg) of the microcapsules was suspended in the release medium (200 ml) and the suspension was stirred with a magnetic stirring bar (3 cm long) at a rate of 200 rpm. One ml samples were withdrawm at predetermined intervals, and in each case one ml of fresh release medium was added to the flask to maintain the original volume. After diluting the sample solution with the same release medium, the drug concentrations were analyzed spectrophotometrically; sulfamethizole in medium No. 1 at 270 nm, or in medium No. 2 at 262 nm, 5-fluorouracil in the physiological saline at 266 nm, and doxorubicin in the saline at 234 nm.

Results and Discussion

Examinations of Microencapsulation Conditions

Based on toxicological considerations[13] ethyl acetate was chosen as a solvent for EC and polylactic acid among several possible solvents for these polymers. The presence of polylactic acid together with EC greatly improved the sustained release characteristics. A polylactic acid level of 50% was more favorable in sustaining the release than one of 20% (Fig. 1), but at polylactic acid levels above 50%, embryonic microcapsules tended to aggregate. Ultrasonification was helpful in adding drug particles to disperse in the solvent. *n*-Pentane was selected as a phase separation agent among various poor solvents for EC based on the easy removal of the solvent from the microcapsules after preparation due to its low boiling point, and toxicological considerations.[14]

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Characteristics of Microcapsules

It is expected that polylactic acid is incorporated into the EC capsular wall, since the weight of dried microcapsules obtained roughly corresponded to the sum of the weights of added EC, polylactic acid, and the drug. Drug contents of the microcapsules are listed in Table I. Scanning electron microscopic observation revealed that the shape of the microcapsules was spherical, and few pores were observed on the surface of the microcapsules (Figs. 2 and 3). The average diameter of unit microcapsules which contained polylactic acid and sulfamethizole corresponding to 50% and 20% of the amount of EC was 16.7 (arithmetic mean) ±14.1 (SEM) μm as measured in photomicrographs.

Release Patterns

Sulfamethizole Microcapsules——The effect of the amount of polylactic acid added to the preparation on the release rate of sulfamethizole from the microcapsules in an acidic solution

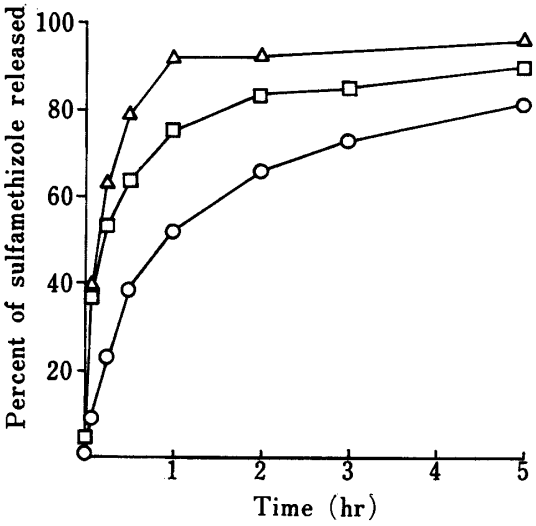

Fig. 1. Comparison of Release Patterns of Sulfamethizole in the Medium at pH 1.2 from Microcapsules prepared in the Presence of Various Amounts of Polylactic Acid

During preparation, the amount of the drug added was kept at 40% while the amounts of polylactic acid were varied: △, 0%; □, 20%; ○, 50%; of the amount of EC. Data are averages of two experimental runs.

Table I. Drug Contents in the Microcapeules,[a] % (w/w)

	Percentage of drug added relative to the amount of EC		
	40(21)[b]	20(12)[b]	8(5)[b]
Sulfamethizole	25	12	—
5-Fluorouracil	—	14	—
Doxorubicin	—	14	4

a) Polylactic acid corresponding to 50% (w/w) of EC was used.
b) Theoretical drug contents assuming that all solids added are incorporated into the microcapsules.

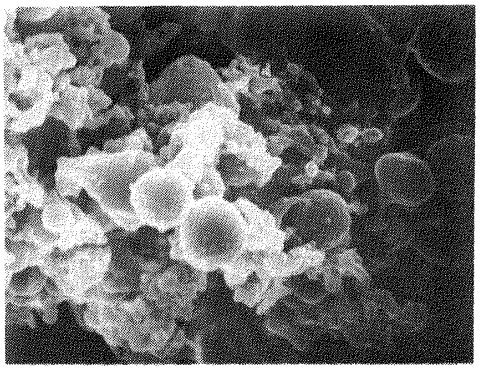

Fig. 2. Scanning Electron Photomicrograph of Sulfamethizole Microcapsules prepared from EC–Polylactic Acid–Sulfamethizole=10: 5: 2 (×2000)

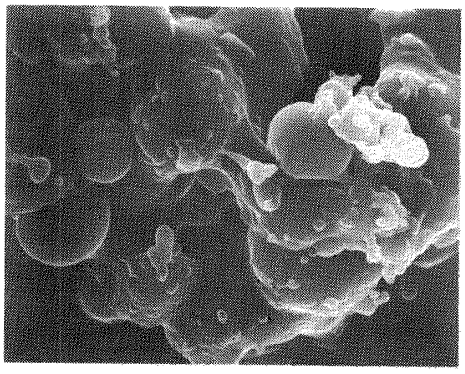

Fig. 3. Scanning Electron Photomicrograph of 5-Fluorouracil Microcapsules prepared from EC–Polylactic Acid–5-Fluorouracil=10: 5: 2 (×2000)

at pH 1.2 is shown in Fig. 1. As the amount of polylactic acid increased, more sustained release from the microcapsules was obtained. The release half-life increased from 9 min in the absence of polylactic acid to 54 min in its presence at the 50% level for the preparation. This may be attributed to incorporation of polylactic acid into an EC film around the drug particles and to its action as a plasticizer. As a result, the numbers of pores in the film are expected to decrease, as the amount of polylactic acid is increased. This hypothesis is supported by the presence of pH dependency in release rates when a smaller amount of polylactic acid was used and its absence when a larger amount of polylactic acid was used (see Fig. 5).

Fig. 4 compares the drug release patterns from microcapsules with different drug contents in acidic solution. Although the absolute rate of release was greater from the capsules with larger drug contents, there was little difference in the relative rate (the rate of release per unit drug content) even if the wall thickness of each microcapsule can be decreased by increasing the total amount of the drug added in the preparation. This aspect requires further examination.

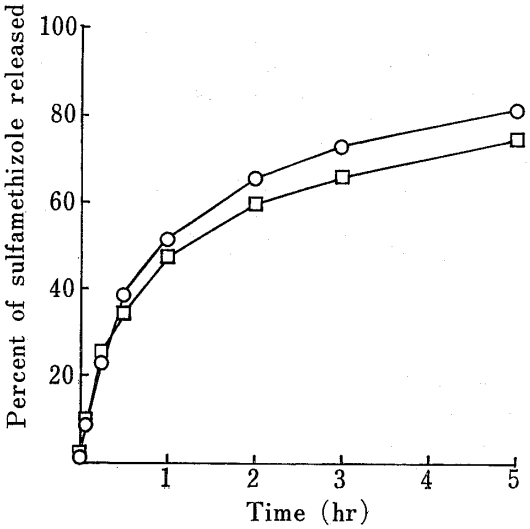

Fig. 4. Comparison of Release Patterns of Sulfamethizole in the Medium at pH 1.2 from Microcapsules prepared in the Presence of Various Amounts of the Drug

During preparation, the amount of polylactic acid added was kept at 50% and the amounts of the drug added were varied: ○, 40%; □, 20% of the amount of EC. Data are averages of two experimental runs.

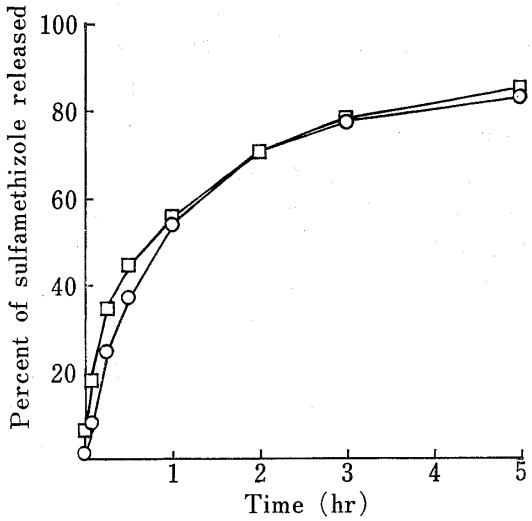

Fig. 5. The Release Patterns of Sulfamethizole from Microcapsules in the Release Medium at pH 1.2 (○) and at pH 7.5 (□)

During preparation, the amounts of polylactic acid and the drug added were 50% and 20% of the amount of EC, respectively. Data are averages of two experimental runs.

Only small differences were observed between the release patterns of the drug from the microcapsules in JP IX disintegration medium No. 1 at pH 1.2 and those in medium No. 2 at pH 7.5, as shown in Fig. 5. The release rate of the drug from the microcapsules prepared from EC and polylactic acid (50% of EC) does not seem to be dependent on the drug solubility in the release media. On the other hand, when a smaller amount of polylactic acid was used (10% of EC) the release rate was much greater at pH 7.5 than at pH 1.2. A similar pH-dependent release of aspirin from EC microcapsules has been reported.[15,16] This suggests a different release mechanism in the case of EC-polylactic acid microcapsules compared with EC microcapsules and hydrogels, in which the release rate is dependent on the solubility of the drug in the release media.[17] In the drug release from EC-polylactic acid microcapsules, permeation of the drug through the membrane can be rate-limiting, whereas in EC microcapsules and hy-

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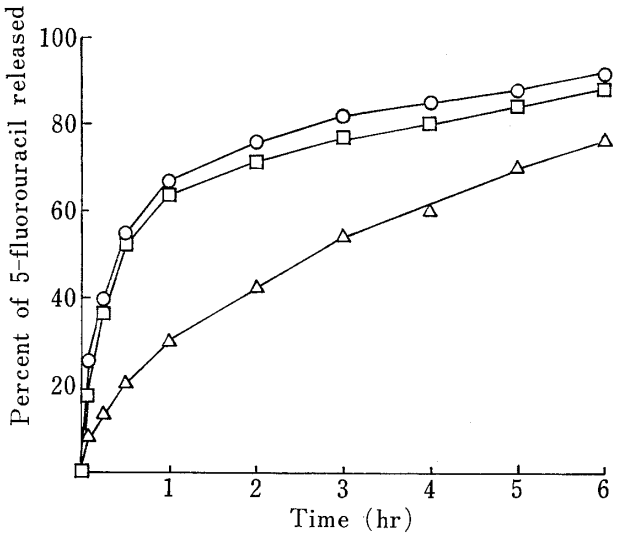

Fig. 6. Release Patterns of 5-Fluorouracil from Microcapsules of Different Sizes in Physiological Saline

During preparation, the amounts of polylactic acid and the drug added were 50% and 20% of the amount of EC, respectively. Key: ○, 105—149 μm; □, 149—177 μm; △, larger than 177 μm in diameter.

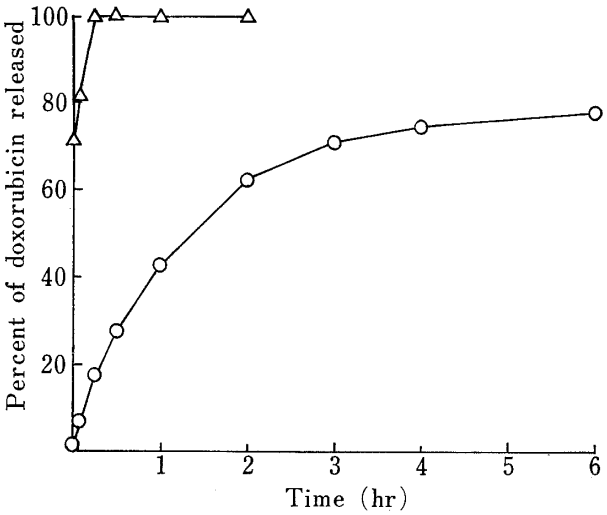

Fig. 7. Comparison of the Release Pattern (○) of Doxorubicin from Microcapsules and the Dissolution Pattern (△) from the Powder in Physiological Saline

During preparation, the amounts of polylactic acid and the drug added were 50% and 8% of the amount of EC, respectively.

drogels, the solubility of the drug in release media can be an important factor if the drug dissolves in solvent which enters through pores and the dissolved drug then emerges through pores.

5-Fluorouracil and Doxorubicin Microcapsules——Fig. 6 shows the release profiles of 5-fluorouracil from microcapsules in three size fractions. Larger microcapsules which have a smaller surface area per unit weight of microcapsules showed slower release than the smaller ones. There was little difference in the drug contents among microcapsules of two size ranges (microcapsules of less than 105 μm and those of 149—177 μm). A release half-life of as long as 2.6 hr was obtained. This size-dependent release rate was noted in the release of phenobarbital sodium from EC microcapsules.[4] Appropriate sizing of microcapsules by sieves may be necessary in order to obtain microcapsules with desirable release rates. The release profile of doxorubicin from its microcapsules is compared with the dissolution profile from the drug powder in Fig. 7.

A release half-life of 84 min was obtained. Sustained release of antineoplastic drugs obtained in the present study may be applicable for the *in vivo* use of antineoplastic drugs in transcatheter arterial embolization therapy.[18,19]

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