Preparations of Agglomerated Crystals of Polymorphic Mixtures and a New Complex of Indomethacin–Epirizole by the Spherical Crystallization Technique

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Abstract □ Agglomerated crystals of indomethacin and epirizole were prepared by the spherical crystallization technique. The solvent used was ethanol–water–chloroform, ethyl acetate–water, or ethyl acetate–aqueous sodium chloride. From the ethanol–chloroform–water system, we obtained agglomerated crystals of a polymorphic mixture of the β form of indomethacin (original form, γ) and amorphous epirizole. When the mole percent of epirizole loaded into the system was less than 63 and 38% for the ethyl acetate–water and ethyl acetate–aqueous sodium chloride systems, respectively, the agglomerated crystals consisted of a polymorphic mixture of the α form of indomethacin and amorphous epirizole. When the respective mole percent of epirizole loaded was more than 65 and 43% in the aforementioned systems, a new complex of indomethacin–epirizole (molecular ratio equal to 2:1) was obtained. Recovery of complex from the drugs loaded in the ethyl acetate–aqueous sodium chloride system was higher than that in the ethyl acetate–water system, as a result of a salting-out effect. The solubility of indomethacin in the agglomerated complex in a solution of 30% aqueous ethanol and in disintegration test solution no. 2 (composition, 0.05 M KH_2PO_4 plus 0.0236 M, NaOH, pH 6.8), specified in the Japanese Pharmacopeia X (JPX), was higher than in the physical mixture (molecular ratio of indomethacin to epirizole equal to 2:1). In the ethanol solution, indomethacin was transformed into the γ form during dissolution, and a decrease in solubility occurred. The process of dissolution of the tablet of the agglomerated complex was described by zero-order kinetics. The rate of release of indomethacin from the complex was three times more rapid than that from the physical mixture in the disintegration test solution. These agglomerated crystals can be compounded directly into pharmaceutical systems without further processing such as granulation.

A novel agglomeration technique that transforms fine crystals directly into compacted spheres during crystallization has been reported.[1–5] The spherically agglomerated crystals can be prepared in tablet form or compounded directly into a pharmaceutical system without further processing such as granulation. In addition, this technique may enable the crystalline form of a drug to convert to a different polymorphic form and thus attain better bioavailability.

We now report a new method for the preparation of the spherically agglomerated crystals of polymorphic or complex forms of indomethacin and epirizole. These two drugs, which exist as polymorphs[6,7] and a hydrate,[8] respectively, are widely prescribed for their anti-inflammatory, antipyretic, and analgesic properties. Tsurumi et al. reported a reduction in the gastric and intestinal ulcerogenicity of indomethacin when co-administered with epirizole.[9] We investigated the formulation parameters for crystallization and determined the crystalline forms of the drugs in the resultant agglomerated crystals. We also assessed the solubility and dissolution rate of the spherically agglomerated crystals. Drug recovery in the agglomerated preparation was given particular attention.

Experimental Section

Spherical Crystallization Process—Spherical crystallization of indomethacin (γ form; supplied by Sumitomo Pharmaceutical Co., Ltd., Osaka, Japan) and epirizole (anhydrous; supplied by Daiichi Seiyaku Co. Ltd., Tokyo, Japan) was carried out by three methods to investigate the polymorphic transformation of the drugs during crystallization. The drugs and crystallization solvents used, ethanol, chloroform, and ethyl acetate, were Japanese Pharmacopeia X (JPX) or chemical reagent grades and were used without further purification.

Method 1—Indomethacin (1.0 g, 20–100 mol %) and epirizole (0–3.0 g, 0–80 mol %) mixtures were dissolved in ethanol (10 mL) in a stoppered test tube thermostatically controlled at 70°C. After the ethanol solution was cooled to room temperature, 5 mL was introduced into a dispersed mixture of chloroform (1.0–1.2 mL) plus water (15 mL), and the preparation was shaken at 200 to 400 strokes per min for 20 min in a horizontal shaker. The crystals produced were simultaneously agglomerated during the shaking and then separated and dried prior to measurement of the physicochemical properties.

Method 2—Indomethacin (1.0 g, 15–40 mol %) and epirizole (0.9–4.0 g, 60–85 mol %) mixtures were dissolved in ethyl acetate (9 mL) thermostatically controlled at 70°C. The ethyl acetate solution was cooled to 10°C and added to distilled water (100 mL), and the preparation was stirred for 20 min at 630 rpm with a turbine-type agitator with six blades. Two milliliters of the solvent sampled from the system was passed through a membrane filter (pore diameter, 0.3 μm) and then diluted with a solution of 30% aqueous ethanol. Indomethacin and epirizole in the diluent were assayed spectrophotometrically. Indomethacin was determined with a spectrophotometer (model 100-60; Hitachi Manufacturing Co., Ltd., Tokyo, Japan) at 320 nm. Epirizole was determined at 251 and 266 nm with a double-wavelength spectrophotometer (double-beam spectrophotometer model 556; Hitachi Manufacturing Co., Ltd.). The agglomerated crystals were separated and dried.

Method 3—Method 3 was a modification of method 2 in which the loadings of indomethacin and epirizole were 1.0 g (37–62 mol %) and 0.4–1.1 g (38–63 mol %), respectively, and the water was replaced with 100 mL of a solution of 10% aqueous sodium chloride.

Measurement of Physicochemical Properties of Agglomerated Crystals—The shape and surface topography of the resultant agglomerated crystals were determined with an optical microscope (model JM; Olympus Optical Co., Ltd., Tokyo, Japan) and a scanning electron microscope (model JSM-S1; Nihon Denshi Co., Ltd., Tokyo, Japan). The particle size of the agglomerated crystals was measured by sieve analysis. The crystalline form of the agglomerate was analyzed by X-ray diffractometry (model JDX; Nihon Denshi Co., Ltd.), infrared spectroscopy (model A-102; Nihon Bunko Co., Ltd., Tokyo, Japan), and diffuse reflectance spectroscopy (DRS) (model MPS-50L; Shimadzu Co., Ltd., Kyoto, Japan). Thermal degradation of the agglomerated crystals was determined by differential scanning calorimetry (DSC) (model DSC; Rigaku Denki Co., Ltd., Tokyo) with a heating rate of 10°C/min. The composition of the drugs in the agglomerated crystals was determined spectrophotometrically after dissolution in a solution of 30% aqueous ethanol, which was also used for assays of the drugs remaining in the crystallization solvent.

Dissolution Behavior of Agglomerated Crystals—The solubilities of the resultant agglomerated crystals in a solution of 30% aqueous ethanol and in the JPX disintegration test solution composed of 0.05 M KH_2PO_4 plus 0.0236 M NaOH adjusted to pH 6.8 were measured by shaking the agglomerated crystals dispersed in each solvent at 37°C. A 3-mL aliquot of the solution was sampled at suitable intervals through a pipette plugged with cotton and passed through a membrane filter (pore diameter, 0.3 μm). The ethanol solution of the drugs was assayed spectrophotometrically as described above. The disintegration of the test solution of the drugs was assayed spectrophotometrically after being diluted with the same

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volume of methanol as that of the solution sampled. A wavelength of 320 nm was used for indomethacin, and wavelengths of 250 and 267.5 nm were used for epirizole to avoid the absorption of indomethacin.

Dissolution Test of Tablets Prepared from Agglomerated Crystals—The dissolution test was conducted with a rotating disk method. Disks of the agglomerated crystals and a mixture of the native drugs were prepared by compression at 500 kg/cm^2 with a single punch tablet machine. A disk (diameter, 1.0 cm) was attached to the center of the lower face of a turbine-type stirrer (diameter, 5.0 cm) with epoxy resin. The surface area of the disk exposed to the dissolution medium was 0.79 cm^2. The stirrer with the disk centrally located 2.5 cm from the bottom of the dissolution test vessel specified in the JPX (corresponding to the paddle method of the U.S. Pharmacopeia) was rotated at 100 rpm in 900 mL of disintegration test solution 2. At suitable intervals, a 3-mL aliquot was passed through a membrane filter (pore diameter equal to 0.3 μm). A preparation of the same volume and the same temperature of the solvent was simultaneously placed in a vessel to maintain a constant volume. The drug content was assayed as described above.

Results and Discussion

Physicochemical Properties and Identification of Agglomerated Crystals—An optical micrograph of the resultant agglomerated crystals prepared in the ethyl acetate–water system (method 2) is shown in Fig. 1a. The double or triple agglomerates suggested that the crystals were formed by random coalescence. Scanning electron micrographs of the agglomerated crystals, indomethacin, and epirizole are shown in Fig. 1b, c, and d, respectively. Fig. 1b shows that these crystals were fine and needle-like, whereas the original indomethacin and epirizole crystals were flat and cubelike with round edges, respectively.

The compositions of indomethacin and epirizole in the agglomerated crystals prepared in the ethanol–chloroform–water system (method 1) were plotted against the loading mole percent of the drugs in the crystallization solvent shown in Fig. 2. The plots deviated from a straight line at a slope of

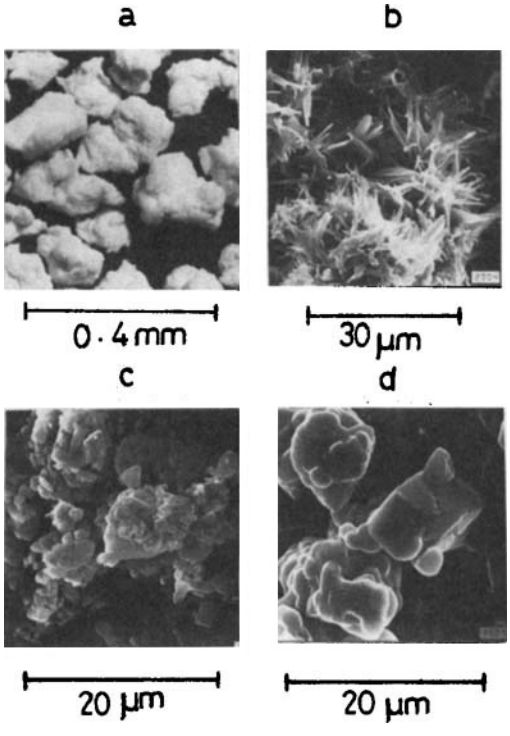

Figure 1—*Optical and scanning electron micrographs of raw drugs and agglomerated crystals prepared by method 2. Key:* (a) *and* (b) *agglomerated crystals;* (c) *indomethacin;* (d) *epirizole.*

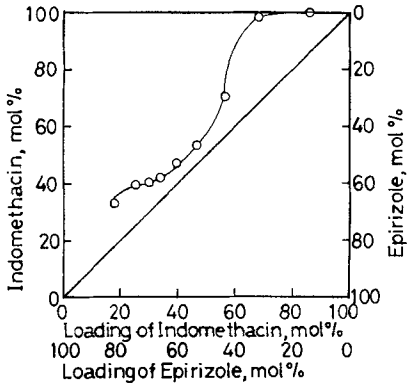

Figure 2—*Relationship between drug loading ratio and content ratio of the drugs in the agglomerated crystals prepared by method 1.*

45°, thereby indicating that the recovery of epirizole was less than that of indomethacin, which was thought to be due to the greater solubility of epirizole. When the loading mole percent of indomethacin exceeded 70%, epirizole was not crystallized. The infrared spectrum (Fig. 3d) and the X-ray diffraction pattern (Fig. 4e) of the agglomerated crystals prepared by method 1 were identical with that of the physical mixture of the β form of indomethacin and epirizole anhydrate in Fig. 3b and that of the β form of indomethacin in Fig. 4b, respectively. Characteristic diffraction peaks of epirizole in Fig. 4d never appeared in the pattern of the agglomerated crystals in Fig. 4e, suggesting that the agglomerated crystals prepared in the ethanol–chloroform–water system were a physical mixture of the β form of indomethacin (original form, γ) and of amorphous epirizole.

The ratios of indomethacin to epirizole in the agglomerated crystals prepared in the ethyl acetate–water solution (method 2) and the ethyl acetate–aqueous sodium chloride solution (method 3) systems were plotted against the loading mole percent of the drugs in the crystallization solvent. The ratios of indomethacin to epirizole in both agglomerated crystals were higher than in the solvents (Fig. 5). When equal loadings were used, we found that the content of epirizole in the agglomerated crystals prepared by method 3 was higher than that in the agglomerated crystals prepared by method 2. In method 3, a solution of 10% NaCl was used to decrease the

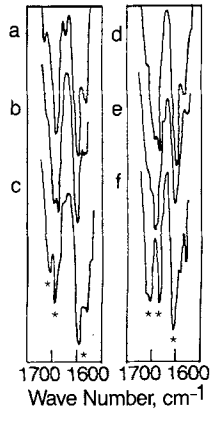

Figure 3—*Infrared spectra. Key:* (a) *physical mixture of α-indomethacin–epirizole;* (b) *physical mixture of β-indomethacin–epirizole;* (c) *physical mixture of γ-indomethacin–epirizole;* (d) *agglomerated crystals prepared by method 1;* (e) *agglomerated crystals prepared by methods 2 and 3 (mole percent of epirizole loaded is ≤63 and ≤38%, respectively);* (f) *agglomerated crystals prepared by methods 2 and 3 (mole percent of epirizole loaded is ≥65 and ≥43%, respectively). The asterisks indicate the characteristic peaks of each compound.*

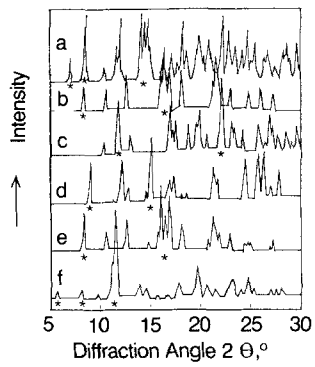

Figure 4—*X-ray diffraction patterns. Key:* (a) *α-indomethacin;* (b) *β-indomethacin;* (c) *γ-indomethacin;* (d) *epirizole;* (e) *agglomerated crystals prepared by method 1;* (f) *agglomerated crystals prepared by methods 2 and 3 (mole percent of epirizole loaded is ≥65 and ≥43%, respectively). The asterisks indicate the characteristic peaks of each compound.*

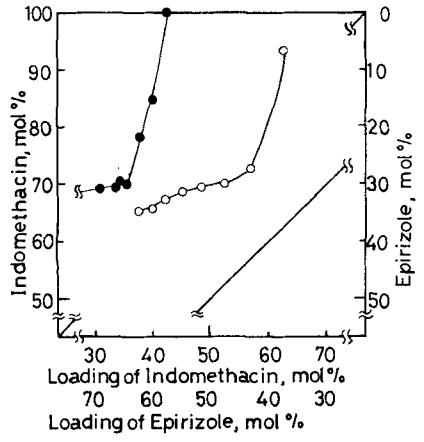

Figure 5—*Relationship between drug loading ratio and ratio of drugs in agglomerated crystals prepared by methods 2 and 3. Key:* (●) *without sodium chloride (method 3);* (○) *with sodium chloride (method 2).*

solubility of epirizole in the solvent by a salting-out effect. The recoveries of the drugs as agglomerated crystals are shown in Fig. 6. The recovery of indomethacin was always 100%, irrespective of the mole percent of the drug loaded in the solvent, whereas that of epirizole depended on the mole percent of the drug loaded. When a solution of aqueous NaCl was used, the recovery of epirizole increased, thereby leading to an increase in the total drug recovery. The curves of the drug composition in the agglomerated crystals versus the mole percent of the drug loaded reached a breakpoint of 63 mol % of epirizole loaded in method 2 and 43 mol % in method 3, respectively (Fig. 5). When the mole percent of epirizole loaded was higher than these values, the drug compositions in the agglomerated crystals were fairly constant. When the mole percent of epirizole loaded was less than these values, however, the content of indomethacin in the agglomerated crystals sharply increased and corresponded with the decrease in the mole percent of epirizole loaded. This finding suggests that the breakpoint in Fig. 5 is critical for determining the physicochemical properties of the agglomerated crystals.

The infrared spectrum in Fig. 3e of the agglomerated crystals prepared in the ethyl acetate–water system (method 2) when the mole percent of epirizole loaded was <63% was identical to that of the physical mixture of indomethacin (α form) and anhydrous epirizole in Fig. 3a. The infrared

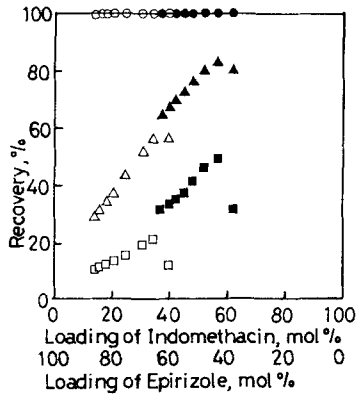

Figure 6—*Relationship between drug loading ratio and drug recovery in agglomerated crystals prepared by methods 2 (without sodium chloride; open symbols) and 3 (with sodium chloride; solid symbols). Key:* (○, ●) *indomethacin;* (□, ■) *epirizole;* (△, ▲) *whole product.*

spectrum of the agglomerated crystals prepared in the ethyl acetate–aqueous sodium chloride system (method 3) when the mole percent of epirizole loaded was <38% was also the same as that of the above physical mixture. X-ray analyses revealed the epirizole crystals to be amorphous.

The infrared spectrum (Fig. 3f) of the agglomerated crystals prepared by methods 2 and 3, when the mole percent of epirizole loaded was higher than 65 and 43%, respectively, differed from that of the other agglomerated crystals and the physical mixtures of polymorphic indomethacin and epirizole. We found that the absorption of the carbonyl stretching vibrations of the carboxyl group at 1715 cm^{-1} and that of the amido group at 1690 cm^{-1} of indomethacin shifted to 1705 and 1670 cm^{-1}, respectively. The absorption band of C = C or C = N stretching vibration of epirizole shifted to 1610 cm^{-1}. The X-ray diffraction pattern in Fig. 4f also differed from that of the other agglomerated crystals and physical mixtures. These findings suggest that the agglomerated crystals might be a new complex or a new polymorphic mixture of indomethacin–epirizole. The mole ratio of indomethacin to epirizole in the agglomerated crystals was approximately 2.

Results of the diffuse reflectance spectra (DRS), of the physical mixtures of γ-indomethacin–epirizole and the agglomerated crystals with a molecular ratio of indomethacin to epirizole of 2:1, prepared by method 2 or 3 are shown in Fig. 7. The DRS peaks of the agglomerated crystals and the physical mixtures appeared at 410 and 360 nm, respectively. The significant difference (>10 nm) in the wavelengths of the two peaks suggested interactions between indomethacin and epirizole molecules in the agglomerated crystals.[10] The agglomerated crystals with a molecular ratio of indomethacin

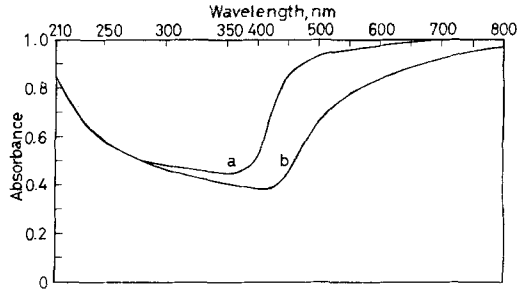

Figure 7—*Diffuse reflectance spectra. Key:* (a) *physical mixture of γ-indomethacin–epirizole;* (b) *agglomerated crystals prepared by methods 2 and 3 (molecular ratio of indomethacin to epirizole equal to 2:1).*

to epirizole of 2:1, prepared by methods 2 or 3, might therefore be a complex of the two drugs. The thermodiagram of the agglomerated crystals prepared by method 2 or 3, obtained with differential scanning colorimetry (DSC), is shown in Fig. 8. When the ratio of indomethacin to epirizole in the agglomerated crystals was 2:1, the wetting and melting points of the crystals coincided at 110°C. The DSC thermograms of the physical mixtures and the agglomerated crystals prepared by the three methods, with a molecular ratio of indomethacin to epirizole of 2:1, are shown in Fig. 9. The agglomerated crystals prepared by methods 2 or 3 exhibited only one endothermic peak in the DSC thermogram, whereas the physical mixtures and the agglomerated crystals prepared by method 1 revealed two endothermic peaks at 60–70°C and 110–120°C. The above finding indicates that the agglomerated crystals, which had a molecular ratio of indomethacin to epirizole of 2:1, were a new complex of indomethacin–epirizole with a melting point of 109 to 113°C.

Dissolution Behavior of the Agglomerated Crystals of a New Complex Prepared by Method 2—The dissolution behavior of indomethacin in the agglomerated crystals of the new complex prepared by method 2 and one physical mixture each of α-, β-, and γ-indomethacin–epirizole (molecular ratio of indomethacin to epirizole equal to 2:1) were investigated in a solution of 30% aqueous ethanol. The apparent solubility of indomethacin in the complex increased rapidly at the initial stage, reached a maximum (424 g/mL) after 30 min, and then decreased gradually to that of the mixture of γ-indomethacin–epirizole (Fig. 10). During the dissolution test, the complex was transformed into the mixture of γ-indomethacin–epirizole, resulting in a decrease in the solubility of indomethacin in the complex, as noted by other workers.[11–13] β-Indomethacin also was transformed to the γ form during the dissolution.

The change in the solubility of indomethacin found in the solution of 30% aqueous ethanol did not appear in JPX disintegration test solution 2. The solubility of indomethacin in the agglomerated complex increased gradually, with a dissolution time of up to 2.5 times that in the physical mixture of γ-indomethacin–epirizole (molecular mixing ratio equal to 2:1). The solubility of epirizole in the agglomerated complex was lower than that in the physical mixture, as shown in Fig. 11.

The drug release patterns of the tablets of the agglomerated complex crystals and the physical mixtures of α-, β-, and γ-indomethacin–epirizole (molecular mixing ratio, 2:1) in JPX disintegration test solution 2 are shown in Fig. 12. The release of indomethacin from the agglomerated crystals and the physical mixtures were described in terms of zero-order kinetics. The rate of release of indomethacin from the agglomerated crystals was three times more rapid that of the physical mixture of γ-indomethacin–epirizole and almost equal to that of the physical mixture of β-indomethacin

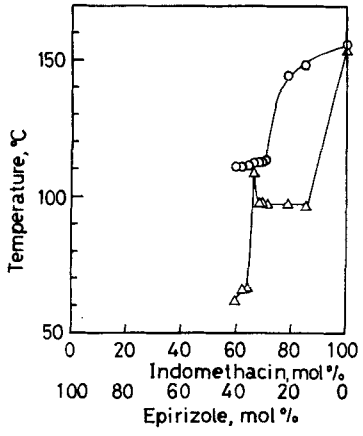

Figure 8—*Phase diagram for agglomerated crystals prepared by method 2, constructed by differential scanning calorimetry. Key:* (○) *end of melt;* (△) *beginning of melt.*

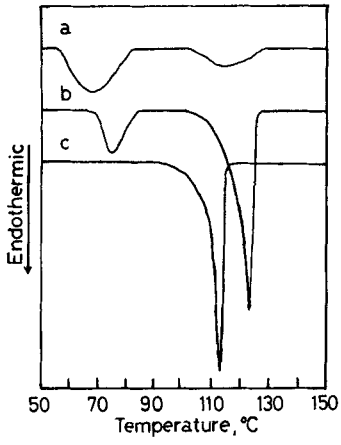

Figure 9—*Differential scanning calorimetry thermograms. Key:* (a) *physical mixture of β-indomethacin–epirizole;* (b) *agglomerated crystals prepared by method 1;* (c) *agglomerated crystals prepared by methods 2 and 3 (molecular ratio of indomethacin to epirizole equal to 2:1).*

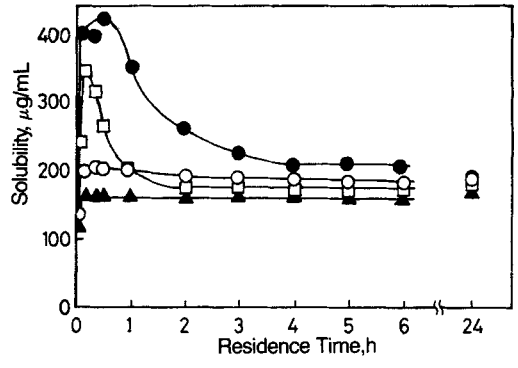

Figure 10—*Solubility of indomethacin in a solution of 30% aqueous ethanol at 37°C. Key:* (○) *physical mixture of α-indomethacin–epirizole;* (□) *physical mixture of β-indomethacin–epirizole;* (▲) *physical mixture of γ-indomethacin–epirizole;* (●) *spherically agglomerated crystals prepared by method 2.*

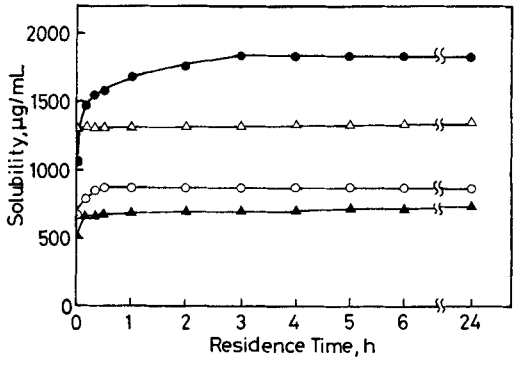

Figure 11—*Solubility of agglomerated crystals and physical mixture in Japanese Pharmacopeia X disintegration test solution 2 at 37°C. Key:* (▲) *indomethacin of physical mixture of γ-indomethacin–epirizole;* (△) *epirizole of physical mixture of γ-indomethacin–epirizole;* (●) *indomethacin of spherically agglomerated crystals prepared by method 2;* (○) *epirizole of spherically agglomerated crystals prepared by method 2.*

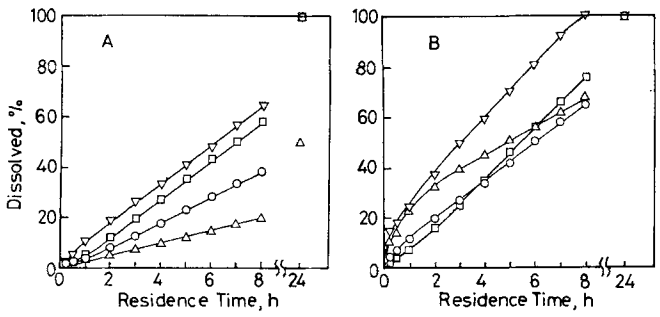

Figure 12—*Dissolution profiles of indomethacin* (A) *and epirizole* (B) *in JPX disintegration test solution 2 at 37°C by a rotating disk method. Key:* (○) *physical mixture of α-indomethacin–epirizole;* (▽) *physical mixture of β-indomethacin–epirizole;* (△) *physical mixture of γ-indomethacin–epirizole;* (□) *spherically agglomerated crystals prepared by method 2.*

(metastable form)–epirizole. The improved bioavailability of indomethacin in the agglomerated complex is probably due to the improved solubility and rate of release. The release of epirizole from the agglomerated crystals followed zero-order release kinetics, whereas that from the physical mixtures did not. The constant release of the new complex shown in Fig. 12 should aid in the avoidance of fluctuating concentrations of drug in blood, which occur as a result of variations in drug dissolution. The therapeutic effect of the complex should be enhanced, as concomitant administration of indomethacin and epirizole reduces the adverse effects of such fluctuations in concentration.[9]

References and Notes

1. Kawashima, Y.; Okumura, M.; Takenaka, H. *Science* **1982**, *216*, 1127–1128.
2. Kawashima, Y.; Aoki, S.; Takenaka, H. *Chem. Pharm. Bull.* **1982**, *30*, 1900–1902.
3. Kawashima, Y.; Naito, M.; Lin, S. Y.; Takenaka, H. *Powder Technol.* **1983**, *34*, 255–260.
4. Kawashima, Y.; Aoki, S.; Takenaka, H.; Miyake, Y. *J. Pharm. Sci.* **1984**, *73*, 1407–1410.
5. Kawashima, Y.; Okumura, M.; Takenaka, H.; Kojima, A. *J. Pharm. Sci.* **1984**, *73*, 1535–1538.
6. Yamamoto, H. *Chem. Pharm. Bull.* **1968**, *16*, 17–19.
7. Pakular, R.; Pichnej, L.; Sypychala, S.; Butkiewics, K. *Pol. J. Pharmacol. Pharm.* **1977**, *29*, 151–157.
8. Mitsui, J.; Kuroda, M.; Tsunakawa, N.; Takeya, Y. *Chem. Pharm. Bull.* **1971**, *19*, 2365–2372.
9. Tsurumi, K.; Kyuki, K.; Fujimura, H. *J. Pharm. Dyn.* **1980**, *3*, 659–666.
10. McGinity, J. W.; Lach, J. L. *J. Pharm. Sci.* **1976**, *65*, 896–902.
11. Higuchi, W. I.; Bernardo, P. D.; Metha, S. C. *J. Pharm. Sci.* **1967**, *56*, 200–207.
12. Nogami, H.; Nagai, T.; Yotsuyanagi, T. *Chem. Pharm. Bull.* **1967**, *17*, 499–509.
13. Griffiths, R. V.; Mitchell, A. G. *J. Pharm. Sci.* **1971**, *60*, 267–270.

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