Molecular Interaction between E-Prostaglandins and Selected Polymers and Its Potential Utilization in Oral Dosage Form Design

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Abstract □ Coacervate formation was observed between some E-prostaglandins and povidone in acetonitrile. This molecular interaction was studied using differential scanning calorimetry, IR spectrophotometry, and light microscopy. The structural requirements for coacervate formation between E-prostaglandins and povidone were investigated. Possible utilization of this molecular interaction in the development of E-prostaglandin formulations was explored. The dissolution rate of some insoluble E-prostaglandin esters increased when they were coprecipitated with povidone and polyethylene glycol. For example, the *p*-hydroxybenzaldehyde semicarbazone ester of 16,16-dimethyldinoprostone dissolved about 200 times faster as a povidone coprecipitate than did the control mixture. Enhancement of the dissolution rate was observed for the povidone coprecipitates of dinoprostone and its *p*-acetylphenyl and β-naphthyl esters but not for the *p*-phenylphenyl ester. Fast dissolving dispersions of the E-prostaglandin esters also could be prepared with the water-insoluble cross-linked polyvinylpyrrolidone. This type of dispersion was nonglassy and easily dispersible in water. Thus, it might have certain advantages over the classical soluble povidone coprecipitates in terms of ease of handling. The degree of enhancement in dissolution of dispersions of cross-linked polyvinylpyrrolidone and E-prostaglandin esters is apparently dependent on the structure of the esters. The potential dissolution enhancement may be related to the strength of the interaction between the macromolecule and the esters, as indicated by the qualitative relationship between the extent of adsorption of the prostaglandins to cross-linked polyvinylpyrrolidone and the dissolution rate enhancement.

Keyphrases □ Prostaglandins, various—molecular interaction with selected polymers, application to oral dosage form design □ Polymers—povidone and cross-linked polyvinylpyrrolidone, molecular interaction with various prostaglandins, application to oral dosage form design □ Molecular interaction—various prostaglandins with selected polymers, application to oral dosage form design

Dinoprostone (prostaglandin E_2) (I) is unstable in aqueous solutions and in the solid state at ambient or elevated temperatures (1–7). Various attempts have been made to improve the solid-state stability of I. These efforts can be broadly classified into two categories: the formulation (or physical admixture) approach and the chemical (or prodrug) approach. Examples of the former technique can be found in the patent literature. It has been claimed that povidone (II) (8), β-cyclodextrin (9), and sodium chloride and succinic acid (10) all stabilize I against dehydration to prostaglandin A_2. The prodrug approach was used to synthesize various higher melting esters; the stability of these derivatives was vastly improved over the parent E-prostaglandins (7).

BACKGROUND

Questions remain regarding the stabilization of I in the solid state. Except for β-cyclodextrin, where clathrate formation appears to be the stabilizing force, the stabilization mechanism for the physical admixture approach is unknown. If I can be stabilized when dispersed in such diverse classes of solids as the ionic sodium chloride, the hydrogen donor succinic acid, and the polymeric dipolar II, then stabilization may be derived from a purely physical dispersion of I in a diluent solid, independent of any chemical interaction between the prostaglandin and the diluent.

The solid-state degradation of I appears to be promoted through its dissolution in its own decomposition product, prostaglandin A_2 (a viscous liquid at room temperature). This mechanism was indicated by the observation of an initial induction period followed by rapid decomposition of I in the solid state (11). The major stabilization mechanism in these systems may then be the physical separation of molecules of I in the diluent so that prostaglandin A_2 can no longer affect the now distant and unreacted I molecules. If this mechanism is correct, then stabilization can, in principle, be achieved with a host of pharmaceutical solids, regardless of their chemical structures. But if stabilization from solid dispersion requires a specific chemical interaction, then the nature of this interaction is of interest so that other nontoxic compounds capable of the same interaction can be systematically screened as I stabilizers.

In the prodrug approach, the improvement of the stability of E-prostaglandins is related directly to the elevation of the melting point of I through prodrug formation: the higher the melting point of the prodrug, the more stable the derivative (7). Unfortunately, a significant increase in the melting point drastically decreases aqueous solubility. This decrease, in turn, probably reduces the dissolution rate, potentially diminishing the *in vivo* oral absorption of the compound.

The following question can then be asked: Is it possible to increase the dissolution rates of the higher melting prodrugs so that both the stability and dissolution criteria can be simultaneously fulfilled? Polymer II is an ideal test compound through which this question may be probed. Its ability to improve the solid-state stability of I is well substantiated (8), and the molecular interaction between II and other organic compounds was studied extensively (12, 13). Also, II can promote the dissolution rate of poorly soluble drugs through the formation of coprecipitates (14). Thus, it appears that II is a unique chemical and may favorably influence both the stability and the dissolution of E-prostaglandins. A cross-linked polyvinylpyrrolidone polymer (III) with extremely limited aqueous solubility is also available. The interaction between this polymer and E-prostaglandins also is of interest.

The present report represents initial attempts to explore the molecular interaction between E-prostaglandins and selected macromolecules. The potential utilization of this interaction in the design of stable and fast dissolving oral dosage forms of E-prostaglandins also was investigated.

EXPERIMENTAL

Reagents and Materials—Methyl (IV), decyl (V), *p*-phenylphenyl (VI), *p*-acetylphenyl (VII), β-naphthyl (VIII), and *p*-hydroxybenzaldehyde semicarbazone (IX) esters of E-prostaglandins were synthesized according to Morozowich (7). The polymers used were II[1], III[2], polyethylene glycol 4000[3], and silanized silica gel 60[4]. The acetonitrile used was distilled in glass[5].

Coacervate Formation—Screening for coacervate formation was carried out by mixing solutions of prostaglandin (10 mg in 1 ml of acetonitrile) and II (100 mg in 1.5 ml of acetonitrile). Coacervate formation was positive when a dense oily layer was formed after mixing. Three different ratios, 1:5, 1:10, and 1:20, of I to II were tested.

Preparation of Coprecipitates of Prostaglandins with II—Coprecipitates of II generally were prepared in a 1:10 ratio of prostaglandin to polymer. The components were separately dissolved in acetonitrile and mixed. The solvent was removed first by a nitrogen stream

[1] Plasdone K 29-32, control No. G-30121B, GAF Corp., New York, NY 10020.
[2] Polyplasdone XL, control No. 906, GAF Corp., New York, NY 10020.
[3] Carbowax 4000, Union Carbide Chemicals, New York, NY 10017.
[4] RP-2, 63–200-μm size, fine free, E.M. Laboratories, Elmsford, NY 10523.
[5] Burdick & Jackson Laboratories, Muskegon, MI 49442.

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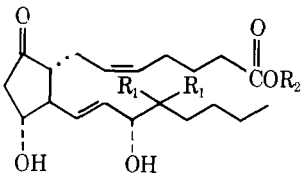
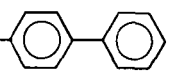


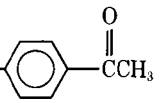
I: $R_1 = R_2 = H$

IV: $R_1 = H$, $R_2 = CH_3$

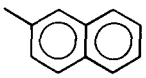
V: $R_1 = H$, $R_2 = (CH_2)_9CH_3$



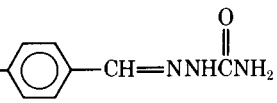
VI: $R_1 = H$, $R_2 =$



VII: $R_1 = H$, $R_2 =$



VIII: $R_1 = H$, $R_2 =$



IX: $R_1 = CH_3$, $R_2 =$

and then in a flash evaporator. The glassy film left was scraped from the container.

Preparation of Dispersions of Prostaglandins in III—Dispersions of III were generally prepared in a 1:100 ratio of prostaglandin to III. Compound III was dispersed in acetonitrile to which a solution of prostaglandin in acetonitrile was added. The solvent was then removed in a flash evaporator.

Dissolution Studies—The dissolution apparatus used for UV-absorbing esters consisted of a jacketed beaker maintained at 37 ± 0.5°. The powder was introduced into the dissolution medium (water), and the mixture was stirred at 150 rpm by a magnetic bar. The solution was removed continuously from the dissolution system through a sintered-glass filter of medium porosity *via* a peristaltic pump, passed through a flowcell *placed inside a spectrophotometer*[6], and returned to the dissolution system. The flow rate was approximately 10 ml/min.

The concentrations of prostaglandin esters were monitored at the following wavelengths: VI, 250 nm; VII, 254 nm; VIII, 273 nm; and IX, 282 nm. No rigorous attempts were made to control the particle size of the powders. For I (which has insignificant UV absorption above 240 nm), manual removal of 2-ml aliquots of the dissolution medium at different times was carried out *via* pipets, the tips of which had been wrapped with

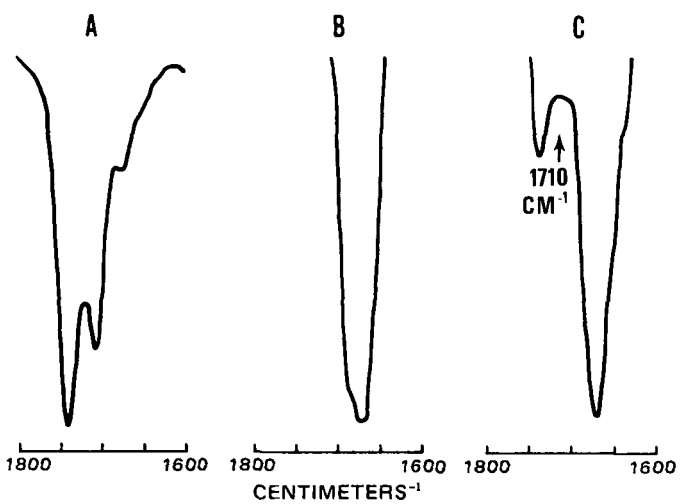


Figure 1—*Absorption peaks due to the carbonyl stretching vibration of I (A), II (B), and a 1:1 coacervate of I and II (C). Spectra were recorded in chloroform.*

[6] Beckman DB-G.

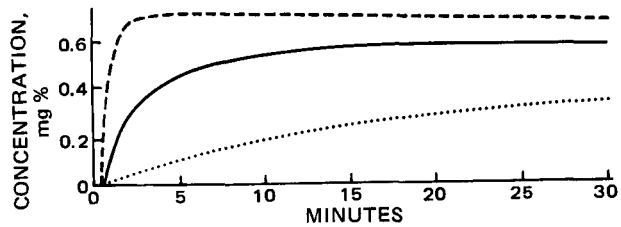


Figure 2—*Dissolution behavior of various forms of VII. Key: - - -, 20.2 mg of a 1:10 coprecipitate of VII and II; —, 20.4 mg of the corresponding physical mixture; and ···, 2.0 mg of pure VII.*

glass wool. When these aliquots were cooled to room temperature, 2 ml of 2 *N* KOH in methanol was added to each tube, and the prostaglandin B_2 formed was measured spectrophotometrically at 280 nm (4). The I concentration was calculated from a calibration plot established with known I concentrations.

Adsorption Studies—Different weights of III were introduced into test tubes or erlenmeyer flasks, and a known volume of the test drug solution in acetonitrile was added and equilibrated with the powder. The equilibrium concentrations of prostaglandin esters, tannic acid, and caffeine in the supernates were determined by UV spectrophotometry. For supernates of I, acetonitrile was removed by a nitrogen stream. The solutions were then reconstituted with methanol and assayed by conversion to prostaglandin B_2 with addition of 2 *N* KOH (4).

RESULTS AND DISCUSSION

Coacervate Formation and Characterization—On mixing solutions of I and II in acetonitrile as described, the mixture became turbid on standing and a dense oily layer settled out. This "oil" could be separated and, by repeated drying in a flash evaporator, "crystallized" into glassy solids. Since the limiting solubility of either I or II in acetonitrile was far in excess over the respective concentration present before mixing, this observation strongly suggested the existence of a molecular interaction between I and II.

The dense oil could be rationalized as being either the stoichiometric complex of I and II or a coacervate formed by the two molecular species. The latter is defined as a colloid-rich layer formed by separation of a macromolecular solution into two phases. A classical example of this phenomenon is the coacervation of gelatin when its solution is mixed with solutions of acacia, alcohol, sodium sulfate, or starch (15). Although the present case involves phase separation from a nonaqueous, albeit polar, medium, the term coacervate is still used because it facilitates conceptualization of the experimental observation.

A number of observations supported the presence of coacervate formation over that of complexation. First, when different weight ratios of I to II (1:5, 1:10, and 1:20) were mixed in acetonitrile, the volume of the dense oil appeared to increase with increasing amounts of II. The actual volumes were much greater than those that could be expected from I and II alone. Thus, the separated phase must have contained significant numbers of solvent molecules, which was consistent with coacervate behavior.

Second, the glassy solids obtained were analyzed with respect to the content of I. The ratios of I to II added to those recovered in the glassy solids were identical within experimental errors; for the ratios added (1:5, 1:10, and 1:20), the ratios recovered were 1:6.6, 1:10.6, and 1:20.0, respectively. If the glassy solids isolated were stoichiometric complexes of I and II, one would reasonably expect a somewhat constant ratio of I to II in the complexes recovered. It could, however, be reasoned that a mixture of stoichiometric complexes was formed and that alteration in the ratio of I to II added would affect the relative contribution of each

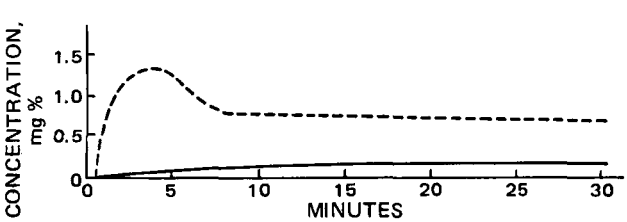


Figure 3—*Dissolution behavior of various forms of VIII. Key: - - -, 21.1 mg of a 1:10 coprecipitate of VIII and II; and —, 22.1 mg of the corresponding physical mixture.*

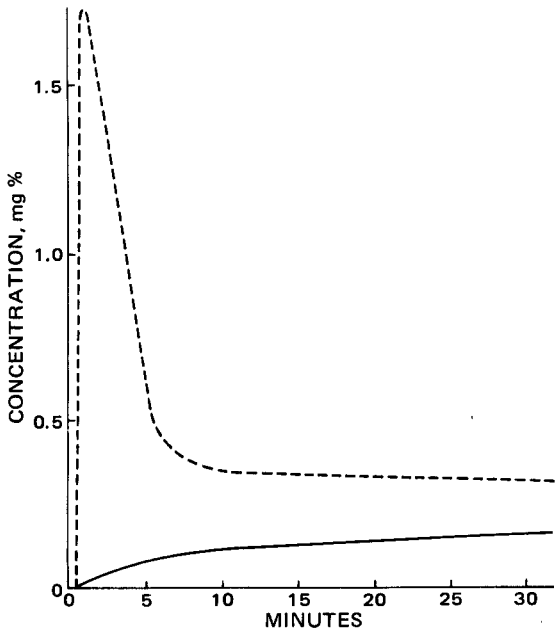


Figure 4—*Dissolution behavior of various forms of IX. Key: - - - , 44.0 mg of a 1:10 coprecipitate of IX and II; and —, 43.8 mg of the corresponding physical mixture.*

stoichiometric complex to the glassy solid, which, in turn, would affect the ratio recovered.

If this were the case, then successive washing of the glassy solids with acetonitrile, particularly those formed from a high ratio of II to I, would change the drug to polymer ratio in the residue solid, because different stoichiometric complexes could be presumed to have different solubilities in acetonitrile. This, however, was not found to be the case. The glassy solid obtained from a 1:20 I to II coacervate was washed with acetonitrile twice. The ratio in the residue solid remained constant at 1:20 after each washing.

Various compounds, including some C_1-esters of I, were tested for coacervate formation with II. Only one weight ratio of test compound to polymer (1:10) was used. Thus, although the presence of coacervation with any one test compound gave a reasonable indication of molecular interaction, the opposite was, of course, not necessarily true. However, because of the similarity in molecular weights of the prostaglandins, those prostaglandins that did form coacervates might have stronger interactions with II than those that did not. Under the conditions used, esters IV–VI and VIII failed to form coacervates with II in acetonitrile while ester IX did.

Nonprostaglandin compounds, such as tannic acid and cholic acid, also

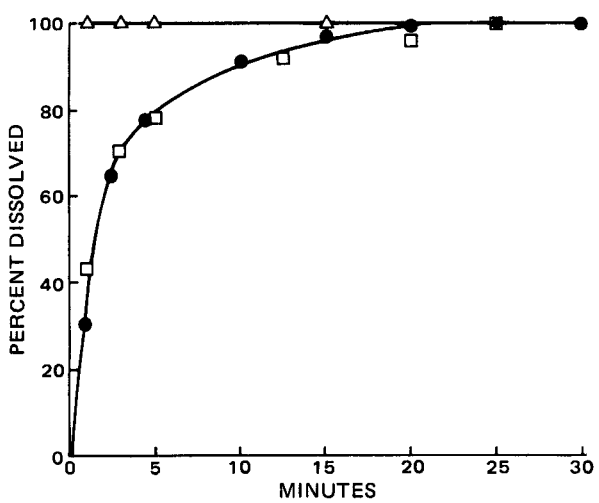


Figure 5—*Dissolution behavior of various forms of I. Key: △, 1.0 g of a 1:100 dispersion of I in III; □, 1.0 g of the corresponding physical mixture; and ●, 10 mg of pure I.*

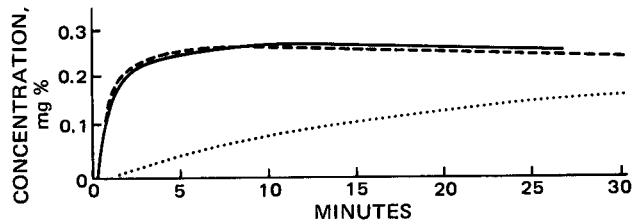


Figure 6—*Dissolution behavior of various forms of VII. Key: - - - , 200 mg of a 1:100 dispersion of VII in III; —, 202 mg of the corresponding physical mixture; and ···, 2.0 mg of pure VII.*

formed coacervates with II in acetonitrile, but acetic acid did not. Since organic acids are known to be strong interactants with II (12), blocking of the COOH group in I by ester formation might have weakened the molecular attraction between the prostaglandin and polymer molecules so that coacervation in acetonitrile was no longer possible. Although IX is also an ester, it possesses three active hydrogen atoms at the semicarbazone moiety. These hydrogen atoms might be able to participate in hydrogen bonding, establishing the interaction of the molecule with II.

Participation of the terminal COOH group of I in the molecular interaction with II was also consistent with the IR data of the I–II (1:1) coacervate. Shown in Fig. 1 are the carbonyl absorption peaks of I, II, and the I–II (1:1) coacervate in chloroform. For I, the absorption peaks due to the carbonyl stretching vibration at 1740 and 1710 cm^{-1} were assigned to the cyclopentanone carbonyl and the carboxylic carbonyl groups of I, respectively (16). The carbonyl group in the pyrrolidone ring of II gave an absorption peak at 1675 cm^{-1}. In the I–II (1:1) coacervate, the carboxylic carbonyl peak at 1710 cm^{-1} became undetectable. This result is consistent with the interpretation that the carboxylic hydrogen participates in the I–II interaction, because proton donation would impart a more carboxylate-like structure to I, shifting the carboxylic carbonyl peak to a lower wave number (17) and causing it to merge with the pyrrolidone peak of II.

The contribution from II toward the molecular interaction can also be examined. Compound II has two potential sites for interaction: the lactam linkage, which is available for hydrogen bonding and dipole–dipole interaction, and the alkyl backbone, which is available for hydrophobic bonding. Caffeine possesses two cyclic amide linkages, and it did not form a coacervate with I. This result is perhaps expected, because caffeine is not a polymer. Differential scanning calorimetry[7] of a 1:1 molar ratio of a I to caffeine mixture and coprecipitate showed, however, that the melting behavior of I was unaffected by caffeine. It appears then that the lactam group in II was not the sole contributor to the I–II interaction; hydrophobic bonding between the alkyl backbones of I and II may also have strengthened the interaction.

From these studies, it is apparent that potential stabilizers of I in the solid state may come from compounds containing a polar portion capable of hydrogen bonding and a nonpolar portion capable of hydrophobic bonding, *e.g.*, dialkyl-substituted amides, long chain esters, and surfactants.

Dissolution of II–Prostaglandin Coprecipitates—Coprecipitates of II and prostaglandin esters were prepared, and their dissolution profiles were determined using the setup described under *Experimental*. Figures 2, 3, and 4 show the representative dissolution curves of the coprecipitates and physical mixtures of II with VII, VIII, and IX, respectively. At least duplicate runs were performed for each compound, and these dissolution curves were all internally consistent. In all dissolution curves, there was an initial lag time of less than 1 min, corresponding to the time required for the dissolution medium to travel from the beaker to the flowcell.

Examination of the dissolution curves shows that, with these esters, the coprecipitates were much faster dissolving than the corresponding control physical mixtures. For example, the dissolution rate of the coprecipitate of IX between 0 and 2 min was about 200 times faster than its physical mixture within the same time interval. The enhancement in dissolution rate was less for esters VII and IX. For VI (absorptivity at 250 nm = 38.0 ml/mg cm in methanol), both the coprecipitate and physical mixture showed negligible absorbance at 250 nm for as long as 40 min after the start of the dissolution runs. The presence of the prostaglandin ester in these samples was confirmed by UV spectrophotometry, suggesting that the lack of absorbance was due to the poor solubility of

[7] DuPont model 900 thermal analyzer.

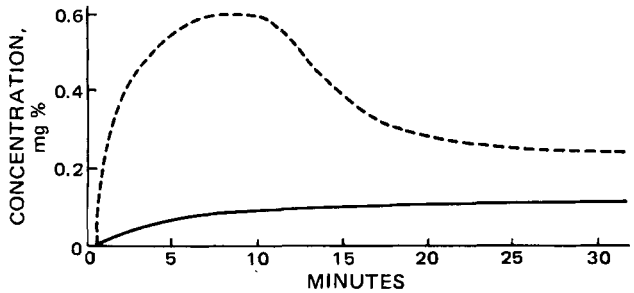


Figure 7—*Dissolution behavior of various forms of IX. Key: - - -, 200 mg of a 1:100 dispersion of IX in III; and —, 202 mg of the corresponding physical mixture.*

this compound even in the coprecipitate rather than to the absence of drug in the samples tested.

The coprecipitate of IX with II was examined under a hot-stage microscope[8], and its melting behavior was compared to its physical mixture. As expected, melting of IX could be distinctly observed in the physical mixture but not in the coprecipitate. The coprecipitate appeared as a homogeneous glassy solid and started to soften at about 180°. These thermal and microscopic data suggested that prostaglandin esters formed solid solutions with II in the coprecipitate. Enhancement of the dissolution of IX also was observed for its coprecipitate with polyethylene glycol 4000 but not for the coprecipitate with silanized silica gel 60[4], a methyl-substituted silica gel used in reversed-phase chromatography (18).

The ineffectiveness of silanized silica gel to improve the dissolution rate of IX suggested that the driving force for dissolution enhancement did not derive entirely from physical adsorption of the prostaglandin ester on the diluent powder matrix. Molecular interactions between the prostaglandin and the diluent, such as those provided by II and polyethylene glycol, appeared to be essential in promoting dissolution of prostaglandin esters in coprecipitates. The influence of particle size on the enhancement of coprecipitate dissolution was not examined.

Dissolution of III–Prostaglandin Dispersions—Polymer III has been used as a tablet disintegrant (19) and as a stabilizer for nitroglycerin in sublingual tablets (20). Unlike ordinary II, a glassy solid very soluble in water and many organic solvents, III is powdery and is insoluble in common solvents. When preparing coprecipitates with II, evaporation of the solvent usually led to formation of a film, which had to be scraped from the surface of the container. The film-like pieces obtained were usually very light and difficult to handle.

In contrast, dispersions of prostaglandins in III were nonglassy and were easier to handle. This type of dispersion has not been reported. It was of interest to explore whether dispersions of prostaglandin esters in III could enhance dissolution as was found for the coprecipitates with II.

Figures 5, 6, and 7 show the dissolution curves of the dispersions and physical mixtures of III with I, VII, and IX, respectively. With I and VII, the dissolution curves of the pure drugs were included for comparison. No appreciable dissolution for pure IX was detected under the experimental conditions. From Fig. 5, it is apparent that dissolution of the dispersion of I with III was extremely fast; complete dissolution was observed at 1 min, which was the first data point obtained. There was essentially no difference between the dissolution profiles of the physical mixture and pure I.

Similarly, the dissolution of the dispersion of IX was significantly enhanced compared to its physical mixture (Fig. 7). The dispersion of IX with III appeared to dissolve more slowly than the coprecipitate of the same drug (Fig. 4). At the same time, however, the III with IX dispersion sample, once dissolved, also precipitated out more slowly than the II with IX system. If the dissolution profiles are indicative of the corresponding absorption rates, then the dispersion of VII with III may be more desirable in oral dosage forms because of its ability to provide a more sustained increase in solubility vis-à-vis the corresponding coprecipitate with II.

In sharp contrast to the dispersions of IX, those of VI, VII (Fig. 6), and VIII provided no noticeable increase in dissolution rates over their respective control physical mixtures. In fact, the dissolution rates of both the dispersions and physical mixtures of VI and VIII were so slow that no detectable absorbance could be measured during the dissolution runs up to 30 min.

[8] Leitz Ortholux microscope equipped with a Mettler FP2 hot stage.

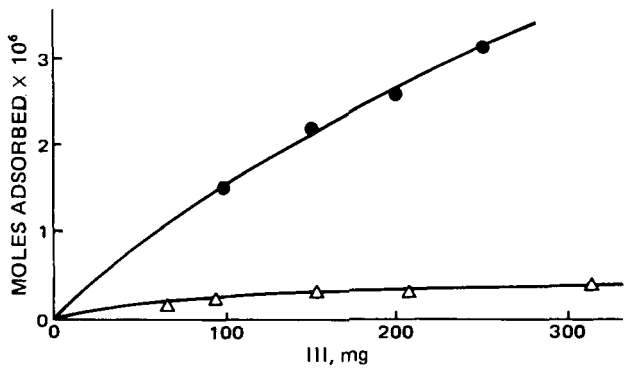


Figure 8—*Adsorption of I and VI–IX as a function of the weight of III in acetonitrile at 25°. Key: ●, adsorption of I; and △, adsorption of IX. Compounds VI–VIII gave no measurable adsorption.*

Correlation between Adsorption of Prostaglandin Esters on III and Dissolution Enhancement—Not all of the coprecipitates and dispersions were capable of enhancing the dissolution of all of the prostaglandins. In coprecipitates with II, the dissolution was not affected in the *p*-phenylphenyl ester. In dispersions with III, the dissolution of three prostaglandin esters (the *p*-phenylphenyl, β-naphthyl, and *p*-acetylphenyl) was not significantly affected by dispersion formation. The substituent apparently played an important role in determining whether coprecipitate and dispersion formulation could affect dissolution favorably. The effect of structure on the dissolution rates of coprecipitates and dispersions has not been reported. This attempt to relate the molecular interactions of various prostaglandin structures with III to the observed dissolution behavior may represent the first study in this area.

Preliminary adsorption studies of prostaglandins with III in acetonitrile revealed that compounds whose dispersions showed enhanced dissolution were also significantly adsorbed onto III. In reverse, prostaglandin esters whose dispersions did not show increased dissolution were not adsorbed by the polymer (Fig. 8). The degree of adsorption was possibly a reasonable reflection of the extent of molecular interaction between the prostaglandins and III because:

1. The adsorption data with III appeared to parallel those on coacervate formation: prostaglandins that showed strong adsorption on III (I and IX) also formed coacervates with II, and vice versa.

2. Tannic acid, a known interactant with II (12), showed extremely strong adsorption on III (Fig. 9) whereas caffeine, a known noninteractant with II (21), did not show any appreciable adsorption.

The adsorption characteristics of tannic acid and I on III (Fig. 9) could be described according to the classical Langmuir adsorption isotherm: $C/N = 1/Km + C/m$, where C is the equilibrium concentration of the adsorbate, N is the number of moles of adsorbate adsorbed per gram of III, K is the Langmuir adsorption constant, and m is the number of moles of adsorbate that 1 g of III can adsorb when the monolayer is complete. The numerical values of K and m were 9.01×10^5 and 1.11×10^{-5} for tannic acid and 1.58×10^4 and 2.34×10^{-5} for I, respectively. Mourey *et al.* (22) recently showed that the molecular interaction between azo

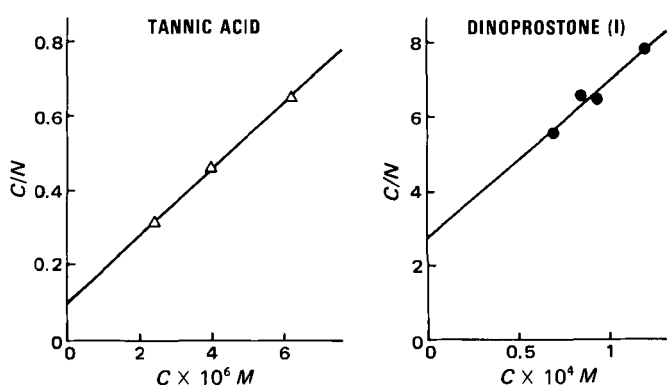


Figure 9—*Langmuir adsorption isotherm of tannic acid and I on III in acetonitrile at 25°.*

compounds and III in aqueous solutions is structure dependent. The presence of polar functional groups (*e.g.*, hydroxyl, amino, carboxylic, and sulfonic groups) in unionized forms facilitates interaction with the polymer.

The thermodynamic driving force for dissolution enhancement in prostanglandin dispersions appeared to derive from molecular interaction (in this case, adsorption) of the prostaglandin with III. This adsorption enabled the breaking up of the crystal lattice of the prostaglandin, thus requiring less energy input for dissolution. When no adsorption took place, the prostaglandin probably precipitated out by itself on evaporation of the acetonitrile, existing as separate particles unassociated with the polymer. These precipitates would essentially have the same physical characteristics as the physical mixtures and, therefore, would not be expected to show enhancement in dissolution.

It appears feasible to predict whether a certain prostaglandin may benefit in dissolution from dispersion formation with III. When a prostaglandin interacts with III, as indicated by significant adsorption, the dispersion formed probably will exhibit an increased dissolution rate. Prostaglandins that show poor interaction with III probably will not improve their dissolution rates through dispersion formation.

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Formation of a Cyclic Derivative of Ethacrynic Acid with Diazomethane

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Abstract □ Samples of ethacrynic acid were treated with methanol–hydrochloric acid or with diazomethane. GLC and mass spectrometric analysis indicated that the methanol–hydrochloric acid reaction gave the expected methyl ester, whereas diazomethane treatment gave a compound containing an additional 14 mass units. Accurate mass measurement and PMR and IR spectra showed that this product was a cyclic derivative of the methyl ester of ethacrynic acid, methyl 4-(2,3-dihydro-4-ethyl-5-furyl)-2,3-dichlorophenoxyacetate. Either derivatization method can be used for development of an assay for ethacrynic acid.

Keyphrases □ Ethacrynic acid—reaction with methanol–hydrochloric acid or diazomethane, PMR, IR, and GLC–mass spectral analyses of products □ GLC—analysis, derivatives of ethacrynic acid □ Derivatization—ethacrynic acid with methanol–hydrochloric acid or diazomethane, PMR, IR, and GLC–mass spectral analyses of products □ Diuretics—ethacrynic acid, reaction with methanol–hydrochloric acid or diazomethane, PMR, IR, and GLC-mass spectral analyses of products

Ethacrynic acid (I) is a potent, orally active diuretic (1–4) for which a suitable GLC procedure was required for investigating its pharmacokinetic profile in humans. Previously reported analytical procedures involved the administration of ^{14}C-I followed by a tedious workup using TLC or column chromatography (4, 5).

Ethacrynic acid is not amenable to direct GLC analysis, and an approach was to convert it into its methyl ester either by treatment with diazomethane or by reaction with

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