

Polymeric Pharmaceutical Coating Materials I

Preparation and Properties

By LEWIS C. LAPPAS and WAYNE MCKEEHAN

Several polymeric materials were prepared for evaluation as potential enteric and sustained-release coatings. Three methods were used to prepare some aliphatic partial esters of copolymers of maleic anhydride with ethylene or vinyl methyl ether. Infrared and electrometric titration techniques were employed for identification and characterization of the raw material and the resultant esters. The physical data presented demonstrate relative differences in solubility and related parameters due to modification of ester chain length and degree of esterification. The relationship of ester chain length and degree of esterification is direct for dissolution pH and inverse for moisture diffusion rate, moisture sorption, and water miscibility. No significant evidence of thermal instability was found after aging at 50°. Preliminary toxicological investigations indicate that these compounds are nontoxic.

CONSIDERABLE evidence exists in the literature to demonstrate the effect of formulation on the efficacy of orally administered pharmaceuticals. Various physical and physicochemical properties of the formulation contribute directly to the release and subsequent absorption of the drug to affect its onset, intensity, and duration of action (1-3).

Schanker has pointed out that drugs are not absorbed uniformly throughout the gastrointestinal tract (4). The dissociation of the drug and/or its solubility in the pH gradient of the gastrointestinal tract influence absorption markedly. To achieve more effective absorption, it appears that some degree of control over the site of drug release is desirable. Such control is particularly important for those drugs which are primarily absorbed in the intestine, *e.g.*, enteric and sustained-release formulations. Many previous publications on enteric formulation did not give adequate consideration to the significance of these factors.

Currently marketed enteric products are designed to release their medicament based on time, pH, or enzymatic action; time is the primary parameter for sustained-release products. Generally, the trend has been to establish a single enteric coating for all types of drugs. Coating ingredients are frequently natural products such as fats, waxes, and cellulose derivatives which do

not readily control drug release in specific areas of the intestinal tract.

In view of these considerations, this work was initiated to develop a series of stable, synthetic films which dissolve at various pH values within the pH range of the intestinal tract. This would permit selection of specific coatings to optimize absorption of drugs from various areas of the intestine. A series of partial esters of maleic anhydride copolymers with ethylene or vinyl methyl ether were prepared. Previous reports have communicated the development and utility of such films as potential enteric and sustained-release coatings (5-9). Relative differences in chemical and hydrophobic properties between these compounds were demonstrated by the measurement of dissolution pH, turbidity point, moisture sorption, and water vapor transmission.

EXPERIMENTAL

Materials.—The two copolymers, poly(ethylene/maleic anhydride) (PEMA) and poly(vinyl methyl ether/maleic anhydride) (PVM/MA), are commercially available from Monsanto Chemical Company and General Aniline and Film Corporation, respectively.¹ Reagent grade materials were used unless noted otherwise. Ethyl alcohol, SD3A absolute, and *n*-hexyl alcohol (b.p. 155-157.5°) were used to prepare the ethyl and *n*-hexyl esters. Potassium nitrate and deionized water were used in preparation of the 92% relative humidity cell (10). Polymer films were cast from acetone solutions onto a triple-distilled C. P. mercury substrate. Anhydrous magnesium perchlorate was used as the desiccant in the water vapor transmission studies.

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¹ Polymer grades used were DX-840-11, -21, -31 for PEMA and Gantrez AN-119, -139, -169 for PVM/MA.

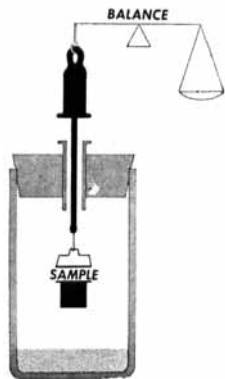


Fig. 1.—Humidity chamber—schematic diagram illustrating weighing position.

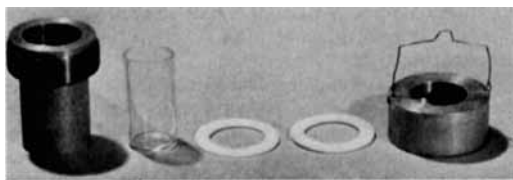


Fig. 2.—Permeation cell—component parts: magnesium cap and body, Teflon gaskets, and glass desiccant vessel.

Moisture Sorption Apparatus.—Figure 1 shows the humidity chamber which is similar to that described by Evans and Critchfield (11). Modification of this apparatus with a glass balance hook and polished glass joint permitted rapid weighing with negligible disturbance of the environment. Platinum wire was used to attach the film to the balance hook.

Water Vapor Transmission Apparatus.—The humidity chamber was the same as that described for the moisture sorption determination. Figure 2 illustrates the permeation cell which is similar to one described by Patel *et al.*, except that it is cylindrical and constructed entirely of magnesium (12). Flat Teflon gaskets, used in place of rubber O-rings, were reamed to exact size to give a diffusion area 1.588 cm. in diameter. A separate magnesium body with holes in the side and bottom was used to dry the films *in vacuo*. The permeation cell was suspended from the hook in the humidity chamber by a platinum wire to permit rapid weighings with minimal environmental disturbance.

Esterification Procedures.—Three general procedures were used to prepare the half-esters of PEMA (DX-840-11, -21, -31) and PVM/MA (Gantrez AN-119, -139, -169). One method consisted of slurrying one part, by weight, of copolymer with four parts, by volume, of *n*-alkanol and refluxing for 3 to 4 hr. After approximately 1 hr. at reflux temperature, an opaque, gelatinous mass formed which became a brilliantly clear, light yellow solution on continued heating. The half-ester was isolated by evaporating the resultant solution to dryness at 60° *in vacuo* in a stainless steel tray. Half-esters from *n*-propyl to *n*-decyl were prepared readily by this method. Synthesis of the one-half ethyl and isopropyl esters required reflux periods of 1–2 days and 4–5 days, respectively.

In the second method, esterification was accomplished by reacting the above ratio of copolymer

to alcohol in a Paar bomb under pressure. Half-esters from ethyl through *n*-octyl and the isopropyl half-ester were produced in 4 hr. at 125°.

The third method consisted of boiling the appropriate alkanol with the copolymer in an open beaker. This method was restricted to *n*-butanol and other higher-boiling alkanols. Complete reaction to the half-ester was achieved in 3 to 4 hr.

Dissolution pH Procedure.²—Twenty-five milliliters of a 5% w/v acetone solution of a partial ester was poured onto a mercury substrate in a 3.5-in. Petri dish and allowed to air-dry 18 hr. at room temperature. The films were removed and dried an additional 18 hr. at 45° *in vacuo*. About 20 mg. of the dried film, which measured approximately 0.006 in. thick, was placed into each of ten 12 × 150 mm. test tubes. Ten milliliters of a citrate–borate–phosphate buffer solution at 0.25 pH-unit increments was added to each tube (13). The tubes were stoppered and stored at room temperature with gentle, periodic agitation for 24 hr. Dissolution pH was determined by noting the lowest pH which dissolved the film completely. The films remained intact at a 0.25 pH unit lower than the dissolution pH.

Turbidity Point Procedure.—A 2.00% solution of the half-ester was prepared using reagent grade acetone. Five milliliters of this solution was pipeted into a 12 × 150 mm. test tube and titrated with deionized water to the production of a stable cloudiness. Turbidity point is defined as the milliliters of water used to attain this endpoint.

Moisture Sorption Procedure.—Films of the PVM/MA half-esters and isopropyl PVM/MA with greater than 40% esterification were prepared by casting 25 ml. of a 5% w/v acetone solution onto a mercury substrate in a 3.5-in. Petri dish. These conditions gave dried films which were approximately 0.006 in. thick. Half-esters of PEMA and isopropyl esters of PVM/MA with less than 40% esterification required film supports. Such supports were made by submerging two 1 × 1.5 in. strips of nylon bolting cloth (170 mesh) in the polymer solution as it was cast onto the mercury substrate. To achieve uniform films with smooth surfaces, the acetone solution was allowed to evaporate slowly at room temperature by placing the Petri dish inside a partially covered 6-in. high glass cylinder. Two 1 × 1.5 in. strips were cut from the center portion of the film while it was still plastic. Each strip was hung lengthwise on the balance hook in separate humidity chambers containing Drierite. The chambers were vented and placed in the oven at 45° *in vacuo* for 24 hr. After cooling to room temperature, the films were transferred to 100% relative humidity chambers for overnight storage at 25°. Then the films were redried over Drierite as before, and an initial dry-weight was obtained for either the film or the film-and-nylon support. For the determination of moisture sorption, the films were placed into 92% relative humidity chambers and stored in a 25 ± 1° room. Daily weighings were made on a balance located in the same room until three successive weights remained the same. Total moisture sorption was expressed as the per cent of the dry-film weight.

² Dissolution pH was defined as the lowest pH at which 20 mg. of polymer film dissolved in 10 ml. of a standard buffer solution.

Next, the polymer was stripped from the nylon support with acetone, and the nylon was dried 18 hr. at 45° *in vacuo*. Using the procedures described above, the moisture sorption of the nylon supports at 92% relative humidity was determined. This value was applied as a correction factor to the results obtained for the nylon-supported films.

Water Vapor Transmission Procedure.—Water vapor transmission (WVT) of *n*-alkanol half-esters of PVM/MA from ethyl through *n*-octyl and the isopropyl ester was measured by a modification of the ASTM method, E96-53T. In an attempt to remove residual alcohol, the polymeric esters were purified further by casting 50 ml. of a 20% w/v acetone solution onto a mercury substrate in a 6-in. Petri dish. After an initial period of air-drying, the films were removed and dried 24 hr. at 45° *in vacuo*. Using this material, film specimens for WVT were prepared by casting 25 ml. of a filtered 6% w/v acetone solution of the polymer onto a mercury substrate in a 4-in. Petri dish and drying as described previously. Two 7/8-in. diameter circles were cut from the middle of the film while it was still plastic, and each was placed in a drying cell between two Teflon disks. The films were air-dried 6 hr. at room temperature and then 24 hr. over Drierite in a vented humidity chamber at 45° *in vacuo*. It was necessary to place the drying cell into a 100% relative humidity chamber for 24 hr. at 25° to remove traces of acetone from the film. The samples were redried over Drierite as described above.

For measurement of WVT, the body of the drying cell was replaced by a solid body (see Fig. 2) containing a glass vial filled with anhydrous magnesium perchlorate to within 3–4 mm. of the film. The permeation cell was placed into the 92% relative humidity chamber, and an initial weight was obtained. Additional weighings were made twice daily for a minimum of 12 days. After redrying over Drierite as described above, the films were removed from the drying cell and examined for possible leaks. Film thickness was measured with a micrometer to 0.0001 in. at five locations.

Stability Procedure.—The stability of some half-esters of PEMA and PVM/MA was investigated in solution and as dry powders. A 13% w/w solution of one-half isopropyl PVM/MA containing 34% isopropanol and 53% acetone was stored in a 1-gal. amber bottle at room temperature. Samples were removed periodically, dried 18 hr. at 45° *in vacuo*, and analyzed by electrometric titration, infrared spectroscopy, and turbidity point determination.

Dry-state stability was studied by storing the polymeric esters in closed bottles at room temperature and 50°. Samples were removed at various intervals and assayed as above.

RESULTS AND DISCUSSION

Materials.—These copolymers and their reaction products have been described by Hanford (14) and Voss and Dickhauser (15). Electrometric titration and infrared spectra of three molecular weights of each copolymer indicated a high degree of uniformity within each copolymer. Smooth, sigmoidal titration curves were obtained which produced two distinct apparent dissociation con-

stants—*viz.*, PEMA: pK_1' —4.90, pK_2' —8.3; PVM/MA: pK_1' —4.85, pK_2' —8.95. The infrared spectrum of a PEMA/mineral oil mull shown in Fig. 3 also is typical of PVM/MA. Measurements of the intensities of the absorption bands at 5.62 and 5.85 μ were used to determine the ratio of anhydride to acid, respectively.

Esterification.—The partial esters evaluated in this study were prepared from the intermediate molecular weight copolymers of PEMA (DX-840-21) and PVM/MA (Gantrez AN-139). Absolute alcohol and greater than a 9:1 anhydride/acid ratio were essential to obtain the half-ester of the low-boiling ethyl and isopropyl alcohols. As the boiling point of the alcohol increased, the anhydride/acid ratio was less critical, and the reaction proceeded to the half-ester with the ratio as low as 7.5:1. Production of less than the half-ester was accomplished through the control of reaction temperature and heating time.

Figure 4 is a typical infrared spectrum of a half-ester of PEMA prepared as a film from acetone solution. Infrared spectra were employed to follow the extent of reaction qualitatively. Direct titration with alkali was used to determine the degree of esterification quantitatively.

The resultant half-esters were colorless to light-yellow and formed clear transparent films when cast from a suitable solvent. These compounds were soluble in highly polar solvents such as ketones and alcohols but insoluble in hydrocarbon and chlorinated hydrocarbon solvents. As the molecular weight of the alcohol increased, the half-ester

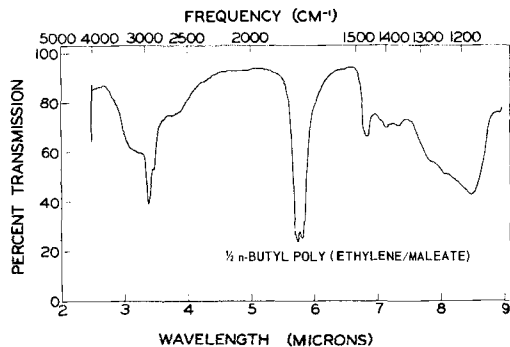


Fig. 3.—Infrared spectrum of PEMA in mineral oil.

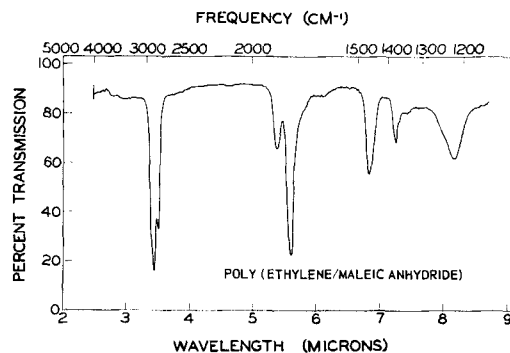


Fig. 4.—Infrared spectrum of 1/2 *n*-butyl PEMA film.

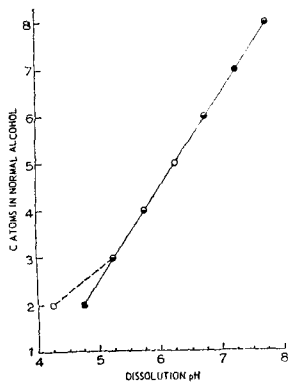


Fig. 5.—Dissolution pH versus number of *n*-alkanol carbon atoms in half-esters of PEMA and PVM/MA. Key: ○, PVM/MA; ●, PEMA; ●, PVM/MA and PEMA.

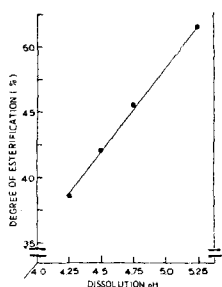


Fig. 6.—Dissolution pH versus degree of esterification of isopropyl PVM/MA partial esters.

films tended to become more plastic. The PEMA half-ester films appeared to be inherently more plastic than the corresponding PVM/MA products.

Apparent Dissociation Constants.—An effort was made to correlate the apparent dissociation constants of the various half-esters with their molecular weight and some solubility-related parameters determined in this study. The pK_a' values could not be determined with sufficient precision to quantitate a relationship. However, the half-esters of PEMA and PVM/MA did exhibit an increase in the pK_a' with an increase in the molecular weight. Apparent dissociation constants of the PEMA half-esters showed a tendency to parallel the slightly higher pK_a' values of the corresponding PVM/MA half-esters.

Dissolution pH.—Visual observation of dissolution and insolubility was facilitated by the use of a 0.25 pH gradient in the buffer solution series. Complete solubility of the film was noted at 0.25 pH unit above the pH at which it remained intact. Figure 5 indicates a direct linear relationship between dissolution pH and the number of alcohol carbon atoms in half-esters of PEMA and PVM/MA. Corresponding esters for each polymer show the same dissolution pH except for one-half ethyl PVM/MA which dissolves 0.5 pH unit lower than one-half ethyl PEMA.

As shown in Fig. 6, a direct linear relationship also exists between the degree of esterification and dissolution pH for various partial esters of isopropyl PVM/MA from 38.8 to 51.5%. Two partial esters 18.5 and 25.5% esterified were water-soluble.

Turbidity Point.—This parameter was used to study the hydrophobic properties of the various half-esters in the solution state. The transition from clarity to cloudiness in an aqueous-acetone solution of the half-esters was distinct and reproducible to within 0.1 ml. of deionized water.

A diffuse and gradual transition to cloudiness was exhibited by the one-half ethyl esters of PEMA and PVM/MA. Partial esters of isopropyl PVM/MA showed distinct turbidity points only when esterification was greater than 45.5%. Although an inverse relationship was noted, sufficient data were not available to quantitate a correlation between degree of esterification and turbidity point. Failure to obtain distinct turbidity points was attributed to the decreased hydrophobicity of these materials.

Figure 7 reveals that turbidity point is inversely related to the chain length of the *n*-alkanol in half-esters of PEMA and PVM/MA. A reversal in the relative hydrophobicity of PEMA and PVM/MA half-esters is noted when the alcohol chains are increased from four to five carbon atoms. Turbidity points of the one-half isopropyl esters of PEMA and PVM/MA were 3.8 and 3.6 ml., respectively, indicating significantly greater hydrophilic properties than the corresponding *n*-propyl derivatives.

Moisture Sorption.—Determinations of apparent moisture sorption equilibria were conducted to investigate the hydrophobic properties of films of various half-esters. Swelling and subsequent drying of the films at 100% relative humidity were necessary to remove traces of solvent which were particularly evident in lower esters such as ethyl and propyl. The plot of alcohol chain length of the half-ester against moisture sorption presented in Fig. 8 is similar to the turbidity relationship described above.

Moisture sorption values of 11.08 and 11.95% for the half-esters of isopropyl PEMA and PVM/MA, respectively, approached those of the corresponding *n*-propyl derivatives. Figure 9 also shows an inverse relationship between degree of esterification and moisture sorption for partial

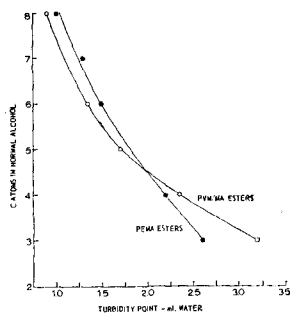


Fig. 7.—Turbidity point versus number of *n*-alkanol carbon atoms in half-esters of PEMA and PVM/MA.

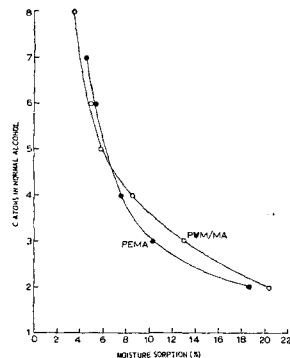


Fig. 8.—Apparent moisture sorption equilibrium versus number of *n*-alkanol carbon atoms in half-esters of PEMA and PVM/MA.

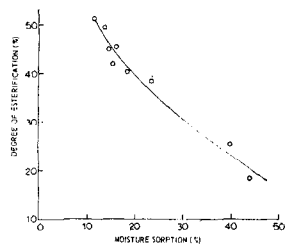


Fig. 9.—Apparent moisture sorption equilibrium versus degree of esterification of isopropyl PVM/MA partial esters.

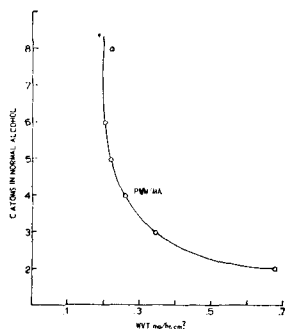


Fig. 10.—Water vapor transmission rate versus number of *n*-alkanol carbon atoms in PVM/MA half-esters.

esters of isopropyl PVM/MA. Although more deviation from the curve is noted, a distinct correlation exists.

Water Vapor Transmission (WVT).—A plot of the number of carbon atoms in the alcohol chain against water vapor transmission for half-esters of PVM/MA is presented in Fig. 10. Each point represents the WVT at 25° for a theoretical film thickness of 0.00525 in. The corrected values for each ester were obtained by plotting the data on Cartesian coordinate paper and extrapolating to 0.00525 in. The figure indicates a rapid decrease in the WVT from the ethyl to the *n*-butyl esters with a gradual decline in the rate of decrease to the *n*-octyl ester. Extrapolation beyond the *n*-octyl ester indicates little or no additional decrease in WVT for pure materials. Deviation of the *n*-octyl value from the curve might be explained by the removal of residual alcohol from the final film leaving it slightly porous. This explanation is consistent with the finding of traces of *n*-octanol in the material prior to the final drying.

Stability.—The stability of a 13% w/w isopropanol-acetone solution of the half-ester of isopropyl PVM/MA was determined after 14 months at room temperature. No visual, chemical, or hydrophobicity changes were noted.

Dry-state stability of the one-half *n*-butyl ester of PEMA was studied over a period of 44 months at 50° and at room temperature. After 49 days at 50°, electrometric titration indicated a decrease in degree of esterification from 48 to 45%. No significant change in the infrared analysis was noted at this time. Periodic analysis during the following 30 months of continuous heating showed no further chemical change. At this time, the turbidity point had increased to 2.6 ml. from an initial value of 2.3 ml. Final analysis after 44 months at 50° revealed a reduction of the degree of esterification to 42% and a slight increase in the anhydride content. No further change in turbidity point was observed. During this period of time, no changes were detected in the room temperature sample.

When stored in the dry state at 50° for 1 year, the one-half *n*-propyl ester of PVM/MA appeared to be chemically stable. Hydrophobic properties of the film as measured by moisture sorption did not show any change.

The one-half isopropyl ester of PVM/MA was analyzed periodically during storage in the dry state at 50° for 33 months. No chemical changes or changes in turbidity point and moisture sorption were found.

Toxicology.—Toxicological data have been obtained for representative samples of these coating polymers. Acute toxicities for three compounds are found in Table I.

Chronic toxicity studies were conducted for 1 year at doses as high as 1 Gm./Kg./day in rats and 100 mg./Kg./day in dogs. No visceral changes attributed to the treatment were observed.

SUMMARY

This investigation was undertaken to develop a series of pharmaceutical coatings which might be used to control the release of drugs upon reaching a specific intestinal pH. Three procedures were described for the preparation of several half-esters of PEMA and PVM/MA. Various partial esters of isopropyl PVM/MA also were prepared. These derivatives form clear, colorless films.

Relative differences in chemical and hydrophobic properties between these compounds were demonstrated by the measurement of dissolution pH, turbidity point, moisture sorption, and water vapor transmission. Significant correlations were found between these parameters and the number of carbon atoms in the *n*-alkanol of the half-ester. An increase in the number of *n*-alkanol carbon atoms produced an increase in dissolution pH and hydrophobicity. An increase in the degree of esterification of isopropyl PVM/MA also increased dissolution pH and hydrophobicity. Evaluation of these parameters for the half-isopropyl esters indicated they were slightly more hydrophilic than the corresponding *n*-propyl derivatives. Turbidity point and moisture sorption data demonstrated a reversal in the relative hydrophobicity of PEMA and PVM/MA half-esters when the *n*-alkanol was increased from four to five carbon atoms.

The stability of representative samples of these compounds was evaluated at room temperature and 50°. Excellent chemical and physical stability was found in the dry state and in solution.

Acute and chronic toxicity studies indicate that these materials are safe for use as pharmaceutical coatings. Additional reports of toxicity and utility of these compounds will be forthcoming.

TABLE I.—ACUTE TOXICITY

Compound	Species ^a	Oral Toxicity ^b
1/2 Isopropyl PVM/MA	Mouse	LD ₅₀ > 10 Gm./Kg.
	Rat	LD ₀ > 10 Gm./Kg.
	Cat	LD ₀ > 2 Gm./Kg.
	Dog	LD ₀ > 2 Gm./Kg.
1/2 <i>n</i> -Butyl PVM/MA	Rat	LD ₅₀ > 10 Gm./Kg.
	Dog	LD ₀ > 2 Gm./Kg.
1/2 <i>n</i> -Butyl PEMA	Mouse	LD ₅₀ > 6.2 Gm./Kg.
	Rat	LD ₀ > 10 Gm./Kg.
	Dog	LD ₀ > 2 Gm./Kg.

^a Fasted animals. ^b Highest dose tested.

REFERENCES

- (1) Wagner, J. G., *THIS JOURNAL*, **50**, 359(1961).
- (2) McKeehan, W., *ibid.*, **52**, 717(1963).
- (3) McKeehan, W., and Lappas, L. C., *ibid.*, **53**, 847 (1964).
- (4) Schanker, L. S., *Ann. Rev. Pharmacol.*, **1**, 29(1961).
- (5) Lappas, L. C., and McKeehan, W., *THIS JOURNAL*, **51**, 308(1962).
- (6) Lappas, L. C., and McKeehan, W., U. S. pat. 3,143,472.
- (7) Nessel, R. J., DeKay, H. G., and Banker, G. S., *THIS JOURNAL*, **53**, 790(1964).
- (8) *Ibid.*, **53**, 882(1964).
- (9) Kleber, J. W., Nash, J. F., and Lee, C.-C., *ibid.*, **53**, 1519(1964).
- (10) Wexler, A., and Hasegawa, S., *J. Res. Natl. Bur. Std.*, **53**, 19(1954) (Res. Paper No. 2512).
- (11) Evans, W. D., and Critchfield, C. L., *ibid.*, **11**, 147 (1933) (Res. Paper No. 583).
- (12) Patel, M., Patel, J. M., and Lemberger, A. P., *THIS JOURNAL*, **53**, 286(1964).
- (13) Carmody, W. R., *J. Chem. Educ.*, **38**, 559(1961).
- (14) Hanford, W. E., U. S. pat. 2,378,629.
- (15) Voss, A., and Dickhauser, E., U. S. pat. 2,047,398.

Rate Studies On the Anaerobic Degradation of Ascorbic Acid III

Rate of Formation of Furfural

By PER FINHOLT, INGER ALSOS, and TAKERU HIGUCHI*

Under acidic and anaerobic conditions, a molecule of furfural appears to be produced from each molecule of ascorbic acid. For nonhydrogen ion-catalyzed reaction, the aldehyde production seems to be substantially less than the mole-for-mole relationship. A pH profile for furfural production has been determined. The rate of disappearance of the aldehyde itself from oxygen-free aqueous solutions also has been studied.

IN PREVIOUS papers, the rate of the anaerobic loss of ascorbic acid (1) and the rate of formation of carbon dioxide by the same degradation (2) were reported. Since furfural is produced at the same time, it was of interest to study the rate of formation of this compound in the same system.

Reichstein and Grüssner (3) were the first to show that furfural was formed by the anaerobic decomposition of ascorbic acid. They heated ascorbic acid with 0.2 *N* hydrochloric acid in the absence of air and identified furfural as one of the decomposition products.

The formation of furfural during anaerobic loss of ascorbic acid was studied more thoroughly by Huelin (4). In his preliminary experiments, solutions of 0.25% ascorbic acid in distilled water were sealed in Florence flasks *in vacuo* and held at 100° for 10 days. During this period, 93% of the ascorbic acid decomposed, and the pH increased from 3.0 to 7.2. For each mole of ascorbic acid destroyed, 0.43 mole of furfural was produced. The furfural was identified by color reaction with aniline acetic acid, melting points

of phenylhydrazone (99°) and dinitrophenylhydrazone (200°), and spectral absorption of dinitrophenylhydrazone in alkaline solution, and was determined as phloroglucide. More detailed measurements were made at 30° with 0.01 *M* ascorbic acid solutions containing buffers. The solutions were kept in sealed glass tubes under nitrogen for 2 years. Furfural was determined colorimetrically with aniline in acetic acid according to the method of Duncan (5). The amount of furfural formed at pH 2.2 was 0.48 mole per mole of ascorbic acid destroyed; at pH 3.0, it was 0.17 mole; and at pH 4.0, it was 0.03 mole. At pH 5.0 and 6.0, no furfural could be detected. The author concludes that a plausible interpretation of the experimental results is that the formation of furfural does not occur in the primary reaction but is determined by the relative rate of various secondary reactions.

Cier *et al.* (6) heated ascorbic acid solutions at pH 2.5–5.5 under anaerobic conditions for 24 hours at 100° and determined the furfural concentration of the heated solutions colorimetrically with benzidine in acetic acid. At pH 5.5, no furfural could be detected. With a decrease in pH, the furfural concentration of the heated solutions increased sharply.

Quite recently, Coggiola (7) isolated the acids formed when a 5% solution of ascorbic acid in water was incubated at 100° under an atmos-

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