Using very fine particle size materials, self-supporting plugs can be formed without the use of any additional binders. We found the addition of a mixture of micronized lactose and 5% magnesium stearate is suitable to improve flow properties.

(1) ASTM-Standard C-204-79 "Standard Test Method for Fineness of Portland Cement by Air-Permeability Apparatus," American Society for Testing Materials, Philadelphia, Pa. 1979.

(2) I. C. Edmundson and J. P. R. Tootil, *Analyst*, **88**, 805 (1963).

(3) I. C. Edmundson, *Analyst*, **91**, 306 (1966).

P. Seth[x]
Mepha Ltd.
CH-4143 Dornach, Switzerland

N. Møller
Royal Danish School of Pharmacy
Department of Pharmaceutics
DK-2100 Copenhagen, Denmark

J. C. Tritsch
A. Stamm
Faculty of Pharmacy
Louis Pasteur University
F-67048 Strasbourg, France

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Physicochemical Interpretation of pH-Stat Titration of Amorphous Aluminum Hydroxycarbonate

Keyphrases □ Amorphous aluminum hydroxycarbonate—pH-stat titration, physicochemical interpretation, fiber optic Doppler anemometer analysis, dissolution data □ pH-stat titration—correlation between the *in vitro* test and *in vivo* acid neutralization, amorphous aluminum hydroxycarbonate, fiber optic Doppler anemometer analysis, dissolution data

To the Editor:

The pH-stat titration of antacids (1) is a valuable *in vitro* test because it has been correlated with *in vivo* acid neutralization (2). In contrast to the acid-neutralizing capacity test and the acid-consuming capacity test, which are essentially indirect assays, the pH-stat titration measures the rate of acid neutralization (3). The pH-stat titration has provided insights into (*a*) the structure of aluminum hydroxycarbonate (4, 5); (*b*) the adsorption of polybasic acids (6), polyols (7), polymers (8), and surface-active agents (8) by aluminum hydroxycarbonate; and (*c*) the interaction of aluminum hydroxycarbonate and magnesium hydroxide (9, 10).

The pH-stat titrigram of aluminum hydroxycarbonate contains three phases rather than the linear rate of acid neutralization expected for an acid–base titration (1). As seen in Fig. 1, the reaction is characterized by an initial rapid reaction (phase I) which abruptly converts into a slow zero-order reaction (phase II) which gradually transforms into a more rapid zero-order reaction (phase III). In contrast, antacids such as sodium bicarbonate, calcium carbonate, magnesium hydroxide, and hydrotalcite exhibit the expected linear pH-stat titrigram (9, 10). The purpose of this communication is to describe the

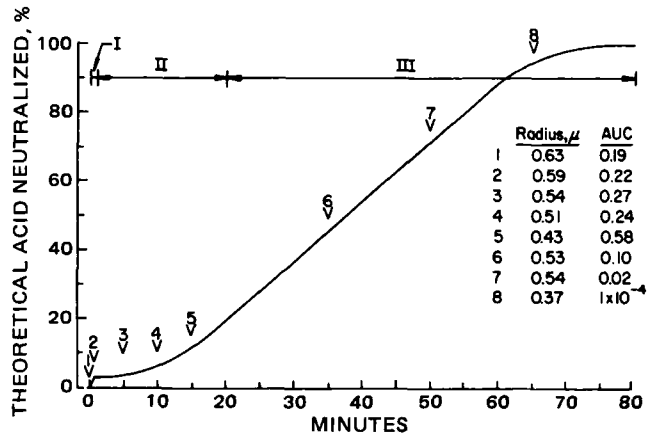

Figure 1—*pH-Stat titration at pH 3, 25° of aluminum hydroxycarbonate gel, which was examined at the indicated points by FODA.*

physicochemical reactions that are responsible for the three phases of the pH-stat titration of aluminum hydroxycarbonate.

The pH-stat titration at pH 3, 25° was interrupted periodically, as indicated in Fig. 1, and the apparent particle size was determined by fiber optic Doppler anemometry (FODA)[1] (11). The apparent mean hydrodynamic radius decreased from an initial value of 0.63 μm to 0.59 μm during phase I. A further decline to 0.43 μm was observed during the second phase. The apparent particle size initially increased during the third phase but decreased at the end of phase III.

The fiber optic Doppler anemometer determines particle size based on the measurement of the Brownian motion of colloidal particles. Thus, particle interactions that reduce the Brownian motion of particles will cause the apparent hydrodynamic radius, as measured by the fiber optic Doppler anemometer, to increase. In fact, if particle interactions cause Brownian motion to cease, the particle(s) will not be detected by FODA. Therefore, the apparent mean hydrodynamic radius data should be interpreted in conjunction with measurements of the total signal intensity. This is readily accomplished by integration of the power spectrum [area under the voltage squared *versus* frequency curve (AUC)] (12, 13). Thus, the AUC will be related to the total number of freely diffusing particles. The AUC increased from 0.19 to 0.22 during phase I and further increased to 0.58 during the slow, second phase. The AUC went to zero during phase III. The AUC data suggest that the number of particles exhibiting Brownian motion increased during phases I and II and decreased sharply during phase III.

It was hypothesized recently that aluminum hydroxide and aluminum hydroxycarbonate are composed of three types of particles: primary particles, secondary particles, and aggregates (14, 15). The primary particle is the basic unit: platy crystallites composed of fused six-membered rings of aluminum joined by double hydroxide bridges. The secondary particles are formed from primary particles arranged in a turbostratic type of arrangement due to the cohesive strength of van der Waals forces. Aggregates are composed of secondary particles formed in response to the balance of attractive and repulsive forces described by

[1] SIRA Institute Ltd., Kent, England.

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DLVO theory. This type of particle structure has also been proposed for carbon black (16).

It is proposed that the pH-stat titrigram can be interpreted in light of the recently proposed particle structure of aluminum hydroxycarbonate (14, 15). The point of zero charge of aluminum hydroxide is 9.6 and ranges from 6 to 9 for aluminum hydroxycarbonate (17). The aluminum hydroxycarbonate gel shown in Fig. 1 had a point of zero charge of 6.5. Acid was consumed very rapidly during the initial phase. Kerkhof *et al.* (1) observed that the rate of acid neutralization during the first phase exceeds the capability of the autoburet to add compensating acid. This rapid proton consumption is primarily due to protonation of the hydroxyl groups and carbonate surface sites as the pH of the system is instantaneously adjusted to 3, well below the point of zero charge. During this phase Al—OH—Al and Al—OCO_2 bonds are not believed to react. Rather, rapid charging due to proton adsorption leads to a positive surface charge. The first phase is very brief, which is expected for a protonation reaction.

The initial phase, which will be referred to as the proton loading phase, quickly shifts into a second phase in which the rate of proton consumption is much slower. The rate of acid neutralization increases gradually during phase II. It is hypothesized that the gradual increase in the rate of acid neutralization is due to an increase in surface area. The increase in surface area is compatible with the hypothesis that aluminum hydroxycarbonate is composed of primary particles, secondary particles, and aggregates. Protonation during phase I caused the development of a strong, positive surface potential. Therefore, large coulombic repulsive forces develop, which are believed to cause the aggregates to disperse into secondary particles and the secondary particles to disperse into primary particles. As peptization proceeds, the exposed surface area will increase, and in turn, the rate of acid neutralization will increase. Therefore, phase II will be termed the peptization phase.

The FODA analysis (Fig. 1) during the first two phases supports the hypothesis that proton loading and peptization occur during phases I and II, respectively. The mean hydrodynamic radius decreased to 0.43 μm by the end of phase II (point 5 in Fig. 1) with the consumption of only 11% of the acid necessary to neutralize the sample. The volume of a 0.43-μm sphere is only 31% of the volume of a 0.63-μm sphere. Therefore, if the decrease in mean hydrodynamic radius had occurred solely by acid dissolution, approximately 70% of the theoretical acid would have been required. Thus, a dispersion effect has taken place.

The decrease in mean hydrodynamic radius during phase I and phase II must be considered in light of the concurrent changes in the AUC: the number of particles exhibiting Brownian movement. The AUC increased by a factor of 3 during phases I and II. Thus, a decrease in mean hydrodynamic radius and an increase in the number of particles exhibiting Brownian motion is occurring simultaneously. In view of the fact that only a small fraction of the acid necessary to dissolve the particles has been added, the observed trends in particle size and AUC are consistent with a proton loading and peptization process.

The rapid rate of acid neutralization which is characteristic of the third phase of the pH-stat titration of aluminum hydroxycarbonate is believed to be due to the high surface area produced by the peptization of the aggregates and secondary particles. Evidence of dissolution is seen in the FODA analysis by the decrease in the AUC during the third phase. The increase in the mean hydrodynamic radius during the early part of phase III (points 6 and 7 in Fig. 1) is believed to arise because of the greater dissolution rate of smaller particles, which causes the mean particle size of the remaining particles to increase. Since the acid dissolution reaction predominates during phase III, it will be referred to as the dissolution phase.

In most aluminum hydroxycarbonate gels, the third phase of the pH-stat titration terminates with the neutralization of the theoretical amount of acid based on the stoichiometric reaction of three protons per aluminum. However, some aluminum hydroxycarbonate gels do not complete the theoretical reaction during phase III, and a very slow rate of acid neutralization proceeds until the reaction is complete. The slow-reacting material is believed to have a very large primary particle size, approaching, in some cases, the crystalline state. On the other hand, an occasional aluminum hydroxycarbonate gel neutralizes more than three protons per aluminum. Gels of this type usually contain a high level of sodium and the superpotency is due to contamination with dawsonite [crystalline sodium aluminum hydroxycarbonate, $NaAl(OH)_2CO_3$] which neutralizes four protons per aluminum (18).

Antacid materials such as sodium bicarbonate, calcium carbonate, magnesium hydroxide, and hydrotalcite which exhibit a linear pH-stat titration are all crystalline materials with a particle size usually in excess of the colloidal range. Therefore, they probably do not exist as primary particles, secondary particles, or aggregates.

An examination of the changes in the pH-stat titrigram during the aging of amorphous aluminum hydroxycarbonate gel further supports the proposed interpretation. IR and X-ray studies indicate that the primary particle size increases during aging (19–21). In some cases the degree of aggregation will also increase during aging due to the consolidating effects of gravitational settling. As seen in Fig. 2, the pH-stat titrigram of a freshly precipitated aluminum hydroxycarbonate gel is dominated by phase I (proton loading) and phase III (dissolution of peptized particles). Most of the aggregates and secondary particles in the freshly precipitated sample are readily dispersed

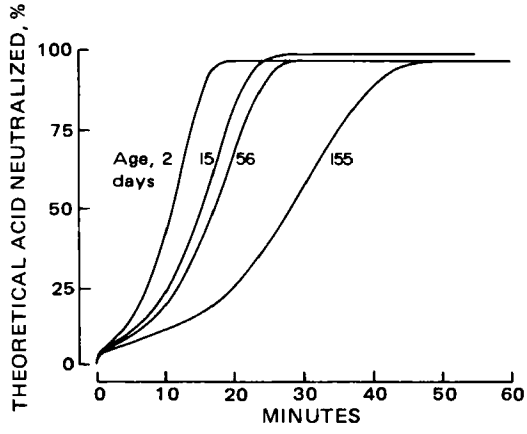

Figure 2—*Effect of aging of aluminum hydroxycarbonate gel at 25° on the pH-stat titration at pH 3, 25°.*

when the pH is lowered to 3. Since no significant particle growth has occurred, the relatively small primary particle size of the fresh gel gives a rapid rate of acid neutralization.

Phase II becomes more evident as the aluminum hydroxycarbonate gel ages. Both the primary particle size and the degree of aggregation increases during aging. Therefore, peptization becomes more important and the slow, second phase becomes more prominent. The rate of acid neutralization during phase III decreases during aging (day 2 *versus* day 155 in Fig. 2), which reflects the increase in primary particle size.

An understanding of the physicochemical processes required for the *in vitro* acid neutralization of aluminum hydroxycarbonate gel is necessary, because similar reactions occur in the GI tract when aluminum hydroxycarbonate gel is used as an antacid.

(1) N. J. Kerkhof, R. K. Vanderlaan, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **66**, 1528 (1977).

(2) J. S. Fordtran, S. G. Morawski, and C. T. Richardson, *N. Engl. J. Med.*, **288**, 923 (1973).

(3) S. L. Hem, J. L. White, J. D. Buehler, J. R. Luber, W. M. Grim, and E. A. Lipka, *Am. J. Hosp. Pharm.*, **39**, 1925 (1982).

(4) N. J. Kerkhof, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **66**, 1533 (1977).

(5) C. J. Serna, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **67**, 1144 (1978).

(6) M. K. Wang, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **69**, 668 (1980).

(7) D. N. Shah, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **70**, 1101 (1981).

(8) M. I. Zapata, J. R. Feldkamp, G. E. Peck, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, in press.

(9) R. K. Vanderlaan, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **68**, 1498 (1979).

(10) R. K. Vanderlaan, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **71**, 780 (1982).

(11) D. A. Ross, H. S. Dhadwal, and R. B. Dyott, *J. Colloid Interface Sci.*, **64**, 533 (1978).

(12) B. J. Berne and R. Pecora, "Dynamic Light Scattering," Wiley, New York, N.Y., 1976, pp. 28, 60.

(13) A. M. Jamieson and M. E. McDonnell, *Adv. Chem. Ser.*, **174**, 163 (1979).

(14) E. C. Scholtz, Ph.D. Thesis, Purdue University, 1981.

(15) J. C. Liu, Ph.D. Thesis, Purdue University, 1982.

(16) F. A. Heckman, *Rubber Chem. Technol.*, **37**, 1245 (1964).

(17) J. R. Feldkamp, D. N. Shah, S. L. Meyer, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **70**, 638 (1981).

(18) C. J. Serna, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **67**, 324 (1978).

(19) S. L. Nail, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **64**, 1166 (1975).

(20) S. L. Nail, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **65**, 231 (1976).

(21) S. L. Nail, J. L. White, and S. L. Hem, *J. Pharm. Sci.*, **65**, 1192 (1976).

Edward C. Scholtz
Mary I. Zapata
Stanley L. Hem[x]
Department of Industrial and Physical Pharmacy
Joseph R. Feldkamp
Joe L. White
Department of Agronomy
Purdue University
West Lafayette, IN 47907

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Simplified Method for Intravenous Dosing and Serial Blood Sampling of Unanesthetized Guinea Pigs

Keyphrases □ Blood sampling—serial collection from unanesthetized guinea pigs □ Guinea pigs—dosing and serial blood sampling without anesthesia □ Vacuum bleeding apparatus—serial blood collection from unanesthetized guinea pigs

To the Editor:

Ascorbic acid (vitamin C) levels can influence oxidative demethylation processes in the guinea pig (1), the only common laboratory animal unable to synthesize ascorbic acid endogenously (2). Thus, the guinea pig is an especially popular model for studying the effects of ascorbic acid on drug metabolism. Because of this, it is important to define methods for drug administration and repetitive blood sampling when using this species for pharmacokinetic studies. The following procedures were developed during a recent investigation of the effect of ascorbic acid on caffeine pharmacokinetics in young and aged guinea pigs.

A large towel was effective in quieting and immobilizing the animals during both drug dosing and blood sampling. The animals were prepared for dosing, by removing the hair over the injection site with an electric clipper. The prominent superficial vein on the medial side of the thigh (the medial saphenous vein) was used for all drug injections. The injection site was dabbed with ethanol and then the vein was enlarged using a finger to block venous return, and a small (21 gauge) needle was inserted in the direction of blood flow. A small quantity of blood was drawn *into the* 1-ml syringe to ensure correct insertion of the needle in the vein. Following a bolus injection of the drug solution, the needle was withdrawn quickly and pressure placed on the puncture with a finger to prevent venous backflow and avoid loss of some of the injected dose.

The method used here for repeated blood sampling was modified from that of Dolence and Jones (3) and found to be highly effective, producing a minimum of stress or pain in the guinea pigs. Similar to the procedure for caffeine dosing, the guinea pigs were wrapped in a towel exposing only the hind legs, which kept them comfortable yet immobilized. The leg not used for drug injection was extended and the hair removed using an electric clipper. A thin layer of silicone grease[1] was applied to the leg to form a tight seal with the vacuum bleeding apparatus and to prevent loose hair from contaminating the blood sample. One of the toenails was clipped, cutting into the vein and producing a flow of blood. Using this method alone only a small quantity of blood could be collected before coagulation stopped the flow. To circumvent this problem the prepared leg was positioned in the bleeding apparatus, as shown in Fig. 1, and the vacuum adjusted to produce a steady blood flow. The blood was collected in a 3-ml heparinized tube attached to the bottom of the vacuum-bleeding apparatus with a plastic[2] collar. The vacuum was adjusted to either increase or decrease the blood flow.

[1] Dow Corning, Midland, MI 48640.
[2] Tygon (R3603) tubing, Cole-Parmer, Chicago, IL 60648.

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