Gastric Acid Secretion Rate and Buffer Content of the Stomach after Eating

RESULTS IN NORMAL SUBJECTS AND IN PATIENTS WITH DUODENAL ULCER

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ABSTRACT New methods are described by which the buffer content and the rate and pattern of net gastric acid secretion in human subjects fed normal meals can be measured by use of sodium bicarbonate infusion to control intragastric pH. With these techniques, it was shown that the rate of acid secretion in response to a steak meal in seven duodenal ulcer patients was twice the rate achieved in six control subjects and that the amount of acid secreted after eating exceeded the peak histamine response in the ulcer patients but not in the controls. Meal-stimulated acid secretion, expressed as a function of the peak histamine response, was roughly correlated with the serum gastrin concentration ($r = 0.45$), but it was concluded that other factors must also contribute to the higher than normal secretory responses to a meal found in duodenal ulcer patients. Measurement of buffer content of the stomach revealed that the duodenal ulcer patients emptied the meal buffer at a much more rapid rate than the normal subjects. By 2 h after eating, the ulcer subjects had less than half as much buffer in their stomachs as the controls. The combination of acid hypersecretion and rapid buffer emptying leads to abnormally high gastric acidity after a meal in duodenal ulcer patients. These results suggest that, in addition to a large parietal cell mass, parietal cell responsiveness to a meal and the rate of buffer emptying may be important in the pathogenesis of duodenal ulcer.

INTRODUCTION

Previous measurements of gastric acid secretion during and after the ingestion of normal food have been made almost exclusively in animals with fundic pouches. The only method applicable to man involves measurement of the base excess in arterial blood, is valid only during the 1st h after eating, and has not been widely used (1, 2). One purpose of the present paper is to describe a new method for measuring the rate and pattern of acid secretion after eating a normal meal. The method involves manipulation of gastric pH and is therefore not entirely physiological; however, the control of gastric pH inherent in this method is a significant advantage in interpreting the results. A second purpose of the present paper is to describe a new method for determining the buffer content of the stomach at intervals after eating. This measurement is of interest for two reasons: first, the buffer remaining in the stomach is one major factor, along with the rate of acid secretion, that determines gastric acidity; and, second, the amount of buffer in the stomach can be used to estimate the rate of gastric emptying. The method we have used to measure buffer content involves rapid in vivo gastric titration with sodium bicarbonate.

These methods have been applied to a group of normal subjects and to a group of patients with duodenal ulcer, and the results are correlated with the peak secretory response to histamine and with serum gastrin levels.

METHODS

Subjects

The study group was composed of six normal subjects, laboratory workers and medical students, with a mean age of 34 yr (range: 23–55) and seven patients with well-established chronic duodenal ulcer disease with a mean age of 38 yr (range: 20–58). The ulcer patients were being treated with noncalcium antacids but had not received anticholinergic drugs. None of the ulcer patients had had prior

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gastric operation or a recent complication and none had clinical or radiographic evidence of impaired gastric emptying. Five of the six normal subjects and six of the seven ulcer patients were men.

The standard meal

The meal consisted of 5 ounces of ground sirloin steak, cooked and seasoned with salt and pepper, two pieces of toast with one teaspoon of butter, and 360 ml water. The subjects were instructed to chew the food thoroughly, and to drink sips of the water after every few bites of solid food. The time required to finish the meal was 20–30 min.

This meal contained 39 g protein, 30 g carbohydrate, 30 g fat, and 546 kcal (estimated from standard dietary tables). The total volume of the meal was approximately 500 ml, and when homogenized, the pH was 5.5 (range 5.40–5.64).

Since the same meal was fed to each subject, the amount of food ingested per pound of body weight varied from subject to subject. However, the weight of the control subjects was on an average equal to that of the ulcer patients at 159 lb each (range control subjects: 105–199, range ulcer patients: 140–187 lb).

Measurement of pH

pH was measured with a Sargent pH meter (Sargent-Welch Co., Chicago, Ill.), using standards of pH 1.07, 2.1, 4.0, and 7.0.

Acid secretion rate after food

Principle. The average pH of the meal, upon homogenization, was 5.5. Before eating, the gastric pH was raised to 5.5 by infusion of 0.3 N sodium bicarbonate. During the time the subject was eating and for the subsequent 4 h, 0.3 N sodium bicarbonate was infused at the rate required to maintain gastric luminal pH at 5.5. The number of milliequivalents of sodium bicarbonate required per hour to keep the pH at 5.5 is equal to the rate of net acid secretion (expressed as milliequivalents HCl per hour).

Procedure. After a 10 h fast, subjects were intubated with a 16 Fr Levin tube, to which was attached a small polyvinyl tube (ID 1 mm). The Levin tube was used for sampling gastric contents, and the polyvinyl tube was utilized for infusing sodium bicarbonate. The opening of the polyvinyl tube was 10 cm proximal to the most proximal opening of the Levin tube. The tubes were positioned under fluoroscopic control so that the Levin tube openings were in the antrum and lower body of the stomach, and the opening of the polyvinyl tube was in the upper part of the body of the stomach.

After the tubes were in place, a 2–3 ml sample of residual gastric content was removed, its pH measured, and then it was returned to the stomach through the Levin tube. If the pH was below 5.5, sodium bicarbonate infusion was begun. The gastric contents were sampled repeatedly (average frequency of sampling before eating was 43.3±4.6 times/h), the pH read quickly, and the samples then returned to the stomach.

After the rate of bicarbonate infusion had been adjusted to achieve a stable gastric pH of 5.5, the subject began to eat the meal. Frequent sampling was continued during and after the meal was eaten, and the rate of bicarbonate infusion was further adjusted every few minutes as necessary to maintain gastric pH at 5.5. After the meal was completed, the subject rested in the recumbent position and the gastric content was sampled at an average rate of 19.8±1.0 times/h, or about once every 3 min. In all cases, the pH of the sample was quickly determined, and then the sample was returned to the stomach.

In order to facilitate mixing of the stomach contents, the subject was shaken manually for about 5 s before the collection of each sample. The manner of shaking was similar to that used for obtaining gastric specimens for cytologic examinations. In addition, the subject frequently changed positions from side to side and from back to front. A final method used to enhance mixing consisted of aspiration of 50–70 ml of gastric fluid into the bulb syringe, mixing thoroughly by agitation, taking 2–3 ml for pH determination, and returning the remainder of the sample to the stomach. All three mixing aids were used in each subject.

The 0.3 N sodium bicarbonate was infused by a Holter pump. By changing the dial settings on this pump, the rate of infusion could be varied from 0.5 to 180 meq of bicarbonate/h, and the pump dial was used to estimate roughly the rate of bicarbonate infusion at any given instant.

The cumulative volume of bicarbonate solution infused was recorded approximately every 3 min (each time a sample was removed from the stomach) by noting the residual volume in the volumetric cylinder which served as a reservoir for the sodium bicarbonate infusion. The exact normality of the sodium bicarbonate solution was determined after each test by titration with an acid standard, and the results of each experiment were then calculated in terms of milliequivalents of bicarbonate infused hourly and cumulatively for 4 h.

Buffer capacity

Principle. After ingestion of the standard meal, the pH of gastric contents was allowed to fall spontaneously (due to acid secretion) or made to fall (by 0.1 N hydrochloric acid infusion) to below 2.5. 2 h after eating, 0.3 N sodium bicarbonate was infused rapidly, and the pH of the gastric content was monitored at frequent intervals until the pH was 6.0. The buffer content of the stomach was estimated from the titration curve.

Procedure. The subjects were intubated with the same tube and in the same manner as described in the previous section, except that intubation was carried out 30 min after the meal was eaten rather than before the meal. Gastric contents were mixed and sampled as described above. If necessary, 0.1 N hydrochloric acid infusion was begun 60 min after the meal and continued for the next hour in order to lower intragastric pH. In all instances the aim was to have gastric pH below 2.5 by the time that gastric buffer content was to be measured at 2 h. At that time the stomach content was rapidly alkalinized by infusion of 0.3 N sodium bicarbonate. Well-mixed gastric samples were obtained as frequently as possible (average sampling frequency, 0.8 times per min), the pH determined and the sample returned to the stomach. The cumulative volume of bicarbonate infused was recorded each time a sample was taken. The average time required to alkalinize the stomach (from pH 2.5 to 5.25) was 7.4±1.8 min.

The titration curve of gastric contents with sodium bicarbonate was then plotted, and buffer capacity was estimated by a technique described in the Results section. In three subjects buffer content of the stomach was also measured at 1 and 3 h after the meal by a similar method.

Serum gastrin concentration

In the experiments in which acid secretion was measured by controlling gastric pH at 5.5, venous blood was collected before and 1, 2, and 3 h after eating began. In two of the duodenal ulcer subjects, the 3 h sample was omitted due to difficulty in obtaining blood specimens. The blood was allowed to clot, and serum was obtained by centrifugation and stored at −20°C until assayed.

Serum gastrin concentrations were measured by radioimmunoassay (3). Porcine gastrin I served as the standard. It was shown previously that samples of synthetic human gastrin and porcine gastrin I with equivalent biologic activity exhibited similar potency in this assay system (4). All samples were assayed in duplicate. The sensitivity of the assay was sufficient to measure a gastrin concentration of 1 pg/ml, and at the 1:10 dilution of serum used in the assay, serum gastrin concentrations of 10 pg/ml or greater could be measured.

Basal and peak histamine response

After discarding the residual gastric content, basal secretion was collected for 60 min through a 16 Fr. Salem sump tube with a Stedman intermittent suction pump (Stedman Foundry & Machine Co., Inc., Aurora, Ind.). 30 min after the basal collection was started, 50 mg of Benadryl (Parke, Davis & Co., Detroit, Mich.) was injected intramuscularly. At the end of the basal period, histamine acid phosphate, 0.04 mg/kg was injected subcutaneously. Gastric content was collected in 15-min intervals for 1 h.

Hydrogen ion concentration was measured by the method of Moore and Scarlata (5).

The peak histamine response was calculated as the sum of the two highest 15-min secretion rates, multiplied by 2 to express the results in milliequivalents per hour.

RESULTS

Acid secretion rate after eating

In Vitro Experiments

Preliminary in vitro studies were carried out with samples of homogenized meal in order to determine if acid added to the meal could be estimated accurately by the amount of sodium bicarbonate necessary to keep the pH constant at 5.5. Such an experiment is summarized in Fig. 1. In this experiment, acid was added at a constant rate of approximately 4 meq/h to 100 ml of the homogenized meal. Mixing was accomplished by a magnetic stirrer (60 rpm). Sodium bicarbonate was added from a burette at a rate necessary to keep the pH at 5.5. At intervals of 10 min, 10 ml of the reaction mixture was discarded to simulate gastric emptying. As shown in Fig. 1, the rate of acid addition was almost exactly equal to the amount of sodium bicarbonate required to keep the pH constant at 5.5. This held true whether the reaction took place in an open system (surface of reaction mixture open to air) or in a closed system that would allow higher P_{CO_2} concentrations to build up. Four additional experiments at pH 5.5 gave the same results.

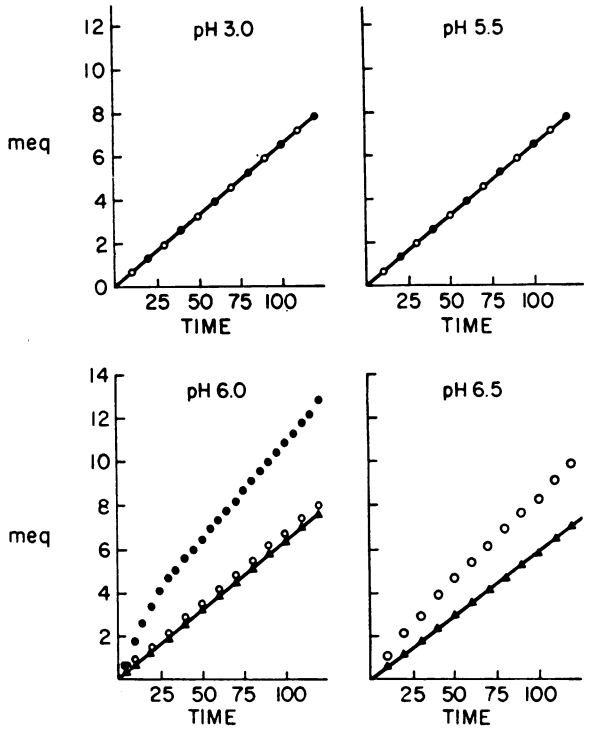

FIGURE 1 Correlation of the milliequivalents of 0.1 N HCl added to a sample of homogenized meal (straight line) and the milliequivalents of 0.3 N $NaHCO_3$ required to maintain the reaction pH at 3.0, 5.5, 6.0, or 6.5 in vitro. As noted in the text, a fraction of the reaction mixture was discarded every 10 min (to simulate gastric emptying). The open circles denote experiments carried out in an open beaker; the closed circles denote experiments carried out in an airtight system that would prevent escape of CO_2 formed as a result of the reaction of HCl and $NaHCO_3$. The triangles indicate experiments carried out in an open beaker that was gassed with air to facilitate the dissipation of CO_2. It is emphasized that the straight line represents the rate of acid addition, and is not a line through the symbols, which indicate the rate of bicarbonate addition.

In a set of similar in vitro experiments, the amount of sodium bicarbonate required to keep the pH at 3.0, 5.0, 6.0, and 6.5 was determined. Interestingly, after the meal pH was adjusted to the end point pH of either 3.0 or 5.0 by the addition of acid, the further *rate* of acid secretion was accurately estimated by the amount of bicarbonate required to maintain pH at the desired level. Fig. 1 shows the results of a typical experiment at pH 3.0. On the other hand, in order to maintain the pH constant at 6.0 or 6.5, more bicarbonate was required than the amount of acid added. This was true in both a closed and an open system. However, if the reaction mixture was gassed with air to aid in the removal of CO_2, the rate of acid addition was accurately reflected by the amount of bicarbonate required to keep the pH constant. This is shown in Fig. 1.

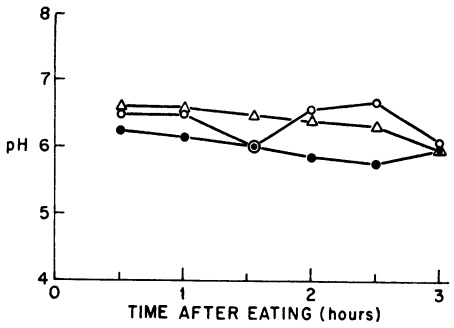

FIGURE 2 pH of gastric contents after eating the steak meal in three patients with pernicious anemia.

These results indicate that high CO_2 concentrations result in an overestimation of acid "secretion" at pH 6.0 and above but not when the pH is 5.5 or below. This is presumably because the excess amount of bicarbonate required to maintain a given pH as P_{CO_2} rises is a log function of the initial bicarbonate concentration, as expressed in the Henderson-Hasselbalch equation. At pH 5.5 the excess bicarbonate is an insignificant amount compared with the total titration whereas at pH 6.0 and 6.5 the required excess is significant. The extra amount of bicarbonate that must be added to maintain a pH of 6.0 or 6.5 results in an overestimation of the rate of acid "secretion".

The osmolality of 0.3 N sodium bicarbonate is 545 mosmol/kg. When this is added to 0.16 N hydrochloric acid (the approximate concentration of acid believed to be secreted by the parietal cells [6, 7]), in an amount required to raise the pH to 5.5, the osmolality of the resulting solution (mainly sodium chloride) is 210 mosmol/kg. This is approximately equal to the osmolality of gastric contents after the ingestion of a steak meal similar to the one used in the present in vivo studies (8).

These in vitro studies suggest that gastric acid secretion can theoretically be determined from the amount of sodium bicarbonate necessary to maintain the intragastric pH at 5.5, 5.0, or 3.0. They also indicate that titration at pH 6.0 or 6.5 with sodium bicarbonate would probably result in an overestimation of acid secretion and emphasize the importance of avoiding gastric pH levels higher than 5.5 when bicarbonate is the alkalinizing agent. Neutralization of gastric acid with 0.3 N sodium bicarbonate would not be expected to alter intragastric osmolality.

IN VIVO STUDIES

Patients with pernicious anemia. Although the meal had an average pH of 5.5, it is not known what the pH of gastric contents would be if no acid were secreted by the stomach, since salivary, pancreatic, biliary, and nonparietal gastric secretions might alkalinize the meal slightly. In order to assess this, three patients with well-documented pernicious anemia, who were proven achlorhydric after 0.04 mg/kg histamine, were studied. They were fed the standard meal, and gastric contents were sampled at 30-min intervals thereafter. Fig. 2 shows the pH of the gastric contents in these patients. The pH stayed around 6.5 in two and varied between 5.8 and 6.3 in the third patient. Thus, the gastric content pH of pernicious anemia patients after eating is higher than the original pH of the meal.

In order to quantitate the amount of the alkaline secretion in pernicious anemia patients, their stomachs were infused with 0.1 N hydrochloric acid at a rate necessary to keep the pH at 5.5 during and after the ingestion of the standard meal. The procedure was exactly the same as described under Methods for normal subjects and patients with duodenal ulcer, except that acid was infused instead of bicarbonate. The results are shown in Fig. 3 for one of these patients, who required 4.8 meq of hydrochloric acid to keep the gastric pH at 5.5, all of which was infused in the 1st h and three-quarters after eating. After that time, the gastric pH remained constant at 5.5, even though no further hydrochloric acid was infused. One of the other patients behaved in an almost identical fashion. The third subject required 14 meq of hydrochloric acid, all of which was infused in the first 2 h.

These studies indicate that from 4.8 to 14 meq of an alkaline secretion mixed with the ingested meal during the first 2 h after eating in these three patients with pernicious anemia. During the next 2 h, there was no evidence of further addition of an alkaline fluid to the gastric content. If this estimate may be extrapolated to normal subjects and to patients with duodenal ulcer, estimation of postcibal acid secretion in such persons by

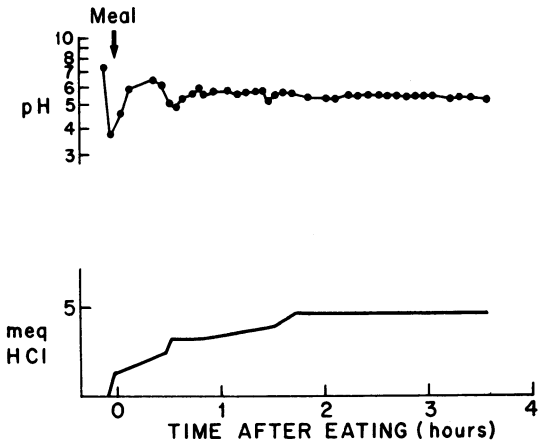

FIGURE 3 Milliequivalents of 0.1 N HCl required to keep gastric pH at 5.5 in a patient with pernicious anemia.

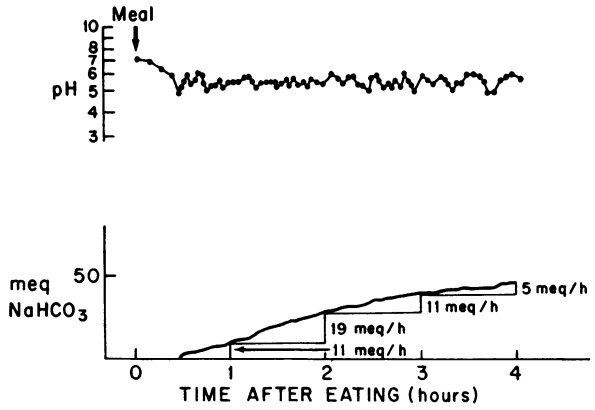

FIGURE 4 Milliequivalents of 0.3 N $NaHCO_3$ required to keep gastric pH at 5.5 in normal subject (C. P.).

titration of gastric pH to 5.5 with sodium bicarbonate will underestimate the true rate of acid secretion by about 2.5 to 7 meq/h in the first 2 h. However, it is not certain that patients without pernicious anemia would have the same amount of alkaline solutions enter the stomach after the meal as those with pernicious anemia. It has, for instance, been suggested that patients with achlorhydria have a greater tendency for duodenal-gastric reflux than other subjects (9) and that patients with pernicious anemia secrete more alkaline fluid into the stomach in response to an acid load than normals (10).

Studies in normal subjects and patients with duodenal ulcer. An example of the results in one normal subject is shown in Fig. 4. This is the only patient in the control or duodenal ulcer group in whom the pH of the gastric fluid before eating was higher than 5.5. Therefore, no bicarbonate was added before eating, and in fact none was added until about 30 min after the meal was begun. The figure shows the milliequivalents of sodium bicarbonate required to keep the gastric pH at 5.5, and it will be noted that 11 meq was required in the 1st h, 19 in the 2nd, 11 in the 3rd, and 5 meq in the 4th and final h.

In carrying out in vivo titration with sodium bicarbonate it is possible to maintain the pH at a level near 5.5, but many samples are obtained whose pH is either below or above this level by one-half pH unit, and occasionally further deviations were encountered. This will result in an error in the calculated rate of acid secretion only to the extent that over- and under-titrated gastric content is emptied from the stomach, and the errors of over- and under-titration will, to some degree at least, cancel each other out.

Reproducibility was assessed in six subjects; two examples of the complete titration are given in Fig. 5. Excellent agreement is evident, and it should be pointed out that the results of the first test cannot bias the second test since the normality of the sodium bicarbonate solution is not determined until the titration is completed. Furthermore, the very nature of these in vivo titrations, with the pump rate changed after every gastric pH determination, virtually precludes subconscious bias.

Fig. 6 shows the results of duplicate tests on all six subjects who were studied on two occasions, expressing the results according to the highest rate of acid secretion observed in any one of the 4-h periods. Again, the agreement between duplicate tests is good, the correlation coefficient being 0.92.

Fig. 7 shows the acid secretion rate in six normal subjects and seven duodenal ulcer subjects, plotted so that the rate of acid secretion in milliequivalents per hour is shown for each 30 min period. Average basal secretion, measured by standard aspiration techniques, was 7.5 and 1.4 meq/h in the ulcer and control subjects, respectively. Acid secretion after the meal, measured by bicarbonate titration, reached a peak at 1 h in the ulcer patients and at 1½ h in the controls. Peak secretion rate was 65.7 meq/h in the ulcer group, 31.8 meq/h in the normal subjects. Acid secretion gradually diminished to 21.4 and 7.7 meq/h in the ulcer and control subjects, respectively, during the 4th h.

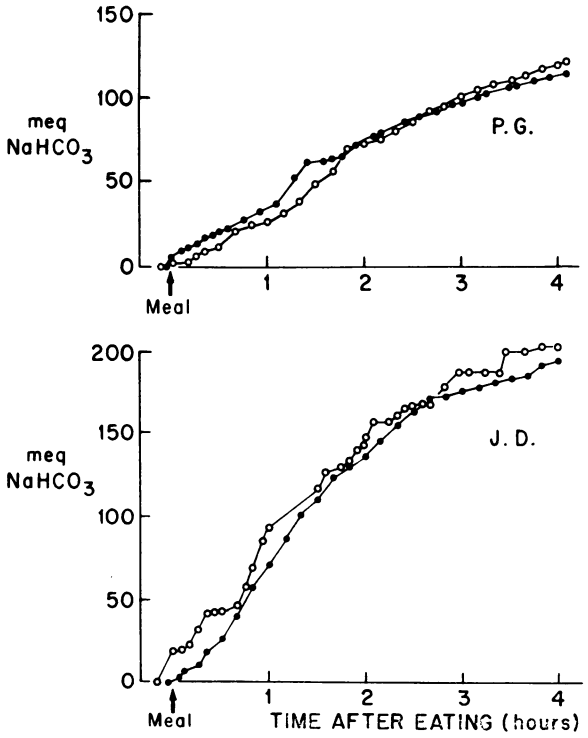

FIGURE 5 Reproducibility of in vivo titration to pH 5.5 in two patients with duodenal ulcer.

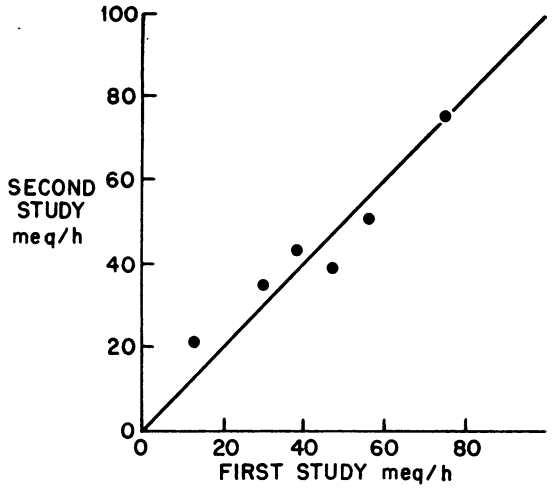

FIGURE 6 Reproducibility of in vivo titration to pH 5.5 in six subjects. Results are the highest rate of acid secretion observed in any of the 4-h periods after the meal was begun. The line is perfect identity, not the regression line of the experimental observations.

Table I shows the peak secretion observed after eating during any 1 h period and the peak histamine response and basal secretion rate for each subject. Most ulcer patients secreted acid after eating at a rate exceeding their peak histamine response (average 116%), whereas most normal subjects secreted less after the meal than after histamine (average 86%). The difference was statistically significant ($P < 0.05$). The correlation of the peak histamine and peak meal response in all subjects is shown in Fig. 8. The correlation coefficient was 0.86.

The individual and mean serum gastrin levels before and at 1, 2, and 3 h after eating are shown in Fig. 9. Although gastric pH was constant at 5.5 in all these experiments, the serum gastrin response was highly variable; postprandial gastrin concentrations rose by a factor of 3 in some and hardly at all in other subjects.

TABLE I

Basal, Peak Histamine, and Peak Meal Acid Secretion in Normal Subjects and Patients with Duodenal Ulcer*

Subject	Basal	Peak histamine	Peak meal	$\frac{\text{Peak meal}}{\text{Peak histamine}} \times 100$
Normals				
C. C.	4.2	40.4	35	87
J. H.	0.0	41.0	41	100
C. P.	0.1	25.0	19	76
B. B.	0.3	40.2	35	87
J. B.	1.4	29.8	17	57
J. R.	2.3	30.8	34	110
Mean ±SE	1.4±0.7	34.5±2.8	30±4.0	86±7.6
Duodenal ulcer patients				
M. C.	20.3	80.6	94	117
J. D.	2.5	49.4	75	152
P. G.	1.0	28.3	39	138
A. C.	10.2	79.0	64	81
X. C.	4.4	54.5	65	119
G. P.	3.7	46.8	51	109
G. W.	10.4	68.8	61	89
Mean ±SE	7.5±2.5	58.2±7.2	64±6.6	116±9.5

* Milliequivalents per hour.

The average fasting and postcibal levels were higher in the duodenal ulcer patients than in the controls, but since the variation within each group was wide the differences are not statistically significant. On the average, peak gastrin level occurred 1 h after the meal and was approximately 2 times the fasting level in the control subjects and 1½ times the fasting level in the duodenal ulcer patients. In all normal subjects except one, the gastrin concentration fell at 3 h to a level lower than at 2 h, whereas in the duodenal ulcer subjects there was a tendency for the gastrin concentration to be as high at 3 h as it was at 2 h. More work obviously needs to be done to see if this difference is real.

In order to better assess the role of serum gastrin concentration in determining the rate of postcibal acid

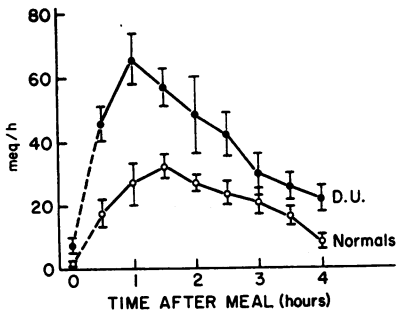

FIGURE 7 Rate of acid secretion after eating in six normal subjects and in seven patients with duodenal ulcer. Mean ±SE. The rate of basal acid secretion, measured by standard methods, is shown for comparison at zero time.

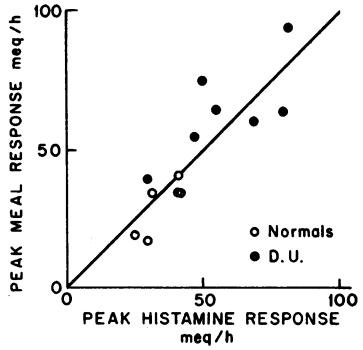

FIGURE 8 Correlation of peak histamine and peak meal response in normal subjects and in patients with duodenal ulcer.

secretion, the results in individual subjects are depicted in Fig. 10. In the top sequence in this figure the serum gastrin level 1 h after the meal, the absolute rise in serum gastrin at 1 h, the peak rise in serum gastrin after the meal, and the maximum percent rise above basal level are plotted against the peak rate of acid secretion after the meal. There is no indication from these results that the serum gastrin level is important in determining the different rates of peak gastric acid secretion noted in different people after the meal.

In the bottom sequence in Fig. 10, the same aspects of the serum gastrin response are correlated with the peak meal response, expressed as a function of the peak histamine response. This method of expression tends to normalize the secretory rate to a constant parietal cell mass. Of the various ways in which the gastrin level was expressed, the best correlation with acid secretion was the gastrin level at 1 h after eating ($r = 0.45$). This correlated better than the absolute rise in serum gastrin or the percent rise above the basal level.

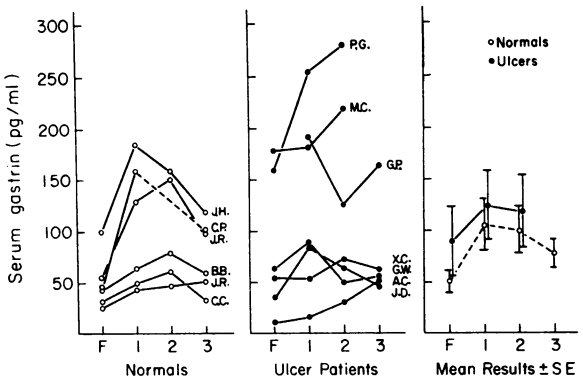

FIGURE 9 Serum gastrin concentration in the fasting state (F) and at 1, 2, and 3 h after the steak meal. In the fasting state, the gastric pH in these subjects was not known but at all times after the meal the gastric pH was kept constant at 5.5. The mean result at 3 h in the ulcer patients is not plotted, since two ulcer patients (P. G. and M. C.) did not have blood drawn for gastrin level at 3 h.

Buffer capacity of the stomach

IN VITRO EXPERIMENTS

Reversibility of the reaction of the meal with acid. Samples of the diluted meal were carefully titrated with concentrated hydrochloric acid to a pH of 1.0 and then back-titrated with concentrated sodium hydroxide. The titration curves were virtually identical, indicating that the reaction of meal with acid is reversible. This point was proven by another experiment in which a sample of the original meal having a pH of 5.55 was acidified

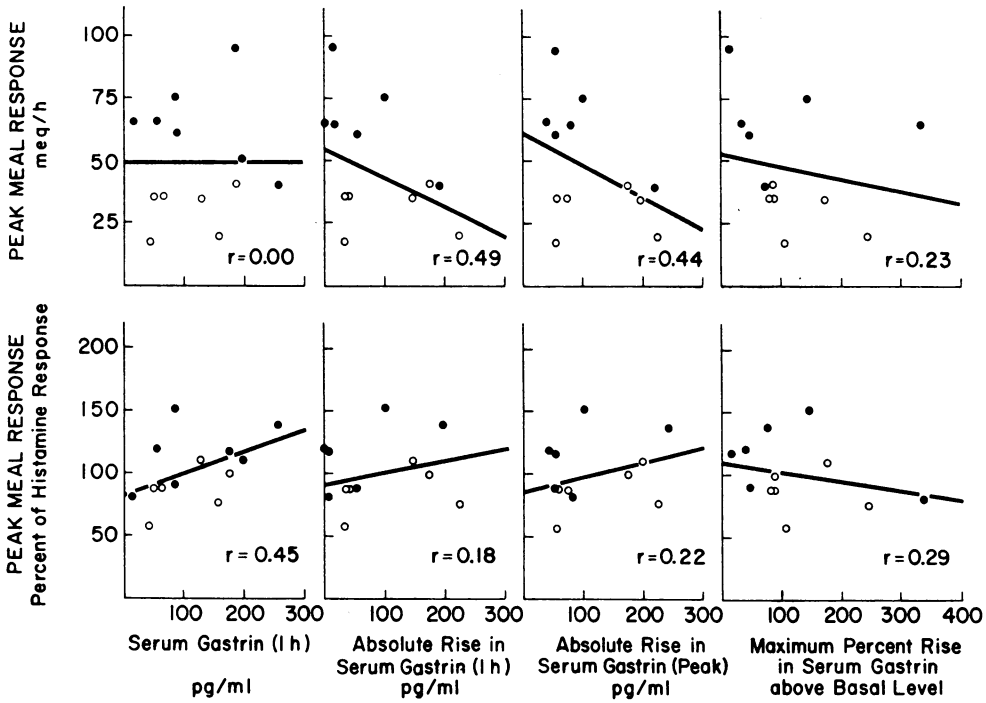

FIGURE 10 Correlation of serum gastrin level and the peak acid secretion rate after the steak meal. In the top sequence the observed peak meal response is correlated with serum gastrin. In the bottom sequence the peak meal response is expressed as a percent of the peak histamine response, which is assumed to normalize the results to a constant parietal cell mass.

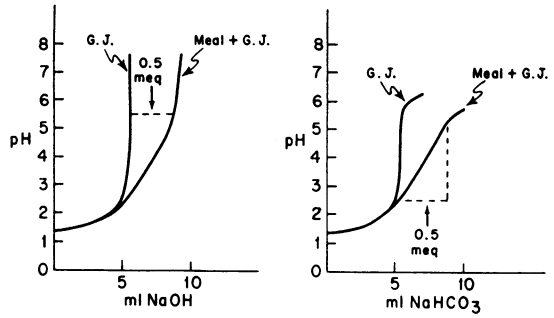

FIGURE 11 Titration of fasting gastric juice (G. J.) and homogenized meal plus gastric juice with NaOH and $NaHCO_3$. In these experiments 2.5 ml of the homogenized meal were diluted with 27.5 ml of gastric juice and titrated with NaOH and $NaHCO_3$. For comparison 27.5 ml of gastric juice plus 2.5 ml of water were similarly titrated.

with hydrochloric acid to pH 1.10. After a 5 min period, sodium hydroxide was added in an amount exactly equivalent to the amount of hydrochloric acid added initially, and the pH was again measured. If food combines reversibly with acid, the pH after addition of sodium hydroxide should be 5.55, whereas if the reaction of food with hydrochloric acid was irreversible (liberation of CO_2 and water, for instance), then the pH after sodium hydroxide was added would be higher than the starting point of 5.55. The actual pH after addition of sodium hydroxide was 5.60, indicating that almost all of the reaction of food buffer and hydrochloric acid is reversible.

Direct and indirect estimates of buffer capacity. Titrations of gastric juice and homogenized meal plus gastric juice with sodium hydroxide and sodium bicarbonate are shown in Fig. 11. With sodium hydroxide the titration curve of gastric juice and meal plus gastric juice was similar up to a pH of about 2.0, after which the curve for gastric juice plus the meal deviated to the right. The amount of buffer in the meal sample was defined as the difference in the amount of sodium hydroxide necessary to titrate a pure gastric juice standard and an equal volume of an unknown from a given pH less than 2.0 to a pH of 5.5 (the original pH of the meal). This is referred to as the direct method of measuring buffer capacity. In the example shown in Fig. 11, this was 0.50 meq of buffer per 2.5 ml of the homogenized meal sample. By this method the buffer content of the entire meal averaged 87 meq.

As shown in Fig. 11, the shape of the titration curves with sodium bicarbonate was different than with sodium hydroxide; with sodium bicarbonate the titration curves with and without food deviated at approximately 2.2 instead of 2.0, and above pH 5.5 the curves deviated sharply rightward. Note that the amount of sodium bicarbonate required to raise the meal sample pH from 2.5 to 5.25 is equal to 0.5 meq, the known amount of buffer in this sample of meal (as defined by the sodium hydroxide titration curves). Estimation of buffer capacity by the amount of sodium bicarbonate necessary to raise the pH from 2.5 to 5.25 is hereafter referred to as the indirect method. (Other points along the titration curve could, of course, also be chosen that would give the correct amount of buffer.) In order to see if the amount of sodium bicarbonate necessary to raise pH from 2.5 to 5.25 could be used as a reliable measure of buffer capacity, samples of diluted meal and samples of postcibal gastric contents from control and duodenal ulcer patients were measured by both the direct and indirect methods. The results are shown in Fig. 12. Note that the agreement is excellent, indicating that buffer content can be estimated from a sodium bicarbonate titration curve alone. This means that the quantity of buffer in the stomach contents can theoretically be accurately estimated by in vivo titration with sodium bicarbonate, even though the volume of the gastric content is unknown (see below).

Generation of buffer by peptic digestion. 10 ml of homogenized meal plus fresh gastric juice was incubated and stirred for 0, 1, and 2 h at 37°C; the samples were then titrated with sodium hydroxide, and buffer content was calculated by the direct method described above and in Fig. 11 (left side). As shown in Fig. 13, the meal plus gastric juice incubated for 0 h had a pH of 1.98, whereas after 1 and 2 h of incubation the pH was raised to 2.27 and 2.70, respectively. The buffer capacity of the 10 ml sample was 1.74 meq at zero time, 2.14 meq at 1 h, and 2.33 meq at 2 h. Thus, incubation of the meal plus fresh gastric juice generated a 33% increase in buffer capacity in 2 h.

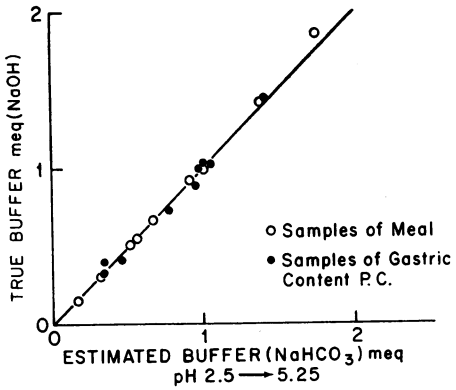

FIGURE 12 Correlation of the "true buffer" as measured by titration with NaOH according to the method shown on the left side of Fig. 11, and "estimated buffer" measured by the amount of $NaHCO_3$ necessary to raise the pH of the sample from 2.5 to 5.25.

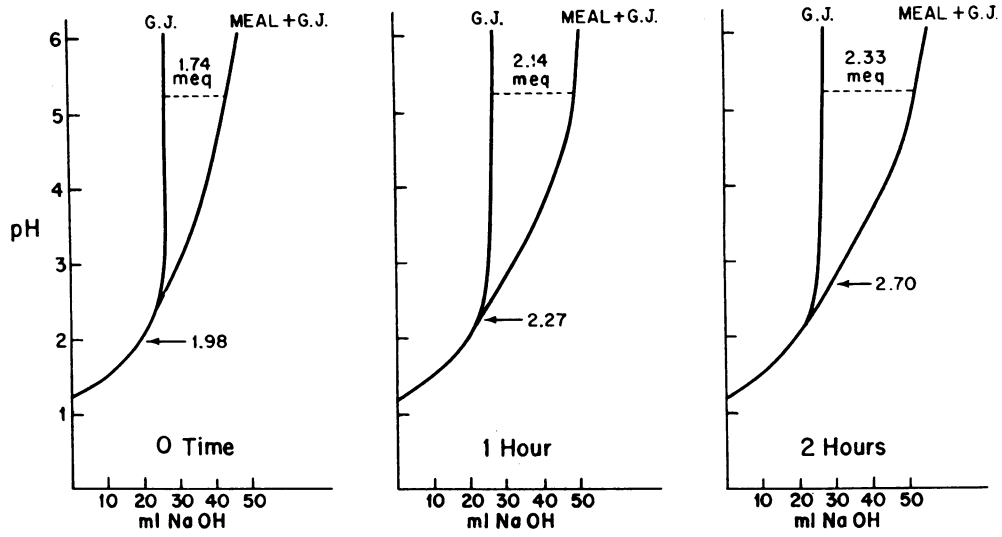

FIGURE 13 Buffer capacity of 10 ml of homogenized meal after incubation with 20 ml fresh gastric juice for 0, 1, and 2 h. The pH of the mixture after each incubation interval (before titration) is indicated by the arrows. Buffer capacity is defined as the difference in the amount of NaOH required to titrate the meal sample and pure gastric juice from a pH of 2.0 to a pH of 5.5.

IN VIVO EXPERIMENTS

Fig. 14 shows the titration of the gastric content with sodium bicarbonate at 1, 2, and 3 h after eating in a patient with duodenal ulcer. Estimated buffer, i.e., the amount of sodium bicarbonate required to bring the pH from 2.5 to 5.25 (see in vitro studies) was 29 meq at 1 h, 5 meq at 2 h, and 1.2 meq at 3 h. For comparison it should be recalled that the buffer capacity of the entire meal was 87 meq.

Hourly buffer capacity measurements such as those shown in Fig. 14 were done in only three subjects. In the rest, buffer capacity was measured only at 2 h, and the results are shown in Fig. 15. Buffer capacity of the stomach at 2 h averaged 23.6 meq in the normal subjects and 9.5 meq in the patients with duodenal ulcer ($P < 0.05$). One of the seven ulcer patients was not studied.

Fig. 16 compares the buffer content of the stomach at 2 h with the peak gastric acid secretion rate after the

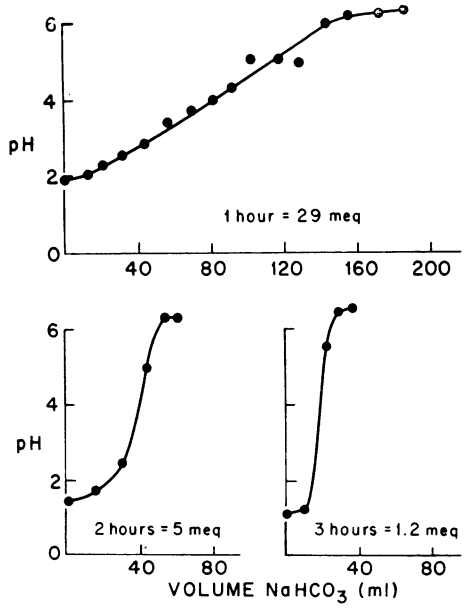

FIGURE 14 Buffer content of the stomach 1, 2, and 3 h after beginning the meal in a duodenal ulcer patient.

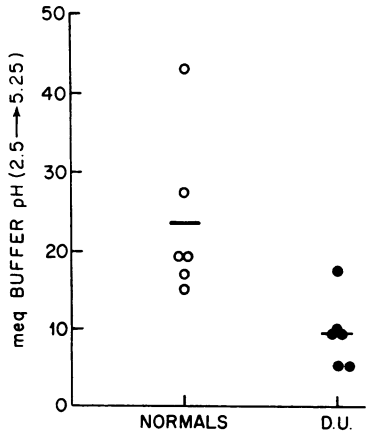

FIGURE 15 Buffer content of the stomach 2 h after beginning the meal in six normal subjects and in six patients with duodenal ulcer.

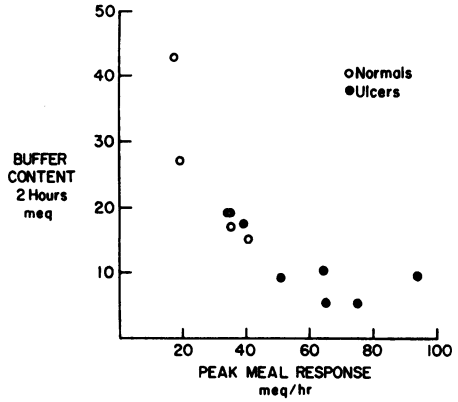

FIGURE 16 Correlation of the peak meal response during any of the 4 h after eating and the buffer content of the stomach 2 h after eating.

meal. There is a distinct tendency for people with high secretory rates to empty the meal more rapidly than people with lower secretory rates, although the correlation appears curvilinear rather than linear on arithmetic scale. Better definition of the type of relationship could probably be obtained by relating the half-life of buffer in the stomach to acid secretion rate, and this is an important subject for future study. The buffer content in the stomach 2 h after eating was also correlated in a negative fashion with the peak histamine response and with the peak meal response expressed as a percentage of the peak histamine response.

As noted previously, a prerequisite for making an estimate of buffer content by the present method is that the gastric fluid pH must be less than 2.5 before titration with sodium bicarbonate. In all of the ulcer patients in whom buffer content was measured 2 h after eating, the gastric fluid pH was less than 2.5 by virtue of their own acid secretion. However, preliminary studies in normal subjects showed that this was frequently not the case; consequently, 0.1 N hydrochloric acid was infused in all of the normal subjects from 1 to 2 h after the meal. Table II shows the pH of the gastric content 1 h after eating, the volume of 0.1 N hydrochloric acid infused before buffer measurement, and the stomach fluid pH and hydrogen ion concentration just before titration with sodium bicarbonate. The average volume of 0.1 N hydrochloric acid infused in the normals was 175 ml, and the average hydrogen ion concentration before alkali titration was 16.8 meq/liter in the normals (which corresponds to a pH of 1.86) and 25.5 meq/liter in the duodenal ulcer subjects (pH 1.68).

TABLE II

Gastric Acidity at 1 h after Eating, Volume of 0.1 N Hydrochloric Acid Infused before 2 h Buffer Content Measurement, and pH and Hydrogen Ion Concentration of Gastric Fluid just before Titration with Sodium Bicarbonate

Subject	Gastric acidity at 1 h		vol 0.1 N HCl infused	Gastric acidity just before titration with $NaHCO_3$	
	pH	$[H^+]$*		pH	$[H^+]$*
		meq/liter	*ml*		*meq/liter*
Normals					
C. C.	2.48	4.0	78	2.03	11.5
J. H.	4.27	0.1	14	1.54	35.8
C. P.	3.61	0.3	192	1.86	16.9
B. B.	3.19	0.9	248	1.80	19.5
J. B.	6.16	0.0	400	2.25	6.9
J. R.	4.18	0.1	118	1.98	12.8
Mean	3.19‡	0.9±0.6	175	1.86‡	16.8±3.5
Duodenal ulcer patients					
M. C.	2.48	4.0		1.41	48.7
J. D.	2.73	2.4		1.86	16.9
P. G.	3.30	0.7		1.88	16.2
A. C.	2.30	6.1		2.03	11.5
X. C.	2.80	1.9		1.67	26.4
G. P.	2.04	11.2		1.57	33.4
Mean	2.34‡	4.5±0.6		1.68‡	25.5±5.6

* By the method of Moore and Scarlata (5).

‡ Value is the pH corresponding to average hydrogen ion concentration, not the average of the pH values.

DISCUSSION

These experiments have shown that acid secretion after a normal meal can be estimated by the amount of sodium bicarbonate required to keep the pH of the stomach contents at 5.5, which is the pH of the meal after homogenization. The accuracy of the method is dependent on the degree to which gastric contents can be mixed. In spite of vigorous efforts to mix the gastric contents, we were not able to maintain gastric pH exactly constant, which indicates that mixing was imperfect. However, this will result in an error in the calculated rate of acid secretion only to the extent that over- and under-titrated gastric content is emptied from the stomach, and errors of over- and under-titration will, to some degree, cancel each other out. The excellent agreement obtained in duplicate tests in six subjects suggests that the method is accurate, although any technique that would improve mixing would enhance accuracy even further.

Acid secretion measured by constant titration (and also that measured by most other tests of acid secretion, including basal and peak histamine secretion) reflects net acid secretion, i.e., true acid secretion minus the rate at which secreted base (salivary, pancreatic, biliary, and nonparietal gastric fluids) is mixed with the gastric contents, and minus the acid loss by back diffusion. The

contribution of secreted base was easily documented by studying patients with pernicious anemia, but the quantity of base secreted into the stomach in these patients probably overestimates that secreted in normal subjects and in patients with duodenal ulcer (9, 10). There is no method available for measuring the rate of base addition to the gastric contents in patients who secrete acid, although by making certain assumptions the rate of parietal and nonparietal secretion can be calculated mathematically (6).

It should be pointed out that acid secretion rates reported in this paper apply to the special circumstance where the pH is maintained constant at an abnormally high level. Based on previous studies in experimental animals and humans, keping the gastric pH at 5.5 (rather than allowing acid secretion to lower gastric pH to below 3) would be expected to have the following effects: First, it would probably speed gastric emptying. Second, it would prevent peptic digestion, which in turn would prevent generation of additional buffer and of polypeptides and amino acids. Third, it would markedly retard back diffusion of hydrogen ions from the gastric lumen to blood. And, fourth, it would permit more antral gastrin release in response to various stimuli (distention, amino acids, dipeptides). Although for these reasons the present method is "unphysiologic," it should be noted that there is some advantage to a method which measures acid secretion and serum gastrin concentration at a constant and controlled level of gastric pH, especially if the results in pathologic conditions such as in patients with duodenal ulcer are compared with results in normal people. In addition, in vitro studies (shown in Fig. 1) suggest that constant titration of the gastric content to any level between 3.0 and 5.5 may be used to measure acid secretion; therefore, the effect of gastric pH (within this range at least, and probably at lower pH levels) on acid secretion can be determined by constant titration in future studies.

With gastric pH maintained constant at 5.5 the net rate of gastric acid secretion in seven patients with duodenal ulcer was found to be approximately twice that in a group of healthy control subjects. Furthermore, there was a tendency for the duodenal ulcer subjects to secrete at a higher percentage of their peak histamine response than the normal subjects. This might have important bearing on the pathogenesis of duodenal ulcer and suggests that the parietal cell responsiveness to a meal as well as the parietal cell mass (as estimated by the peak histamine response) may determine the amount of acid secreted by a given individual. The recent study of Isenberg, Best, and Grossman (11) and of Richardson and Fordtran [1] suggesting that patients with duodenal ulcer are hyper-responsive to small doses of pentagastrin and histamine, would be compatible with this concept. Another possible explanation for the greater responsiveness of the ulcer patients is that they received larger volumes of alkali (to match their higher rates of acid secretion) than the controls, and this may have augmented their acid secretory response to the meal. On the other hand, three of the control subjects (C. C., J. H., and B. B.) had peak histamine responses averaging 40.5 meq/h, and these can be compared with three of the ulcer patients (J. D., P. G., and G. P.) whose average peak histamine response was 41.5 meq/h. In these three control subjects, the average percentage of acid secretion after the meal compared with the peak histamine response was 91.3%, while the similar value for the three ulcer patients was 133%. This suggests that the tendency for duodenal ulcer patients to secrete a greater percentage of their peak histamine response after eating than normal subjects is probably not due simply to the fact that larger volumes of alkali were required to neutralize the secreted acid. The problem is obviously not completely settled, however, and in the future it will be important to control the variable of volume by infusing a more concentrated alkaline solution in high secretors than in low secretors, so that the volume of intragastric infusion in different groups will be more comparable.

The pattern of acid secretion after eating is of some interest. Acid secretion did not become maximum until the second 30 min period in ulcer patients and not until the third 30 min period in the control group. After reaching a peak, secretion gradually diminished over the next 2–3 h period. 4 h after eating, secretion rate was about one-third maximal in the duodenal ulcer patients and about one-fourth maximal in the control subjects; this difference is probably due to the same unknown factors which are responsible for the higher ratio of basal to peak histamine secretion in the duodenal ulcer subjects compared with the normal group (as calculated from the data in Table I).

The serum gastrin concentrations after eating, with gastric pH maintained constant at 5.5, were no higher than in a previous study when subjects were fed a beef meal and gastric pH allowed to seek its natural level (12). When gastric pH was not manipulated, the peak serum gastrin occurred about 30–60 min after eating, probably before gastric contents were significantly acidified. For this reason, it is perhaps not surprising that maintaining gastric pH at 5.5 made little or no difference in peak levels of gastrin. However, it is surprising that neutralization did not raise gastrin to higher levels at 2 and 3 h after the meal. This observation suggests that antral pH in humans may not have the important

[1] Richardson, C. T., and J. S. Fordtran. 1973. Histamine dose response curves: correlation with basal gastric secretion, Ewald meal response, gastric emptying, serum gastrin and parotid salivary flow. Unpublished results.

physiologic regulatory influence on protein-stimulated antral gastrin release that has been predicted from physiologic studies in animals. Further study correlating variations in gastric pH, acid secretion rate and serum gastrin concentrations in the same subjects will be necessary to settle this point with any degree of certainty.

In the present studies, mean fasting and postcibal serum gastrin concentrations were higher in the duodenal ulcer subjects than in our normals. Wide difference in the level of serum gastrin concentrations were evident within each group, however, and the difference between groups in mean gastrin concentrations was not statistically significant. On the other hand, there was a rough positive correlation between the absolute level of gastrin 1 h after eating and the peak secretory response expressed as a percentage of the maximum histamine response ($r = 0.45$). The absolute and percent postcibal rise above fasting gastrin concentration correlated less well with the secretory response than the absolute concentration of gastrin 1 h after the meal. We conclude that the higher meal response of the duodenal ulcer patients than normals, relative to their peak histamine response, may in part be related to higher gastrin levels after the meal, but that other factors are almost certainly involved.

A second objective of this study was to measure buffer capacity of the stomach contents after eating a meal. In vitro experiments showed that the reaction of meal buffer with acid was reversible, and that acidified buffer remaining in the stomach after a meal could therefore be measured by back-titration with alkali. Additional experiments revealed that buffer capacity of any given sample of the meal, of the meal plus gastric juice, or of any sample of the postcibal stomach content could be determined from its titration curve with sodium bicarbonate, equating the amount of bicarbonate necessary to raise the pH from 2.5 to 5.25 with buffer capacity. It was also shown that on incubation of the meal and fresh gastric juice at 37°C, a 33% increment in buffer capacity is generated due to peptic digestion.

The buffer capacity of the meal used in these studies was 87 meq after homogenization. The buffer which entered the stomach was probably less than 87 meq since normal chewing does not disperse the meal as well as homogenization. Buffer content of the stomach at hourly intervals after eating was studied in three subjects (an example is shown in Fig. 14), and rather rapid decrease in buffer capacity during this period was noted in each subject. Buffer content was measured at 2 h after eating in all six of the normal subjects and in six of the seven duodenal ulcer subjects. The ulcer patients had less than half as much buffer in their stomachs 2 h after eating as the normal subjects. Furthermore, when the two groups were considered together, there appeared to be an inverse correlation between the rate of acid secretion and buffer content in the stomach at 2 h (Fig. 16), i.e., patients with higher rates of acid secretion had less buffer remaining in their stomachs and vice versa. It should be noted that more rapid buffer emptying in high secretors probably cannot be explained simply by higher gastric volumes in the hypersecretors, since in the low secretor group 0.1 N hydrochloric acid was infused into the stomach to lower gastric pH before the time that buffer capacity was measured. It is also unlikely that this difference in buffer emptying can be explained by the lower gastric pH in high secretors during the 1st h after eating (Table II), since high acidity is generally assumed to reduce the rate of gastric emptying.

While no previous workers have, as far as we are aware, specifically measured gastric buffer content after a meal, several investigators have estimated gastric emptying of test meals in normal subjects and in patients with duodenal ulcer. Shay used a barium and water test meal and by X-ray found that patients with duodenal ulcer have rapid gastric emptying (13). Hunt used test meals of saline, glucose, and glucose plus acid and found that gastric emptying as measured by the serial sampling technique was the same in patients with duodenal ulcer and in normal students (14). Brömster, Carlberger, and Lundh found no difference in ulcer and control subjects in the half-life in the stomach of a liquid test meal as measured by external isotope counting (15), while Griffith, Owen, Campbell, and Shields, using a similar method, found that a meal of porridge, eggs, milk, and bread was emptied more rapidly in duodenal ulcer patients than in controls (16). Finally, George recently reported that by a double sampling technique and water as a test meal, duodenal ulcer and normal subjects have similar rates of gastric emptying (17). Thus, the results of previous studies are contradictory and have not established whether or not duodenal ulcer subjects have normal or increased rates of gastric emptying. It is interesting, however, that in the only previous study which employed a normal type meal (16), gastric emptying was more rapid in duodenal ulcer patients than in controls, and this would agree with our finding of less gastric buffer capacity after a steak meal in duodenal ulcer patients than in controls. Nevertheless, we wish to emphasize that our method measures only the emptying rate of buffer and not necessarily the emptying rate of the total mass in the stomach. The mechanisms of emptying of buffer may not be the same as for total mass.

Combining the acid secretion and buffer emptying data presented in this paper,[2] three tentative conclusions

[2] It is emphasized that before estimation of buffer content the gastric pH was 2.5 or lower, whereas when acid secretion was measured gastric pH was maintained at 5.5. Emptying and secretion were not, therefore, studied under

seem appropriate. First, it is apparent that the steak meal used in these experiments elicited a higher secretory fraction of the peak histamine response in the duodenal ulcer patients (116%) than in the normal subjects (86%), suggesting that parietal cell responsiveness to a meal is greater in duodenal ulcer patients than in controls. Second, the duodenal ulcer patients emptied the meal's buffer much more rapidly than normal subjects, which would result in reduced efficiency of the buffer in preventing a rise in gastric acidity. (The importance of these two abnormalities under normal physiologic conditions is evident from the fact that gastric acidity, when allowed to seek its natural level, was much higher 1 h after the meal in the ulcer patients than in the control subjects. Furthermore, acidity was higher 2 h after eating in the ulcer patients than in the normals, in spite of exogenous gastric acid infusion in the normal subjects [Table II].) Third, the duodenal ulcer patients continued to secrete acid at high rates even after more than 90% of the meal's buffer had been emptied from the stomach, and these continued high rates of acid secretion after almost all buffer has been emptied from the stomach seem physiologically inappropriate. All three of these factors may be important in the pathogenesis of duodenal ulcer, and all three could be manifestations of a single underlying abnormality, such as increased vagal tone or a defect in hormonal inhibition of gastric motility and secretion by duodenal factors.

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the same conditions of intragastric pH, and this qualification should be kept in mind when acid secretion and buffer content are compared.

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