

Effect of food on serum gastrin evaluated by radioimmunoassay

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SUMMARY The effect of food on serum gastrin in normal man has been evaluated by radioimmunoassay. Protein and amino acids produced up to a five-fold increase in serum gastrin levels, followed in potency by alcohol, fat, and glucose. Distilled water did not stimulate the release of gastrin. An injection of atropine sulphate (0.6 mg) augmented the response to all good stimuli but in itself did not affect serum gastrin levels in the basal state.

In animals the release of gastrin in response to feeding involves both vagal impulses (Uvnäs, 1942) and the activation of a local mechanism in the antrum (Woodward, Lyon, Landor, and Dragstedt, 1954; Baugh, Bravo, Barcena, and Dragstedt, 1956; Elwin and Nilsson, 1963).

Sensitive radioimmunoassay now permits the direct measurement of gastrin levels in serum, and rises in immunoreactive gastrin have been reported following both feeding (Hansky and Cain, 1969; McGuigan and Trudeau, 1970) and insulin hypoglycaemia (Korman, Soveny, and Hansky, 1971).

This study examines the effect of standard meals and individual food constituents on serum immunoreactive gastrin with and without prior atropinization.

Material and Methods

Normal healthy volunteers were investigated after an overnight fast. These comprised 20 males and three females between the ages of 20 and 30 years. Subjects were investigated in groups of at least four, and informed consent was obtained from each subject. A 19-gauge needle was inserted into a forearm vein and patency ensured by frequent flushing with a solution of heparin, 1,000 units in 20 ml of 0.9% sodium chloride. This solution has been shown not to affect serum gastrin levels.

Three groups had a meal which was high in either protein, carbohydrate, or fat, and blood was drawn for gastrin estimation at -90, -60, -30, 0, 15, 30, 45, 60, 75, 90, 105, and 120 minutes. The test was

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repeated in each subject with intramuscular atropine sulphate 0.6 mg 30 minutes before the meal.

PROTEIN MEAL

Ten subjects had a meal comprising steak and eggs, cheese and milk. Contents of the meal were protein 60g, fat 51g, and carbohydrate 11g.

FAT MEAL

Eight subjects had a meal comprising eggs scrambled in butter, pears and cream. Contents of the meal were protein 27.5g, fat 61g, and carbohydrate 11.2g.

CARBOHYDRATE MEAL

Five subjects had a meal comprising cereal, sugar and milk, toast and honey. Contents of the meal were protein 18g, fat 9.5g, and carbohydrate 103g.

A further five groups were investigated after the ingestion of a specific food constituent. These were alcohol, casilan, glycine, cream, and glucose. Blood was drawn at -60, -30, 0, 5, 10, 15, 20, 30, 45, 60, and 90 minutes. The study was repeated with intramuscular atropine sulphate, 0.6 mg, given 30 minutes before the food.

ALCOHOL

Four subjects had a drink of 250 ml of vodka equivalent to 10g of alcohol.

CASILAN

Four subjects had a drink comprising 60g casilan in milk. The contents of this are no carbohydrate, 1.4g fat, and 52g protein.

GLYCINE

Four subjects had a drink of 250 ml of 0.3 M glycine.

CREAM

Four subjects had a drink of 250 ml of fresh cream. This contains 7g protein, 99g fat, and 9g carbohydrate.

GLUCOSE

Four subjects had a drink of 80g glucose in 250 ml of water.

Two further groups of subjects were studied and blood was drawn for gastrin estimation at -60, -30, 0, 15, 30, 45, 60, 75, 90, and 120 minutes. Four subjects had a drink of 250 ml of distilled water and six subjects were given 0.6 mg of atropine sulphate intramuscularly.

Of the 23 subjects studied, five had tests involving four or more stimuli, three had three stimuli, and the remaining 15 subjects had one or two stimuli.

The gastrin concentration in 0.5 ml serum was estimated in duplicate by radioimmunoassay (Hansky and Cain, 1969). Since the original report modifications have improved both sensitivity and reproducibility and these will be briefly described. Initially the incubation period for labelled gastrin, antiserum, and unknown was 24 hours which gave 40% binding in the absence of unlabelled gastrin. More recently, a 72-hour incubation has given 50% binding in the absence of unlabelled gastrin. Standard curves (Figure 1) were obtained by the assay of from

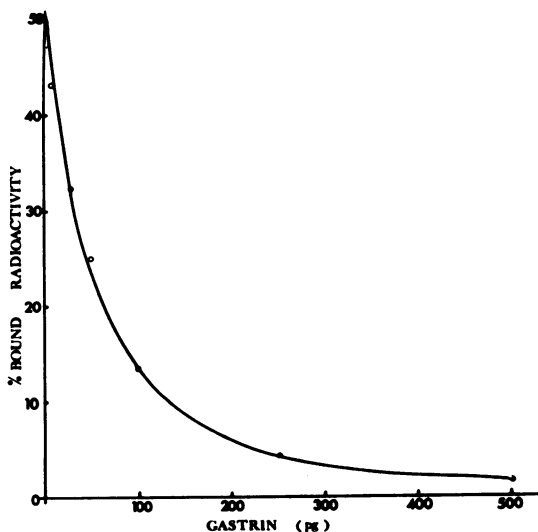


Fig. 1. Calibration curve for radioimmunoassay with antibody (dilution 1/5000) showing percentage antibody-bound ^{125}I -gastrin plotted against increasing amounts of unlabelled gastrin.

0 to 500 pg human gastrin I (HGI) in 0.5 ml of serum freed of gastrin by prior charcoal absorption. The sensitivity of the assay system is 5 pg per ml. Reproducibility was assessed by freezing aliquots of serum with a gastrin concentration in the middle of the normal range (≈ 50 pg/ml). These were assayed serially in lots of five triplicates over a number of weeks so that eight sets of 18 determinations were made. For each group of five triplicates the coefficient of variation was 2.6% (within-assay variation) and for the eight sets of sera assayed at three-day intervals the coefficient of variation was 7.3% (between-assay variation).

The group means were analysed by Student's *t* test on an Olivetti table computer using standard formulae.

Results

PROTEIN MEAL

This (Fig. 2) caused a significant rise in serum gastrin from a mean (\pm SEM) fasting level of 15 ± 2.3 pg/ml to 78 ± 16.0 pg/ml at 45 minutes after ingestion ($P < 0.005$). When repeated with a prior injection of atropine, there was a rise from 22 ± 3.1 to 88 ± 10.4 pg/ml at 45 minutes after ingestion ($P < 0.005$). The magnitude of the response was

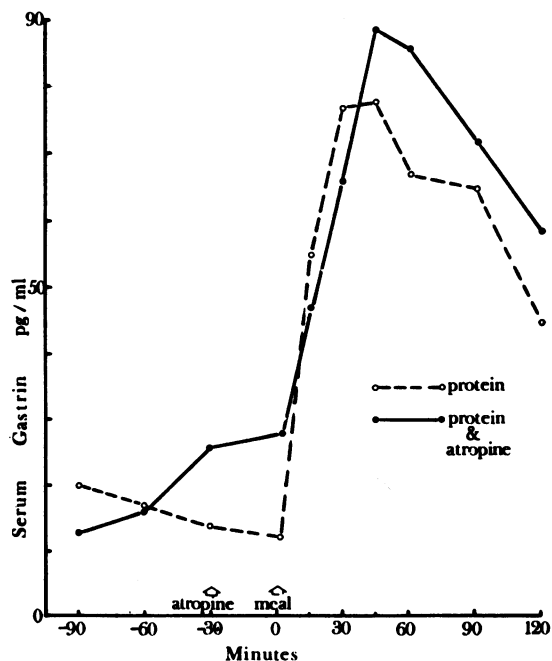


Fig. 2. Serum gastrin response to protein meal with and without prior intramuscular injection of atropine sulphate 0.6 mg.

similar in both studies although the response after atropine was prolonged.

FAT MEAL

This (Fig. 3) caused a significant rise in serum gastrin from a basal level of 24 ± 8.4 pg/ml to 52 ± 19.8 pg/ml at 30 minutes ($P < 0.05$).

Prior atropinization caused a greater and more significant rise from 25 ± 6.0 pg/ml to 78 ± 11.4 pg/ml at 90 minutes ($P < 0.005$). The peak response occurred later with prior atropinization and the duration of the response was prolonged.

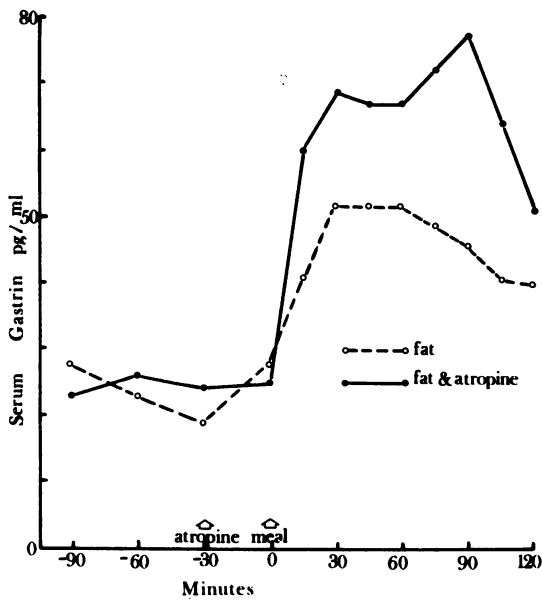


Fig. 3. Serum gastrin response to fat meal with and without prior intramuscular injection of atropine sulphate 0.6 mg.

CARBOHYDRATE MEAL

This (Fig. 4) caused a significant rise in serum gastrin from 20 ± 3.4 pg/ml to 57 ± 11.5 pg/ml at 45 minutes ($P < 0.005$). Prior atropinization caused a similar rise from 15 ± 2.9 pg/ml to 52 ± 13.9 pg/ml at 90 minutes ($P < 0.005$). The peak response was delayed by prior atropinization.

ALCOHOL

This (Fig. 5) did not cause a significant rise in gastrin (13 ± 5.6 to 27 ± 16.3 pg/ml at 10 minutes, $P = 0.15$), but prior atropinization produced a significant rise in gastrin from 31 ± 6.9 pg/ml to 73 ± 32.8 pg/ml at 15 minutes ($P < 0.05$).

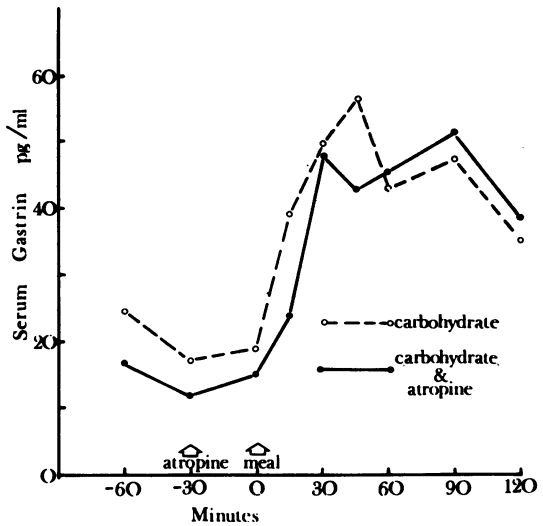


Fig. 4. Serum gastrin response to carbohydrate meal with and without prior intramuscular injection of atropine sulphate 0.6 mg.

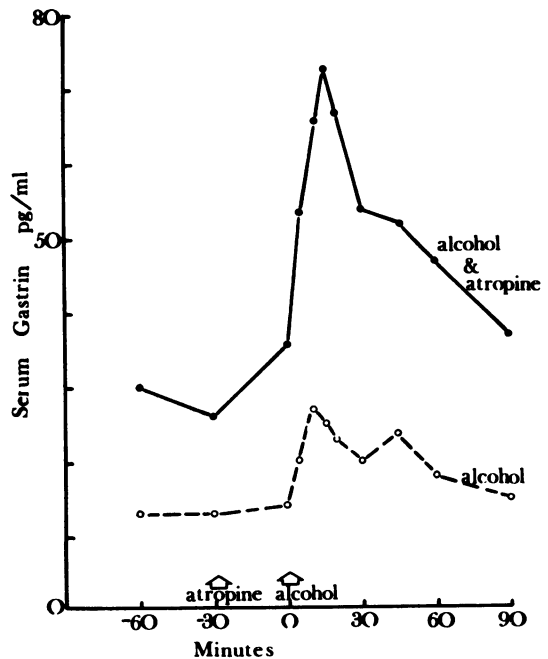


Fig. 5. Serum gastrin response to alcohol with and without prior intramuscular injection of atropine sulphate 0.6 mg.

CASILAN

Casilan (Fig. 6) caused a rise from 31 ± 9.8 pg/ml to 78 ± 36.3 pg/ml at 15 minutes after ingestion ($P = 0.05$). Prior atropinization caused a greater response from 48 ± 10.9 to 135 ± 79.4 pg/ml at 45 minutes ($P = 0.05$). This response was of greater magnitude and more prolonged after atropine.

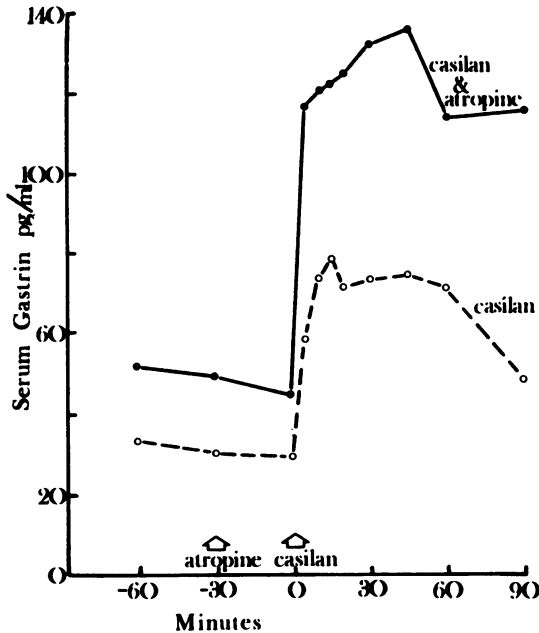


Fig. 6. Serum gastrin response to casilan with and without prior intramuscular injection of atropine sulphate 0.6 mg.

GLYCINE

This (Fig. 7) caused a rise from 12 ± 3.7 pg/ml to 40 ± 26.8 pg/ml at 30 minutes ($P < 0.05$). When repeated with atropine, a greater and significant rise occurred (17 ± 4.8 to 88 ± 29.3 pg/ml at 15 minutes, $P < 0.005$). The response after atropine was prolonged.

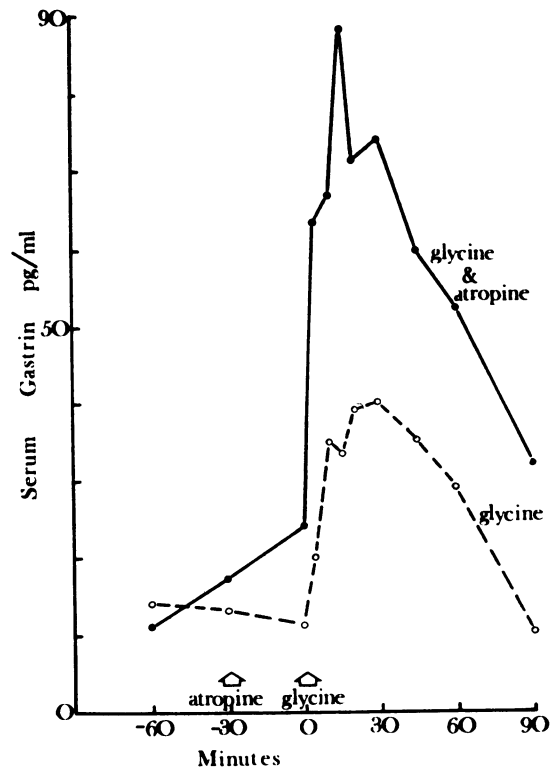


Fig. 7. Serum gastrin response to glycine with and without prior intramuscular injection of atropine sulphate 0.6 mg.

CREAM

The ingestion of cream (Fig. 8) did not cause a significant rise in serum gastrin (32 ± 4.5 to 44 ± 13.8 pg/ml at 15 minutes, $P = 0.25$). When repeated after prior atropinization, it rose from 23 ± 15.1 to 57 ± 13.0 pg/ml at 30 minutes but again the rise was not significant ($P < 0.15$). However, the response was delayed and of greater magnitude than without atropine.

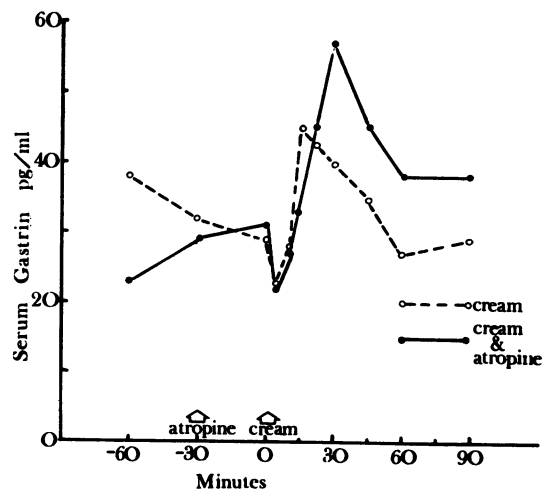


Fig. 8. Serum gastrin response to cream with and without prior intramuscular injection of atropine sulphate 0.6 mg.

GLUCOSE

Glucose (Fig. 9) produced a rise from 1 ± 0.7 to

10 ± 3.8 pg/ml at 10 minutes ($P < 0.01$). These figures are close to the limits of sensitivity of the assay and interpretation of the significance of this rise is questionable. Prior atropinization caused a significant rise from 8 ± 3.3 to 22 ± 8.5 pg/ml at 10 minutes ($P < 0.025$).

Figure 10 shows the response to atropine and distilled water alone. With distilled water, gastrin was unchanged (19 ± 2.6 to 24 ± 1.8 pg/ml, $P = 0.1$) and similarly with atropine there was no significant alteration in gastrin levels (28 ± 5.9 to 35 ± 18.1 pg/ml, $P = 0.3$).

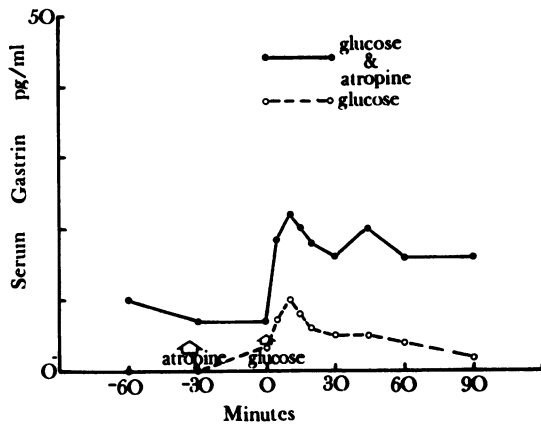


Fig. 9. Serum gastrin response to glucose with and without prior intramuscular injection of atropine sulphate 0.6 mg.

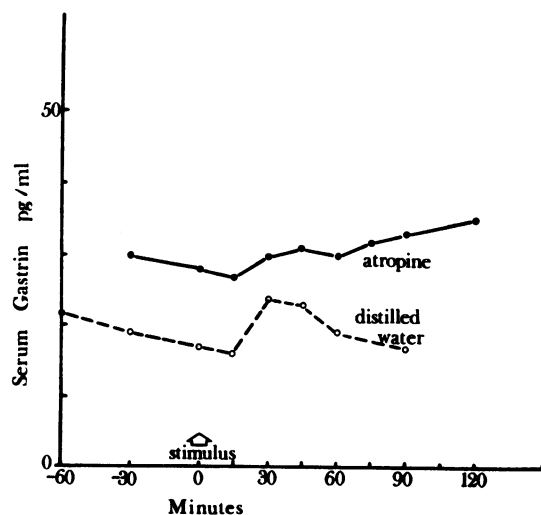


Fig. 10. Serum gastrin response to 250 ml of distilled water and to the intramuscular injection of atropine sulphate 0.6 mg.

Discussion

These studies indicate that in normal man the ingestion of various foods elicits a rise in serum gastrin. As judged by the time of occurrence of the peak response this is probably due to local stimulation of gastrin release. That there is a vagal component as well is suggested by the commencement of the response within 10 minutes of ingestion. Although comparisons of stimuli were not always made in the same subjects, a number had their response studied following four or five different stimuli. The responses of these individuals were similar to the mean of each group studied, and hence it was felt that valid conclusions could be drawn in the comparison of different foods.

Cooke and Grossman (1968) compared stimulants of antral release of gastrin in dogs and found that acetylcholine was most powerful followed by glycine, and that ethanol, meat extracts, and sodium bicarbonate were weak stimulants. In the present study it was found that proteins and amino acids were the most potent releasers of gastrin. In terms of increase of gastrin above fasting levels, the figures were 63 pg/ml for a protein meal, 47 pg/ml for casilan, and 28 pg/ml for glycine. Predominantly fat or carbohydrate meals which contained a significant amount of protein raised the serum gastrin level by 28 pg/ml and 37 pg/ml respectively. However, pure fat (cream) and glucose produced increments of only 12 and 9 pg/ml respectively. It is therefore evident that fat and carbohydrates as such are poor stimulants of gastrin release and stimulation by meals containing predominantly these constituents is in the main due to their protein content.

Although there is general agreement about protein and amino acids, Woodward, Robertson, Ruttenberg, and Shapiro (1957) showed that dilute alcohol was a powerful stimulant of the antral gastrin mechanism and this effect was abolished by acidification of the alcohol to a pH of 1.2. In this study alcohol produced a rise in serum gastrin of 14 pg/ml above basal levels. This was not significant and suggests that in man, unlike the dog, alcohol is a poor stimulant of gastrin release.

Emås and Grossman (1969) found that truncal vagotomy caused an increase in the response of Heidenhain pouches to a feeding meal. They suggested that although the vagal component of gastrin release was eliminated, these procedures produced stasis of food in the stomach and increased the contact time with antral mucosa. The present results after atropine form an analogous situation. Here, casilan, glycine, and a protein meal produced the greatest increase above basal levels (87 pg/ml, 71

pg/ml, and 66 pg/ml respectively), followed by alcohol (42 pg/ml), cream (34 pg/ml), and glucose (14 pg/ml). It is of some interest that predominantly liquid meals produced a greater rise in serum gastrin above basal levels in the atropinized state compared to the situation without prior atropine. This suggests that the serum gastrin response to these liquid stimuli is modified by their rapid passage into the intestine and decreased time of contact with antral mucosa, a situation which is partly remedied by atropine. In agreement with Walsh, Yalow, and Berson (1970), atropine alone was shown to have no effect on serum gastrin in normal man in the absence of a chemical stimulant.

It is of interest to compare our results with those of two other series published recently. McGuigan and Trudeau (1970) showed that the peak serum gastrin response to feeding meals, glycine, and sodium bicarbonate occurred from 20 to 40 minutes after ingestion. These are similar to the findings in the present study where the peak response occurred 30 to 45 minutes after ingestion. However, Byrnes, Young, Chisholm, and Lazarus (1970a), studying both normal subjects and patients with duodenal ulcer, in response to a liquid protein meal found a rapid peak response within 10 minutes which fell to normal levels within 30 minutes. They also reported levels of up to 26 ng/ml which are well above those found in the present study or in that by McGuigan and Trudeau (1970).

The time differences of these responses may be explained on the basis that the groups are measuring different components of gastrin (Byrnes, Lazarus, and Young, 1970b). The present assay and that of McGuigan and Trudeau (1970), based on antibodies to human gastrin I, measure gastrin I, gastrin II, and probably 'big' gastrin (Yalow and Berson, 1970), and these may be the forms of 'gastrin' released by local stimulation. Byrnes *et al* have an assay based on pentagastrin antibodies which measures all peptides having the C terminal tetrapeptide sequence of gastrin. Some peptide having this C terminal group, related to but not gastrin I or gastrin II, may be the form of 'gastrin' predominantly released by vagal stimulation (Hansky, Korman, and Soveny, 1970; Byrnes *et al*, 1970b). If this hypothesis is true, it would be expected that Byrnes would obtain less stimulation after giving atropine before food whereas we find augmentation of response to food. Further, Byrnes, Lazarus, and Young (1970c) find that atropine causes a fall in basal gastrin levels whereas the present study shows no change. These differences open up a most interesting vista as to the form in which gastrin is secreted in response to different stimuli and inhibitors.

These studies do indicate that there may be some physiological basis for cream, white meat, bread and butter diets in the therapy of duodenal ulceration. Indeed if gastrin has any acid stimulatory capacity then pure protein feedings would be least desirable and pure fat or carbohydrate feedings most desirable. However, any therapeutic applications of the present results would have to be tempered by the assessment of the acid neutralizing properties of substances studied as well as of their ability to release gastrin.

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