# Effect of Bile and Fat on Gastric Motility under the Influence of Various Stimulants

J. J. Anderson, M.D., R. J. Bolt, M.D.,* B. M. Ullman, M.P.H., and P. Bass, Ph.D.

Experimental studies in rats, dogs, and humans have demonstrated the inhibitory effects of intraduodenal fat on spontaneous gastric motility.[1–3] Most of these studies have been done with open-tip and balloon-pressure recordings. The experiments in animals are frequently done under acute conditions or with the gastrointestinal tract altered by fistulas. To overcome unreliability in the results obtained by these experimental methods, we recorded contractility of circular and longitudinal smooth muscle layers from the gastric body and antrum by using extraluminal force transducers. The size of the recording units and the implant procedure did not interfere with intraluminal contents and allowed chronic recordings without altering the physiologic environment.[4–6]

Since spontaneous gastric motility is intermittent, the inhibitory effect of fat was also tested against various stimulants. Fat inhibition of gastric motility is mediated through a humoral pathway.[7] Frazer emphasized the importance of the physical state of fat for inhibition of gastric motility. Micellar fat is presumably the physical-chemical state under which lipids are most readily absorbed by the small bowel mucosa.[9] For the above reasons, we used oleic acid, glycerol-1-monooleate, and bile under conditions conducive to micellar formation.[10] While this study was in progress, Swan *et al.* reported that micellar fat, placed in a duodenal loop of dogs, exerted no inhibitory effect on the spontaneous motility of Heidenhain pouches.[11]

The purpose of this study was to determine, using extraluminal force transducers, (1) the effect on gastric contractility when saline, bile, or micellar fat was placed in a duodenal Thiry loop; (2) to see if the preparation in the duodenum differentially affected the gastric body or gastric antrum; (3) to determine if longitudinal or circular muscle layers were more susceptible to the inhibiting influence of substances placed in the duodenum; and (4) to ascertain whether fat inhibition of motility was effective when stomach contractility was driven by various stimuli.

---

From the University of Michigan Medical Center and Parke, Davis and Company Research Laboratories, Ann Arbor, Mich.

*Present address: Division of Medical Sciences, University of California School of Medicine, Davis, Calif.

## METHODS

### Animal Preparations

The technic of constructing extraluminal force transducers has been described.[12] These were implanted into 6 healthy beagle dogs weighing approximately 10 kg. and the following surgical procedure was performed (Fig. 1). The proximal duodenum was transected just below the common and

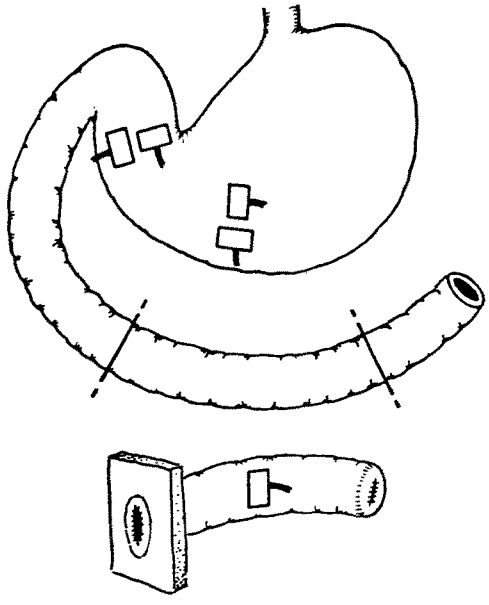

**Fig. 1.** Schematic representation of arrangement of extraluminal force transducers on body and antrum of stomach and on Thiry loop of duodenum. Transducers are arranged to record from either transverse or longitudinal axis of organ. Tracings taken from duodenal loop not included in this study.

pancreatic ducts. A 12-cm. Thiry loop was made with the stoma formed from the caudad end near the ligament of Treitz. The duodenum was anastomosed end to end.

### Experimental Procedure

Gastric contractility was monitored for a 30-min. period while under the influence of stimuli and with "inhibiting" substances in the duodenal Thiry loop. The gastric stimuli used were: (1) food, 200 gm. of horse meat* (analysis: protein—11.5%, fat—4.0%, fiber—1.0%); (2) acetylcholine (Ach) given intravenously at 2 concentrations: 50 μg./kg./min. and 200 μg./kg./min., and (3) 5-hydroxytryptophane (5 HTP) given intravenously at a concentration of 600 μg./kg./min. At the start of each experiment the stoma of the Thiry loop was sealed with a No. 16 Foley catheter and 10 ml. of one of the following substances was instilled after adjustment to pH 7: (1) 0.9% NaCl; (2) micellar fat, consisting of 2.5 mM of glycerol-1-monooleate and 5 mM of oleic acid in 100 ml. of dog bile diluted with 0.9% sodium chloride in a ratio of 1:10; and

*Country Best Foods, Agway Inc., Syracuse, N. Y.

(3) dog bile diluted with 0.9% sodium chloride in a ratio of 1:10. The substances were placed in the Thiry loop 1–3 min. before administering the gastric stimulants.

A total of six animals were prepared for this study. Each of 5 dogs was subjected to the following 18 randomized treatments. Four treatments replicated twice consisted of either instilling 0.9% sodium chloride into the Thiry loop and infusing Ach (50 or 200 μg./kg./min.) or 5-HTP (600 μg./kg./min.) intravenously for 30 min., or feeding horse meat (200 gm.). Two experiments consisted of placing saline in the loop and not administering gastric stimulants. The remaining 8 procedures consisted of challenging the gastric stimulants with bile in the loop in 4 experiments and micellar fat in the loop in 4 experiments. The sixth dog was used for establishing the dose response levels of the stimulants and determining whether bile or micellar fat in the loop would antagonize spontaneous activity.

## Measurement and Analysis of Data

The recorded tracings obtained from each muscle layer were analyzed for number and amplitude of contraction in a 30-min. period. Contractions of 5 gm. force or greater were the only ones considered for measurement. Anything less was not easily differentiated from base line artifact. The amplitude was assessed using a "motility index" system.[13] This was obtained by placing a calibrated (gram of force) transparent grid over the record and counting the number of contractions which fell within specific levels of force. Each contraction received a weighted score of 1, 2, 4, 8, or 16, corresponding to recorded amplitudes of 5–10 gm., 10–20 gm., 20–40 gm., 40–80 gm., and > 80 gm., respectively. These scores were added together to provide an index for the 30-min. period. This approach considers the height of the deflection, the calibration of the sensors, the gain of the polygraph, and the entire period of observation.

The average height of contraction from each site monitored in a given 30-min. period was also calculated by dividing the motility index by the number of contractions. This is referred to as the mean motility index.[14] Analysis of variance was performed on these data, as well as on the number of contractions and on the motility index.

To assess the effectiveness of each of the inhibitors with each of the stimulants, a factorial design was utilized for the experiments. Since, however, certain cells had more replications than others, approximations in the analysis of variance were necessary to adjust for this. A measure of over-all effectiveness for the entire stomach was constructed by combining the corresponding measures for each of the 4 areas monitored. Further, to evaluate differences of muscle layers in both gastric body and antrum and between these 2 areas, the 3 corresponding paired differences also were subjected to a factorial analysis of variance. Significance was taken to be the .05 level. The

terms "trend" or "tend to" are used when a numerical difference appeared which was not statistically significant.

## RESULTS

Since the spontaneous contractile activity of the antrum and body is variable, the ability of bile or micellar fat to influence spontaneous contractility could not be assessed. A single dog study confirmed the variability of the control pattern and the "false" impression that bile and micellar fat could inhibit gastric contractility (Fig. 2). An attempt to obtain a continuous motor pattern was made by administering stimulants.

General effects on the number of contractions in 30 min. under the various conditions studied are seen in Table 1. It should be emphasized that the expected number of contractions (150/30 min.) was not obtained since no contractions below 5 gm. of force were tabulated. The analysis of variance

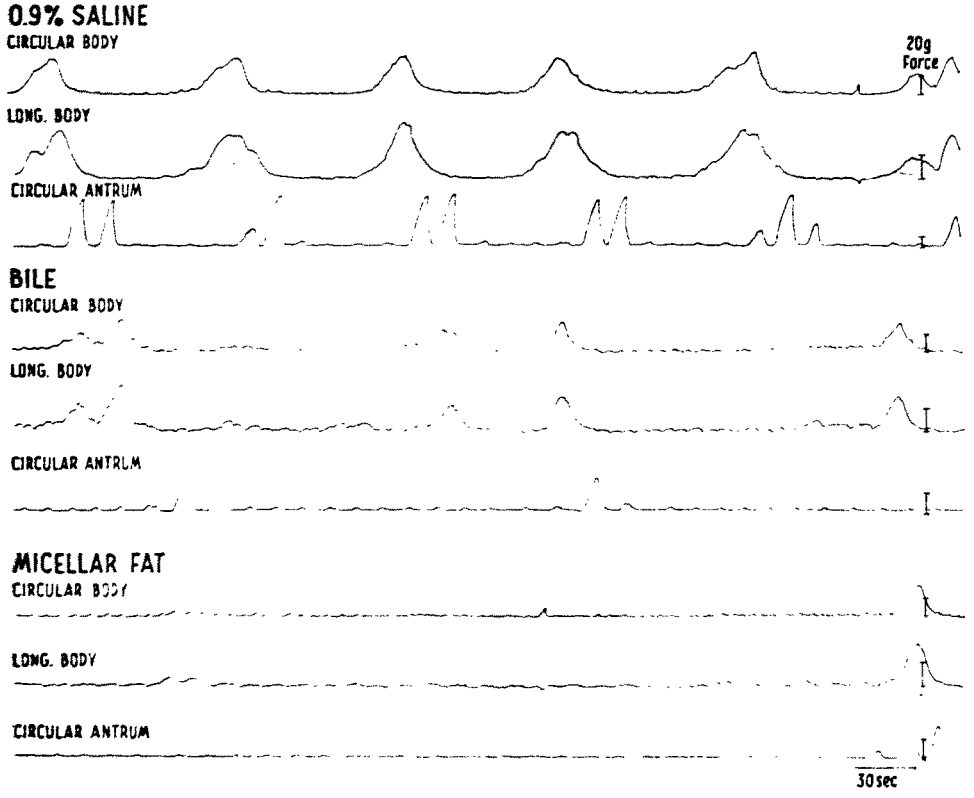


**Fig. 2.** Tracings of spontaneous response of 3 areas of stomach when saline, bile, or micellar fat are instilled into Thiry loop of duodenum. Did latter two substances inhibit contractility or were these "normal" periods of quiescence? Tracings obtained from same animal on different days. Force bars correspond to 20 gm. of force. Time bar equals 30-sec. duration.

TABLE 1. MEAN NUMBER OF CONTRACTIONS IN 30 MINUTES

| Stimulant | Substance in Thiry loop | No. contractions: Gastric body, Circ. axis | Gastric body, Long axis | Gastric antrum, Circ. axis | Gastric antrum, Long axis | Mean | Over-all mean |
|---|---|---|---|---|---|---|---|
| Ach | Micellar fat | 32.8 | 6.6 | 14.8 | 16.6 | 17.7 | |
| (50 μg./kg./min.) | Bile | 23.4 | 2.4 | 19.8 | 30.4 | 19.0 | 20.0 |
| | Saline | 24.6 | 5.4 | 30.8 | 31.8 | 23.2 | |
| Ach | Micellar fat | 20.6 | 8.2 | 62.0 | 68.4 | 39.8 | |
| (200 μg./kg./min.) | Bile | 25.4 | 6.6 | 52.2 | 52.2 | 34.1 | 37.6 |
| | Saline | 23.4 | 8.0 | 62.8 | 60.8 | 38.8 | |
| 5-HTP | Micellar fat | 107.0 | 27.8 | 92.2 | 92.4 | 79.8 | |
| (600 μg./kg./min.) | Bile | 127.8 | 69.0 | 131.0 | 134.0 | 115.4 | 99.8 |
| | Saline | 120.8 | 54.4 | 117.2 | 124.0 | 104.1 | |
| Food | Micellar fat | 69.6 | 18.6 | 53.2 | 30.2 | 42.9 | |
| (200 gm.) | Bile | 51.0 | 14.2 | 56.4 | 48.2 | 42.4 | 41.7 |
| | Saline | 62.0 | 15.6 | 45.0 | 36.4 | 39.8 | |
| MEAN | | 57.4 | 19.7 | 61.4 | 60.4 | | |
| OVER-ALL MEAN | | 38.6 | | 60.9 | | | |

indicates that there was no statistically significant change in the number of contractions induced by the 3 stimuli—acetylcholine (2 doses), 5-HTP, and food—when either bile, micellar fat, or saline were in the loop. The number of contractions induced by 5-HTP in comparison with saline tended to be decreased with micellar fat (79.8 vs. 104.1 contractions) but not with bile (115.4 vs. 104.1 contractions) in the loop (Table 1, Fig. 3). Under the influence of acetylcholine (50 μg./kg./min.) the number of contractions in the antrum in both muscle layers tended to be decreased by micellar fat in the loop (Table 1, 14.8 vs. 30.8 and 16.6 vs. 31.8). Food is capable of producing a continuous motor pattern at all 4 sites monitored. This motor pattern was not altered by the various substances placed in the duodenal loop (Table 1 and Fig. 4).

In a paired comparison there were significantly more contractions in the circular muscle layer than in the longitudinal muscle layer of the body (57.4 vs. 19.7; $F = 2.85$). In a paired comparison between the total number of contractions in the body vs. the antrum, there were significantly more contractions in the antrum (38.6 vs. 60.9; $F = 11.66$). When combining the 4 sites monitored, the number of contractions under the various stimuli were significantly different from each other (20.0, 37.6, 99.8, and 41.7; $F = 35.64$) with 5-HTP being the most patent stimulant.

The data related to the number of contractions have also been expressed as per cent of activity (Fig. 5), taking 150 contractions as a maximal number of contractions in a 30-min. period. This method of arranging the data indicates that even when stimulants are administered, it is difficult to obtain continuous activity against which to assess inhibitors of gastric motility. The

standard errors of the means have been plotted as an indication of the variability among the 5 dogs when subjected to various experimental procedures. One may note the overlap of the standard errors by observation.

The amplitude of contractions under the various conditions are seen in Table 2. The values obtained are tabulated and expressed as "motility index." The analysis of variance indicates that neither bile nor micellar fat significantly decreased the amplitude of contractions induced by the stimuli. Though not statistically significant, the amplitude of contraction induced by 5-HTP or the lower dose of acetylcholine tended to be decreased by the micellar fat in the loop (Table 2 and Fig. 3). Food generates a continuous motor pattern of relatively low amplitude which could not be markedly reduced further with the substances placed in the loop (Table 2 and Fig. 4). Figure 4 demonstrates why 5 gm. of force was selected as the lowest measurable amplitude.

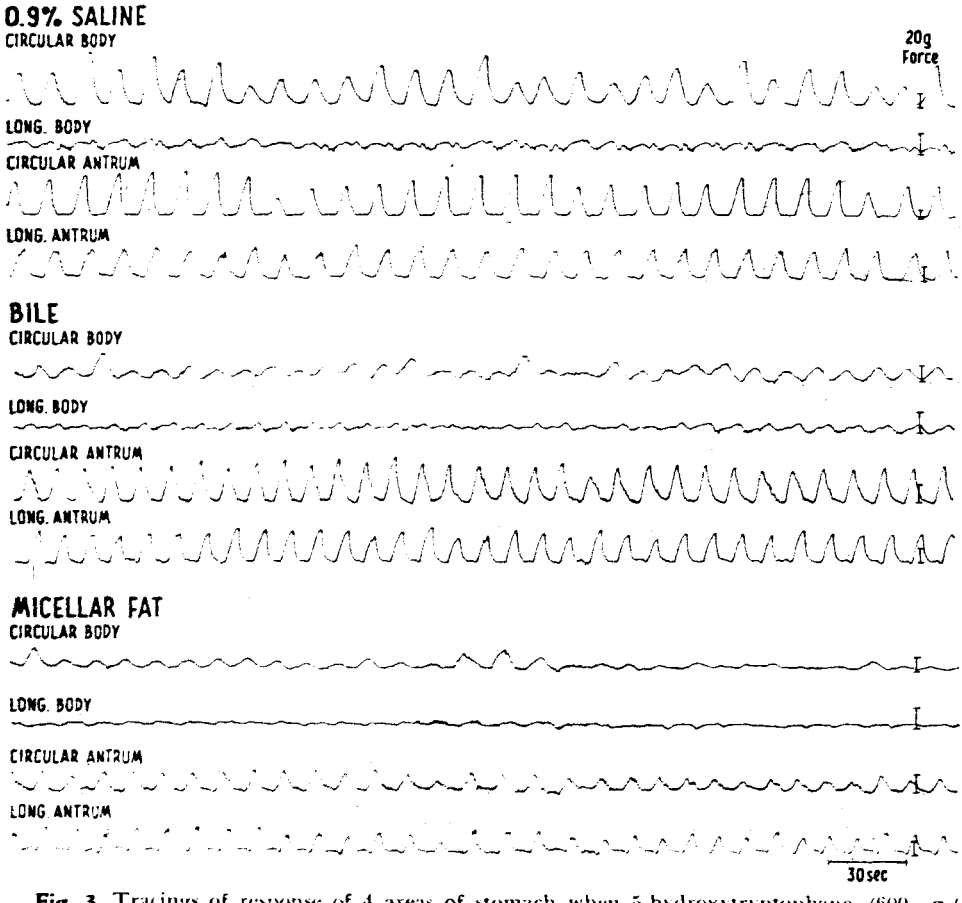


**Fig. 3.** Tracings of response of 4 areas of stomach when 5-hydroxytryptophane (600 μg./kg. min.) infused I.V. after saline, bile, or micellar fat were placed in duodenal Thiry loop. Tracings obtained from same animal on different days. Trend of micellar fat to inhibit this type of activity not significant. Calibration as in Fig. 2.

Contractions lower than 5 gm. could not be clearly differentiated from baseline artifact.

In a paired comparison, the amplitude of the circular muscle layer of the body was significantly greater than that of the longitudinal muscle (98 vs.

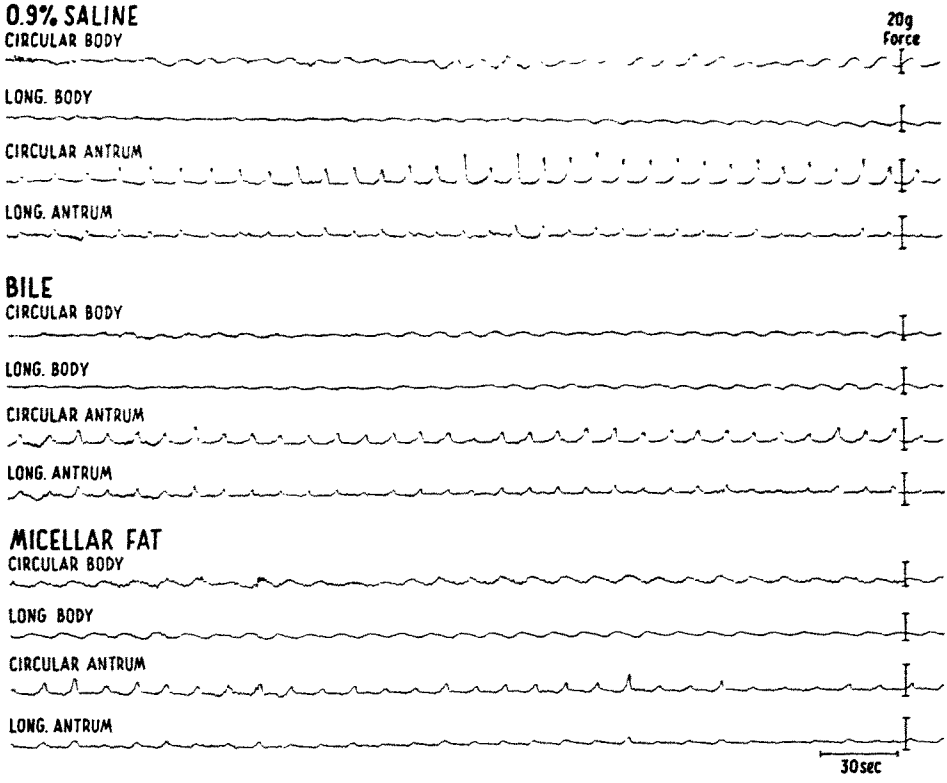


**Fig. 4.** Tracings of response of 4 areas of stomach following ingestion of horse meat (200 gm.) after saline, bile, or micellar fat were placed in duodenal Thiry loop. Tracings obtained from same animal on different days. Calibration as in Fig. 2.

25.2; $F = 24.92$). A similar significant difference was found between the two muscle layers of the antrum (205.6 vs. 185.4; $F = 4.63$). Comparing the total amplitude of contractions of the body vs. the antrum, the latter was significantly greater (61.6 vs. 195.5; $F = 20.30$). Like the frequency results, the amplitude of contractions vary significantly under the influence of the different stimuli. For the over-all effect, we obtained a motility index of 38.7, 108.0, 311.7, and 55.9, yielding $F = 30.97$ with 5-HTP again being the most effective stimulant for contraction.

## DISCUSSION

In a normal physiologic environment one can obtain many hours of recording from multiple leads. To quantify these records it is necessary to use

some form of conversion from an analog signal to a digital form. Once in a digital format the data lend themselves to statistical analysis rather than to a simple description of the recorded motor patterns. Since multiple recordings are obtained from adjacent sites, a paired comparison among the various sites

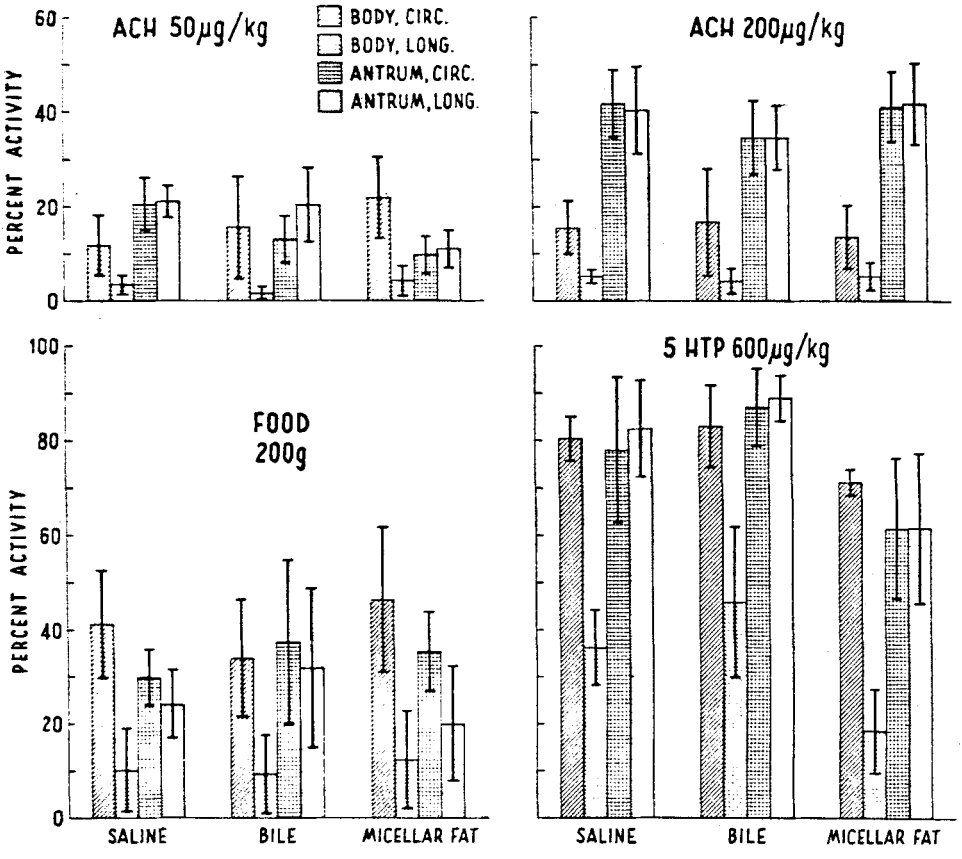


**Fig. 5.** Percentage of activity from 4 areas of stomach in response to I.V. administration of acetylcholine (50 or 200 μg./kg./min.), 5-hydroxytryptophane (600 μg./kg./min.), or ingestion of food (200 gm.) after saline, bile, or micellar fat were instilled into duodenal Thiry loop. Bars represent standard errors of mean.

can be utilized in an analysis of variance. The contractions are relatively easy to enumerate. They may be quantified by expressing activity as the number of contractions in a given time period, or expressed as a percentage of activity of the expected number of contractions. The theoretic maximal frequency is readily available since the rate of gastric contraction for the same preparation is remarkably stable. For example, it was possible to obtain 150 contractions in a 30-min. period.[12]

The amplitude of contraction is an indication of the amount of work per-

TABLE 2. AMPLITUDE OF CONTRACTIONS IN 30 MIN. EXPRESSED AS MOTILITY INDEX

| | | *No. contractions* | | | | | |
|---|---|---|---|---|---|---|---|
| | | *Gastric body* | | *Gastric antrum* | | | |
| *Stimulant* | *Substance in Thiry loop* | *Circ. axis* | *Long axis* | *Circ. axis* | *Long axis* | *Mean* | *Over-all mean* |
| Ach | Micellar fat | 39.8 | 7.2 | 24.2 | 27.0 | 24.6 | |
| (50 μg./kg./min.) | Bile | 27.0 | 2.4 | 44.4 | 57.4 | 32.8 | 38.7 |
| | Saline | 45.6 | 6.6 | 112.8 | 69.6 | 58.6 | |
| Ach | Micellar fat | 30.2 | 9.2 | 224.8 | 163.8 | 107.0 | |
| (200 μg./kg./min.) | Bile | 34.4 | 9.6 | 217.2 | 99.6 | 90.2 | 108.0 |
| | Saline | 45.6 | 8.4 | 285.2 | 168.0 | 126.8 | |
| 5-HTP | Micellar fat | 187.4 | 32.0 | 361.6 | 301.2 | 220.6 | |
| (600 μg./kg./min.) | Bile | 278.0 | 97.6 | 539.2 | 598.0 | 378.2 | 311.7 |
| | Saline | 297.2 | 79.2 | 447.2 | 521.6 | 336.3 | |
| Food | Micellar fat | 73.4 | 19.2 | 66.8 | 63.6 | 55.8 | |
| (200 gm.) | Bile | 52.6 | 14.2 | 91.0 | 102.2 | 65.0 | 55.9 |
| | Saline | 64.6 | 17.2 | 52.6 | 52.6 | 46.8 | |
| MEAN | | 98.0 | 25.2 | 205.6 | 185.4 | | |
| OVER-ALL MEAN | | 61.6 | | 195.5 | | | |

formed by the muscle and can be obtained by measuring the height of the contraction. The assessment of these data can be a tedious task and we have limited it to selected sampling periods. The problems of assessing motility tracings have been expressed by others.[15–17]

To take into account both the amplitude and the frequency of contractions, we utilized the motility index approach of Jacoby and Brodie.[13] The principle of this method is to enumerate contractions of similar amplitudes and assign weighted scores.

Using these methods, we could not demonstrate a statistically significant motor inhibition of the stomach when fat was present in the duodenum. This is not in accord with data obtained by Farrell and Ivy[7] and Quigley, Zettelman, and Ivy[1] in the dog or by Menguy[3] in the rat. Our results are more in agreement with studies on humans performed by Smith and Code.[2] The latter authors observed that the ingestion of a fatty meal leads to a decrease in the amplitude and an increase in the frequency of contraction. The discrepancy between other workers and our results may be due to several factors. The Thiry loop utilized in this study, although consisting of half the duodenum, may not provide sufficient surface to initiate the inhibitory effect of fat. The amount and type of fat utilized in the loop may not be of sufficient quantity or in the proper physical-chemical form. The motor stimulants used may have been too potent to be inhibited by the substances in the duodenum. In our feeding studies, the presence of the various nutrients could have overridden any influence of materials placed in the loop. The variable response among the dogs could have masked the response exerted by the materials in the loop.

Finally, the variance may be due to the different methods of assessing gastric contractility. The utilization of acute procedures, surgical alterations, and intraluminal devices could alter the physiologic environment. In our experience, the normal stomach in an interdigestive state is active for only one-third of the time. The inhibition reported by various investigators could conceivably have been a variable of the above time factor, or the alteration of gastric motility induced by various intraluminal devices could be more readily inhibited by substances in the small bowel than is the motility induced by Ach, 5-HTP, or food.

## SUMMARY

This study was initiated to evaluate the effect on contractility of the longitudinal and circular muscles of the canine gastric body and antrum when micellar fat or bile was instilled in a Thiry loop of duodenum. These studies were performed while the animals were stimulated by food, acetylcholine, and 5-hydroxytryptophane (5-HTP). The latter two chemicals were infused for a 30-min. period. Micellar fat in the duodenum tended to decrease the frequency and amplitude of contractions only in those experiments using 5-HTP or the lower dose of acetylcholine (50 μg./kg./min.) as stimulants. This trend was not seen when bile alone or saline was placed in the duodenal loop. The data were evaluated by a motility index technic and analyzed by a paired comparison method.

P. B.
*Parke, Davis and Company*
*2800 Plymouth Rd.*
*Ann Arbor, Mich.*

## REFERENCES

1. Quigley, J. P., Zettelman, H. J., and Ivy, A. C. Analysis of the factors involved in gastric motor inhibition by fats. *Amer J Physiol 108:*643, 1934.
2. Smith, A. W. M., and Code, C. F. The effect of an ordinary and of an excessively fatty breakfast on human gastric antral motility. *Gastroenterology 35:*398, 1958.
3. Mengly, R. Role of biliary and pancreatic secretions in the inhibition of gastric motility by fat in the intestine. *Amer J Dig Dis 5:*792, 1960.
4. Jacoby, H. I., Bass, P., and Bennett, D. R. In vivo extraluminal contractile force transducer for gastrointestinal muscle. *J Appl Physiol 18:*658, 1963.
5. Bass, P., and Wiley, J. N. Electrical and extraluminal contractile-force activity of the duodenum of the dog. *Amer J Dig Dis 10:*183, 1965.
6. Reinke, D. A., Rosenbaum, A. H., and Bennett, D. R. Patterns of dog gastrointestinal contractile activity monitored in vivo with extraluminal force transducers. *Amer J Dig Dis 12:*113, 1967.
7. Farrell, J. I., and Ivy, A. C. Studies on the motility of the transplanted gastric pouch. *Amer J Physiol 76:*227, 1926.
8. Frazer, A. C. "Some Surface Phenomena in the Study of the Absorption of Fat from the Small Intestine." In *Surface Phenomena in Chemistry and Biology*, Danielli, J. F., Parkhurst, K. G. A., and Riddiford, E. C., Eds. Pergamon, New York, 1958, pp. 299–307.
9. Hofmann, A. F. A physicochemical approach to the intraluminal phase of fat absorption. *Gastroenterology 50:*56, 1966.
10. Konturek, S., and Grossman, M. J. Effect of perfusion of intestinal loops with acid, fat, or dextrose on gastric secretion. *Gastroenterology 49:*481, 1965.

11. SWAN, K. G., KONTUREK, S. J., JACOBSON, E. D., and GROSSMAN, M. I. Inhibition of gastric secretion and motility by fat in the intestine. *Proc Soc Exp Biol Med 121*:840, 1966.
12. ANDERSON, J. J., BOLT, R. J., ULLMAN, B. M., and BASS, P. Differential response to various stimulants in the body and antrum of the canine stomach. *Amer J Dig Dis 13*:147, 1968.
13. JACOBY, H. I., and BRODIE, D. A. Gastrointestinal actions of metoclopramide. *Gastroenterology 51*:676, 1967.
14. LUDWICK, J. R., WILEY, J. N., and BASS, P. Electrical and extraluminal contractile-force activity of reversed canine duodenum. *Gastroenterology* (In press).
15. VANTRAPPEN, G., HELLEMANS, J., and VANDENBROUCKE, J. A method for analysis of intestinal motility records. *Amer J Dig Dis 10*:449, 1965.
16. CONNELL, A. M., TEXTER, E. C., and VANTRAPPEN, G. Classification and interpretation of motility records. *Amer J Dig Dis 10*:481, 1965.
17. ROSENBAUM, A. H., REINKE, D. A., and BENNETT, D. R. In vivo force, frequency, and velocity of dog gastrointestinal contractile activity. *Amer J Dig Dis 12*:142, 1967.

## Postgraduate Course on Gastroenterology

A postgraduate course titled "Frontiers in Gastroenterology" will be held on May 12–15, 1968, at the Bellevue-Stratford Hotel, Philadelphia, Pa. The course is sponsored by the American College of Physicians and The American Gastroenterology Association. Co-chairmen are Henry Tumen and James Roth.

There will be 8 sessions covering the following topics: The Sequelae of the Surgical Treatment of Duodenal Ulcer; Adverse Drug Reactions on the Gastrointestinal Tract; Chronic Pancreatitis; Granulomatous Colitis–Entity or Enigma?; Gastrointestinal Microbiology; Mesenteric Vascular Disease; Chronic Active Hepatitis; and Motor Disorders of the Esophagus.

For further information, please contact The American College of Physicians, 4200 Pine St., Philadelphia, Pa. 19104.