The Presence of Spirochetes In Human Gastric Mucosa

By

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IN 1893 Bizzozero (1) reported the presence of spirochetes in the gastric mucosa of the dog and stressed the occurrence of these organisms in the parietal cells. Later reports agreed with these observations (2, 3). Krienitz (4) in 1906 found spirochetes in the stomach contents of patients with ulcerating gastric carcinoma. This was confirmed by Luger and Neuberger (5) who in addition noted the rarity of these organisms in the gastric mucosa and gastric juice of normal individuals. Recently Doenges (6) reported that spirochetes could be found in the gastric glands of 43 per cent of human stomachs examined in the routine of autopsies. The question arises whether these organisms are natural inhabitants of human gastric mucosa or are just post-mortem or agonal saprophytic invaders.

Therefore we confined ourselves to a histologic investigation of human gastric mucosa made available from gastric tissue resected for duodenal or gastric ulcers or for carcinoma.

The purpose of this paper is to report the results of our studies.

METHODS OF INVESTIGATION

Gastric tissue was obtained from thirty-five patients who were subjected to partial gastric resection; nineteen had carcinoma of the stomach; fourteen had duodenal and two had gastric ulcers. The Wassermann and Kahn reactions of the blood were negative in all patients. The tissue was stained with hematoxylin and eosin, and in addition by the silver impregnation methods of deGalantha (7) and DaFano (8). The latter, while ordinarily used to demonstrate the Golgi apparatus, was extremely satisfactory for outlining the spirochetes in the gastric mucosa of the dog (9). The results with the three different staining techniques were similar except that the silver impregnation methods outlined the spirochetes more clearly.

In order to study the influence of room temperature as well as body temperature on the rate of growth of spirochetes in the mucosa, tissue was obtained from two stomachs, one of which had chronic gastritis associated with duodenal ulcer, and the other had carcinoma at the pylorus. In both cases tissue was removed from the pylorus as well as from three and nine cm. proximal to this. These various segments were prepared for microscopic study by three different techniques: (1) immediate fixation and staining by the hematoxylineosin, and the silver impregnation methods of deGalantha and DaFano; (2) tissue was exposed to room temperature for three, seven, eleven and twenty-one hours respectively before fixation and staining; (3) tissue was incubated at 37 degrees centigrade for the same periods just noted and stained by the same techniques.

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RESULTS

In the gastric mucosa obtained from 35 patients, spirochetes were found in 13 patients, an incidence of 37.1 per cent. This approximates the figures published by Doenges (43 per cent) (6). However the incidence of these organisms in stomachs with benign and malignant ulcerations was greater than in those in which there was no ulceration. Thus organisms were found in only 14.2 per cent in non-ulcerating stomachs as compared with 52.3 per cent where ulceration was present. Since only two cases of benign gastric ulcers were studied, we can attribute no special significance to the high percentage noted (Table I).

TABLE I

Diagnosis	Number of Cases	Cases Positive	% Cases Positive
Carcinoma	19	9	47.3%
Gastric ulcer	2	2	100%*
Duodenal ulcer associated with chronic gastritis	14	2	14.2%

*Average of 21 gastric ulcerations (malignant and benign), 11 positive — 52.3%.

TABLE II

Diagnosis	Number of Cases	Number of Slides	Positive Results Number of Slides	Percentage Positive Slides
Gastric carcinoma	18	90	16	17.7
Gastric ulcer	2	16	3	18.7
Duodenal ulcer associated with chronic gastritis	13	54	2	3.7

Although 52.3 per cent of the gastric ulcerations revealed the presence of spirochetes, it should be emphasized that the organisms were found with difficulty and frequently required long and careful search through many slides. Thus from tissue obtained from 18 out of 19 cases of gastric carcinoma, 90 slides were prepared. Spirochetes were found in only 16 slides or 17.7 per cent (Table II). The greatest number of spirochetes were invariably present either in areas of

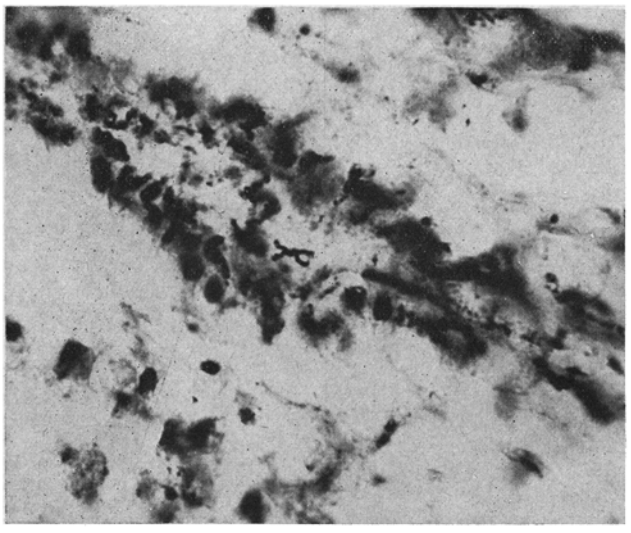

Fig. 1. Silver impregnation, DaFano technique. 970x. Spirochetes in the lumen of a gastric gland. The tissue was obtained from a case of gastric carcinoma.

extensive necrosis, or in the lumina of glands of contiguous mucosa but never within the cellular portion of the tumor (Fig. 1).

Gastric mucosa from 13 cases of chronic gastritis associated with duodenal ulcer was prepared for microscopic study and fifty-four slides were satisfactory. Spirochetes were found in only two slides or 3.7 per cent (Table II). In both instances the organisms were found only in the lumina of the glands in the most superficial layers of the mucosa, in contrast to the observations made in the dog in which the spirochetes were found throughout the thickness of the mucosa as well as within the canaliculi of the parietal cells (Fig. 2).

Eighty-one slides were prepared from tissue which was left at room temperature and incubated at 37 degrees centigrade respectively for three, seven, eleven and twenty-one hours. One case had carcinoma of the pylorus and the other had duodenal ulcer associated with mild chronic gastritis. Twenty-four slides were made directly from the carcinoma. In this group spirochetes were noted in eleven slides, 45.8 per cent. Twelve slides prepared from three and nine cm. proximal to the carcinoma were entirely negative for spirochetes. In the case of duodenal ulcer also used in this study, 45 slides were prepared out of which only four were positive, 8.9 per cent. It was noted here that spirochetes were only found after the tissue had been incubated or kept at room temperature for seven, eleven, or twenty-one hours (Fig. 3).

Sixteen slides were prepared from two cases of benign gastric ulcer. Three slides were positive, 18.7 per cent. In each instance the spirochetes were found in the tissue at the periphery of the gastric lesion.

COMMENT

The presence of spirochetes in the gastric mucosa of certain laboratory animals is now established. In 1919 Kasai and Kobayashi (3) noted these organisms in the dog, cat and monkey. Edkins (10) found a high frequency of spirochetes in the gastric mucosa of the cat, but noted a marked decrease in the number of organisms in the fasting state. This observation may help to explain the necessity for prolonged search through numerous slides in the detection of organisms in surgical specimens. The prolonged preparation preceding gastric surgery such as abstinence from food, strict attention to oral hygiene, and frequent gastric lavages may not only interfere with the growth of the organisms but may actually eliminate them from the gastric contents.

From our results it does not seem likely that spirochetes are natural inhabitants of normal human gastric mucosa. Although 37.1 per cent of the total number of patients revealed the presence of organisms, they were not found with the same abundance as noted in the gastric mucosa of the dog. In our material it very often was necessary to search through many slides prepared from the same specimen in order to find one slide which revealed organisms, and very often this slide contained only a few spirochetes. This is substantiated by the data in Table II which indicated that the number of slides containing spirochetes in relation to the total number of slides examined is extremely low. Furthermore these organisms in our experience were only seen within the lumina of the glands in the most superficial portion of the mucosa. None were noted in the canaliculi of the parietal cells.

Our results would seem to indicate that spirochetes are found with increasing frequency in the presence of necrotizing malignant or benign ulceration. The converse is true; in the absence of ulceration, spirochetes are rarely found. There was no evidence in our specimens indicating that these organisms have any pathogenic significance.

In the tissue subjected to incubation and exposure to room temperature for varying periods of time, very little influence was noted on the growth and number of organisms. In the stomach with carcinoma, spirochetes were found only in those sections taken directly from the tumor. In sections taken three and nine cm. away, that is, microscopically normal mucosa, spirochetes could not be demonstrated.

In the other case, in a search through 45 slides, only four revealed the presence of organisms, approximating the results obtained in unincubated similar tissue (Table II).

In a survey of the illustrations noted in the various reports, the spirochetes in the mucosa of the experi-

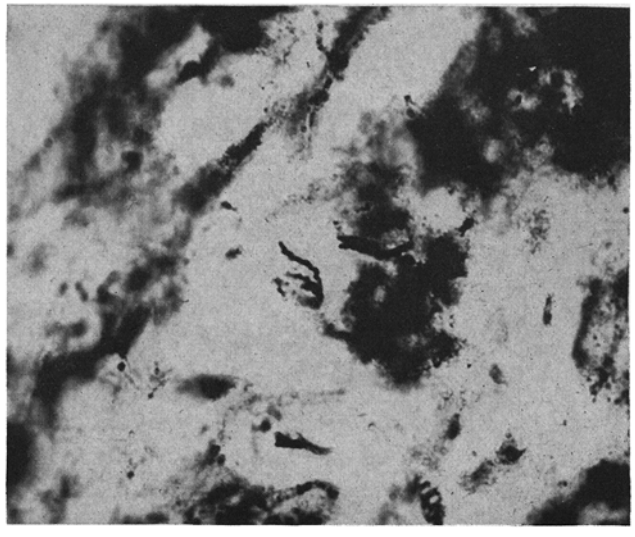

Fig. 2. Untoned silver impregnation, DaFano technique. 970x. Spirochetes in the gastric mucosa of a dog.

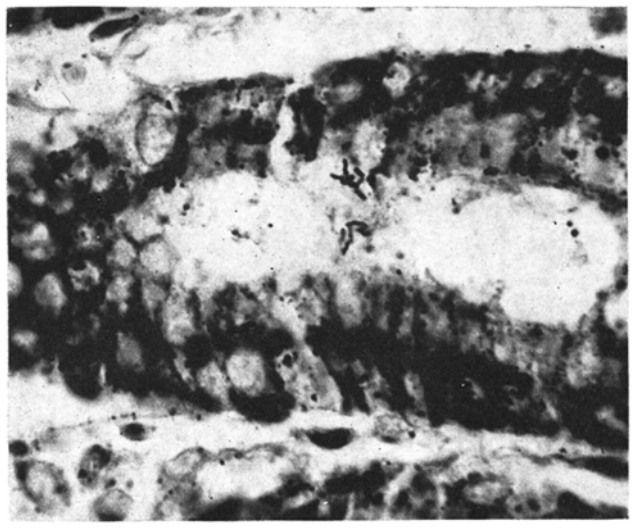

Fig. 3. Silver impregnation, DaFano technique. 970x. The tissue was obtained from a case of chronic gastritis and was incubated at 37 degrees centigrade for 11 hours.

mental animals leave no question as to their identity. This was not always the case with the illustrations purporting to demonstrate spirochetes in human gastric mucosa. This was successfully obviated by the silver impregnation methods referred to previously.

CONCLUSIONS

1. Spirochetes are rarely found in the mucosa of gastric tissue resected for duodenal ulcer without attendant gastric ulcerations.

2. They are frequently found in those stomachs whose mucosa is diseased by malignant or benign ulcerations. In these instances they are found in or close to the lesion.

3. The silver impregnation method of DaFano will demonstrate spirochetes in the gastric tissue of the dog and human.

4. Incubation and exposure to room temperature for varying periods seemed to exert very little influence on the number of spirochetes in human gastric mucosa.

We wish to express our appreciation to Dr. Monroe J. Schlesinger, in whose department this work was carried out. Mrs. Edith Herman rendered valuable technical assistance.

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DISCUSSION

DR. FRANK D. GORHAM (St. Louis, Mo.): Dr. Barron's and Dr. Freedberg's report of their results in a search for spirochetes in the gastric mucosa of pathologic human stomachs is interesting.

More and more there is a tendency to separate the possible causative factors of acute cyclic peptic ulcer from the factors of chronicity of some peptic ulcers that refuse to heal under usual ulcer management. In this latter group I have used over a period of some ten years bismuth (Bismoid), administered intramuscularly, with encouraging results.

Some four years ago, when searching for a pharmacological explanation of the therapeutic action of bismuth in chronic ulcer, one of the possibilities which suggested itself was that in chronic ulcer one of the factors of chronicity might be an associated infection by an organism thriving best in an acid (hydrochloric acid) medium. At that time my attention was directed to some investigations of Dr. Cowdry, of the Washington University School of Medicine.

Cowdry and his coworkers, Scott and Doenges, were finding in the stomachs of Macacus rhesus monkeys, spirochetes in the cytoplasm of the parietal cells and in the lumen of the gastric glands, and in some instances associated with localized inflammatory changes.

I obtained for Dr. Cowdry some human stomachs which he studied. Later Doenges examined some 242 well preserved stomachs removed at autopsy, finding spirochetes in some 43 per cent. These spirochetes were not found in the glands of the intestinal mucosa, suggesting that their habitat was specifically the stomach. Also this organism was not Vincent's, neither was it Spirochaeta pallida, but an unidentified spirochete.

From their investigation, Dr. Cowdry and his coworkers could not definitely establish any etiological relationship between the presence of the spirochetes in the mucosa of the stomach and serious gastric pathology.

All investigations to date tend to corroborate Dr. Barron's and Dr. Freedberg's conclusions that the role of spirochetes in gastric disease has not been established; however, I believe that a further search should be made for an organism thriving in hydrochloric acid medium (and variations of hydrochloric acid are normal in all stomachs) as a possible factor of chronicity, if not an etiological factor, in peptic ulcer.

DR. LOUIS E. BARRON (Boston, Mass.) (closing the discussion): The results of our studies on resected stomachs were similar in many ways to those reported by Dr. Doenges for stomachs removed at autopsy. There are, however, certain minor differences.

In our experience spirochetes were found with great difficulty. Frequently it was necessary to prepare a large number of sections before one or two revealed organisms.

Furthermore, in order to prevent erroneous interpretation of artifacts, our slides were examined by other members of the laboratory and a unanimous opinion was reached before a slide was considered positive for spirochetes.

We cannot substantiate the view that spirochetes, stained by the routine hematoxylin-eosin stains, can be detected with ease. In our experience it was necessary to use the silver impregnation techniques to outline the organisms.