these animals. The wave-like cycle of these chemical and bacterial changes reported here are characteristic of autonomic nervous system disturbances.

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Influence of Gastric Acidity upon Viability of Bacteria in Isolated Stomach.

A. J. NEDZEL AND LLOYD ARNOLD.

From the Department of Bacteriology and Preventive Medicine, University of Illinois, College of Medicine, and Research Laboratories of the Illinois Department of Public Health, Chicago.

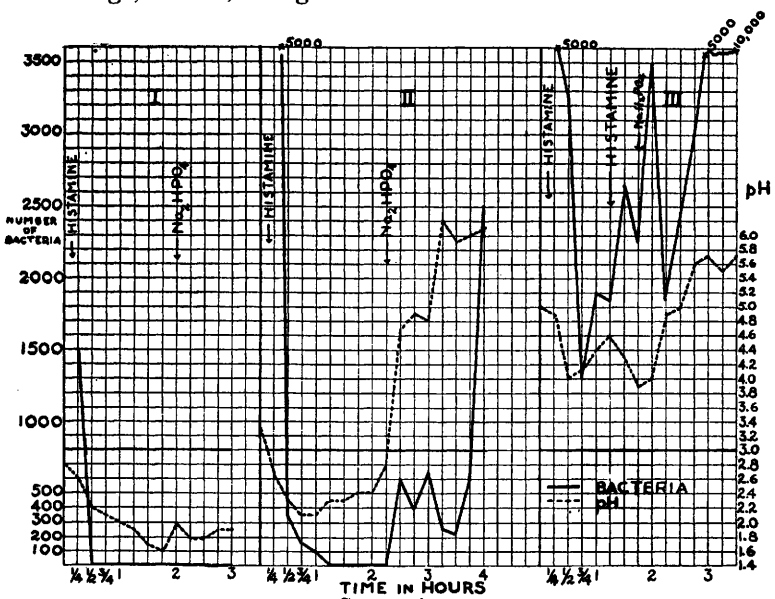
Johnson and Arnold[1] in a series of experiments on dogs with a non-leaking gastric fistula have found that exogenous bacteria (*B. prodigiosus*) lose their viability in the presence of free hydrochloric acid and when the gastric contents were alkalinized with Na_2HPO_4 buffer solution these previously non-viable exogenous bacteria regained their viability. This phenomenon was so constant that we decided to set up acute experiments in which we could entirely separate the stomach from the duodenum. By this technic we could completely exclude the possibility of any regurgitation from the intestine.

The experiments on dogs were conducted in the following manner. The abdomen of an animal, under nembutal anesthesia (1 cc. of 3% solution per kilo intraperitoneally), was opened and the pyloric part of the stomach with the duodenum brought up to the surface. A series of ligatures was put around the pyloric end of the duodenum through the wall of the duodenum to avoid a hemorrhage into the stomach and then the duodenum was severed about 1 cm. below the ligatures. In such a manner we produced a wide and non-bleeding opening into the stomach which facilitated the securing of specimens from the gastric contents. All the specimens were taken by sterile Wright pipettes.

First, we determined the pH (LaMotte hydrogen-ion testing set) of the gastric contents, then introduced into the stomach a suspension of *B. prodigiosus* (washings of a 24-hour growth on agar plate in 20 cc. sodium chloride solution) and simultaneously injected 1.5 mg. of a histamine intravenously. At certain time intervals (see

[1] Johnson and Arnold, PROC. SOC. EXP. BIOL. AND MED., 1931, **29**.

chart) a specimen from the stomach was taken and tested for pH and for the presence of viable *B. prodigiosus* (one drop of the stomach contents spread on an agar plate, readings in 24 hours). At the same time slides were made from the stomach specimens and stained by the Gram method. The slides were always positive for the presence of Gram negative bacillus. Ten dogs were used, averaging 35 pounds in weight. The stomach of each dog reacted differently depending on the functional status of the stomach. The results can be divided into 3 groups, I and III representing the average of 3 dogs, and II, 4 dogs.



GRAPH 1.

Ordinate: Left, number of viable *B. prodigiosus* per cc. of gastric contents. Right, pH of gastric contents.

Abscissa: time in hours or fraction of hours.

Solid line represents the curve of viable bacteria in gastric contents. Broken line represents the pH of the gastric contents.

Group I (Chart I). The stomach contained free hydrochloric acid. The injection of histamine increased the concentration of acid. Na_2HPO_4 buffer solution (20 cc.) was introduced into the gastric lumen; no change in the h-ion concentration could be demonstrated. Numerous Gram negative bacilli were present in smears; were non-viable on nutrient agar.

In the second group (Chart II) where there was no free hydrochloric acid, the histamine stimulated acid secretion and the *B. prodigiosus* became non-viable. But the addition of Na_2HPO_4 buffer solution brought the pH up to 4.7 and higher. Simultaneously with

this change toward acid-deficit the *B. prodigiosus* again became viable, as is clearly shown on the chart (II).

In the third group (Chart III) the gastric mucosa was less active. In this case the injection of the same dose of histamine did not cause the stomach to produce the free acid. The addition of Na_2HPO_4 buffer solution was followed by an increase in the number of viable bacteria. The introduced exogenous bacteria (*B. prodigiosus* in our case) remained viable throughout the experiment.

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Has the Free Gastric Acidity Bactericidal or Bacteriostatic Power?

TITUS M. JOHNSON AND LLOYD ARNOLD.

From the Department of Bacteriology and Preventive Medicine, University of Illinois, College of Medicine, and Research Laboratories of the Illinois Department of Public Health, Chicago.

Viable bacteria were introduced into the stomach of a series of non-leaking gastric fistula dogs and cultures made at certain time intervals along with titrations of the free acid and acid-deficit. One agar plate of *B. prodigiosus* growth was washed off in 100 cc. of sterile saline. All animals were fasted 20 to 24 hours before experiments. When 100 cc. of a heavy *B. prodigiosus* suspension was placed in a gastric lumen containing free acid secretion, the subsequent specimens removed and cultured upon nutrient agar were sterile. If there was an absence of free acid in the stomach and the same procedure was followed, the nutrient agar plates were so overgrown that dilutions had to be made to estimate the bacterial content. Some experiments were carried out over 3 and 4 hours of time. It was observed that so long as free H-ions were present no viable bacteria could be demonstrated. But if an acid-deficit developed during the course of the experiment, we observed a sudden appearance within the lumen of the stomach of many viable *B. prodigiosus* and they persisted so long as free acid was absent. Test bacteria were only introduced at the beginning of the experiment, their presence or absence within the lumen of the stomach determined by subculturing on nutrient agar, could be correlated with the acid-base balance existing at the time within the cavity of this organ.

Over 300 experiments have been carried out, *B. prodigiosus, B.*