

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  $\text{Na}_2\text{HPO}_4$  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?

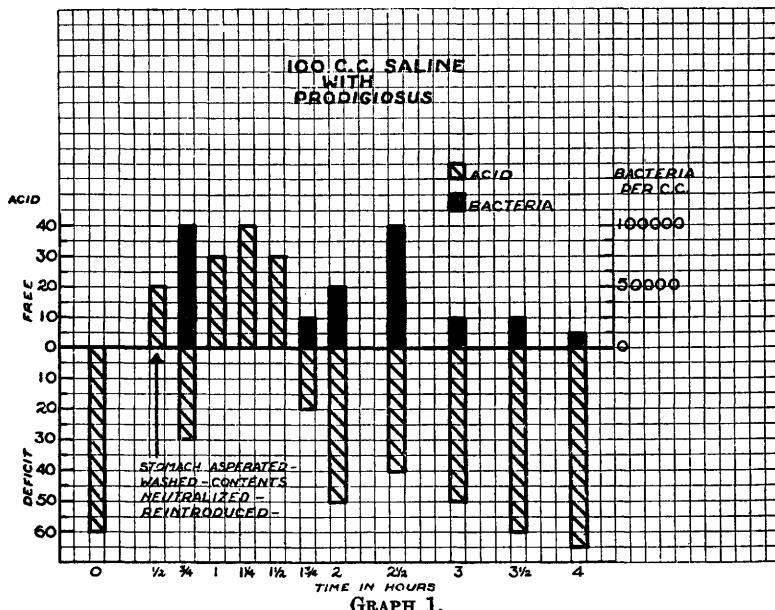
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Viable bacteria were introduced into the stomach of a series of non-leaking gastric fistula dogs and cultures made at certain time intervals along with titrations of the free acid and acid-deficit. One agar plate of *B. prodigiosus* growth was washed 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.*

*coli*, and *B. murii* have been used as test bacteria. The results are the same with each bacteria. The test bacteria have been suspended in saline, milk, phosphate buffered solutions, and meat. Graph 1 illustrates an example where there was an acid-deficit at the beginning of the experiment and it persisted for one hour. Then free acid appeared. One hundred fifty thousand bacteria per cc. of gastric contents were present in a viable condition during the period of acid-deficit. As soon as free acid was detected no viable bacteria could be demonstrated. Sterile alkaline phosphate solution was introduced to neutralize the free acid. The original test bacteria reappeared and persisted during the period of acid-deficit. Graph 2 shows an experiment where saline and *B. prodigiosus* were introduced into an acid-deficit gastric lumen. Free acid appeared within a half hour; no viable bacteria could be demonstrated. The gastric contents were then removed through the fistula, cultured, neutralized with sodium hydroxide, again cultured, then replaced within the lumen of a second dog that had not been used for 2 weeks and whose stomach contained free acid. The neutralized gastric contents did

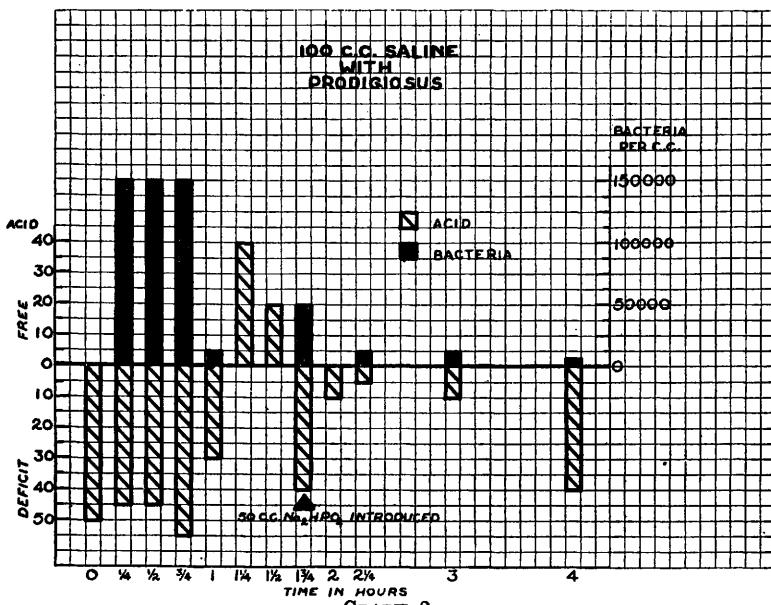


GRAPH 1.

Ordinate: Left, free and acid-deficit in clinical units in gastric contents. Right, Number of bacteria per cc. of gastric contents.

Abscissa: time in hours or fraction of hour from the beginning of the experiment.

One agar plate of *B. prodigiosus* suspended in 100 cc. saline was introduced into the stomach through fistula at the beginning of the experiment. Fifty cc. of sterile alkaline phosphate buffered solution introduced after one hour and 45 minutes at the time of free gastric acidity.



Ordinate: Left, gastric acidity expressed in clinical units. Right, bacteria per cc. of gastric contents.

Abscissa: time in hours.

One agar plate of *B. prodigiosus* suspended in 100 cc. saline and introduced through fistula into the stomach. After free acid appeared ( $\frac{1}{2}$  hour) the stomach was emptied. Cultures proved the absence of viable *B. prodigiosus*. The gastric contents were neutralized and introduced into the stomach of a second dog. Cultures again showed no viable *B. prodigiosus* present. Specimen removed from stomach showed heavy growths of *B. prodigiosus* during period of acid deficit.

not contain viable bacteria. One hundred thousand viable bacteria could be demonstrated per cc. of the gastric contents in the second dog during the initial period of acid-deficit. When free acid appeared, the contents were again sterile when put on agar. When an acid-deficit developed (after  $1\frac{3}{4}$  hours) the original test bacteria reappeared in viable form. In all of our experiments many stainable bacteria were present in the gastric contents which resembled the test bacteria. We could not demonstrate any difference in relative numbers of stainable bacteria in the gastric contents of free acid and acid-deficit animals. Berkefeld filtration of the gastric contents apparently removes the bacteria; we were unable to demonstrate them in experiments similar to that in Graph 2, where the gastric contents were filtered before introducing into the second dog's stomach.