

TABLE I. The Effect of Hexamethonium (C6) on Histamine and Urecholine Stimulated Secretion in Dogs with Vagotomized Total Pouches and in Dogs with Gastric Fistulas (Vagally Innervated). Four dogs in each experiment; 2 tests on each dog.

Type of dog	Stimulant	Control secretory rate free HCl, mM/hr	% inhibition— Hr after C6		
			1	2	3
Gastric fistula	H*	5.70 ± 1.1	47 ± 11.5	63 ± 6.3	68 ± 10.3
	U	2.70 ± .9	70 ± 7.6	97 ± 1.4	96 ± 1.8
Total pouch	H	2.50 ± .6	41 ± 1.3	31 ± 11	37 ± 5.5
	U	1.81 ± .6	44 ± 13.9	73 ± 19.9	68 ± 12.8

* H = Histamine, U = Urecholine.

by the preganglionically denervated ganglion cells. Since cholinergic stimuli potentiate the response of the parietal cell to other direct acting stimuli(7,8), a decrease in response would result. The ability of hexamethonium to suppress the spontaneous activity of preganglionically denervated ganglion cells is attested by the fact that it inhibits certain of the spontaneous movements of the isolated ileum(2).

Hexamethonium resembles atropine(9) in that both have a greater inhibitory activity in the vagally innervated than in the vagally denervated stomach. Atropine acts by blocking the effect of acetylcholine, hexamethonium, according to the present hypothesis, by depressing acetylcholine synthesis. If it is assumed that the basal rate of acetylcholine synthesis is greater in the vagally innervated than in the vagally denervated stomach then this difference in effectiveness of hexamethonium and atropine in these two types of stomach preparations would be explicable.

Summary. In dogs with gastric fistulas

(vagally innervated) and in dogs with pouches of the entire stomach (vagally denervated), the secretion of acid in response to Urecholine or histamine is depressed for at least 3 hours following intravenous administration of 4 mg/kg of hexamethonium bis-trimethylammonium chloride. The inhibition is greater in the vagally innervated than in the vagally denervated stomach preparations.

1. Paton, W. D. M., and Zaimis, E. J., *Brit. J. Pharm.*, 1949, v4, 381.
2. Feldberg, W., *J. Physiol.*, 1951, v113, 483.
3. Kay, A. W., and Smith, A. N., *Gastroenterology*, 1951, v18, 503.
4. Grossman, M. I., *Methods in Medical Research*, 1951, v4, 173.
5. Antia, F., Rosiere, C. E., Robertson, C. R., and Grossman, M. I., *Am. J. Physiol.*, 1951, v166, 470.
6. Burn, J. H., *Physiol. Rev.*, 1950, v30, 177.
7. Robertson, C. R., and Grossman, M. I., *Fed. Proc.*, 1948, v7, 103.
8. Grossman, M. I., *Physiol. Rev.*, 1950, v30, 33.
9. Code, C. F., Hightower, N. C., and Hallenbeck, G. A., *Gastroenterology*, 1951, v19, 254.

Received January 14, 1952. P.S.E.B.M., 1952, v79.

Effect of Antibacterial Substances on Fecal Clostridia Populations.* (19332)

NORA L. LARSON AND LAWRENCE E. CARPENTER. (Introduced by T. H. Jukes.)

From the Hormel Institute of the University of Minnesota, Austin, Minn.

The reports of marked increase in the rate of gain of chicks, turkey poult and weaned pigs fed diets supplemented with antibiotics

have aroused interest in the possible role of intestinal microflora on growth. In an effort to obtain information on the action of antibiotics, McGinnis and coworkers(1) have observed that the growth of *C. perfringens* is inhibited in the ceca of turkey poult fed

* Supported by grants from the Hormel Foundation and Lederle Laboratories Division of the American Cyanamid Company, Pearl River, N. Y.

diets containing 100 p.p.m. of penicillin and terramycin. Fecal samples from pigs fed diets containing 15 mg of terramycin per kg contained less than 10 *C. perfringens* per g as compared with 34 to 70 million for samples from control pigs.

The studies reported herein were conducted with weaned pigs to observe the effect of dietary antibiotics and 3-nitro-4-hydroxy phenyl arsonic acid on the population of clostridia in feces.

Procedure. Two separate experiments were conducted on the effect of dietary antibiotics and 3-nitro-4-hydroxy phenyl arsonic acid on the population of *C. perfringens* in the feces of weaned pigs. The basal diet employed for both experiments had the following composition: ground yellow corn 41%, ground oats 20%, tankage (60%) 10%, soybean oil meal 7%, alfalfa leaf meal 10%, wheat middlings 10%, and mineral 2%. In the *first experiment*, 28 weaned Chester White pigs from the Hormel Foundation herd weighing between 34 and 58 lbs were divided into 7 groups of 4 pigs each and were fed the following dietary supplements:

Group	Supplement to 100 lb of basal	
1	0	g
2	1.14	3 nitro-4-hydroxy phenyl arsonic acid
3	4.54	3 nitro-4-hydroxy phenyl arsonic acid
4	.25	procaine penicillin
5	2	chloramphenicol
6	.25	aureomycin hydrochloride
7	2	g

The pigs were housed in a constant temperature farrowing barn in pens 6 x 15 ft with concrete floors and glazed tile walls. The floors were washed with water once daily, and excreta were removed at least 6 times daily. Fresh water and feed were supplied at all times. The pigs were weighed weekly for 4 weeks. All pigs were fed the basal diet during the first week and the supplemented diet during the last 3 weeks. Two fecal samples were collected from each pen 4 times a week, after the floors had been washed down with water. The fresh samples were weighed, diluted with water, and used immediately for bacterial studies.

Clostridia populations were determined by the method of Wilson and Blair(2) as modified by Thompson(3). Verification of colonies for clostridia was carried out by subculturing in iron-milk. Stormy fermentation was produced by about 80% of the colonies. In the *second experiment*, carried out over a 5-week period, 20 weaned Chester White pigs weighing between 28 and 48 lbs were divided into 5 groups of 4 pigs each. The management conditions were the same as in the first experiment. All pigs were fed the basal diet for one week during which time fecal samples were collected. Starting the second week, the pigs were fed the following dietary supplements for a period of 4 weeks:

Group	Supplement to 100 lb of basal	
1	0	g
2	4.54	3-nitro-4-hydroxy phenyl arsonic acid
3	2	procaine penicillin
4	2	chloramphenicol
5	2	aureomycin hydrochloride

Fecal samples were collected from individual pigs 5 days each week by inducing defecation by exercise or by inserting a suppository into the rectum. The bacteriological methods were the same as in the first experiment.

Results and discussion. The average daily gain and the clostridia populations of fecal samples from the first experiment are given in Table I. The pigs fed the diet containing a high level of arsonic acid (4.54 g per 100 lb of feed) gained an average of 0.93 lb per day compared to 0.78 lb for the control pigs, even though the fecal clostridia populations were essentially the same for the two groups. The clostridia populations of the feces of the group fed the low level of arsonic acid (group 7) were also in the same range and the growth rate was the same as that of the control pigs. The animals fed diets containing penicillin (groups 4 and 5) and 2.0 g aureomycin per 100 lb of feed (group 2) had the lowest populations of fecal clostridia during the last 2 weeks, but they did not gain any faster than the pigs fed the high level of arsonic acid.

The results of the second experiment are given in Table II. All the supplemented groups made better weight gains than the control group.

TABLE I. Effect of Dietary Antibiotics and 3-Nitro-4-Hydroxy Phenyl Arsonic Acid on Population of *C. perfringens* in Feces of Pigs.

Group No.	Supplement per 100 lb basal diet (g)	Avg daily gain,* lb	Avg No. of <i>C. perfringens</i> per g fresh feces ($\times 10^3$)			
			1st wk	2nd wk	3rd wk	4th wk
1	0	.78	487	90	19	17
2	2 aureomycin hydrochloride	.87	53	30	1.9	4.3
3	.25	.81	373	13.5	7.6	13.6
4	2 procaine penicillin	.90	20	1.4	.4	.1
5	.25	.89	3046	18	.3	2.3
6	4.54 3-nitro-4-hydroxy phenyl arsonic acid	.93	500	31	69	12
7	1.14	.77	40	17.5	22	17

* During the last 3-week period when supplements were fed.

TABLE II. Effect of Dietary Antibiotics and 3-Nitro-4-Hydroxy Phenyl Arsonic Acid on Population of *C. perfringens* in Feces of Pigs.

Group No.	Supplement per 100 lb basal diet (g)	Avg daily gain,* lb	Avg No. of <i>C. perfringens</i> /g fresh feces ($\times 10^3$)				
			1st wk	2nd wk	3rd wk	4th wk	5th wk
1	0	1.05	2960	614	29.4	61	126.4
2	2 aureomycin hydrochloride	1.19	3975	798	8.9	15	3.5
3	2 procaine penicillin	1.38	5900	18900	325.9	31.9	3.8
4	2 chloramphenicol	1.23	134	6051	15	25.8	21.5
5	4.54 3-nitro-4-hydroxy phenyl arsonic acid	1.31	3280	639	22.1	20	30

* During last four wk of experiment when supplements were fed.

In general the population of clostridia in the feces decreased for all groups during the test period and there was no apparent correlation between the rate of gain and population of the fecal samples.

There was considerable variation in the population of the fecal samples collected from the same lot of pigs on different days. Also, there was day-to-day variation in the samples from the same animal. The average daily gains for the pigs in each group varied considerably, in some cases. Because the number of pigs per group was small, the differences in rates of gain of pigs in the supplemented lots over those of control pigs were not significant except for group 6 of Table I. In most other cases, the differences approached significance at the 5% level.

Summary. The results in 2 separate experi-

ments suggest that populations of fecal clostridia can be altered by feeding antibiotics, but growth of the host animal is not directly related to the population of fecal clostridia.

We are indebted to Dr. Salsbury's Laboratories of Charles City, Ia., for the supply of 3-nitro-4-hydroxy phenyl arsonic acid, to Lederle Laboratories Division of American Cyanamid Co., for the supply of aureomycin hydrochloride, and to Parke, Davis & Company of Detroit, for the supply of chloramphenicol.

1. Sieburth, John McN., Gutierrez, Jose, McGinnis, James, Stern, Joel R., and Schneider, B. H., *Proc. Soc. Exp. Biol. and Med.*, 1951, v76, 15.

2. Wilson, W. J., and Blair, E. Maud M'V., *J. Path. Bact.*, 1924, v27, 119.

3. Thompson, Luther, *Am. J. Clin. Path.*, 1939, v9, 173.

Received January 14, 1952. P.S.E.B.M., 1952, v79.