

contracted the disease in Gurabo, moved into the camp while still in the acute stage of the disease. The latrines were in very bad condition and drained into a pool, the water of which was used for drinking purposes. There was an abnormal number of flies, and there were abundant opportunities for direct contact. In Bayamón, contrary to the distribution in the first epidemic area, practically all of the cases occurred in the urban zone and the mortality rate was very low. The epidemic also made its appearance in Manatí and Camuy. In these last two towns, no detailed epidemiological study was carried out.

In San Lorenzo and Gurabo the disease acquired such epidemic proportions that it became necessary to establish emergency hospitals.

The epidemiologic survey carried out in the affected area, consisted in the localization of cases by field inspectors and the taking of a careful history of every case. The symptomatology and the relation which the disease might have had with food products was carefully recorded.

A total of 4,639 cases was studied, distributed as follows: urban, 32.74 per cent; rural, 66.8 per cent; males 49.17 per cent, females 50.59 per cent. Table No. 2 gives details of the cases studied and those reported by physicians.

TABLE No. 2  
DISTRIBUTION OF CASES OF DYSENTERY STUDIED IN THE 1932 SURVEY,  
AND CASES REPORTED BY PHYSICIANS DURING THE YEAR 1932-33

Town	Cases Studied in the Survey				Cases reported during fiscal year 1932-33	
	Number	Percentage Distribution		Percentage of cases which took to bed	Number	Percentage of total
		Urban	Rural			
San Lorenzo.....	818	40.1	59.9	38.5	1,274	15.2
Gurabo.....	708	22.6	77.4	39.1	830	9.9
Juncos.....	582	41.37	58.6	24.7	706	8.2
Caguas.....	694		100	51.9	1,058	12.6
Humacao.....	920	29.6	70.4	49	996	11.9
Bayamón.....	719	70.4	29.6	45.1	961	11.5
Other towns.....	198	8.6	91.4	33.3	2,519	30.2
Total.....	4,639	32.74	66.8	42.12	8,344	.....

If the clinical course of the disease were benign, the malady would, on an average, last from four to six days. Two thousand, six hundred and seventy-five of the cases



studied (57.8%) were ambulatory, the other 1,954 (42.12%) were severe enough to take to their beds. The symptomatology was characteristic throughout the area involved. The disease was initiated by abdominal pain accompanied in a great number of the cases by malaise and headache. There was tenderness on pressure over the whole abdomen. The bowel movements were liquid or semi-liquid. Diarrhea with blood and mucus, tenesmus and burning on defecation, were rather constant manifestations. The symptomatology varied with the severity of the individual case, the number of bowel movements being as a rule ten to twelve per day, although cases were recorded with one hundred movements in twenty-four hours. Vomiting was not a constant manifestation but fever was present in the majority of the cases. Many of the affected individuals gave a history of having had the same disease after the hurricane of 1928 and some during the period intervening between the two storms. Unfortunately no accurate history as to recurrences of the disease was taken during the survey. Table No. 3 shows a relation of the percentage frequency of the different symptoms as compared with other similar epidemics of dysentery studied in Puerto Rico.

TABLE No. 3

FREQUENCY (IN PERCENTAGE) OF IMPORTANT SYMPTOMS DURING DYSENTERY EPIDEMICS OF 1928 AND 1932 AND THE INSTITUTIONAL OUTBREAK OF 1930

Symptoms	Epidemic of 1928		Penitentiary Outbreak in 1930 Costa and Garrido	Epidemic of 1932
	Costa and Garrido	Lavandero		
Fever.....	77.8	21.7	73.6	66.28
Malaise.....				89.15
Headache.....	66.34		73.6	73.37
Vomiting.....	36.11		39.4	29.92
Diarrhea.....	83.97	98.8	76.3	88.05
Mucus (stool).....	87.39	89.8	97.3	93.81
Blood (stool).....	73.93	76	95.2	68.63
Abdominal pain.....	92.52	62	97.3	93.31
Tenesmus.....	73.5	62.6	97.3	86.67
Burning on defecation.....				90.62
Cases Studied.....	936	253	46	4,639

The distribution of the cases by age groups conforms with the distribution in the general population, indicating that the

disease spread evenly throughout the whole population affecting all groups and classes of the population alike (See table No. 4).

TABLE No. 4

DISTRIBUTION OF CASES BY AGE GROUPS IN THE DYSENTERY EPIDEMICS OF 1928 AND 1932 AND PERCENTAGE DISTRIBUTION BY AGE GROUPS OF THE POPULATION IN PUERTO RICO DURING THE SAME YEARS

Age Groups	Percentage Distribution by age groups in the population of Puerto Rico estimated on Jan. 1		Percent of cases studied		
	1933	1929	Epidemic of 1928		Epidemic of 1932
			Costa und Garrido	Lavandero	
Under 5 years.....	14.5	14.7	19.5	26.5	21.25
Under 1 year.....	2.75	2.9	4.38	7.1	2.67
1 to 4 years.....	11.75	11.84	15.14	19.2	18.58
5 to 9 years.....	14.4	14.56	12.1	10.3	7.64
10 to 14 years.....	12.9	12.91	8.5	7.5	8.94
15 to 19 years.....	12.56	11.81	8.4	7.8	8.59
20 to 29 years.....	15.88	16.24	14.7	15.8	18.67
30 years and over.....	29.73	29.71	36.4	32	34.88
Total Number of Cases Studied.....			936	253	4,639

The epidemic of 1932 invaded a smaller area than that of 1928; however, the number of cases reported was larger, which might be accounted for by better reporting by physicians, better organized work of the field personnel of the Department of Health and the better recognition of clinical cases of the disease, by physicians.

There seemed to be two distinct areas of epidemic, the first and larger, in the eastern and central portion of the Island; the second, which became affected later, in the northern section, where the disease did not appear in epidemic form during the epidemic of 1928.

The morbidity rate in 1928 was 425.8 per 100,000 population, as compared to 518.4 for 1932; 70 per cent of all the cases of dysentery reported during the year 1932-33 were from the six municipalities where the epidemic was most intense.

The mortality rate for the whole Island was 47.4 in 1928 as compared to 18.3 in 1932. In spite of a larger morbidity rate, the mortality was much lower during 1932. The dysentery mortality curve of 1932, if compared to the morbidity

curve, shows certain characteristics worth while discussing (See Fig. 1). The peak of the epidemic was in October and November, when the largest number of cases occurred. The mortality from dysentery increased considerably during those months as compared to the preceding two months, but the peak of the mortality occurred in February when the number of cases was much lower. The ascent curve of the mortality was very gradual, the number of deaths proportionally smaller than in 1928; there was a second upward trend in mortality after the curve had begun to decline, and the second descent was very gradual. Both high morbidity and mortality rates persisted for a longer period after the peak than in 1928, that is, the descent curve of the 1932 epidemic was much more gradual than in the 1928 epidemic (See table No. 5).

TABLE No. 5  
MORBIDITY AND MORTALITY RATES (PER 100,000  
POPULATION, COMPUTED BY MONTHS ON AN-  
NUAL BASIS) FROM DYSENTERY DURING  
THE YEARS 1928-29 AND 1932-33, WHEN  
EPIDEMICS OF THE DISEASE WERE  
REGISTERED

	1928-29		1932-33	
	Deaths	Cases	Deaths	Cases
July.....	9.3	24.9	7.3	24.8
August.....	9.3	14.0	8.0	13.1
September.....	5.6	24.1	8.3	30.9
October.....	12.4	144.6	11.0	370.1
November.....	110.1	2,099.5	18.1	1,898.2
December.....	146.2	1,660	15.3	1,430.2
January.....	93.3	471.9	24.9	582.3
February.....	52.5	207.4	31.2	786.4
March.....	39.6	138.4	22.7	482.8
April.....	36.1	107.3	19.6	219.2
May.....	28.8	97.2	27.1	226.8
June.....	24.9	114.8	26.5	190.5
Year.....	47.4	425.8	18.3	518.4

The mortality in the towns affected also shows certain characteristics. In San Lorenzo, where the epidemic first made its appearance, the highest rate was registered. In Bayamón, where 11.5 per cent of the total cases of the Island were recorded, the mortality was practically negligible. In the other towns affected the mortality was higher, but the rate was in every case much lower than in the 1928 epidemic.

Table No. 6 shows the mortality rate in various municipalities where the disease appeared in an epidemic form in 1928 or in 1932.

TABLE No. 6  
DEATH RATES (PER 100,000 POPULATION) FROM DYSENTERY FOR  
CERTAIN MUNICIPALITIES DURING THE YEARS FROM  
1927-28 TO 1932-33

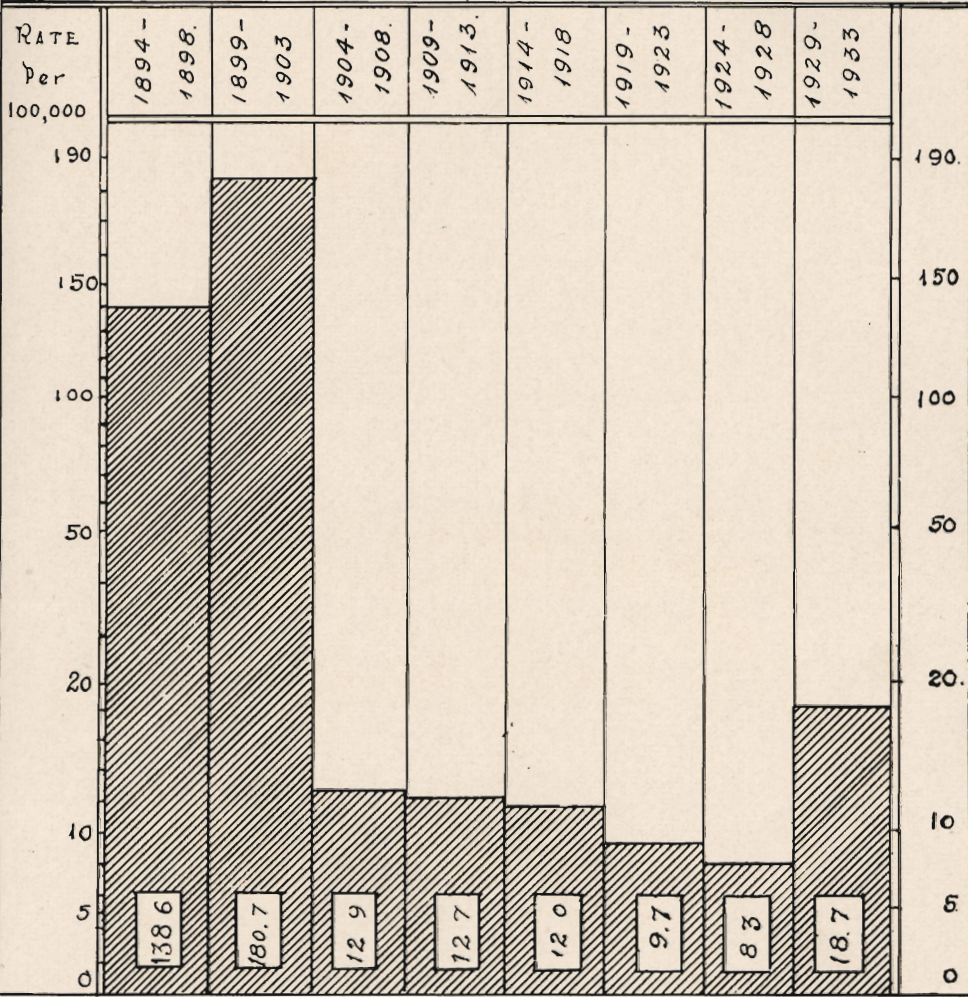
	1927-28	1928-29	1929-30	1930-31	1931-32	1932-33
PUERTO RICO.....	4.9	47.4	14.7	6.5	6.6	18.3
Aguas Buenas.....		142.6		7.7	7.5	
Caguas.....		116.7	8.4	2.0	8.0	31.4
Cayey.....		245.0	10.5			29.8
Cidra.....	10.8	257.0				
Comerio.....		182.1				11.6
Gurabo.....	6.8	87.7	46.5		19.4	38.2
Humacao.....	3.6	41.9		6.6	6.4	15.4
Juncos.....	28.2	69.9	69.2	27.4	6.8	27.0
San Lorenzo.....	13.4	232.2	21.4	4.2	12.3	148.5
Yabucoa.....	28.0	161.8	86.9	22.6	13.4	66.6
Bayamón.....	3.5	6.9	13.6			3.2
Camuy.....						90.0
Loíza.....		70.6	42.8	68.5	10.4	51.1
Luquillo.....		13.1				73.0
Manatí.....	12.6	24.7	8.1	4.0	46.8	160.9
All other towns.....	4.4	29.5	13.5	5.9	5.8	10.

Table No. 7 gives the mortality rate from dysentery in the island of Puerto Rico since 1890 to the present time.

TABLE No. 7  
DEATH RATES IN PUERTO RICO (PER 100,000 POPULATION) FROM  
DYSENTERY DURING THE YEARS FROM 1890 TO 1933

Year	Rate	Year	Rate	Year	Rate
1890.....	61.9	1904-05	2.2	1919-20	8.8
1891.....	44.3	1905-06	11.5	1920-21	8.8
1892.....	64.5	1906-07	18.2	1921-22	11.1
1893.....	46.2	1907-08	13.5	1922-23	9.3
1894.....	74.4	1908-09	2.2	1923-24	8.2
1895.....	107.5	1909-10	13.6	1924-25	8.7
1896.....	94.8	1910-11	20.4	1925-26	9.8
1897.....	212.8	1911-12	16.8	1926-27	10.0
1898.....	203.4	1912-13	10.5	1927-28	4.9
1899.....	374.3	1913-14	5.9	1928-29	47.4
1900.....	278.3	1914-15	10.4	1929-30	14.7
1900-01.....	186.9	1915-16	9.1	1930-31	6.5
1901-02.....	30.1	1916-17	17.9	1931-32	6.6
1902-03.....	34.0	1917-18	16.7	1932-33	18.3
1903-04.....	19.0	1918-19	10.3		

Fig IV Average death rate (5 year periods) from dysentery in PUERTO RICO.



Rates are for fiscal years (July - June) except quinquennium 1894-1898 which is in calendar years.

In Fig. No. 2 is presented a map of Puerto Rico, showing the towns which were affected to the greatest degree during each of the three big epidemics of dysentery following the hurricanes of 1899, 1928 and 1932. Figure No. 4 gives the average annual death rate since 1894 in five-year periods.

II. BACTERIOLOGICAL STUDY. 1. *Collection of the Samples.*—The stools were collected by field inspectors working both in the urban and rural districts. Instructions were given by these to the patients themselves as to how to collect the specimens and the amount and type of material to be placed in the containers, it being impossible for the inspectors themselves to do the work on account of the large extent of territory to be covered in the investigation. The feces were collected in 30 per cent glycerine and sent to the Biological Laboratory at San Juan; in the great majority of the cases the sample was more than twelve hours old when received.

2. *Technique.*—Upon arrival, the sample was plated on Endo's and Eosine methylene blue agar plates, incubated for 24 hours at 37°C, when the plates were examined and suspicious colonies picked out under the microscope and transferred to tubes of Russell's double sugar, which were incubated at 37°C for 24 hours. All Gram negative bacilli which produced acidity or acid and gas in the butt, but no reaction in the slant, were considered as suspicious and transferred to the routine differential media in use in the laboratory which consisted of:

*Carbohydrates:* 1 per cent solution (lactose, sucrose, glucose, mannite, maltose, and xylose) in Dunham's peptone. The media was then incubated at 37°C for two weeks and the results recorded every day.

The ability of the organism to produce indol was determined by cultivation for four days in Dunham's peptone and testing with Ehrlich's reagent.

The production of hydrogen sulphide was evidenced by a brown coloration in stab cultures in tubes of Difco lead acetate agar. The reaction in Difco purple milk and liquefaction in Difco gelatin were also tried.

The suspicious dysentery bacilli were agglutinated with stock Flexner's antiserum. Early in the study antiserum was produced with native strains isolated; thereafter both



kinds of serums were used in the agglutination reactions in dilutions of 1:100-200-400 and 800.

The results of the agglutination reactions together with the cultural reactions formed the criterion for the preliminary classification of the cultures studied.

The percentage occurrence of organisms of possible etiological importance isolated from the 318 feces examined, can be seen in Table No. 8.

Other organisms of undetermined pathogenic action were isolated; *Bacillus proteus* in sixteen cases (5.1%), *Bacillus fecalis alcaligenes* in 8 cases (2.5%) and late fermenters of lactose (gas) in 12 cases (3.7%).

### III. STUDY OF NATIVE STRAINS OF BACTERIUM DYSENTERIAE.\*

In the bacteriological examination of feces from cases of epidemic dysentery in previous studies in the island of Puerto Rico, carried on by González Martínez in 1912<sup>15</sup>, Costa Mandry in 1927<sup>16-17</sup> and Costa Mandry and Garrido Morales in 1931<sup>6</sup>, no attempt was made to definitely classify the individual organisms isolated, but they were placed in one of the then well-known subgroups (the Flexner or mannite-fermenters in the instances cited) depending upon the morphology and cultural characteristics of the bacterium.

In the present study we are attempting the individual classification of a number of dysentery organisms isolated during the 1932 epidemic, together with other organisms isolated since then from sporadic cases of the same disease. We have included in the group selected, representative cultures from each town where cases of the disease were studied by us (Table 16). Any culture which in the preliminary study suggested a dysentery bacillus but showed atypical characteristics, was purposely selected.

The cultures, when this study was commenced, had been kept in agar slants in the ice box (with monthly transplants) from 5 to 8 months since the time they were first isolated.

Known dysentery bacilli used as controls in the grouping

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\* We are specially indebted to the International Health Division of the Rockefeller Foundation for their invitation to us to visit the Bacteriological Laboratory of the University of Chicago under Prof. Jordan, and other laboratories in the United States to observe methods employed in the study of organisms of the Eberthella and Salmonella groups.

We also wish to express our appreciation to Miss Olga Ortiz and Mr. Saldaña for valuable technical assistance.

and classification of our native strains were obtained from varied sources, as can be seen from Table No. 9.

TABLE No. 8

MICROORGANISMS OF POSSIBLE ETIOLOGIC IMPORTANCE ISOLATED  
FROM 318 FECAL SPECIMENS OF CASES OF DYSENTERY DURING  
THE POST CYCLONIC EPIDEMIC OF 1932

Organisms	Frequency	
	Number	Percentage
Flexner group.....	99	31.1
Schmitz bacillus.....	5	1.57
Sonne bacillus.....	2	0.62
Dispar group.....	6	1.88
<i>Salmonella Morgagni</i> .....	1	0.31
<i>Salmonella Paratyphi</i> .....	2	0.62
<i>Salmonella Schottmülleri</i> .....	4	1.25

TABLE No. 9

## SOURCES OF STOCK DYSENTERY CULTURES

Number	Organism	Source
F 6 Y...	Flexner.....	Department of Hygiene and Bacteriology, University of Chicago. (1933)
S B.....	Sonne Brown.....	
D.....	Dispar.....	
Sh.....	Shiga.....	
W.....	Cable.....	London School of Tropical Medicine, Department of Bacteriology. (1933)
V.....	Lentz.....	
X.....	Hughes.....	
Y.....	Lister 17.....	
Z.....	Whittington.....	
H D.....	Hiss Russell.....	Detroit City Laboratories (1933)
F D.....	Flexner.....	
F R.....	Flexner.....	Virginia State Laboratory, Richmond, (1933)
H R.....	Hiss Russell.....	
F N.....	Flexner.....	Division of Lab. and Res., N. Y. Dept. of Health, Albany (1931)

BIOCHEMICAL CHARACTERISTICS.—The organism to be studied was plated out on Levine's eosine methylene blue (Difco) agar, and after incubation at 37°C for twenty-four hours, *smooth* colonies were picked out under the microscope and transferred to slant tubes of Russell's double sugar agar. From here the organism was inoculated into a tube of plain broth, incubated at 37°C for twenty-four hours and trans-

planted thence to the different media. From the original Russell tube, subcultures were made on plain agar slants, incubated for 18 to 24 hours at 37°C and kept in the ice box for future use.

*Fermentation Tests:* The carbohydrates employed in this study were prepared as follows: The peptone base consisted of 1 per cent Bacto peptone and 0.5 per cent sodium chloride in water, adjusted to pH. 7.2, and sterilized at 15 lbs. pressure for twenty minutes. The stoppered tubes and vials for the finished carbohydrate media were prepared and sterilized beforehand. When ready for use, brom cresol purple (0.04%) was added to the peptone base in a sufficient amount to give a good color; the carbohydrate was then added in the concentration to be used; the media was then tubed and sterilized at 10 pounds pressure for fifteen minutes. Lactose, sucrose, maltose, mannite and dextrose were added in one per cent concentration, the other carbohydrates in 0.5 per cent. The organism was transplanted in the carbohydrates and kept at 37°C for twenty-one days, the change in reaction being observed and recorded daily.

MISCELLANEOUS TESTS.—*Hydrogen sulphide:* The ability of the organism to produce hydrogen sulphide was tested in lead acetate agar (Difco). Inoculation was made by stabbing the media; the production of hydrogen sulphide was detected by means of a brown discoloration diffused throughout the line of growth.

*Indol:* The presence of indol was detected by the addition of Ehrlich's reagent to four-days old culture (37°C) in Dunham's peptone and to two-days old culture in tryptophane broth (Difco).

*Gelatin:* The liquefaction of gelatin was detected by growing the organisms in ten per cent gelatin with 0.3 per cent beef extract and 0.5 per cent peptone, the media, after inoculation was kept in the incubator at 37°C.

*Milk:* The reaction of the organism in milk was determined by growing it in Difco purple milk.

AGGLUTINATION REACTIONS.—The macroscopic test tube method was employed throughout. Eighteen to twenty-four-hour old agar slant cultures of the organisms were suspended in saline solution to the proper density. The mixture of

serum and bacterial suspension was incubated for three hours at 37°C. and then placed over night in the ice box, after which the presence or absence of agglutination was determined by examination under a magnifying hand lens. The total volume of fluid in each tube was one cubic centimeter.

AGGLUTININ ABSORPTION.—All our antiserums were prepared in rabbits, using both dead and live cultures of the organisms. Before use, each serum was carefully titrated. In the absorption tests, the following technique was employed:

Five cc of a 1:50 dilution of the serum in normal salt solution was placed over a slant agar culture of the organism (18–24 hours old) to be tested, the culture was washed off and the suspension delivered to a tube which was placed in the incubator at 37°C for three hours, after which the suspension was centrifuged at high speed for one half hour and the supernatant fluid removed. With this fluid the same process was repeated four or five times, depending on the results previously obtained with the serum against its homologous culture.

#### DISCUSSION

The dysentery group of bacilli, besides the well known and classical types, comprises a number of organisms of different characteristics and undetermined pathologic specificity. The various organisms of the group have been the subject of numerous studies in attempts to simplify the recognition and classification of each type by their agglutinating characteristics, bio-chemical reactions, or the combined results of both methods<sup>22, 23, 24, 25, 26.</sup>

It is a well known fact that certain organisms when freshly isolated are inagglutinable, and this is specially noted in the typhoid-dysentery group, and is a source of difficulty to investigators interested in these problems. Boyd<sup>27</sup> in India, found 30 per cent of freshly isolated strains of organisms, which from their bio-chemical reactions resembled true dysentery bacilli, to be inagglutinable.

In studies carried out at the Division of Laboratories and Research of the New York State Department of Health at Albany<sup>28</sup>,—

“Microorganisms which closely resemble *B. typhosus* or *B. dysenteriae* in their morphological, cultural, and biochemical reactions, but which are not

agglutinated in antisera prepared with these species nor in the patient's sera, are isolated with relative frequency from fecal or urinary specimens submitted for release, from patients who have had bacteriologically proved typhoid fever or bacillary dysentery; for diagnosis or release, from clinical cases of typhoid fever or dysentery not confirmed by bacteriological findings; and from suspected carriers. They are usually nonmotile, ferment rhamnose, and produce indol. In certain instances, the clinical and epidemiological data indicate very strongly that these are inagglutinable variants of true typhoid or dysentery bacilli."

The lack of knowledge as to the true etiological agents of a number of acute and chronic diarrheal conditions in Puerto Rico makes it necessary that all organisms with a possible etiological relationship be thoroughly studied and classified.

Following the well-known early classification of dysentery bacilli according to their action on certain carbohydrates, we separated our organisms into four large groups, each of which was again subdivided according to the serologic characteristics of each individual bacterium (Table 15).

*Group A.*—Organisms which do not attack lactose or mannite (5 subcultures studied).

*Group B.* (Flexner group).—Organisms which do not attack lactose but produce definite acidity in mannite (27 subcultures studied).

*Group C.* (Sonne Group).—Composed of organisms which produce acidity (as a rule delayed) in lactose, no indol and no action on xylose (2 subcultures studied).

*Group D.*—Composed of a varied number of organisms which produce acidity in lactose and other carbohydrates and produce indol (8 subcultures studied).

Organisms isolated during the dysentery epidemic of 1932 and other cultures from sporadic cases of the same disease studied here and there since the epidemic, were primarily grouped into one of the four classifications according to their biochemical reactions (Table No. 10). The biochemical characteristics of the stock dysentery organisms used for control in this study can be seen in Table No. 11.

Individual typing of each organism according to its agglutinating (Table No. 12) and absorption (Table No. 13 and 14) characteristics was then carried out.

In the typing of the organisms, reciprocal absorption was performed whenever we possessed both serum and or-

TABLE No. 10

## RESULTS OF FERMERTATION TESTS AND OTHER MISCELLANEOUS PROCEDURES OF NATIVE DYSENTERY BACILLI

Group	Culture Number	Milk	Indol (Tryp)	Indol (Pep)	Hydrogen Sulphide	Gelatin Liquefaction	Glucose	Lactose	Sucrose	Mannite	Maltose	Xylose	Salicin	Sorbitol	Rhamnose	Arabinose	Dulcitol	Trehalose	Inosite	Levulose	Galactose	Dextrin	Melezitose	Inulin	Glycerine	
A	61	+	+	+	-	-	+	-	-	-	14	-	-	12	+	-	-	+	-	+	+	-	0	-	+	
	127	+	+	+	-	-	+	-	-	-	-	-	-	+	+	-	-	+	-	+	+	-	0	-	+	
	47	+	+	+	-	-	+	-	-	-	-	-	-	9	+	-	-	+	-	+	+	-	0	-	9	
	195	-	+	+	-	-	+	-	6	-	-	-	-	-	-	-	-	-	-	+	+	-	-	-	+	
	55	-	+	+	-	-	+	-	6	-	-	-	-	-	-	-	-	-	-	+	+	-	-	-	+	
B I	88	+	+	+	-	-	+	-	-	+	5	-	-	-	-	8	-	+	-	+	+	14	-	-	-	
	98	+	+	+	-	-	+	-	-	+	12	-	-	-	-	10	-	+	-	+	+	-	-	-	-	
	13	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	+	-	-	-	
	279	+	+	+	-	-	+	-	-	+	-	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
	77	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
	286	+	+	+	-	-	+	-	-	+	+	-	-	-	-	-	-	+	-	+	+	0	-	-	-	
	160	+	+	+	-	-	+	-	-	+	-	-	-	-	-	-	-	+	-	+	+	-	0	-	-	
	175	+	+	+	-	-	+	-	-	+	10	-	-	-	-	-	-	+	-	+	+	-	-	-	-	
	264	+	+	+	-	-	+	-	-	+	12	-	-	-	-	+	-	+	-	+	+	-	0	-	-	-
	B II	90	+	+	+	-	-	+	-	-	+	10	-	-	-	-	+	-	+	-	+	+	-	-	-	-
P4		+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	-	-	-	
215		+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
P2		+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	0	-	-	
233		+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	-	-	-	
B III	305	+	+	+	-	-	+	-	-	+	+	-	-	+	-	10	-	+	-	+	+	-	-	-	-	
	309	+	+	+	-	-	+	-	-	+	+	-	-	+	-	5	-	+	-	+	+	-	0	-	-	
	303	+	+	+	-	-	+	-	-	+	+	-	-	5	-	+	-	+	-	+	+	-	-	-	-	
	288	+	+	+	-	-	+	-	-	+	+	-	-	5	-	+	-	+	-	+	+	-	0	-	-	
	202	+	+	+	-	-	+	-	-	+	-	-	-	+	-	+	-	-	-	+	+	-	-	-	-	
	283	+	+	+	-	-	+	-	-	+	12	-	-	+	5	+	-	+	-	+	+	-	-	-	-	
B IV	P3	+	+	+	-	-	+	-	-	+	+	-	-	-	+	-	+	-	+	+	-	-	-	-	-	
	238	+	+	+	-	-	+	-	-	+	+	-	-	-	+	-	-	-	-	+	+	-	-	-	-	
	16	+	+	+	-	-	+	-	-	+	+	-	-	-	+	-	-	-	-	+	+	-	-	-	-	
	40	+	+	+	-	-	+	-	-	+	+	-	-	2	2	+	-	+	-	+	+	-	-	-	-	
	250	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	-	-	-	
B V	17	+	+	+	-	-	+	-	-	+	+	-	-	6	-	-	-	12	-	+	+	-	-	-	-	
B VI	M	+	+	+	-	-	+	-	-	+	+	-	-	-	+	-	+	-	+	+	-	-	-	-	-	
B VII	79	+	+	+	-	-	+	-	-	+	+	-	-	-	+	-	-	+	-	+	+	-	0	-	-	
	33	+	+	+	-	-	+	-	-	+	-	-	-	-	+	-	-	-	-	+	+	+	-	-	-	
C	151	+	-	-	-	-	+	6	7	+	+	-	-	-	+	+	-	+	-	+	+	0	0	-	-	
	221	+	-	-	-	-	+	8	7	+	+	-	-	-	+	+	-	+	-	+	+	-	-	-	-	
D 1	129	+c	+	+	-	8	+	+	+	+	+	+	-	-	+	+	-	+	-	+	+	-	-	-	6	
	170	+c	+	+	-	10	+	6	6	+	+	+	-	+	+	+	5	+	-	+	+	-	-	-	+	
D 2	83	+	+	+	-	-	+	+	-	+	+	+	8	+	+	+	10	+	-	+	+	-	-	-	5	
D 3	146	+	+	+	-	-	+	+	+	+	+	+	+	+	+	6	+	-	+	+	-	+	-	+	+	
D 4	Rob	+	+	+	-	-	+	+	10	+	+	+	-	+	10	+	+	+	-	+	+	-	-	-	+	
	189	+c	-	-	-	-	+	-	-	-	-	-	-	-	-	+	-	-	-	+	+	-	-	-	-	

+ Acid Production within 4 days  
 - Negative Reaction  
 C Coagulation

Figure — Days when Reaction became evident  
 0 — Initial Acidity with Reversion to Neutrality

TABLE No. 10

RESULTS OF FERMERTATION TESTS AND OTHER MISCELLANEOUS PROCEDURES OF NATIVE DYSENTERY BACILLI

Group	Culture Number	Milk	Indol (Tryp)	Indol (Pep)	Hydrogen Sulphide	Gelatin Liquefaction	Glucose	Lactose	Sucrose	Mannite	Maltose	Xylose	Salicin	Sorbitol	Rhamnose	Arabinose	Dulcitol	Trehalose	Inosite	Levulose	Galactose	Dextrin	Melezitose	Inulin	Glycerine	
A	61	+	+	+	-	-	+	-	-	-	14	-	-	12	+	-	-	+	-	+	+	-	0	-	+	
	127	+	+	+	-	-	+	-	-	-	-	-	-	+	+	-	-	+	-	+	+	-	0	-	+	
	47	+	+	+	-	-	+	-	-	-	-	-	-	9	+	-	-	+	-	+	+	-	0	-	9	
	195	-	+	+	-	-	+	-	-	6	-	-	-	-	-	-	-	-	-	+	+	-	-	-	+	
	55	-	+	+	-	-	+	-	-	6	-	-	-	-	-	-	-	-	-	+	+	-	-	-	+	
B I	88	+	+	+	-	-	+	-	-	+	5	-	-	-	-	8	-	+	-	+	+	14	-	-	-	
	98	+	+	+	-	-	+	-	-	+	12	-	-	-	-	10	-	+	-	+	+	-	-	-	-	
	13	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	+	-	-	-	
	279	+	+	+	-	-	+	-	-	+	-	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
	77	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
	286	+	+	+	-	-	+	-	-	+	+	-	-	-	-	-	-	+	-	+	+	0	-	-	-	
	160	+	+	+	-	-	+	-	-	+	-	-	-	-	-	-	-	+	-	+	+	-	0	-	-	
	175	+	+	+	-	-	+	-	-	+	10	-	-	-	-	-	-	+	-	+	+	-	-	-	-	
	264	+	+	+	-	-	+	-	-	+	12	-	-	-	-	+	-	+	-	+	+	-	0	-	-	-
B II	90	+	+	+	-	-	+	-	-	+	10	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
	P4	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	-	-	-	
	215	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
	P2	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	0	-	-	
	233	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	-	-	-	
B III	305	+	+	+	-	-	+	-	-	+	+	-	-	+	-	10	-	+	-	+	+	-	-	-	-	
	309	+	+	+	-	-	+	-	-	+	+	-	-	+	-	5	-	+	-	+	+	-	0	-	-	
	303	+	+	+	-	-	+	-	-	+	+	-	-	5	-	+	-	+	-	+	+	-	-	-	-	
	288	+	+	+	-	-	+	-	-	+	+	-	-	5	-	+	-	+	-	+	+	-	0	-	-	
	202	+	+	+	-	-	+	-	-	+	-	-	-	+	-	+	-	-	-	+	+	-	-	-	-	
	283	+	+	+	-	-	+	-	-	+	12	-	-	+	5	+	-	+	-	+	+	-	-	-	-	
B IV	P3	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
	238	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	-	-	-	
	16	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	-	-	-	
	40	+	+	+	-	-	+	-	-	+	+	-	-	2	2	+	-	+	-	+	+	-	-	-	-	
	250	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	-	-	+	+	-	-	-	-	
B V	17	+	+	+	-	-	+	-	-	+	+	-	6	-	-	-	12	-	+	+	-	-	-	-	-	
B VI	M	+	+	+	-	-	+	-	-	+	+	-	-	-	-	+	-	+	-	+	+	-	-	-	-	
B VII	79	+	+	+	-	-	+	-	-	+	+	-	-	-	+	-	-	+	-	+	+	-	0	-	-	
	33	+	+	+	-	-	+	-	-	+	-	-	-	-	+	-	-	-	-	+	+	+	-	-	-	
C	151	+	-	-	-	-	+	6	7	+	+	-	-	-	+	+	-	+	-	+	+	0	0	-	-	
	221	+	-	-	-	-	+	8	7	+	+	-	-	-	+	+	-	+	-	+	+	-	-	-	-	
D I	129	+c	+	+	-	8	+	+	+	+	+	+	-	-	+	+	-	+	-	+	+	-	-	-	6	
	170	+c	+	+	-	10	+	6	6	+	+	+	-	+	+	+	5	+	-	+	+	-	-	-	+	
D 2	83	+	+	+	-	-	+	+	-	+	+	+	8	+	+	+	10	+	-	+	+	-	-	-	5	
D 3	146	+	+	+	-	-	+	+	+	+	+	+	+	+	+	6	+	-	+	+	-	+	-	+	+	
D 4	Rob	+	+	+	-	-	+	+	10	+	+	+	-	+	10	+	+	+	-	+	+	-	-	-	+	
	189	+c	-	-	-	-	+	-	-	-	-	-	-	-	-	+	-	-	-	+	+	-	-	-	-	

+ Acid Production within 4 days  
 - Negative Reaction  
 C. Coagulation

Figure — Days when Reaction became evident  
 O — Initial Acidity with Reversion to Neutrality

ganism of a particular strain. We worked with one organism and its homologous serum of Group A; 12 organisms and their homologous sera of Group B; 2 organisms and their homologous sera of Group C and 2 organisms and their homologous sera of Group D besides organisms and sera for control purposes obtained from diverse sources. From the antisera we attempted to absorb the agglutinins with the other unknown cultures and tested the absorbed serum with the absorbing culture, the agglutinin producing culture and the other cultures. Constant difficulty was encountered throughout the work on account of the marked antigenic similarity between the different cultures, especially the Flexner Group (B).

Simple agglutinations were performed with the serum of each individual subgroup against representative cultures of all the subgroups as can be seen in Table 12. The dilutions of serum used were as follows: 1 in 100, 200, 500, 1,000, 2,000, 5,000, 10,000, 15,000 and 20,000.

The characteristics of the individual strains as grouped by us follow:

*Group A—Non-fermenters of lactose or mannite.*

In this group, generally recognized as the Shiga group, are included the true Shiga bacillus and the Schmitz or ambiguous bacillus. These two organisms although belonging in the same group are different in their pathologic, serologic and cultural characteristics. The Schmitz bacillus is the less toxic and produces indol; the Shiga bacillus is the most toxic of the whole group producing the most severe type of dysentery, it does not produce indol, and neither of the two ferment mannite. In our study we have not encountered any strains of true Shiga bacillus but three of the organisms isolated by us were similar in their cultural characteristics to what has been described as Schmitz bacillus by Gardner<sup>29</sup>, Koser et al<sup>24</sup> and Johnston, Brown and Kaake<sup>30</sup>. These three organisms (Nos. 61, 47, and 127) belonged to the same serologic sub-type. Milk was acidified but not coagulated, indol production was definite, there was no production of hydrogen sulphide in lead acetate agar and no liquefaction of gelatin. Acid production was definite in glucose, sorbitol, trehalose, rhamnose, levulose, galactose and glycerine. One of the organisms, No. 61, produced a slight