

# A Throat Culture Survey of Troops Stationed in Puerto Rico<sup>1</sup>

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THROAT CULTURES for beta-hemolytic streptococci were made of 1,490 soldiers stationed in the City of San Juan and vicinity. Seven hundred and eighty-five of these soldiers came from various parts of the continental United States and had been in this tropical environment from four months to two years; the vast majority of them had been in Puerto Rico for about a year. Seven hundred and five were island troops, of whom only eight had ever been off the Island. The living conditions of both groups were similar.

The pour-plate method, three plates for each individual, was utilized. Details of this procedure have already been described in a previous publication.<sup>2</sup>

Cultures were taken from June 6 to December 20, 1943 and from November 13 to December 20, 1944. The hemolytic streptococci isolated were grouped according to Lancefield's method,<sup>3</sup> using a formamide extract recommended by Fuller.<sup>4</sup> Two hundred and two strains (13.5 percent) were isolated from the 1,490 soldiers studied.

Twenty-seven soldiers (1.8 percent) had group A, 104 (7 percent) had group C, and 69 (4.6 percent) had group G streptococci. From one individual a beta-hemolytic streptococcus, which remained unclassified, was isolated; from another a group B organism was obtained.

Of the 785 continental soldiers examined, 79 (10 percent) gave throat cultures positive for beta-hemolytic streptococci. The 705 island soldiers studied gave 123 (17.4 percent) positive cultures. Table 1 shows group distribution of all these strains.

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1. Received for publication June 26, 1945. This investigation was carried out in collaboration with the Commission on Epidemiological Survey, Board for the Investigation and Control of Influenza and other Epidemic Diseases in the Army, Preventive Medicine Service, Office of the Surgeon General, U. S. Army.

2. A. Pomales Lebrón, A study of hemolytic streptococci as found in the tropical island of Puerto Rico. *Puerto Rico J. Pub. Health & Trop. Med.*, 16:66-133, 1940.

3. R. C. Lancefield, A serological differentiation of human and other groups of hemolytic streptococci. *J. Exp. Med.*, 57:571-595, 1933.

4. A. T. Fuller, The formamide method for the extraction of polysaccharides from hemolytic streptococci. *Brit. J. Exp. Path.*, 19:130-139, 1938.

TABLE 1

Beta-Hemolytic Streptococci Isolated from Throat Cultures of Continental and Island Troops Stationed in Puerto Rico

Date of Culture	Continental Troops Positive					Unclas-sified	Island Troops Positive					Unclas-sified
	Number Examined	A	B	C	G		Number Examined	A	B	C	G	
1943												
June 2	40	2	0	0	0	0	17	1	0	2	2	0
June 15	41	0	0	1	1	0	20	1	0	1	1	0
June 17	57	0	0	2	2	0	4	0	0	0	0	0
June 24	42	3	0	0	2	0	0	0	0	0	0	0
June 30	68	1	0	6	1	0	0	0	0	0	0	0
July 15	23	2	0	2	1	0	49	0	0	2	4	0
July 22	30	1	0	1	1	0	26	0	0	0	3	0
August 5	63	1	0	1	3	1	0	0	0	0	0	0
August 17	1	0	0	1	0	0	63	1	0	5	6	0
September 15	25	0	0	1	1	0	0	0	0	0	0	0
September 23	25	2	0	0	0	0	0	0	0	0	0	0
October 6	25	2	0	2	1	0	0	0	0	0	0	0
October 20	25	1	0	3	1	0	0	0	0	0	0	0
October 27	25	0	0	1	0	0	0	0	0	0	0	0
November 4	25	0	0	1	0	0	0	0	0	0	0	0
November 23	0	0	0	0	0	0	45	0	0	10	1	0
December 1	0	0	0	0	0	0	50	0	0	3	5	0
December 6	0	0	0	0	0	0	49	0	0	7	3	0
December 8	0	0	0	0	0	0	50	0	0	10	3	0
December 15	0	0	0	0	0	0	51	1	0	10	5	0
December 20	0	0	0	0	0	0	49	0	0	9	2	0
1944												
November 13	85	0	1	3	5	0	50	2	0	0	3	0
November 16	101	3	0	5	7	0	40	2	0	6	2	0
November 22	52	0	0	2	0	0	43	0	0	3	0	0
December 6	27	0	0	0	1	0	49	1	0	1	1	0
December 20	5	0	0	0	0	0	50	0	0	3	1	0
Totals	785	18	1	32	27	1	705	9	0	72	42	0

It must be stated here that 9 (1.2 percent) group A strains were isolated from island troops while 18 (2.3 percent) came from the continental soldiers. Of the 79 positive cultures obtained from this latter group, 18 (22.7 percent) belonged to group A; of the 123 positive cultures from the Puerto Rican troops, 9 (7.2 percent) were A strains.

One hundred and seventy-nine island soldiers were examined during the months of June, July, and August of 1943. Positive cultures were obtained from 29. Sixty-nine positive cultures were obtained from the 294 Puerto Rican soldiers studied during the months of November and December of the same year. From previous studies of a similar nature, conducted in Puerto Rico and in

other tropical regions,<sup>5</sup> the conclusion had been reached that a significant seasonal variation in the proportion of hemolytic streptococci, obtained from the throats of healthy persons, did not exist in tropical environments. This seemed to also hold true for the present studies, as a 10 percent gross carrier rate was obtained during these same months for both continental and island troops examined in 1944. It is interesting to note that of the 69 strains isolated from the 294 island troops examined during November and December 1943, only one strain belonged to group A.

Table 2 summarizes the data concerning the relative numbers of hemolytic streptococci among the continental and island soldiers from whom positive cultures were obtained. The condition of the throat at the time of culture is also indicated.

The high proportion of C and G organisms, especially among island troops, is striking.

Of the 27 soldiers from whom group A streptococci were cultured, 15 had no throat symptoms at the time the culture was taken; no history was obtained from 4, but 8 were suffering from an acute sore throat. In only one case with a normal throat did the cultures yield many group A organisms. If the group A cultures, from individuals with a sore throat, are not taken into account, the group A carrier rate among the continental Americans would be 1.8 percent and for the Puerto Ricans, 0.7 percent.

In two instances many group C colonies, but no group A organisms, were found in individuals suffering from an acute sore throat. Previous studies carried out in Puerto Rico<sup>6</sup> and elsewhere<sup>7</sup> have demonstrated that beta-hemolytic streptococci not belonging to Lancefield's group A have been found associated with pathological conditions. The same (Puerto Rico)<sup>8</sup> study showed that beta-hemolytic streptococci were cultured from 33 percent of excised tonsils. Out of 14 strains grouped, 9 were A and 5 were C.

An examination of Table 2 shows no significant difference be-

5. A. Pomales Lebrón, A bacteriological study of normal throats, pathological throats, and excised tonsils, made in Puerto Rico. Puerto Rico J. Pub. Health & Trop. Med., 11:512-532, 1936.

D. F. Milam and W. G. Smillie, Bacteriological study of "colds" on isolated tropical island (St. John, United States Virgin Islands, West Indies). J. Exp. Med., 53:733-751, 1931.

P. Morales Otero, Brief summary of findings on flora of normal throats in San Juan, Porto Rico, June 1929-30. Porto Rico J. Pub. Health & Trop. Med., 7:377-380, 1932.

A. F. Coburn, The Factor of Infection in the Rheumatic State (Baltimore: The Williams and Wilkins Company, 1931).

6. A. Pomales Lebrón, *op. cit.* (5).

7. S. M. Wheeler and G. E. Foley, A note on non-group A streptococci associated with human infection. J. Bact. 46:391-392, 1943.

8. A. Pomales Lebrón, *op. cit.* (5).

Island (705)

Continental (785)

No. of Colonies	A	B	C	G	Un.	A	B	C	G	Un.	A	B	C	G	Un.
Very few (1-5 colonies)	0000 +++		000000 +	000000 0		00		000000 +	000000 0		00		000000 0	000000 +++	
Few (5-20 colonies)	00000 +		000000	00000 0000 ++		0 + +		000000	00000 0000 ++		0 + +		00000 00000 0000 000 ++	00000 00000 000 +	
Some (20-30 colonies)	00 +		000000 0	00000 0	0	+		000000 0	00000 0	0	+		00000 00000 00000 ++	00000 000 +	
Many (50-100 colonies)	0 +	0	0000	00		++		0000	00		++		00000 00000 0000 ++	000 +++	
Numerous (More than 100 colonies)	+		000 +	0				000 +	0				00000 00000 00000 +++	00000 +	
	18	1	32	27	1	9	None	72	42	None	9	None	72	42	None
						79 (10%)					123 (17.4%)				

0 = No throat trouble when culture was taken.  
 † = No definite information concerning condition of throat.  
 + = Acute sore throat.  
 Un. = Unclassified.

tween island and continental troops as regards the relative numbers of streptococci of the various groups obtaining in their throats.

The typing of 23 of the 27 group A strains was attempted, utilizing the precipitin method described by Swift, Wilson, and Lancefield.<sup>9</sup> Of 7 strains from Puerto Ricans, 4 were negative with all the serums.<sup>10</sup> The other three were types 1, 33, and 41, respectively. Of 16 cultures from American soldiers, 7 were negative; 3 were type 19, 2 type 1, 2 type 44, and the other two strains were types 36 and 14, respectively. The majority of these cultures were tested about ten months after isolated. This fact, and the further one that we were dealing mainly with organisms from normal throats, may explain the negative results obtained with eleven strains.

STREPTOCOCCAL ANTIHEMOLYSIN

Antihemolysin determinations were made on serums from 962 soldiers of the 1943 series, of whom 486 were continental North Americans and 476 were Puerto Ricans. Samples of blood were taken at the same time that the throats were cultured, or within one week after. The same hemolytic filtrate was utilized in the majority of the determinations. Todd's strain Aronson (Schnitzer) was used for the production of the hemolysin. The methods employed for the test have been described already,<sup>11</sup> and the results summarized in Table 3.

TABLE 3  
Median Antihemolysin Values

	Number of Cases	Median Values in Units
Continental and island troops <sup>a</sup>	962	73.0
Continental troops <sup>a</sup>	486	69.2
Island troops <sup>a</sup>	476	73.0
Continental and island troops harboring hemolytic streptococci (groups A, C, and G)	138	81.0

<sup>a</sup>These groups are composed of all the men, irrespective of whether they gave a positive or a negative throat culture.

The antihemolysin values varied from 13 to 793 units among the continental troops, and from 13 to 631 units in the island troops.

9. H. F. Swift, A. T. Wilson, and R. C. Lancefield, Typing of group A hemolytic streptococci by M precipitin reactions in capillary pipettes. *J. Exp. Med.*, 78:127-133, 1943

10. These serums were kindly supplied to us by Dr. R. C. Lancefield.

11. S. M. Wheeler and G. E. Foley, *op. cit.*

As shown in Table 3, the median value for troops, both continental and Puerto Rican, having positive cultures, was higher (81 units) than that for soldiers in general (73 units), irrespective of whether they gave a positive or a negative culture.

## DISCUSSION

Numerous studies have been made of the beta-hemolytic streptococcus carrier rates for various parts of the world. These studies have shown that such carrier rates vary widely. Investigations of this nature, conducted in Puerto Rico, indicate a moderate fluctuation in the proportion of healthy carriers of beta-hemolytic streptococci. During the year 1931,<sup>12</sup> 200 throats were cultured and beta-hemolytic streptococci isolated in 4 percent of them. In 1935, 416 throats were examined and a gross carrier rate of 9 percent was found. These figures contrast strongly with the 17.4 percent of positive cultures obtained from the 705 island soldiers studied in the present series. It may be of interest to point out again that most of these strains belonged to groups C and G. The low proportion obtained in the 1931 series may be explained, at least in part, by the fact that the streak-plate method was then utilized and that only persons without histories of sore throat during the previous two years were included. Both men and women were included in that series.

The 1935 series consisted of male workers in camps situated in the interior of Puerto Rico and of dairy farm male personnel near San Juan. In some respects, this last group is comparable to the island troops studied in the present investigation. The troops, however, were on the average younger men, but both groups were studied at the same time of the year.

Although the Puerto Rican and continental groups are not exactly comparable as regards time, it is interesting to observe that the proportion of positive cultures among the island troops (17.4 percent) was higher than that obtained among the continental soldiers (10 percent). The proportion of group A streptococci in the strains isolated from both continental and insular troops was 22.7 percent and 7.3 percent, respectively.

In this connection reference is made to a study of the incidence of beta-hemolytic streptococci in the throats of normal monkeys that arrived in Puerto Rico from Calcutta, via New York, in November

12. A. Pomales Lebrón, *op. cit.* (5).

1938. These monkeys were examined one week after arrival and at long intervals thereafter. The results obtained are summarized in Table 4 for the purpose of comparison.

TABLE 4  
*Incidence and Group Distribution of Beta-Hemolytic Streptococci from the Throats of Normal Monkeys*

Date	Number of Animals Examined	Number of Animals with a Positive Culture	Distribution Among Lancefield Groups			
			A	B	C	G
Nov.-Dec. 1938	172	22 (13%)	9 (41%) <sup>a</sup>	0	8 (37%)	5 (22%)
Jan.-Feb. 1940	309	39 (13%)	0	0	26 (67%)	13 (33%)
Sept. 1941	146	11 (7.5%)	1 (9%)	0	4 (36%)	6 (55%)
Jan. 1943	70	32 (45.7%)	0	1 (3%)	23 (72%)	8 (25%)

<sup>a</sup>These percentages refer to the proportion of strains of any particular group among the total number of strains isolated.

An examination of Tables 1 and 4 shows that the results from a single study of the throat flora of given populations, even though carried out at the same time of the year, may be entirely misleading. This is made evident when we observe that the hemolytic streptococcus gross carrier rate for 294 island troops was 23.4 percent during the months of November and December 1943 as compared with one of 10 percent for 232 Puerto Rican soldiers examined during the same months of 1944. The same fluctuation may be seen among the monkeys with a gross carrier rate of 13 percent for 1938 and 1940 and one of 45 percent for 1943. These findings suggest that factors other than variations in temperature and overcrowding may fundamentally affect the hemolytic streptococcus carrier rate.

Another point of interest is the high proportion of groups C and G strains obtaining in the throats of the soldiers and of the monkeys, contrasting strongly with the low proportion of group A streptococci found. It must be noted that 41 percent of the strains obtained from the monkeys in 1938 belonged to group A, while none of this group was obtained in 1940 and 1943 when the gross carrier rates were the

same, or much higher. In 1941 only one strain out of 11, isolated from 146 animals, belonged to group A. The disappearance of A organisms from the throat of these animals, when the gross carrier rate was maintained at the same level, or much higher, is interesting.

We have seen that the proportion of group A streptococci among Puerto Ricans (1.2 percent) and among continental North Americans (2.3 percent), after the latter had been on the Island for about one year, is low even when the gross carrier rate was relatively high. Whether or not the latter's carrier rate would fall to a lower level after a prolonged sojourn in the tropics is, of course, a matter of conjecture. There exists, however, the possibility of a high group A carrier rate having been present among the continental troops on arrival so that a carrier rate of 2.3 percent may have represented a significant decrease.

Hare<sup>13</sup> summarizes the work done by various investigators in different parts of the world (London, Edinburgh, United States, Japan, Hong-Kong, Melbourne). Of 3,102 throats examined by these workers, 574 were positive for beta-hemolytic streptococci, a gross carrier rate of 18.5 percent. Two hundred and sixteen of the persons examined harbored group A organisms, with a group A carrier rate of 7 percent. Of the total number of strains isolated, 37.6 percent belonged to group A. These figures contrast with our findings wherein a 13.5 percent gross carrier rate and a group A carrier rate of 1.8 percent were found, with a proportion of 13.3 percent of the total number of organisms (from both continental and island troops) belonging to group A.

Schwentker<sup>14</sup> examined the throats of soldiers at several army camps, Chanute Field and Scott Field, in Illinois, and Fort Knox, Kentucky, during the winter months and found a group A carrier rate of 3 percent, 5 percent, and 7 percent, respectively, giving an average carrier rate of 5 percent. The group A carrier rate obtained in Puerto Rico was much lower than that found in temperate regions.

Schwentker also found that during an epidemic of scarlet fever the gross group A and scarlatinal type carrier rates "roughly parallel each other." He further states: "The changes in the gross carrier rate are due to variation in the group A rate which, in turn, are dependent on the changes in rate for the scarlatinal type." This is in marked contrast with the complete lack of parallelism between the

gross and group A carrier rates in the scarlet fever-free groups studied here, in which the increase in the gross carrier rate was due to a corresponding increase in the group C and G organisms.

Of interest also was the observation that the antihemolysin content of the blood of both island and continental troops was not significantly different.

#### SUMMARY

1. Throat cultures from 1,490 soldiers stationed in Puerto Rico—785 continental North Americans and 705 Puerto Ricans—were performed and gross hemolytic streptococcus carrier rates of 10 percent and 17.4 percent obtained for the continental and island troops, respectively. A group A carrier rate of 2.3 percent for continental troops and one of 1.2 percent for Puerto Ricans was found. A gross carrier rate of 13.5 percent and a group A carrier rate of 1.8 percent were also found for the entire group of 1,490 soldiers.

Parallelism between group A and gross carrier rates was completely lacking, the majority of strains belonging to Lancefield's groups C and G. The group A carrier rate obtained in temperate regions was higher than that found in Puerto Rico.

2. Typing was attempted in 23 group A strains. Of 7 Puerto Rican strains, 4 were negative. The remaining belonged to types 1, 33, and 41. Of the 16 cultures from North American soldiers, 7 were negative. Three belonged to type 19, two to type 1, two to type 44, and the other two to types 14 and 36, respectively.

3. The median antihemolysin values in the serums from 473 Puerto Ricans (73.0 units) and 486 North Americans (69.2 units) did not vary significantly.

13. R. Hare, Sources of hemolytic streptococcal infection of wounds in war and in civil life. *Lancet*, 1:109-122, 1940.

14. F. F. Schwentker, Survey of hemolytic streptococci in certain army camps. *Army Med. Bul.*, 65:94-104, 1943.