STUDY OF PARACOLON ORGANISMS WITH THE MAJOR ANTIGEN OF SHIGELLA SONNEI, FORM I

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Received for publication December 14, 1953

Ferguson and co-workers (Ferguson and Henderson, 1947; Ferguson *et al.*, 1947) reported the isolation of a motile, anaerogenic paracolon organism which they designated C 27, sharing the somatic antigen with *Shigella sonnei*, form I. A similar strain was isolated by Wheeler (personal communication, 1947).

In Ceylon, the first strain of this type was isolated during an investigation of the carrier rate of Salmonellae in domestic animals (Schmid and Velaudapillai, 1953), and within the course of one year nine C 27 strains were isolated from man and animals.

The only other country from which it is known that such strains have been isolated is the Belgian Congo (Ewing, personal communication, 1953), where Vande Pitte (personal communication, 1953) isolated four C 27 strains from man and animals.

The original C 27 strain was isolated from the feces of a patient of whom no clinical history was available to Ferguson and Henderson (1947). Three of the four C 27 strains from the Belgian Congo were isolated from diarrhetic patients, one (included in this study) as a mixed infection with *Shigella flexneri* var. *Manchester* and a true *S. sonnei*; the fourth strain was isolated post mortem from the spleen of a chimpanzee, the kidney of which yielded *S. flexneri*, strain 4a (Vande Pitte, personal communication, 1953).

MATERIAL AND METHODS

All our isolations were effected from direct platings on SS agar (Difco), with subsequent inoculation on a combined Kligler-urea medium (Schmid *et al.*, 1953), in which these strains produced a Shigella-like reaction. The strains were motile and gave a good slide agglutination with *S. sonnei* antiserum.

The five C 27 strains isolated from man in Ceylon were derived from 4 diarrhetic children (1 fatal case) and one adult from which no clinical history was available. In all isolations the C 27 strains were the only findings.

TABLE 1

Biochemical activity of C 27 strains of paracolon organisms

	TYPE			
	1	2	3	4
Adonitol	−	−	−	−
Dulcitol	−	−	−	+
Sorbitol	−	−	−	−
Arabinose	−	−	−	−
Xylose	−	−	−	−
Rhamnose	−	−	−	−
Maltose	+	+	+	+
Salicin	$+^{7-9}$	−	$+^{11}$	$+^{8}$
Inositol	$+^{1-2}$	$+^{1-3}$	+	+
Trehalose	+	+	+	+
Lactose	$+^{1-9}$	$+^{2-3}$	−	$+^{2}$
Sucrose	−	−	−	−
Mannitol	−	−	−	−
Glucose	±	±	±	±
Stern's glycerol fuchsin	−	−	−	−
Indole production	+	+	+	+
H_2S production	−	−	−	−
Gelatin liquefaction	−	−	−	−
Ammonium glucose	$+^{1-3}$	$+^{1-3}$	$+^{3}$	$+^{2}$
Ammonium citrate	−	−	−	−
KNO_3 reduction	+	+	+	+
Voges-Proskauer	−	−	−	−
Methyl red	+	+	+	+
Urea	−	−	−	−
D-Tartrate	−	−	−	−
L-Tartrate	−	−	−	−
Sodium citrate	−	−	−	−
Mucate	−	−	−	−

Key: + positive after one day (glucose: ± acid, no gas)
$+^{1-9}$ positive after 1–9 days
− negative after 14 days (gelatin: not liquefied after 60 days).

Type 1: 253-50, 515/C, 517/C, 512/C; Type 2: 602-52, 511/C, 513/C, 514/C, 516/C, 518/C; Type 3: 990-53; Type 4: 523/C.

The Ceylon animal strains were all isolated from apparently healthy animals. The following

TABLE 2

Serological characteristics of C27 strains of paracolon organisms

STRAIN	SERA											
	253-50	602-52	990-53	511/C	512/C	513/C	516/C	517/C	518/C	523/C	514/C	515/C
253-50												
602-52												
990-53												
511/C												
512/C	5,120-10,240	5,120-10,240	2,560-5,120	1,280-2,560	5,120-10,240	5,120-10,240	5,120-10,240	2,560-5,120	5,120-10,240	1,280-2,560	640-1,280	1,280-2,560
513/C												
516/C												
517/C												
518/C												
523/C												
514/C	160	1,280	320	0	1,280-2,560	2,560	320	160	1,280	320	2,560	2,560
515/C												

Key: 0 = no agglutination in a dilution 1:40.

TABLE 3

Serological characteristics of C27 strains of paracolon organisms

SERUM	STRAINS											
	A										B	
	253-50	602-52	990-53	511/C	512/C	513/C	516/C	517/C	518/C	523/C	514/C	515/C
A	+	+	+	+	+	+	+	+	+	+	−	−
B	−	−	−	−	−	−	−	−	−	−	+	+

C 27 strains were investigated: 253-50[2] C 27 original; 990-53[1] and 602-52[2] isolated from man in Africa (Belgian Congo); 515/C, 516/C, 517/C, and 523/C isolated from man in Ceylon; 511/C isolated from a sheep in Ceylon; 512/C isolated from a goat in Ceylon; 513/C isolaled from a polecat (*Pardoxurus hermaphroditus hermaphroditus*) in Ceylon; 514/C isolated from a cow in Ceylon.

These cultures were studied biochemically and serologically. Rabbit sera for somatic and flagellar antigens were produced according to standard methods for each C 27 strain and for somatic antigens of *S. sonnei*, form I.

[1] Number Institut de Médecine Tropicale, Léopoldville, Belgian Congo.

[2] Number Communicable Diseases Center, Chamblee, Georgia.

We wish to thank Drs. W. H. Ewing (Chamblee) and J. M. Vande Pitte (Léopoldville) for making these strains available for our study.

RESULTS

Biochemical behavior. All C 27 strains were anaerogenic and identical in their macroscopical and microscopical behavior as described by Ferguson and Henderson (1947).

The biochemical activity, however, was not uniform. By means of fermentation of dulcitol, lactose, and salicin four different patterns were recognized (table 1).

Somatic antigens. All C 27 strains agglutinated to titer in each C 27 serum and also in *S. sonnei*, form I, serum. In cross adsorption tests with all strains and sera, complete removal of somatic agglutinins was obtained in all sera (titer < 1:40). Hence the strains investigated are identical in their somatic antigens.

Flagellar antigens. Contrary to the uniformity of somatic agglutination, flagellar agglutination revealed two distinctly different patterns of results. Two groups of 10 and 2 strains, respectively, were recognized.

The sera of one group (10 strains) agglutinated the other group (2 strains) approximately to one-tenth of the titers only, whereas the sera of the group of 2 strains did not show such a marked difference of agglutination.

Table 2 shows these results giving lowest and highest titers for each serum and group of strains as there was not always uniform agglutination, but the differences did not exceed one dilution.

Cross adsorption tests with strains and sera within each group were able to exhaust the respective sera (titer < 1:40). Cross adsorption with strains and sera of different groups left residual titers of 1:604 to 1:1,280 for either one or the other group. These adsorbed sera reacted specifically with strains of the homologous group only which were provisionally labeled as "A" (10 strains) and "B" (2 strains) as shown in table 3.

DISCUSSION

The result of Ferguson and Henderson (1947) that the C 27 strains possess the heat stable somatic antigen of *S. sonnei*, form I, was confirmed on 11 other strains, irrespective of the source or country of isolation.

Although the material of this investigation was limited to twelve C 27 strains, it permitted to recognize four biochemical patterns within this group of *Enterobacteriaceae*. No conclusion as to a predominance of one or the other pattern in correlation to source or country of isolation was possible (table 1).

The results of flagellar agglutination indicate that the antigens of C 27 strains are not identical as two strains isolated from man and animal in Ceylon are distinctly different in flagellar agglutination (tables 2 and 3).

As to the composition of the flagellar antigens, these results suggest that the C 27 strains may behave like Salmonellae and, though probably diphasic, may occur in stable monophasic variants like *Salmonella cholerae suis* var. *kunzendorf* (VI, VI, – :1,5), or may also share antigenic factors common to both phases like *Salmonella salinatis* (IV, XII, d, e, h:d, e, n, z_{15}).

The clinical material is too small as to assess a possible pathogenic role of C 27 strains in man. From a practical point of view, however, it is emphasized that all routine isolations of *S. sonnei* be examined for motility to uncover a possible occurrence of C 27 strains.

SUMMARY

Twelve C 27 strains, motile paracolon organisms, possessing the major somatic antigen of *Shigella sonnei*, form I, were compared biochemically and serologically. Four different biochemical patterns were recognized. All C 27 strains were identical in their somatic antigens, whereas two different patterns of flagellar agglutination were found.

It is recommended that all routine isolations of *S. sonnei* be examined for motility.

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