## STUDIES ON HEMOLYTIC STREPTOCOCCI

# VII. DISTINGUISHING CHARACTERS OF THE LACTOSE-NEGATIVE SPECIES OF LANCEFIELD'S GROUPS A AND C

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Since 1916, when Holman described *Streptococcus equi* as lacking in ability to ferment lactose, strains of that character have been designated as *S. equi* indiscriminately. There are, however, in Lancefield's streptococcal groups A and C, three sub-groups, the strains of which may be lactose-negative. Their distinguishing characters are given in table 1, and for comparison there are included in the table the characters of *Streptococcus pyogenes*, the streptococcus which most commonly infects man, and those of the streptococcus which most commonly infects man, and those of the streptococcus which most commonly infects animals, known as "animal pyogenes." The recognition of three separate species of lactose-negative streptococci of groups A and C would serve the convenience of medical bacteriologists, because distinctive characters *in vitro* are correlated with susceptibility of host species, as shown in table 1.

Frost and Engelbrecht proposed the name Streptococcus equisimilis for the beta-hemolytic streptococci which ferment trehalose but not lactose or sorbitol. They did not distinguish between the precipitin groups A and C, and they were unaware that the strains they described are capable of causing human infections. The name S. equisimilis is acceptable for the group which includes trehalose-positive strains of group C, because it appears that Frost and Engelbrecht were dealing with strains of group C. They identified S. equisimilis with type B of Ogura, which Edwards placed in group C, and two strains (1253 and 1254) labeled S. equisimilis received from Dr. Frost were found to belong to group C. One of them (1253) possessed the differential characters of the majority of strains of the group accepted as S. equisimilis. (See table 2 of the accompanying paper.)

In the 5th edition of "Bergey's Manual of Determinative Bacteriology," S. equi is recognized as a distinct species, and S. equisimilis is recognized as an unnamed, "poorly defined species whose taxonomic relationships are not clear." (Species 1 of the appendix, p. 346 of the Manual.) Sherman was of the opinion that there are sufficient reasons for placing in a separate species those strains of group C which are capable of causing human disease, and Brown recognized the trehalose-positive, sorbitol-negative strains of group C as a separate species. He suggested the possibility that the new species might be identified with S. equisimilis of Frost and Engelbrecht.

One character included in table 1—sensitivity to bacteriophage—was not recognized as a differential character in the Manual. Nevertheless it is a highly significant character, and the test for sensitivity to phage is simple and easily applied in any laboratory. The routine worker who would rely on sensitivity to phage B to identify strains of groups A and C would rarely be misled. The close correlation between the grouping according to precipitative reactions and sensitivity to phage B was shown in an earlier paper (Evans and Verder). It was found that the strains of group C are sensitive to filtered phage B, whereas the strains of group A are resistant to it, although they are sensitive to nascent phage B. There are few exceptions to that general rule with most of the discrepancies occurring among the strains of S. equisimilis, which is a transitional species with some of the characters of group A (including ability to cause human infection), although the precipitative reaction and, in the case of most of the strains, the bacteriophagic reaction conforms with that of strains of group C.

Among the characters listed in table 1, only one, fermentation of trehalose, distinguishes S. equi from S. equisimilis. This appears to give an accepted basis for differentiation of species, for in the case of many genera, as outlined in Bergey's Manual, differentiation of species depends on reaction with a single

 TABLE 1

 Characters of the lactose-negative species of beta-hemolytic streptococci of groups A and C

DESIGNATION	DIFFERENTIAL CHARACTERS					
	Precipi- tin group	Sensitiv- ity to phage B filtrate	Fermentation of:			CAUSES TRANSMISSIBLE DISEASE IN:
			Treha- lose	Sorbi- tol	Lactose	
<i>S</i> . —•	С	+	-	+	+	Various species of domestic animals
S. equi	C	+	. —	_	-	Horses
S. equisimilis	C	+*	+	_	- or $+$	Various species of domestic animals and man <sup>†</sup>
<i>S</i> . – <sup>b</sup>	A	-	+	-	- or $+$	Man
S. pyogenes	A	-	+	-	+	Man

\* Exceptions to the general rule are shown in table 2 of the accompanying paper.

<sup>†</sup> See table 2 of the accompanying paper for data on host species.

• The unnamed species known as "animal pyogenes".

<sup>b</sup> Lactose-negative strains of group A.

fermentable substance. Actually, however, there are other less precise, or less readily measurable, points of distinction between S. equi and S. equisimilis, such as vigor or growth in plain infusion broth, sensitivity to bile, lysis of human fibrin, and susceptibility of host species. (See the accompanying paper for a detailed description of S. equisimilis, and see the third paper of this series for a description of S. equi.)

The lactose-negative strains of group A differ from those of group C in their lack of sensitivity to phage B filtrate, as well as in precipitative reactions. Since those tests are not utilized in every laboratory, the distinctions may not be readily acceptable to all bacteriologists. However, students of beta-hemolytic streptococci regard the precipitative reaction as the most significant test available to give practical information concerning the source and pathogenic properties of an unknown strain, and, as already stated, differences indicated by precipitative reactions are confirmed by bacteriophagic reactions. Further study should be made before completing the differentiation of strains of group A into species, but it appears reasonable to state at this time that the lactose-negative strains of group A should not be included in the species S. equisimilis.

The confusion which hitherto existed in the classification of lactose-negative streptococci, with the result that strains of groups A and C were placed together in the species *S. equi*, derived from the assignment of too great significance to the fermentation of lactose, a character which in the genus *Streptococcus* is subordinate to the precipitative reaction and to sensitivity to phage B. A more logical classification would be to divide groups A and C into sub-groups on the basis of precipitative reactions and sensitivity to phage B, and further subdivide into species on the basis of fermentative reactions.

The following key is suggested for the differentiation of those streptococci of the pyogenic group which do not hydrolyze sodium hippurate, to replace that given on page 324 of the 5th edition of Bergey's Manual:

- a. Precipitin group C (sensitive to filtered phage B).
  - b. Sorbitol fermented.
    - 1. Trehalose not fermented, lactose fermented. Streptococcus sp. (Animal pyogenes)
  - bb. Sorbitol not fermented.
    - 2. Trehalose and lactose not fermented. Streptococcus equi.
  - 3. Trehalose fermented, lactose may or may not be fermented, S. equisimilis.
- aa. Precipitin group A (resistant to filtered phage B). Trehalose fermented, sorbitol not fermented. (Further study is required before the subdivision of group A into species may be completed.)

#### SUMMARY

The differential characters of S. equisimilis are defined, and a key is proposed for the identification of the species of beta-hemolytic streptococci of group C.

#### REFERENCES

See the list of references of the accompanying paper.