Rejection of the Type Strain of *Streptococcus mitis* (Andrewes and Horder 1906)

Request for an Opinion

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The type strain of *Streptococcus mitis*, strain NCTC 3165 (= ATCC 33399), does not fit the description of the species, and it is genetically unrelated to reference strains that do conform to the description. Strain NCTC 3165, which was deposited as *S. mitis* in 1930, does fit the description of *Streptococcus sanguis* White and Niven (1946) and was recognized as a *S. sanguis* strain by authors of reports that were published prior to 1980, when this reference strain was elevated to the type strain of *S. mitis*. *Streptococcus oralis* Bridge and Sneath (1982) fits the description of *S. mitis*, and its type strain, strain NCTC 11427, is genetically homologous with reference strains that fit the description of *S. mitis*. *S. oralis* has the same cell wall characteristics that have been described for *S. mitis* ("S. mitior"). I propose that *S. oralis* be considered a later synonym of *S. mitis* and that the type strain of *S. oralis* be adopted as the type strain of *S. mitis*.

Streptococcus mitis was first described by Andrewes and Horder in 1906 (1). These authors noted the frequent occurrence of this organism in saliva and intestinal contents and its lack of association with disease. It did not clot milk and fermented sucrose and lactose, but it rarely fermented raffinose or inulin. Sherman et al. (16) regarded *S. mitis* as a "rather ill defined and heterogeneous group" but showed that it was clearly different from *Streptococcus salivarius*, another common oral commensal, and listed several characteristics by which these two species could be differentiated. Among these characteristics were the inability of *S. mitis* to ferment inulin, its alpha-hemolysis, and the overall tendency of *S. mitis* to be less active against several substrates, such as esculin, arginine, raffinose, and trehalose. *S. mitis* never fermented arabinose, mannitol, or sorbitol. Sherman et al. also noted that *S. mitis* was easily distinguished from *Streptococcus bovis*, *Streptococcus equinus*, and *Streptococcus thermophilus*, yet it had no characteristics "uniquely its own" which would facilitate its identification. In 1972, Colman and Williams (5) described a group of alpha-hemolytic streptococci that were distinguishable by cell walls that contained ribitol teichoic acid but little or no rhamnose. These bacteria seldom hydrolyzed esculin or arginine and usually failed to ferment salicin and trehalose. Colman and Williams preferred the name "*S. mitior*" for the species because the epithet *mitis* connoted an ill-defined group and the name *mitior* had been used previously by Schottmüller (15) and predated the publication of Andrewes and Horder (1). Nevertheless, these streptococci were similar, if not identical, to the *S. mitis* of Andrewes and Horder (1) and Sherman et al. (16). In addition, Colman and Williams noted that certain strains of *Streptococcus sanguis* (20) that failed to hydrolyze arginine and esculin and had the characteristic "*S. mitior*" cell wall, but produced a glucan (or dextran) like *S. sanguis*, could be considered dextran-positive "*S. mitior*" strains. *S. sanguis* NCTC 7864 (= ATCC 10557) was an example.

Also in 1972, Schleifer and Kandler (12) showed that certain strains of streptococci had peptidoglycan with direct L-lysine cross-links. Among these strains was strain NCTC 7864 (= ATCC 10557).

Comparisons of deoxyribonucleic acid (DNA) base sequence homologies among strains of *S. sanguis* showed that those strains that did not hydrolyze arginine or esculin and therefore resembled *S. mitis* (or "*S. mitior*") were indeed unrelated to other more typical *S. sanguis* strains (7). Among these unrelated strains was *S. sanguis* ATCC 10557. A later study (6) showed that the streptococci whose biochemical characteristics resembled those of *S. mitis* but that made a glucan like *S. sanguis* were indeed genetically identical to strains that were considered typical of "*S. mitior*" or *S. mitis*. (Some other strains of *S. mitis* or "*S. mitior*" form a separate genetic group [6].)

Thus, by 1980, research on the streptococci had shown that *S. mitis* (or "*S. mitior*") comprised a species characterized by alpha-hemolysis, the failure to hydrolyze arginine or esculin, and fermentation of few sugars. This species had cell walls lacking rhamnose but containing ribitol and a peptidoglycan with direct lysine links. Some strains produced glucan from sucrose. This organism was genetically distinct from *S. sanguis*. In 1980 *S. mitis*, not "*S. mitior*", was included on the Approved Lists of Bacterial Names (17). The official description is that given in *Bergey's Manual of Determinative Bacteriology*, 8th ed. (8); this description was based on the data of Sherman et al. (16). Strain NCTC 3165 was chosen as the type strain (17).

Strain NCTC 3165^T (T = type strain) was deposited in the National Collection of Type Cultures in 1930, prior to the description of *S. sanguis* and at a time when human nonhemolytic streptococci were usually called *S. mitis* if they were not *S. salivarius*. However, strain NCTC 3165^T has characteristics of *S. sanguis*, such as hydrolysis of arginine and esculin and fermentation of inulin. Colman and Williams (5) and Cole et al. (3) recognized this strain as *S. sanguis* if the species was expanded to include strains that did not make dextran (glucan) (5), and Colman and Williams (4) showed in 1965 that strain NCTC 3165^T had cell walls like those of *S. sanguis* and not like those of "*S. mitior*" (*S. mitis*). The reason for selecting this strain in 1980 as the type strain for *S. mitis* is unclear.

Since 1980, several investigators (9, 10, 13, 14, 19; Coykendall, in *Proceedings of the Xth Lancefield Meeting*, in

TABLE 1. Relationships among type and reference strains of *S. mitis*, *S. sanguis*, and *S. oralis*

Organism(s)	Hydrolysis of:		Acid from inulin	Cell wall sugars ^a				Peptidoglycan type ^b	DNA homology with ^c :		
	Esculin	Arginine		Rhamnose	Glucose	Galactose	Ribitol		<i>S. oralis</i> ATCC 11427 ^T	<i>S. sanguis</i> ATCC 10556 ^T	<i>S. sanguis</i> ATCC 10558
<i>S. mitis</i> type strain (NCTC 3165)	+	+	+	+	+	+	—	Lys-ala	Very low (24)	Low	High (93)
Strains that fit the description of <i>S. sanguis</i> ^d											
Type strain ATCC 10556	+	+	+	+	+	—	—	Lys-ala	Low (41)	High (100)	Low (50)
Strain ATCC 10558	+	+	+	+	+	+	—	Lys-ala	Low (39)	Low (47)	High (100)
<i>S. oralis</i> type strain NCTC 11427	—	—	—	(+)	(+)	+	+	Lys-direct	High (100)	Very low	Very low
Strains that fit the description of <i>S. mitis</i> , such as strain ATCC 10557 ^d	—	—	—	—	—	+	+	Lys-direct	High (87)	Very low	Very low

^a See references 3, 4, and 10. +, Present; —, absent; (+), small amount.

^b Lys-direct, Direct lysine links; Lys-ala, lysine-to-alanine links (10, 12, 14).

^c Data compiled from references 6, 7, 10, and 14. The numbers in parentheses are percentages (Coykendall, in press).

^d Descriptions from reference 6.

press) have confirmed that *S. mitis* NCTC 3165^T splits arginine and esculin and ferments raffinose and inulin. The peptidoglycan of this strain has lysine-alanine cross-links (10, 14). Streptococci with these characteristics fit the description of *S. sanguis* (8) much better than the description of *S. mitis*.

The results of DNA hybridization experiments confirm that *S. mitis* NCTC 3165^T belongs to one of two genetic groups that conform to the description of *S. sanguis* (7). One group includes type strain ATCC 10556. The members of this group conform to the description of the species and have a lysine-alanine peptidoglycan. The second group of strains that fits the description of *S. sanguis* includes *S. sanguis* ATCC 10558. Although genetically distinct, this group is phenotypically nearly identical to the other *S. sanguis* group. The type strain of *S. mitis* is a member of this second group based on phenotypic characteristics and DNA hybridization data (9, 14; Coykendall, in press). These two genetic groups (or genospecies) have not been divided because they do share some DNA base sequences (7) and because they are so difficult to separate by conventional biochemical tests. Maintenance of these two genetic groups within the single species *S. sanguis* is consistent with the recent report of the Ad Hoc Committee on Reconciliation of Approaches to Bacterial Systematics (18), which recommended that "genospecies that cannot be differentiated from another genospecies on the basis of any known phenotypic property not be named until they can be differentiated by some phenotypic property." (It has been determined that the members of the second group usually produce alkaline phosphatase and do not ferment starch; members of the other group [including the type strain] have the opposite reactions. Kilian et al. [9] have found additional differences in the glycoside hydrolases and immunoglobulin A proteases of the two groups.)

The taxonomy of *S. mitis* is further complicated by *Streptococcus oralis* (2). This species was described in 1982 and comprised a rather heterogeneous group of strains, some of which fermented ribose and mannitol, while others resembled *S. mitis* ("S. mitior") and had been shown previously to be phenotypically and genetically related to "S. mitior" (6). The species description was emended in 1985 (10). The type strain of this species (strain NCTC 11427) fits the description of *S. mitis* and is genetically homologous with strains, such as strain ATCC 10557 (= NCTC 7864), which fit the descrip-

tion of *S. mitis*. (As expected, the DNA from the type strain of *S. oralis* is not homologous with DNA from the type strain of *S. mitis* because the *S. mitis* strain is related to *S. sanguis*.)

Kilpper-Bälz et al. (10) and Schmidhuber et al. (14) analyzed the relationships among type and reference strains of *S. sanguis*, *S. mitis* ("S. mitior"), and *S. oralis*. These authors showed that the cell walls and phenotypic characteristics of these bacteria and their genetic relationships were as described above and showed that *S. oralis* cell walls were the type described for *S. mitis*. Their data also showed that the *S. mitis* type strain was more closely related to the type strain of *S. sanguis*, strain ATCC 10556 (= NCTC 7863), than to reference strains that resemble *S. mitis*, such as strain ATCC 10557 (= NCTC 7864) and *S. oralis* NCTC 11427. This is consistent with the results of previous DNA hybridizations, which showed that the two genetic groups of *S. sanguis* are more closely related to each other than to *S. mitis* ("S. mitior") (7). Except for some biochemical traits, the data of Kilpper-Bälz et al. and Schmidhuber et al. confirmed the genetic, cell wall, and phenotypic groups perceived by other workers (4, 5, 7, 9), but these authors chose to name the groups strictly according to the names of the type strains within each group. Thus, the *S. sanguis* strains that were related to the type strain of *S. mitis* were called *S. mitis* although they fit the description of *S. sanguis*, and the strains that fit the description of *S. mitis* were called *S. oralis*.

Assignment of the name *S. mitis* to streptococci that fit the description of *S. sanguis* would produce several problems. The descriptions of both *S. sanguis* and *S. mitis* would need to be rewritten. Considerable basic and clinical literature on *S. sanguis* and *S. mitis*, including taxonomic work, would be rendered uninterpretable.

I propose that the valid species *S. mitis*, with its valid description, be maintained. The present type strain should be rejected because it does not conform to the description of the species and its DNA is not related to the DNAs of streptococci that do conform to the description (see Rule 18i of the *International Code of Nomenclature of Bacteria* [11]).

Strains of *S. oralis*, including the type strain, fit the description of *S. mitis*, especially if one considers their cell walls and their DNA base sequence homologies with strains that fit the *S. mitis* description. Therefore, I also propose

that *S. oralis* be considered a later synonym of *S. mitis* and that its type strain (strain NCTC 11427) become the type strain of *S. mitis*.

Table 1 summarizes the relationships among *S. mitis*, *S. sanguis*, and *S. oralis*.

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