16S Ribosomal DNA Sequences of Anaerobic Cocci and Proposal of *Ruminococcus hansenii* comb. nov. and *Ruminococcus productus* comb. nov.

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The 16S ribosomal DNA sequences of representative members of the family *Peptococcaceae* were determined. The members of the family examined were divided into the following four phylogenetic groups: *Peptococcus niger* ATCC 27731^T (T = type strain), the *Sarcina*-*Peptostreptococcus anaerobius* group, the ruminococcus-coproccoccus group, and the peptostreptococcus group. *Peptococcus niger*, the type species of the family, was not related to other members of the family. *Peptostreptococcus anaerobius* ATCC 27337^T, the type strain of the type species of the genus *Peptostreptococcus*, was closely related to *Clostridium sordellii* NCIB 10717^T (level of sequence similarity, 85%). *Sarcina ventriculi* GIFU 7886, a spore-forming anaerobic gram-positive coccus, clustered with *Clostridium perfringens* ATCC 13124^T at a similarity value of 91%. Members of the *Sarcina*-*Peptostreptococcus anaerobius* group clustered with clostridia at similarity values ranging from 85 to 91%. The type strains of *Peptostreptococcus prevotii*, *Peptostreptococcus asaccharolyticus*, *Peptostreptococcus micros*, and *Peptostreptococcus magnus* clustered at levels of sequence homology of 84 to 93%. This cluster was not included in the *Peptococcus niger* group or the *Peptostreptococcus anaerobius* group. Thus, these members of the genus *Peptostreptococcus* should be separated from the other members of the genus and also from members of the family *Peptococcaceae*. The sequence of *Peptostreptococcus productus* ATCC 27340^T was different from the sequences of *Peptostreptococcus anaerobius* and *Peptococcus niger*. The sequence of *Streptococcus hansenii* ATCC 27752^T, a strictly anaerobic strain, was different from the sequences of other streptococci; this strain clustered with *Peptostreptococcus productus*, coprococci, and ruminococci. Several phenotypic characteristics of *Streptococcus hansenii* ATCC 27752^T were similar to characteristics of ruminococci. These organisms require fermentable carbohydrates and do not produce butyric acid from glucose. Thus, we propose that *Peptostreptococcus productus* and *Streptococcus hansenii* should be transferred to the genus *Ruminococcus*.

The anaerobic gram-positive cocci are classified in a single family, the *Peptococcaceae* (12). This family contains five genera, *Peptococcus*, *Peptostreptococcus*, *Ruminococcus*, *Sarcina*, and *Coprococcus* (6). These five genera were placed in the family *Peptococcaceae* simply because their members are gram-positive anaerobic cocci. However, it has been suggested that the members of this family are heterogeneous (2, 7, 10, 11). The family name *Peptococcaceae* was not included in *Bergey's Manual of Systematic Bacteriology*, vol. 2 (14), and the members of the family were placed in "other genera" in Section 12, the gram-positive cocci.

The 16S rRNA sequences of most anaerobic cocci have not been determined. Therefore, the phylogenetic positions of these organisms are not yet clear. Woese (18) extensively analyzed the sequences of bacteria and reported that the members of the division *Firmicutes* (4) should be divided into two major phylogenetic groups (18). One group consisted of organisms having low moles percent G+C, and the members of other group had high G+C contents. The anaerobic gram-positive cocci have low G+C contents, and thus we expected them to belong to the low-G+C-content group. When we determined partial sequences of anaerobic cocci and streptococci (1), we found that the anaerobic cocci did not cluster with other members of the low-G+C-content group. Some anaerobic strains of streptococci clustered with

peptostreptococci and ruminococci. Other workers have reported that the members of the genus *Peptostreptococcus* are heterogeneous (7). The levels of RNA-DNA homology among six members of the genus *Peptostreptococcus* showed that these organisms should be divided into at least five generic groups. Our sequence data supported this observation (unpublished data).

To elucidate the phylogenetic relationships among members of the family *Peptococcaceae*, we performed additional sequence analyses of 16S ribosomal DNAs from representative anaerobic gram-positive cocci.

MATERIALS AND METHODS

The bacterial strains used and the accession numbers of the 16S ribosomal DNA sequences deposited in the DNA Data Bank of Japan are shown in Table 1. The sequences of the other species used (Table 2) were obtained from GenBank.

Peptostreptococci and strains of anaerobic streptococci were grown in GAM anaerobic medium (Nissui Corp., Tokyo, Japan) or brucella HK semisolid medium (Kyokuto Corp., Tokyo, Japan). The 16S rRNA genes of these organisms were amplified by using the PCR method and universal primers originally described by Lane et al. (8, 20). The sequences were determined by using both dye primers and the dye terminator method performed with an automatic sequencer (Applied Biosystems, Foster, Calif.). The se-

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TABLE 1. Strains used to determine 16S rRNA sequences

Species	Strain	Status	DDBJ no. ^a
<i>Peptostreptococcus anaerobius</i>	ATCC 27337 ^T	Type strain	D14150
<i>Peptostreptococcus asaccharolyticus</i>	ATCC 14963 ^T	Type strain	D14138
<i>Peptostreptococcus prevotii</i>	ATCC 9321 ^T	Type strain	D14139
<i>Peptostreptococcus tetradius</i>	GIFU 7672 ^T	Type strain	D14142
<i>Peptostreptococcus micros</i>	GIFU 7701	Human clinical isolate	D14143
<i>Peptostreptococcus magnus</i>	ATCC 15794 ^T	Type strain	D14149
<i>Peptostreptococcus productus</i>	ATCC 27340 ^T	Type strain	D14144
<i>Sarcina ventriculi</i>	GIFU 7886	Human stomach isolate	D14151
<i>Coprococcus eutactus</i>	ATCC 27759 ^T	Type strain	D14148
<i>Ruminococcus torques</i>	ATCC 27756 ^T	Type strain	D14137
<i>Ruminococcus gnavus</i>	ATCC 29149 ^T	Type strain	D14136
<i>Streptococcus hansenii</i>	ATCC 27752 ^T	Type strain	D14155

^a DDBJ, DNA Data Bank of Japan.

quences from position 8 to position 1392 (*Escherichia coli* numbering) were determined.

The sequences of the other organisms used for alignment and for the similarity matrix were obtained from previously published data available from the DNA Data Bank of Japan. The ODEN program package of the DNA Data Bank of Japan was used to align the sequences, and phylogenetic distances were calculated by using both the unweighted pair group method and the neighbor-joining (NJ) method (13).

RESULTS

Alignment of the sequences of all of the anaerobic cocci tested revealed that these sequences have a unique 25-base deletion between position 445 and position 470 (*E. coli* numbering). The sequences of representative species around the deleted area are shown in Fig. 1. The peptostreptococci, ruminococci, sarcinas, coprococci, and clostridia shared this deletion; however, *Peptococcus niger* ATCC 27731^T (T = type strain), the streptococci, the enterococci, the lactococci, and the staphylococci did not have this deletion.

A data set for representative species belonging to the domain *Bacteria* (19) was prepared, and the phylogenetic positions of the anaerobic cocci tested were determined by using both the unweighted pair group method and the NJ method. Similar results were obtained from both calculations, and only the results of the NJ method are shown in Fig. 2.

The anaerobic cocci had low G+C ratios and were expected to belong to the low-G+C-content group; however,

these organisms belonged to independent branches of both the high- and low-G+C-content groups (Fig. 3).

A different data set that included data for low-G+C-content organisms was prepared, and the sequences were aligned. The phylogenetic distances were calculated by the NJ method, as shown in Fig. 2. When homology values greater than 84% were used (Fig. 2), the anaerobic cocci were separated into five major clusters. The type species of the genus *Peptococcus*, the genus *Peptostreptococcus*, and the genus *Sarcina* belong to independent clusters. *Sarcina ventriculi* GIFU 7886, the spore-forming anaerobic cocci, and *Clostridium perfringens* ATCC 13124^T formed one cluster at a level of sequence similarity of 91%. Most other members of the genus *Peptostreptococcus* belonged to another independent branch (Fig. 2, 4, and 5); the levels of homology of these organisms ranged from 84 to 93%.

Coprococcus eutactus ATCC 27759^T, *Ruminococcus gnavus* ATCC 29149^T, *Ruminococcus torques* ATCC 27756^T, *Streptococcus hansenii* ATCC 27752^T, and *Peptostreptococcus productus* ATCC 27340^T all belonged to major branch (Fig. 3 through 5); however, the similarity values of these strains ranged from 84 to 89% (Fig. 2).

Peptostreptococcus productus ATCC 27340^T and *R. gnavus* ATCC 29149^T formed a branch at a homology level of 86.70%. These two species also formed a branch with *Streptococcus hansenii* ATCC 27752^T and *Coprococcus eutactus* ATCC 27759^T (Fig. 2 and 3).

The cell wall peptidoglycan types of the anaerobic cocci correlated with the results of the phylogenetic analysis of 16S rRNAs (Table 2). Characteristics that differentiate *Pep-*

TABLE 2. Characteristics that differentiate members of the genus *Ruminococcus* from other anaerobic cocci

Taxon	G+C content (mol%)	Peptidoglycan ^a		Peptone used as a major energy source	Growth stimulated by carbohydrates	Isocaproic acid production	Butyrate production
		Position 3	Interpeptide				
<i>Peptococcus niger</i>	50-51	Lys	D-Asp	+	—	+	+
<i>Peptostreptococcus anaerobius</i>	33-35	Lys	D-Asp	+	—	+	+
Other peptostreptococci	28-37	Lys, Orn	D-Glu, D-Asp, Gly	+	—	—	D ^b
<i>R. productus</i> comb. nov.	43-45	m-Dpm	None	—	+	—	—
<i>R. hansenii</i> comb. nov.	39-42	m-Dpm	None	—	+	—	—
<i>Ruminococcus</i> spp.	39-46	m-Dpm	None	—	+	—	—
<i>Coprococcus</i> spp.	39-44	m-Dpm	None	—	+	—	+
<i>Sarcina</i> spp.	28-31	LL-Dpm	Gly	—	+	—	D

^a D-Asp, D-aspartate; m-Dpm, meso-diaminopimelic acid; Lys, lysine; D-Glu, D-glutamate; Gly, glycine; Orn, ornithine; LL-Dpm, LL-diaminopimelic acid.

^b D, different reactions in different species.

<i>P. niger</i>	ACTGAGACACGGCCCAAACCTCTACGGGAGGCAGCAGTGGGGAATCTTCCGCAATGGGCG	395
<i>St. sanguis</i>	ACTGAGACACGGCCCAAGCTCTACGGGAGGCAGCAGTGGGGAATCTTCCGCAATGGGCG	379
<i>E. coli</i>	ACTGAGACACGGTCCAGACTCTACGGGAGGCAGCAGTGGGGAATATTGCACAATGGGCG	372
<i>Ps. anaerobius</i>	ACTGAGACACGGTCCAAACTC-TACGGGAGGCAGCAGTGGGGAATGTTGCACAATGGGCG	373
<i>R. gnavus</i>	ACTGAGACACGGCCCAAACCTCTACGGGAGGCAGCAGTGGGGAATATTGCACAATGGGCG	388
<i>Ps. asaccharo.</i>	ACTGAGAAACGGTCCAAACTCTACGGGAGGCAGCAGTGGGGAATATTGCACAATGGGCG	385
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<i>P. niger</i>	CAAGCCTGACGGAGCAATGCCGCGTGAGTGAAGAAGGCCTTCGGGTTGTAAACTCTGTCT	455
<i>St. sanguis</i>	CAACCTTGACCGAGCAACGCCGCGTGAGTGAAGAAGGTTTTCGGATCGTAAAGCTCTTTT	439
<i>E. coli</i>	CAAGCCTGATGCAGCCATGCCGCGTGATGAAGAAGGCCTTCGGGTTGTAAAGTACTTTC	432
<i>Ps. anaerobius</i>	CAAGCCTGATGCAGCAACGCCGCGTGAACGATGAAGGCTTTCGGATCGTAAAGTCTGTT	433
<i>R. gnavus</i>	AAACCTTGATGCAGCGACGCCGCGTGAGCGAAGAAGTATTTCGGTATGTAAAGCTCTATC	448
<i>Ps. asaccharo.</i>	AAACCTTGATGCATCGACGCCGCGTGAGCGGAGAAGGTTTTCGAGTCGTAAAGCTCTGTCT	445
** *		
<i>P. niger</i>	CTCATCCAAGAG-AGGGG--AAGGTAGTAACCTGACCTTTCC--AGGACGGTAGATGAGGA	510
<i>St. sanguis</i>	GTAAGAGAAGAACGGGTGTGAGAGTGGAAGT--TCACACT--GTGACGGTACCTTACCA	495
<i>E. coli</i>	AGCGGGGAGGAA-GGGAGTAAAGTTAATACCT--TTGCTCA--TTGACGTTACCCGCAGA	487
<i>Ps. anaerobius</i>	GCAGGGGAAGA-TA-----ATGACGGTACCTGTGA	463
<i>R. gnavus</i>	ATCAGGGGAAGA-A-----ATGACGGTACCTGACTA	478
<i>Ps. asaccharo.</i>	CTATGGGAAGA-TA-----ATGACGGTACCATACGA	475
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<i>P. niger</i>	GGAAGCCCCGGCTAACT-ATGTGCCAGCAGCCGCGGTAAACATAGG-GGGCAAGCGTTG	568
<i>St. sanguis</i>	GAAAGGGACGGCTAACT-ACGTGCCAGCAGCCGCGGTAAACATAGG-TCCCAAGCGTTG	553
<i>E. coli</i>	AGAAGCACCGGCTAACT-CCGTGCCAGCAGCCGCGGTAAACATAGG-GTGCAAGCGTTA	545
<i>Ps. anaerobius</i>	GGAAGCCCCGGCTAACT-ACGTGCCAGCAGCCGCGGTAAACATAGGAGGGGCTAGCGTTA	522
<i>R. gnavus</i>	AGAAGCACCGGCTAAAT-ACGTGCCAGCAGCCGCG-CACTGCGGTTG-GTGCAAGCGTTA	535
<i>Ps. asaccharo.</i>	GGAAGCCCCGGCTGAATAACGTGCCAGCAGCCGCGGTAAACATAGG-GGGCGAGCGTTG	534
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<i>P. niger</i>	TCCGGAATCACTGGGCGTAAAGGGCGCGCAG-GCGGTCTGTAAAGTCAGATGTGAAAGGT	627
<i>St. sanguis</i>	TCCGGATTTATGGGCGTAAAGCGAGCGCAG-GCGGTAGATAAGTCTGAAGTTAAAGGC	612
<i>E. coli</i>	ATCGGAATTACTGGGCGTAAAGCGCACGAG-GCGGTTGTAAAGTCAGATGTGAAATCC	604
<i>Ps. anaerobius</i>	TCCGGATTTACTGGGCGTAAAGGGTGCGTAG-GTGGTCTTCAAGTCGGTGGTTAAAGGC	581
<i>R. gnavus</i>	TCCGGATTTACTGGGCGTAAAGGGAGCGTAGCAGCGATGGGCAAGTCTGATGTGAAAACC	595
<i>Ps. asaccharo.</i>	TCCGGGATCACTGGGCGTAAAGGGTTCCGAG-GCGGTTAGAAAGTCAGATGTGAAATGC	593
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FIG. 1. Sequences of an approximately 25-base deletion found between positions 445 and 470: alignment between position 312 and position 604 of the 16S rRNA gene. The sequences of other peptostreptococci listed in Table 1 had the same deletion in the same position. *P. niger*, *Peptococcus niger* ATCC 27731^T; *St. sanguis*, *Streptococcus sanguis* ATCC10556^T; *E. coli*, *E. coli* ATCC 11775^T; *Ps. anaerobius*, *Peptostreptococcus anaerobius* ATCC 27337^T; *R. gnavus*, *R. gnavus* ATCC 29147^T; *Ps. asaccharo.*, *Peptostreptococcus asaccharolyticus* ATCC 14963^T.

FIG. 2. DNA sequence similarity matrix for gram-positive low-G+C-content organisms: results of a comparison of the sequences between positions 8 and 1392. Values greater than 84% are shaded. *Ps.*, *Peptostreptococcus*.

FIG. 3. Phylogenetic positions of anaerobic cocci in the division *Eubacteria*. Distances were calculated by the NJ method. *Ps.*, *Peptostreptococcus*. Shading indicates gram-positive organisms.

peptostreptococcus productus and *Streptococcus hansenii* from other ruminococci are shown in Table 3.

DISCUSSION

Members of the family *Peptococcaceae* did not cluster in a single group (Fig. 2 through 4). *Peptococcus niger*, the type species of the family, was not related to other genera in the family. *Peptostreptococcus anaerobius*, the type species of the genus *Peptostreptococcus*, was not related to the other members of the genus; it was more closely related to *Clostridium sordellii* (Fig. 2).

When a similarity value greater than 84% was used, the anaerobic cocci were separated into the following four groups; *Peptococcus niger*, the ruminococcus-coprococcus group, the *Sarcina*-*Peptostreptococcus anaerobius* group, and the *peptostreptococcus* group (Fig. 2).

To compare levels of genetic relatedness among previously established groups of low-G+C-content organisms, *Listeria monocytogenes*, *Bacillus thuringiensis*, *Lactococcus lactis*, *Staphylococcus aureus*, and *Streptococcus sanguis* were included in the same data set (Fig. 2). These aerobic gram-positive organisms clustered in a single group at similarity values greater than 84% (Fig. 2), although they belonged to different families.

The data described above indicate that five currently

recognized genera of the family *Peptococcaceae*, including the genera *Peptostreptococcus*, *Sarcina*, *Ruminococcus*, and *Coprococcus*, should be removed from this family. At the moment, we do not propose new families for these taxa, because there is no commonly accepted concept of family. However, the data clearly indicate that members of the family *Peptococcaceae* should be divided into different phylogenetic groups in the future.

Members of the genus *Peptostreptococcus* did not cluster with the type species of the genus (Fig. 2 through 5). Previously (3), we proposed that most members of the genus *Peptococcus* should be transferred to the genus *Peptostreptococcus* simply because their G+C contents were more similar to the G+C content of the type species of the genus *Peptostreptococcus*, *Peptostreptococcus anaerobius*. The RNA sequence data revealed that the transferred members of the genus *Peptostreptococcus* are not related to the type species of the genus *Peptostreptococcus*, *Peptostreptococcus anaerobius*. We recently completed a sequence analysis of all of the members of the genus *Peptostreptococcus*, including three new members (9), and determined the new taxonomic positions of these organisms (unpublished data).

Coprococci and ruminococci are isolated from humans and animal rumina. These organisms have similar chemotaxonomic traits. Their G+C contents range from 39 to 46 mol%. They require fermentable carbohydrates. Position 3

FIG. 4. Phylogenetic tree of gram-positive organisms having low DNA G+C contents in the division *Firmicutes*. Distances were calculated by the NJ method by using the data set shown in Fig. 2. *Ps.*, *Peptostreptococcus*. Shading indicates members of the family *Peptococcaceae*.

in their cell wall peptidoglycan contains *meso*-diaminopimelic acid (Table 2). The only difference between the two genera is their metabolic end products in peptone-yeast extract-glucose medium. Coprococci produce butyric acid, and ruminococci produce formic, acetic, propionic and often succinic acids (6). These data suggest that the members of the genera *Coprococcus* and *Ruminococcus* could be reclassified in a single genus. However, we have studied only three species belonging to the two genera. The levels of homology among the five species studied ranged from 84 to 89%. These values are not high enough to conclude that these organisms belong to a single genus. A new proposal should be made after all members of the genera *Coprococcus* and *Ruminococcus* are sequenced.

Peptostreptococcus productus and *Streptococcus hansenii* clustered with the ruminococci and coprococci and were placed in the ruminococcus-coprococcus group in Fig. 2. The phenotypic and chemotaxonomic traits of *Peptostreptococcus productus* resemble the traits of ruminococci and coprococci (Table 2). Members of the genus *Peptostreptococcus* are asaccharolytic. They use peptones as their major energy sources (5, 6, 12); however, *Peptostreptococcus productus* is not proteolytic but it is saccharolytic. Carbohydrates stimulate the growth of *Peptostreptococcus productus*. Also, the G+C content of *Peptostreptococcus productus* is similar to the G+C contents of ruminococci and coprococci.

Streptococcus hansenii ATCC 27752^T is strictly anaerobic, and its G+C content ranges from 39 to 42% mol%. It

ferments glucose. Carbohydrates stimulate the growth this organism, and the organism produces lactic acid as a major metabolic product in peptone-yeast extract medium (5).

Cell wall peptidoglycan types (16, 17) offer additional evidence. *Streptococcus hansenii*, *Peptostreptococcus productus*, coprococci, and ruminococci have similar structures. They all have *meso*-diaminopimelic acid in position 3 of the peptidoglycan (15). None of the other members of the genus *Peptostreptococcus* have this peptidoglycan type.

The genetic and phenotypic data indicate that *Peptostreptococcus productus* and *Streptococcus hansenii* should be classified together with the ruminococcus-coprococcus group. At the moment, the only difference between the genus *Coprococcus* and the genus *Ruminococcus* is the metabolic products obtained in peptone-yeast extract medium. Ruminococci do not produce butyric acid, but coprococci do produce this acid. *Streptococcus hansenii* and *Peptostreptococcus productus* do not produce butyric acid; this phenotypic difference between the two taxa may be a minor difference and should not be used to reclassify the two taxa in a single genus.

However, for the reasons described above, we propose that *Peptostreptococcus productus* and *Streptococcus hansenii* should temporarily be placed in the genus *Ruminococcus*. *Ruminococcus hansenii* comb. nov. has been described previously (14). This organism is differentiated from other members of the genus *Ruminococcus* by its metabolic products and by its carbohydrate fermentation pattern. Charac-

FIG. 5. Phylogenetic tree of gram-positive organisms having low DNA G+C contents in the division *Firmicutes*. Distances were calculated by the unweighted pair group (UPG) method by using the data set shown in Fig. 2. *Ps.*, *Peptostreptococcus*. Shading indicates members of the family *Peptococcaceae*.

teristics that differentiate *R. hansenii* comb. nov. from other ruminococci are shown in Table 3.

Description of *Ruminococcus productus* (Prévot 1941) comb. nov. Strictly anaerobic gram-positive cocci. Cells are often elliptical and occur singly, in pairs, and in short chains.

Surface colonies on blood agar are 0.5 to 1.0 mm in diameter, circular, entire, low convex, grayish white, and smooth. Growth is stimulated by fermentable carbohydrates and carbon monoxide. Cellobiose, arabinose, glucose, lactose, maltose, mannitol, mannose, melibiose, sorbitol, sucrose,

TABLE 3. Characteristics that differentiate *Ruminococcus* species from each other^a

Species	Cells having oval to pointed ends	Major fermentation product(s)	Hydrolysis of:				Fermentation of:					
			Cellulose	Starch	Arabinose	Cellobiose	Fructose	Lactose	Maltose	Mannitol	Raffinose	Sucrose
<i>Ruminococcus productus</i>	+	Acetate	—	—	+	+	+	+	+	D ^b	+	+
<i>Ruminococcus hansenii</i>	—	Lactate	ND ^c	—	ND	—	—	+	+	—	+	—
<i>Ruminococcus flavescens</i>	—	Succinate, lactate	D	—	D	D	—	D	—	—	—	—
<i>Ruminococcus albus</i>	—	Acetate, ethanol	D	—	D	D	D	D	—	—	—	D
<i>Ruminococcus bromii</i>	—	Ethanol	—	+	—	—	D	—	+	—	—	—
<i>Ruminococcus callidus</i>	+	Succinate	ND	—	—	+	—	D	+	—	+	+
<i>Ruminococcus torques</i>	+	Lactate, ethanol	—	—	—	—	+	+	—	—	—	—
<i>Ruminococcus gnavus</i>	+	Ethanol	—	D	+	—	+	D	+	—	+	D
<i>Ruminococcus lactaris</i>	+	Lactate	—	—	—	—	+	+	D	+	—	—
<i>Ruminococcus obeum</i>	+	Ethanol	—	—	+	—	+	+	+	D	+	+

^a Most of the data were obtained from references 5 and 14.

^b D, different reactions in different strains.

^c ND, no data available.

and xylose are fermented. The major metabolic products in peptone-yeast extract-glucose medium are acetate and formate; occasionally trace amounts of succinate, lactate, and ethanol are produced. Amino acids and peptones are not major energy sources. Nitrate reduction, catalase, and indole production negative. Gas production in glucose deep agar varies from nonexistent to abundant. β -Galactosidase and α -glucosidase are produced, but alkaline phosphatase and β -glucuronidase are not produced. Strains are isolated from human feces. This organism is one of the dominant members of the human intestinal flora. Occasionally isolated from human blood cultures. Characteristics that differentiate *R. productus* from other ruminococci are shown in Table 3.

The type strain is ATCC 27340. The G+C contents of the DNAs of five strains range from 43 to 45 mol%. The peptidoglycan in position 3 in *meso*-diaminopimelic acid.

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REFERENCES

1. Ezaki, T., H. Oyaizu, and E. Yabuuchi. 1991. The anaerobic gram-positive cocci, p. 1879–1892. In A. Balows et al. (ed.), *The prokaryotes*, 2nd ed. Springer-Verlag, New York.
2. Ezaki, T., E. Yabuuchi, K. Kaneda, and I. Yano. 1985. Extractable cellular lipids of Gram-positive anaerobic cocci. *Proc. Jpn. Assoc. Anaerob. Infect. Res.* 15:115–117.
3. Ezaki, T., H. Yamamoto, K. Ninomiya, S. Suzuki, and E. Yabuuchi. 1983. Transfer of *Peptococcus indolicus*, *Peptococcus asaccharolyticus*, *Peptococcus prevotii*, and *Peptococcus magnus* to the genus *Peptostreptococcus* and proposal of *Peptostreptococcus tetradius* sp. nov. *Int. J. Syst. Bacteriol.* 33: 683–698.
4. Gibbons, N. E., and R. G. E. Murray. 1978. Proposals concerning the higher taxa of bacteria. *Int. J. Syst. Bacteriol.* 28:1–6.
5. Holdeman, L. V., E. P. Cato, and W. E. C. Moore. 1977. *Anaerobe laboratory manual*, 4th ed. Virginia Polytechnic Institute and State University, Blacksburg.
6. Holdeman, L. V., and W. E. C. Moore. 1974. New genus, *Coproccoccus*, twelve new species, and emended description of four previously described species of bacteria from human feces. *Int. J. Syst. Bacteriol.* 24:260–277.
7. Huss, V. A. R., H. Festl, and K. H. Schleifer. 1984. Nucleic acid hybridization studies and deoxyribonucleic acid base composition of anaerobic gram-positive cocci. *Int. J. Syst. Bacteriol.* 34:95–101.
8. Lane, D. J., B. Pace, G. J. Olsen, D. A. Stahl, M. L. Sogin, and N. R. Pace. 1985. Rapid determination of 16S ribosomal RNA sequences for phylogenetic analysis. *Proc. Natl. Acad. Sci. USA* 82:6955–6959.
9. Li, N., Y. Hashimoto, S. Adnan, H. Miura, H. Yamamoto, and T. Ezaki. 1992. Three new species of the genus *Peptostreptococcus* isolated from humans: *Peptostreptococcus vaginalis* sp. nov., *Peptostreptococcus lacrimalis* sp. nov., and *Peptostreptococcus lactolyticus* sp. nov. *Int. J. Syst. Bacteriol.* 42:602–605.
10. Ludwig, W., M. Weiznegger, S. Dorn, J. Andreesen, and K. H. Schleifer. 1990. The phylogenetic position of *Peptococcus niger* based on 16S rRNA sequence studies. *FEMS Microbiol. Lett.* 71:139–144.
11. Paster, B. J., J. B. Russell, C. M. J. Yang, J. M. Chow, C. R. Woese, and R. Tanner. 1993. Phylogeny of the ammonia-producing ruminal bacteria *Peptostreptococcus anaerobius*, *Clostridium sticklandii*, and *Clostridium aminophilum* sp. nov. *Int. J. Syst. Bacteriol.* 43:107–110.
12. Rogosa, M. 1974. *Peptococcaceae*, p. 517. In R. E. Buchanan and N. E. Gibbons (ed.), *Bergey's manual of determinative bacteriology*, 8th ed. The Williams & Wilkins Co., Baltimore.
13. Saitou, N., and M. Nei. 1987. The neighbor-joining method: a new method for reconstructing phylogenetic trees. *Mol. Biol. Evol.* 4:406–425.
14. Schleifer, K. H. 1986. Section 12. Gram-positive cocci, p. 999–1103. In P. H. A. Sneath, N. S. Mair, M. E. Sharpe, and J. G. Holt (ed.), *Bergey's manual of systematic bacteriology*, vol. 2. The Williams & Wilkins Co., Baltimore.
15. Schleifer, K. H., and O. Kandler. 1972. Peptidoglycan types of bacterial cell walls and their taxonomic implications. *Bacteriol. Rev.* 36:245–258.
16. Schleifer, K. H., and E. Nimmerman. 1973. Peptidoglycan types of strains of the genus *Peptococcus*. *Arch. Microbiol.* 93:245–258.
17. Weiss, N. 1981. Cell wall structure of anaerobic cocci. *Rev. Institut. Pasteur Lyon* 14:53–59.
18. Woese, C. R. 1987. Bacterial evolution. *Microbiol. Rev.* 51:221–271.
19. Woese, C. R. 1991. Towards a natural system of organisms: proposal for the domains *Archaea*, *Bacteria*, and *Eucarya*. *Proc. Natl. Acad. Sci. USA* 87:4576–4579.
20. Yabuuchi, E., Y. Kosako, H. Oyaizu, I. Yano, H. Hotta, Y. Hashimoto, T. Ezaki, and M. Arakawa. 1992. Proposal of *Burkholderia* gen. nov. and transfer of seven species of the genus *Pseudomonas* homology group II to the new genus, with the type species *Burkholderia cepacia* (Palleroni and Holmes 1981) comb. nov. *Microbiol. Immunol.* 36:1251–1275.