Emendation of the Genus *Cytophaga* and Transfer of *Cytophaga agarovorans* and *Cytophaga salmonicolor* to *Marinilabilia* gen. nov.: Phylogenetic Analysis of the *Flavobacterium*-*Cytophaga* Complex

YASUYOSHI NAKAGAWA* AND KAZUhide YAMASATO†

*Institute of Applied Microbiology, The University of Tokyo,
Bunkyo-ku, Tokyo 113, Japan‡*

A 16S rRNA sequence analysis revealed that the genera *Cytophaga*, *Flavobacterium*, and *Flexibacter* are all polyphyletic and should be redefined and reorganized. *Cytophaga hutchinsonii*, the type species of the genus *Cytophaga*, belongs to a lineage that also contains *Cytophaga aurantiaca*. The genus *Cytophaga* is emended so that it contains only these two species, which decompose distinctly crystalline cellulose. *Cytophaga salmonicolor* and *Cytophaga agarovorans* form a lineage which is intermediate between other aerobic species and anaerobic bacteroides. Phenotypically, these organisms are characterized by being facultative anaerobes, inhabiting marine environments, and containing menaquinone-7 and spermidine. We propose that *C. salmonicolor* and *C. agarovorans* should be transferred to the genus *Marinilabilia* gen. nov. as *Marinilabilia salmonicolor* comb. nov. and *Marinilabilia agarovorans* comb. nov., respectively.

The genus *Cytophaga* was first described for aerobic cellulose-degrading bacteria by Winogradsky (31). Subsequently, this taxon was expanded to include organisms which degrade other biomacromolecules, such as agar and/or chitin (17, 23–26). However, several taxonomic investigations (3, 4, 8, 14, 21) revealed unreasonable overlapping of phenotypic and chemotaxonomic characteristics between members of the genera *Cytophaga* and *Flavobacterium*, which formed a taxon that came to be called the *Flavobacterium*-*Cytophaga* complex. The genera *Cytophaga* and *Flavobacterium*, along with the genera *Bacteroides* and *Flexibacter* and other genera, constitute one phylum of bacteria (33) as determined by cataloging (16) and sequencing (31) of 16S rRNA. Intra- and intergeneric relationships between members of the *Flavobacterium*-*Cytophaga* complex have gradually been elucidated by studying 16S rRNA sequences (6, 12, 13, 29, 34) and DNA-rRNA hybridization data (1, 19, 20). These studies have revealed that phylogenetically the genera *Cytophaga* and *Flavobacterium* could be divided into several groups, some of which included both *Cytophaga* and *Flavobacterium* species and some of which were elevated to genus rank. Vandamme et al. (28) proposed the genus *Chryseobacterium* for the *Flavobacterium gleum* group, the genus *Bergeyella* for *Weeksella zoohelcum*, and the genus *Empedobacter* for *Flavobacterium breve*. However, many misclassified cytophagas and flavobacteria remain. In this paper, the basis of the results of a study on comparative analysis of 16S rRNA sequences and a phenotypic examination of 62 strains belonging to the *Flavobacterium*-*Cytophaga* complex and related genera, we propose that the genus *Cytophaga* should be emended and that *Cytophaga salmonicolor* and *Cytophaga agarovorans* should be transferred to the genus *Marinilabilia* gen. nov. as *Marinilabilia salmonicolor* comb. nov. (the

type species) and *Marinilabilia agarovorans* comb. nov., respectively.

Most of the strains which we used are type strains; the only exceptions were *Cytophaga* sp. strain L-43-1-N and *Flexibacter maritimus* JCM 8137. *Cytophaga* sp. strain L-43-1-N and *Flexibacter maritimus* No. 449^T (T = type strain) were provided by H. Kojima (Japan Leather Research Institute, Tokyo, Japan) and H. Wakabayashi (The University of Tokyo, Tokyo, Japan), respectively. The other strains which we used were obtained from culture collections. The procedures used to determine the quinone systems and the sequences of 16S rRNAs have been described previously (12). Our sequencing experiments generated a continuous stretch of sequence from position 100 to position 1375 (*Escherichia coli* numbering system). The 16S rRNA sequences which we determined and other sequences were compared as follows. Sequences determined in this study and previous studies (12, 13) and obtained from previously published papers (6, 15, 29, 31, 34) were aligned with the sequence of *Cytophaga hutchinsonii* DSM 1761, the type strain of the type species of the genus *Cytophaga*. *Escherichia coli* (2) was included as an outgroup. The software package Clustal V (9) was used to generate similarity values and evolutionary distances (K_{nuc} values) (11) and to reconstruct a phylogenetic tree by the neighbor-joining method (18) from the K_{nuc} values. Positions at which the secondary structures varied in the strains (positions 179 to 220, 447 to 487, 837 to 849, 991 to 1045, and 1134 to 1140) were not included in the analysis. The total number of nucleotides compared was 835 after we eliminated all sites which were not determined in any sequence. The topology of the tree was evaluated by the bootstrap resampling method (5) with 1,000 replicates. If the confidence limit for a group as determined by the bootstrap resampling method was more than 95%, the group was considered statistically significant (5).

The respiratory quinones identified in this study were menaquinone 6 (MK-6) and MK-7; MK-6 was found in *Bergeyella zoohelcum* NCTC 11660^T, *Flavobacterium branchiophilum* No. 270^T, and *Flexibacter maritimus* No. 449^T, and MK-7 was found in *Flexibacter elegans* IFO 15055^T, *Flexibacter filiformis* IFO 15056^T, and *Flavobacterium yabuuchiae* IFO 14975^T. The iso-

* Corresponding author. Present address: Institute for Fermentation, Osaka, 17-85 Juso-honmachi 2-chome, Yodogawa-ku, Osaka 532, Japan. Phone: 81-6-300-6555. Fax: 81-6-300-6814.

† Present address: Culture Collection Center, Tokyo University of Agriculture, 1-1, Sakuragaoka 1-chome, Setagaya-ku, Tokyo 156, Japan.

‡ The present name of the former Institute of Applied Microbiology is Institute of Molecular and Cellular Biosciences.

TABLE 1. Characteristics of the genus *Marinilabilia* and allied aerobic and facultatively anaerobic genera^a

Genus	Habitat(s)	Oxygen requirement ^b	Pigmentation	G+C content (mol%)	Polyamine ^c	Quinone(s)
<i>Marinilabilia</i>	Marine	F	+	37-41	Spd	MK-7
<i>Bergeyella</i>	Parasitic, saprophytic	A	—	35-37	ND ^d	MK-6
<i>Capnocytophaga</i>	Parasitic	F	+	33-41	ND	MK-6
<i>Chryseobacterium</i>	Parasitic, free living	A	+	33-38	Hspd	MK-6
<i>Cytophaga</i>	Soil	A	+	39-42	Hspd	MK-7
<i>Empedobacter</i>	Parasitic, free living	A	+	31-33	Hspd	MK-6
<i>Flavobacterium</i>	Soil, freshwater	A	+	32	Hspd	MK-6
<i>Flexibacter</i>	Soil, freshwater	A	+	29-48	Hspd	MK-6, MK-7
<i>Ornithobacterium</i>	Parasitic	F	—	37-39	ND	MK-7
<i>Microscilla</i>	Marine	A	+	32-44	ND	ND
<i>Riemerella</i>	Parasitic	A	—	29-35	ND	MK-7
<i>Sphingobacterium</i>	Parasitic, free living	A	+	39-42	Hspd	MK-7
<i>Weeksella</i>	Parasitic, saprophytic	A	—	35-38	ND	MK-6

^a Data from this study and references 7, 10, 12 to 14, 19, 22, 27 to 29, and 35.^b F, facultative anaerobes; A, strict aerobes.^c Spd, spermidine; Hspd, homospermidine.^d ND, not determined.

prenoid quinones of the members of the *Flavobacterium-Cytophaga* complex and related genera which are characterized by containing MK-6 and/or MK-7 are summarized in Table 1. The strains of *Bacteroides* species are characterized by containing MK-6, MK-7, MK-9, MK-10, MK-11, MK-12, or MK-13, and *Porphyromonas* strains are characterized by containing MK-9 or MK-10 (35). The major isoprenoid quinones of *Bacteroides splanchnicus* and *Rikenella microfus* are MK-9 and MK-8, respectively.

The reconstructed phylogenetic tree revealed the extreme heterogeneity of this group of bacteria (Fig. 1). The genera *Cytophaga*, *Flavobacterium*, and *Flexibacter* all are polyphyletic and should be redefined and divided into several genera based on phylogenetic relationships. The species assigned to each of these genera belong to several lineages which are only remotely related to each other at the genus level. Once discriminating phenotypic characteristics have been identified, these phylogenetic lineages should be reclassified in new or previously described genera.

The only species that was closely related to *Cytophaga hutchinsonii*, the type species of the genus *Cytophaga*, was *Cytophaga aurantiaca* (Fig. 1). Our bootstrap analysis resulted in a confidence value of 100% for the group containing these two species. The level of 16S rRNA sequence similarity for these two species was 98.9%, and the levels of 16S rRNA sequence similarity between them and other members of the complex were less than 87.3% (Table 2). In addition to constituting an independent phylogenetic lineage, these two species have similar phenotypic characteristics; they both contain MK-7, inhabit terrestrial environments, and are able to degrade crystalline cellulose, which is the defining characteristic given in the original description of the genus (32). On the basis of the facts described above, the genus *Cytophaga* should be confined to these two species, which have the distinguishing phenotypic feature of the genus, the ability to degrade cellulose as Reichenbach (17) discerningly suggested.

Cytophaga salmonicolor, *Cytophaga agarovorans*, and *Cytophaga fermentans* are marine, facultatively anaerobic species that contain MK-7. They are also characterized by having spermidine as their major polyamine, in contrast to most other species belonging to the complex, which contain homospermidine (7). Phylogenetically, *Cytophaga salmonicolor* and *Cytophaga agarovorans* occupy an independent position in this group of bacteria (Fig. 1). They have exhibit a high level of 16S

rRNA sequence similarity with each other (98.9%), but lower levels of similarity with *Cytophaga fermentans* (around 92%) (Table 2). The grouping of *Cytophaga salmonicolor* and *Cytophaga agarovorans* is supported by a confidence value of 100%.

On the basis of phylogenetic, chemotaxonomic, and physiological data, we propose that the genus *Cytophaga* should be emended and that *Cytophaga salmonicolor* and *Cytophaga agarovorans* should be transferred to the genus *Marinilabilia* gen. nov. as *Marinilabilia salmonicolor* (Veldkamp 1961) comb. nov. and *Marinilabilia agarovorans* (Reichenbach 1989) (*Cytophaga salmonicolor* var. *agarovorans* Veldkamp 1961) comb. nov., respectively, in order to partially solve the taxonomic confusion surrounding this group of bacteria. Characteristics of the genus *Marinilabilia* and allied bacteria are shown in Table 1.

Emended description of the genus *Cytophaga* Winogradsky 1929, 577^{AL}, emend. Cells are short to moderately long flexible rods that are 0.3 to 0.5 µm wide and 2 to 10 µm long and have slightly tapered ends. Resting stages are absent. Motile by gliding. Gram negative. The cell mass is usually pigmented yellow to orange because of cell-bound carotenoids, flexirubin type pigments, or both. When covered with alkali (e.g., a 20% KOH solution [flexirubin reaction]), colonies may reversibly change color from yellow to purple- or red-brown. Oxidase positive and catalase negative. Strict aerobes. Chemoorganotrophs. Metabolism is respiratory. Organic acids may also be produced during growth. All strains decompose crystalline cellulose (filter paper). The optimum temperature is 20 to 30°C. The optimum pH is around 7. Common in soil. The major polyamine is homospermidine. The quinone is MK-7. The G+C content of the DNA is 39 to 42 mol%.

The type species is *Cytophaga hutchinsonii*. The other species in the genus is *Cytophaga aurantiaca*. These two species can be differentiated by the color of the cell mass, the flexirubin reaction, and the optimum temperature (17).

Description of *Marinilabilia* gen. nov. *Marinilabilia* (Ma.ri.ni.la'bi.li.a. L. adj. *marinus*, marine, pertaining to the sea; L. adj. *labilis*, gliding; M. L. fem. n. *Marinilabilia*, marine gliding organisms). The description of the genus *Marinilabilia* gen. nov. below is taken from the data of Reichenbach (17) and Veldkamp (30) and our own data. Cells are short to very long flexible rods with rounded or slightly tapered ends and are 0.3 to 0.5 µm wide and 2 to 50 µm long. Resting stages are absent. Motile by gliding. Gram negative. The cell mass is pale yellow to salmon (usually pink to salmon). Chemoorganotroph. Me-

FIG. 1. Phylogenetic tree derived from 16S rRNA sequences for the members of the *Flavobacterium*-*Cytophaga* complex and related genera. All of the sequences except the *Cytophaga* sp. strain L-43-1-N and *Flexibacter maritimus* JCM 8137 sequences are sequences of type strains. *Escherichia coli* was used as the root organism. Scale bar = 0.01 K_{nuc} in nucleotide sequences. The lengths of the vertical lines are not significant. The numbers on the branches are the confidence limits (expressed as percentages) estimated by a bootstrap analysis of 1,000 replicates. Confidence limits less than 60% are not shown. Abbreviations: *Es.*, *Escherichia*; *Cy.*, *Cytophaga*; *Fx.*, *Flexibacter*; *Sp.*, *Sphingobacterium*; *Fl.*, *Flavobacterium*; *Ma.*, *Marinilabilia*; *Ba.*, *Bacteroides*; *Po.*, *Porphyromonas*; *Ri.*, *Rikenella*; *We.*, *Weeksella*; *Em.*, *Empedobacter*; *Ch.*, *Chryseobacterium*; *Be.*, *Bergeyella*; *Or.*, *Ornithobacterium*; *Ca.*, *Capnocytophaga*.

tabolism is respiratory and fermentative. Catalase positive. All strains decompose several kinds of biomacromolecules. Marine organisms requiring elevated salt concentrations. The optimum temperature is 28 to 37°C. The optimum pH is around 7. The major polyamine is spermidine. The quinone is MK-7. The G+C content of the DNA is 37 to 41 mol%. The type

species of the genus is *Marinilabilia salmonicolor*. The other species which belongs to the genus is *Marinilabilia agarovorans*.

The descriptions of *Marinilabilia salmonicolor* comb. nov. and *Marinilabilia agarovorans* comb. nov. are the same as those given by Reichenbach (17). These two species can be differentiated on the basis of the ability to degrade agar.

TABLE 2. 16S rRNA sequence similarity matrix for members of the *Flavobacterium-Cytophaga* complex and related genera

Species	% Similarity																													
	<i>Cytophaga diffluens</i>	<i>Flexibacter aggregans</i>	<i>Cytophaga aprica</i>	<i>Flexibacter flexilis</i>	<i>Cytophaga hutchinsonii</i>	<i>Cytophaga aurantiaca</i>	<i>Flexibacter elegans</i>	<i>Flexibacter canadensis</i>	<i>Sphingobacterium spiritivorum</i>	<i>Cytophaga arvensicola</i>	<i>Marinilabilia agarovorans</i>	<i>Marinilabilia salmonicolor</i>	<i>Bacteroides splanchnicus</i>	<i>Porphyromonas asaccharolyticus</i>	<i>Bacteroides fragilis</i>	<i>Rikenella microfusis</i>	<i>Cytophaga fermentans</i>	<i>Weeksella virosa</i>	<i>Empedobacter brevis</i>	<i>Chryseobacterium meningosepticum</i>	<i>Chryseobacterium gleum</i>	<i>Bergeyella zoohelcum</i>	<i>Ornithobacterium rhinotracheale</i>	<i>Capnocytophaga ochracea</i>	<i>Flexibacter maritimus</i>	<i>Cytophaga uliginosa</i>	<i>Cytophaga latercula</i>	<i>Flavobacterium odoratum</i>	<i>Flavobacterium aquatile</i>	
<i>Flexibacter aggregans</i>	89.8																													
<i>Cytophaga aprica</i>	86.2	84.7																												
<i>Flexibacter flexilis</i>	86.8	84.9	83.0																											
<i>Cytophaga hutchinsonii</i>	86.7	85.6	84.1	87.3																										
<i>Cytophaga aurantiaca</i>	86.4	85.3	83.7	86.8	98.9																									
<i>Flexibacter elegans</i>	87.7	87.3	83.7	86.7	85.9	85.6																								
<i>Flexibacter canadensis</i>	87.2	88.1	82.5	86.5	87.3	86.8	86.1																							
<i>Sphingobacterium spiritivorum</i>	86.7	85.4	84.6	84.6	85.0	84.9	86.2	91.1																						
<i>Cytophaga arvensicola</i>	85.0	85.4	81.9	83.2	83.6	83.6	85.3	86.7	85.0																					
<i>Marinilabilia agarovorans</i>	87.5	84.6	83.4	84.9	85.4	85.4	85.8	87.7	87.1	84.9																				
<i>Marinilabilia salmonicolor</i>	88.1	85.0	84.0	85.4	85.9	86.1	88.1	87.2	85.2	98.9	98.9																			
<i>Bacteroides splanchnicus</i>	85.6	84.7	80.8	82.8	82.6	82.2	83.0	84.9	84.2	82.5	88.7	89.2	83.4																	
<i>Porphyromonas asaccharolyticus</i>	81.8	79.3	81.2	80.4	80.7	80.8	83.2	83.2	81.7	81.6	85.8	86.1	83.4	86.6																
<i>Bacteroides fragilis</i>	85.4	83.6	80.7	81.6	83.4	84.3	84.6	86.6	86.5	82.0	89.3	89.8	84.6	83.7	85.6															
<i>Rikenella microfusis</i>	85.2	83.5	82.9	83.8	84.4	84.3	84.6	86.6	86.5	82.0	89.3	89.8	84.6	83.7	85.6	85.6														
<i>Cytophaga fermentans</i>	88.1	85.4	84.0	84.4	85.4	85.2	85.9	88.0	87.3	86.0	91.7	92.3	87.4	85.0	87.3	89.9	87.7													
<i>Weeksella virosa</i>	84.0	83.5	81.0	82.5	84.0	83.4	84.2	84.4	83.6	84.2	84.3	84.6	82.9	82.0	82.4	84.2	84.2	86.8	92.7											
<i>Empedobacter brevis</i>	85.0	85.2	81.6	83.7	83.8	83.2	83.5	85.5	84.0	83.6	85.5	85.3	82.5	81.6	82.2	84.2	86.8	92.7	91.3											
<i>Chryseobacterium meningosepticum</i>	86.4	84.7	81.4	84.6	84.9	84.3	85.2	86.8	86.0	84.0	87.0	86.8	84.3	82.3	84.0	84.7	87.5	89.2	91.3	95.0										
<i>Chryseobacterium gleum</i>	86.4	84.0	81.2	84.4	84.0	83.5	86.7	84.9	85.4	84.6	86.7	86.7	84.2	82.0	83.4	85.5	87.0	89.2	90.2	95.0	96.3									
<i>Bergeyella zoohelcum</i>	85.3	84.6	81.0	84.6	83.6	83.5	86.5	85.2	85.4	82.2	86.7	86.7	83.5	82.2	83.5	86.1	87.0	89.3	89.7	93.5	96.3	90.7								
<i>Ornithobacterium rhinotracheale</i>	85.8	84.9	82.6	83.8	84.4	84.1	84.7	85.9	84.8	83.7	87.0	87.1	84.6	82.2	81.4	85.9	88.5	90.8	91.9	90.8	90.8	90.7	88.3							
<i>Capnocytophaga ochracea</i>	83.7	82.9	80.8	81.0	82.9	82.4	82.8	84.3	82.5	82.2	85.2	85.2	83.7	83.4	81.9	83.4	87.7	88.3	89.0	86.7	86.1	85.8	89.0	89.0						
<i>Flexibacter maritimus</i>	84.6	83.5	82.8	80.7	82.0	81.4	83.1	84.6	82.3	82.5	83.8	83.8	83.1	82.0	81.6	83.8	87.0	86.2	87.1	87.1	86.4	85.6	86.7	89.0	90.4					
<i>Cytophaga uliginosa</i>	84.2	85.8	81.9	81.6	82.0	81.9	83.8	85.3	83.7	83.5	84.9	84.8	82.9	82.8	83.8	84.2	87.5	87.4	88.3	86.5	86.2	87.0	87.8	90.1	90.4					
<i>Cytophaga latercula</i>	82.8	82.8	81.9	80.5	82.6	82.3	81.1	85.2	83.7	83.5	85.2	85.2	83.0	81.3	82.8	84.4	87.8	87.2	86.8	86.2	86.7	85.6	87.1	90.4	90.8	90.2				
<i>Flavobacterium odoratum</i>	84.7	84.1	83.5	82.9	83.8	83.2	83.6	85.0	84.3	83.7	86.6	86.7	83.4	82.4	82.8	84.9	89.1	89.0	88.9	87.3	87.1	86.1	88.9	89.7	90.3	90.7				
<i>Flavobacterium aquatile</i>	83.7	83.8	81.3	81.7	83.1	82.4	84.0	84.4	82.8	83.1	84.8	84.7	83.4	82.3	82.9	84.1	89.1	89.6	89.0	87.1	87.5	87.1	89.9	91.3	91.3	91.4	91.5	93.3		
<i>Escherichia coli</i>	80.1	80.0	78.1	78.0	78.9	79.0	79.2	79.0	76.9	78.1	77.6	78.1	76.5	74.4	76.1	77.6	77.6	77.7	78.3	77.4	77.3	77.1	76.4	75.6	76.4	76.5	76.9	76.5	76.8	

Nucleotide sequence accession numbers. The 16S rRNA sequence data which we determined have been deposited in the DDBJ, EMBL, GSDB, and NCBI nucleotide databases under the following accession numbers: D14016 for *Chryseobacterium balustinum* IFO 15053^T, D14018 for *Chryseobacterium meningosepticum* IFO 12535^T, D14022 for *Empedobacter brevis* IAM 14197^T, D14017 for *Flavobacterium branchiophilum* No. 270^T, D14019 for *Flavobacterium odoratum* JCM 7458^T, D14021 for *Flavobacterium yabuuchiae* IFO 14975^T, (*Flavobacterium yabuuchiae* is now recognized as a junior subjective synonym of *Sphingobacterium spiritivorum* [27]), D14023 for *Flexibacter maritimus* No. 449^T, D14026 for *Sphingobacterium spiritivorum* JCM 1277^T, D14024 for *Sphingobacterium mizutae* IFO 14946^T, D14025 for *Sphingobacterium multivorum* IFO 14947^T, and D14020 for *Sphingobacterium thalpophilum* IFO 14963^T.

We thank the American Type Culture Collection (Rockville, Md.), the Deutsche Sammlung von Mikroorganismen und Zellkulturen GmbH (Braunschweig, Germany), the Institute of Applied Microbiology, the University of Tokyo (Tokyo, Japan), the Institute for Fermentation, Osaka (Osaka, Japan), the Japan Collection of Microorganisms (Wako, Japan), the National Collection of Industrial and Marine Bacteria, Ltd. (Aberdeen, United Kingdom), and the National Collection of Type Cultures, Public Health Laboratory Service (London, United Kingdom) for kindly supplying bacterial cultures. We are grateful to T. Sakane (Institute for Fermentation, Osaka) for offering every convenience and A. Nakagiri (Institute for Fermentation, Osaka) for suggesting the name *Marinilabilia*.

This work was supported in part by Grant-in-Aid for Co-operative Research 03304017 from the Ministry of Education, Science and Culture of Japan.

REFERENCES

- Bauwens, M., and J. De Ley. 1981. Improvements in the taxonomy of *Flavobacterium* by DNA:rRNA hybridizations, p. 27–31. In H. Reichenbach and O. B. Weeks (ed.), *The Flavobacterium-Cytophaga* group. Verlag Chemie, Weinheim, Germany.
- Brosius, J., M. L. Palmer, P. J. Kennedy, and H. F. Noller. 1978. Complete nucleotide sequence of a 16S ribosomal RNA gene from *Escherichia coli*. *Proc. Natl. Acad. Sci. USA* **75**:4801–4805.
- Callies, E., and W. Mannheim. 1978. Classification of the *Flavobacterium-Cytophaga* complex on the basis of respiratory quinones and fumarate respiration. *Int. J. Syst. Bacteriol.* **28**:14–19.
- Christensen, P. J. 1977. The history, biology, and taxonomy of the *Cytophaga* group. *Can. J. Microbiol.* **23**:1599–1653.
- Felsenstein, J. 1985. Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* **39**:783–791.
- Gherna, R., and C. R. Woese. 1992. A partial phylogenetic analysis of the "flavobacter-bacteroides" phylum: basis for taxonomic restructuring. *Syst. Appl. Microbiol.* **15**:513–521.
- Hamana, K., Y. Nakagawa, and K. Yamasato. 1995. Chemotaxonomic significance of polyamine distribution patterns in the *Flavobacterium-Cytophaga* complex and the related genera. *Microbios* **81**:135–145.
- Hayes, P. R. 1977. A taxonomic study of flavobacteria and related gram negative yellow pigmented rods. *J. Appl. Bacteriol.* **43**:345–367.
- Higgins, D. G., A. J. Bleasby, and R. Fuchs. 1992. CLUSTAL V: improved software for multiple sequence alignment. *Comput. Appl. Biosci.* **8**:189–191.
- Holt, J. G., N. R. Krieg, P. H. A. Sneath, J. T. Staley, and S. T. Williams (ed.). 1994. *Bergey's manual of determinative bacteriology*, 9th ed. Williams & Wilkins Co., Baltimore.
- Kimura, M. 1980. A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. *J. Mol. Evol.* **16**:111–120.
- Nakagawa, Y., and K. Yamasato. 1993. Phylogenetic diversity of the genus *Cytophaga* revealed by 16S rRNA sequencing and menaquinone analysis. *J. Gen. Microbiol.* **139**:1155–1161.
- Nakagawa, Y., and K. Yamasato. 1993. The molecular systematics of *Cytophaga* species based on the 16S rRNA sequences, p. 163–170. In P. J. Jooste (ed.), *Advances in the taxonomy and significance of Flavobacterium, Cytophaga and related bacteria*. University Press, University of the Orange Free State, Bloemfontein, South Africa.
- Oyaizu, H., and K. Komagata. 1981. Chemotaxonomic and phenotypic characterization of the strains of species in the *Flavobacterium-Cytophaga* complex. *J. Gen. Appl. Microbiol.* **27**:57–107.
- Paster, B. J., F. E. Dewhirst, I. Olsen, and G. J. Fraser. 1994. Phylogeny of *Bacteroides*, *Prevotella*, and *Porphyromonas* spp. and related genera. *J. Bacteriol.* **176**:725–732.
- Paster, B. J., W. Ludwig, W. G. Weisburg, E. Stackebrandt, R. B. Hespell, C. M. Hahn, H. Reichenbach, K. O. Stetter, and C. R. Woese. 1985. A phylogenetic grouping of the bacteroides, cytophagas, and certain flavobacteria. *Syst. Appl. Microbiol.* **6**:34–42.
- Reichenbach, H. 1989. Genus 1. *Cytophaga*, p. 2015–2050. In J. T. Staley, M. P. Bryant, N. Pfennig, and J. G. Holt (ed.), *Bergey's manual of systematic bacteriology*, vol. 3. The Williams & Wilkins Co., Baltimore.
- Saitou, N., and M. Nei. 1987. The neighbor-joining method: a new method for reconstructing phylogenetic trees. *Mol. Biol. Evol.* **4**:406–425.
- Segers, P., W. Mannheim, M. Vancanneyt, K. De Brandt, K.-H. Hinz, K. Kersters, and P. Vandamme. 1993. *Riemerella anatipestifer* gen. nov., comb. nov., the causative agent of septicemia anserum exsudativa, and its phylogenetic affiliation within the *Flavobacterium-Cytophaga* rRNA homology group. *Int. J. Syst. Bacteriol.* **43**:768–776.
- Segers, P., P. Vandamme, P. L. Steyn, W. Mannheim, H. Willekens, M. Bauwens, J. De Ley, and K. Kersters. 1993. Phylogenetic studies of *Flavobacterium* and related organisms by DNA-rRNA hybridizations, p. 129–136. In P. J. Jooste (ed.), *Advances in the taxonomy and significance of Flavobacterium, Cytophaga and related bacteria*. University Press, University of the Orange Free State, Bloemfontein, South Africa.
- Shewan, J. M., and T. A. McMeekin. 1983. Taxonomy (and ecology) of *Flavobacterium* and related genera. *Annu. Rev. Microbiol.* **37**:233–252.
- Staley, J. T., M. P. Bryant, N. Pfennig, and J. G. Holt (ed.). 1989. *Bergey's manual of systematic bacteriology*, vol. 3. The Williams & Wilkins Co., Baltimore.
- Stanier, R. Y. 1940. Studies on the cytophagas. *J. Bacteriol.* **40**:619–635.
- Stanier, R. Y. 1941. Studies on marine agar-digesting bacteria. *J. Bacteriol.* **42**:527–559.
- Stanier, R. Y. 1942. The *Cytophaga* group: a contribution to the biology of myxobacteria. *Bacteriol. Rev.* **6**:143–196.
- Stanier, R. Y. 1947. Studies on nonfruiting myxobacteria. I. *Cytophaga johnsonae* n. sp., a chitin-decomposing myxobacterium. *J. Bacteriol.* **53**:297–315.
- Takeuchi, M., and A. Yokota. 1992. Proposals of *Sphingobacterium faecium* sp. nov., *Sphingobacterium piscium* sp. nov., *Sphingobacterium heparinum* comb. nov., *Sphingobacterium thalpophilum* comb. nov. and two genospecies of the genus *Sphingobacterium*, and synonymy of *Flavobacterium yabuuchiae* and *Sphingobacterium spiritivorum*. *J. Gen. Appl. Microbiol.* **38**:465–482.
- Vandamme, P., J.-F. Bernardet, P. Segers, K. Kersters, and B. Holmes. 1994. New perspectives in the classification of the flavobacteria: description of *Chryseobacterium* gen. nov., *Bergeyella* gen. nov., and *Empedobacter* nom. rev. *Int. J. Syst. Bacteriol.* **44**:827–831.
- Vandamme, P., P. Segers, M. Vancanneyt, K. van Hove, R. Mitters, J. Hommez, F. Dewhirst, B. Paster, K. Kersters, E. Falsen, L. A. Devriese, M. Bisgaard, K.-H. Hinz, and W. Mannheim. 1994. *Ornithobacterium rhinotracheale* gen. nov., sp. nov., isolated from the avian respiratory tract. *Int. J. Syst. Bacteriol.* **44**:24–37.
- Veldkamp, H. 1961. A study of two marine agar-decomposing, facultatively anaerobic myxobacteria. *J. Gen. Microbiol.* **26**:331–342.
- Weisburg, W. G., Y. Oyaizu, H. Oyaizu, and C. R. Woese. 1985. Natural relationship between bacteroides and flavobacteria. *J. Bacteriol.* **164**:230–236.
- Winogradsky, S. 1929. Études sur la microbiologie du sol. Sur la dégradation de la cellulose dans le sol. *Ann. Inst. Pasteur (Paris)* **43**:549–633.
- Woese, C. R. 1987. Bacterial evolution. *Microbiol. Rev.* **51**:221–271.
- Woese, C. R., D. Yang, L. Mandelco, and K. O. Stetter. 1990. The flexibacter-flavobacter connection. *Syst. Appl. Microbiol.* **13**:161–165.
- Yokota, A., M. Akagawa-Matsushita, A. Hiraishi, Y. Katayama, T. Urakami, and K. Yamasato. 1992. Distribution of quinone systems in microorganisms: gram-negative eubacteria. *Bull. Jpn. Fed. Culture Collections* **8**:136–171.