International Committee on Systematics of Prokaryotes

Subcommittee on the taxonomy of *Flavobacterium* and *Cytophaga*-like bacteria

Minutes of the meetings, 16 and 17 August 1999, Sydney, Australia

Session 1. Closed meeting

Minute 1. Call to order. The closed meeting was called to order by the Chairman, B. Holmes, at 09:15 on 16 August 1999 in the Sydney Convention and Exhibition Centre, Darling Harbour, Sydney, Australia.

Minute 2. Record of attendance. The members present were J.-F. Bernardet (Secretary), B. Holmes (Chairman), Y. Nakagawa and E. Yabuuchi.

Minute 3. Apologies for absence. Apologies were received from B. Bruun, K. A. Malik, W. Reichardt, P. Segers, J. Ursing and H. Wakabayashi for being unable to attend.

Minute 4. Approval of agenda. The written agenda was presented and approved. The minutes of the last meeting, which was held on 19 August 1996 in Jerusalem, Israel, had been approved by mail and published [*Int J Syst Bacteriol* **48** (1998), 621].

Minute 5. Changes in membership and officers. Resignations had been received from T. A. McMeekin in 1996, H. Reichenbach and P. J. Jooste in 1998, and W. Reichardt in 1999. R. P. Burchard and J. Ursing had resigned following their retirement in 1998 and 1999, respectively. Elected to full membership were J. Bowman (Hobart, Tasmania, Australia) and C. Hugo (Bloemfontein, South Africa), subject to approval by the Secretary for Subcommittees of the International Committee on Systematics of Prokaryotes (ICSP; formerly International Committee on Systematic Bacteriology). A copy of the list of the current members was given to the members present.

Minute 6. Current membership. The current voting membership of the subcommittee is as follows: J.-F. Bernardet (Secretary), Jouy-en-Josas, France; B. Bruun, Copenhagen, Denmark; B. Holmes (Chairman), London, UK; K. A. Malik, Braunschweig, Germany; Y. Nakagawa, Osaka, Japan; P. Segers, Ghent, Belgium; H. Wakabayashi, Tokyo, Japan; and E. Yabuuchi, Osaka, Japan.

Minute 7. Secretary's report on minimal standards. J.-F. Bernardet presented the draft document he had drawn up and gave a copy to the members present; the document was also subsequently circulated to those members who were unable to attend. The end of October was suggested as a time limit for members to send their comments to the Secretary. Y. Nakagawa proposed inclusion in the minimal standards of the 16S rRNA sequence signatures as a method to differentiate the genera within the family *Flavobacteriaceae* and kindly offered to identify these signatures. Subsequently, Y. Nakagawa observed that many published sequences were ambiguous because of the technique used (RNA direct sequencing) and it was thus decided not to include 16S rRNA sequences in the minimal standards. On the other hand, it was decided that the complete 16S rRNA dendro-

grams of the family and of the whole phylum provided by Y. Nakagawa be incorporated into the final document. The changes recently proposed concerning the deposition of patent strains were also added to the manuscript. E. Yabuuchi raised the question of strains that are currently unavailable because they were not deposited in any recognized culture collection. B. Holmes noted the need for vigilance on the part of the editors of the IJSEM regarding this problem. A discussion arose on the name of the subcommittee. The term *Cytophagales*, which could represent the whole phylum, is not well defined and includes some taxa that fall under the responsibility of another subcommittee (see Minute 14). The minimal standards manuscript was subsequently submitted to the Secretary for Subcommittees of the ICSP for publication in the IJSEM, after the suggestions and amendments made by all members were taken into consideration [Bernardet *et al.*, *Int J Syst Evol Microbiol* **52** (2002), 1049–1070].

Minute 8. Plan for future activities. J.-F. Bernardet will carry on his current research on *Chryseobacterium* sp. strains isolated from diseased fish. Y. Nakagawa will continue his current work on the members of the phylum that contain menaquinone 7 (see Minute 18).

Minute 9. Date of next meeting. The next subcommittee meeting will be held at the next congress of the International Union of Microbiological Societies, to be held in Paris, France, in 2002.

Minute 10. Adjournment. The closed meeting was adjourned at 11:30.

Session 2. Open meeting

Minute 11. Call to order. The open meeting was called to order by the Chairman, B. Holmes, at 13:40 on 17 August 1999 in the Sydney Convention and Exhibition Centre, Darling Harbour, Sydney, Australia.

Minute 12. Record of attendance. The members present were J.-F. Bernardet (Secretary), B. Holmes (Chairman), Y. Nakagawa and E. Yabuuchi. J. Bowman, M. Vancanneyt and M. Suzuki were welcomed to the meeting.

Minute 13. Summary of activities. The Chairman gave a summary of the closed meeting for those participants who did not attend it, especially mentioning the changes in membership and the minimal standards draft. B. Holmes pointed out that the subcommittee is not currently facing any serious nomenclatural problems in the *Flavobacterium*–*Cytophaga* group.

Minute 14. Taxonomic developments. J.-F. Bernardet presented an overview of taxonomic and nomenclatural changes in the *Flavobacterium*–*Cytophaga* group published since the

last meeting of the subcommittee, together with the corresponding dendrograms. Several new species and new genera have been described; some of them were retrieved from unusual environments (hot springs, marine and freshwater algae, polar environments ...) and even some intracellular symbionts of insects have been shown to belong to the phylum. A note by J. P. Euzéby [*Int J Syst Bacteriol* **48** (1998), 611–612] was discussed; according to certain Rules of the *Bacteriological Code* (1990 Revision), the epithet *Flavobacterium* is mis-spelled and should be corrected to *Flavibacterium*. However, J. P. Euzéby himself did not support this change for several reasons, and the members of the subcommittee present (as well as B. Bruun in a letter to the Secretary) agreed that the change was unnecessary. J.-F. Bernardet also raised the question of the status of the order *Cytophagales* following recent phylogenetic investigations; it could probably encompass the whole phylum but M. Vancanneyt suggested that, if possible, the definition of the order should be based on both molecular and chemotaxonomic characteristics. It was noted by J.-F. Bernardet that some branches of the phylum are not represented by any current member of the subcommittee. Benefit could be gained from the experience of P. Vandamme on the genus *Capnocytophaga*; M. Vancanneyt agreed to ask P. Vandamme whether he would be willing to join the subcommittee or to suggest the name of another specialist. The *Bacteroides* branch falls under the responsibility of the Subcommittee on the taxonomy of Gram-negative anaerobic rods; previous attempts by B. Holmes to organize joint meetings with this subcommittee have unfortunately failed, but he agreed to try again.

Minute 15. Nomenclature in databases. Nomenclatural problems in 16S rRNA sequence databases were discussed. Confusion arises from the deposition of sequences in databases long before the new species is actually validly published, and from the frequent use of provisional Latin names that are subsequently modified in the formal description. It was noted by B. Holmes that these 'species' have no standing in nomenclature until they are published in the IJSEM. J. Bowman explained how the time that elapsed between the deposition of the sequence and publication was largely determined by the length of the evaluation process of the manuscript; if the latter has to be emended and re-evaluated, then the delay is further increased. M. Suzuki felt it important that, in spite of these problems, new sequences and names be rapidly deposited in databases before being published. J.-F. Bernardet observed that databases are encumbered by 'species' that are merely represented by their sequence because their description has never actually been published. Another major problem is the complete lack of updating of nomenclature in databases; provisional or obsolete names thus remain listed and may be the source of confusion and errors. Several examples may be found in the *Flavobacterium–Cytophaga* group. One is the persistence in databases of two distinct sequences for the synonyms *Cytophaga marina* and *Flexibacter maritimus*. A second is the very different locations in dendrograms of the two *Flexibacter elegans* strains due to the fact that one of them actually became the type strain of another species, *F. filiformis* (ex Solntseva 1940) Reichenbach 1989. J. Bowman also observed that there is a 10% difference between the two sequences of *Cytophaga marinoflava* deposited by different authors.

Minute 16. Single-strain taxa. A discussion arose about the description of new taxa based on a single strain. In a letter to the Secretary, J. Ursing explained that this represents a

major problem for the Editors of the IJSEM. Some of them feel that a species should not be proposed unless more than one strain is available because the study of a single strain gives no idea about the diversity within the taxon. Others, however, think that anything that could be added to our knowledge of biological diversity is important (even a single strain) and that a name is essential for reference purposes. B. Holmes agreed with the former opinion but admitted that practical problems could make it difficult to retrieve several strains. M. Suzuki and M. Vancanneyt felt that a single strain could be sufficient to describe a new species provided it is fully characterized and located on a distinct branch of the 16S rRNA dendrogram. This was also J. Bowman's opinion, who noted that the description of a single-strain species could be emended when new strains became available. Conversely, several strains are necessary if the new taxon is close to previously described species; it was noted by M. Vancanneyt that DNA homology studies between a new taxon and its close neighbours are more relevant when several strains of the new taxon are studied. E. Yabuuchi felt that, when a single strain is available, it should be given a Latin name anyway. This opinion differs from P. Segers' who suggested (in a letter to the Secretary) that a code (such as CDC group ...) would be better attributed to poorly studied and single-strain taxa pending full characterization or description of additional strains. With regard to the drawing up of minimal standards, the general opinion of the members and guests present was that the description of new taxa based on a single strain should not be encouraged. However, when the isolation of several strains was not achievable, an extensive phenotypic, genomic and phylogenetic study of the single strain will be necessary to demonstrate that it differs significantly from all related species.

Minute 17. Ribotyping. The proposal by K. A. Malik (in a letter to the Secretary) to use the Riboprinter (an apparatus that automatically determines the ribotype of bacterial strains) recently acquired by the DSMZ to build a database of the ribotypes of all identified species in the *Flavobacterium–Cytophaga* group, was discussed. Given the large number of species currently included in the phylum, it was felt that this would represent a vast amount of work for the DSMZ. M. Vancanneyt confirmed that ribotyping is indeed an excellent method to check the authenticity of bacterial strains in culture collections. However, due to its high resolution (intra-species to strain level), it was questioned whether this approach would also be successful as an identification tool (generation of species-specific patterns) for new isolates. J.-F. Bernardet reported his experience on the ribotyping of 85 strains of a single species (*Flavobacterium psychrophilum*); the critical factor proved to be the choice of the restriction enzymes, since the very different profiles generated by the 23 enzymes previously tested on a few strains resulted in different clustering. Before each particular study, it is thus necessary to select the enzyme(s) that generate(s) the most interesting clustering from a practical point of view. Since the Riboprinter usually works with one enzyme only, it would probably be difficult to select the enzyme that would generate useful profiles for all species in the phylum. The general opinion on K. A. Malik's proposal was that all interested members of the subcommittee should contact him for ribotyping a group of carefully selected strains or species, preferably after several enzymes have been tested on a few strains.

Minute 18. Reports. Four members or guests very kindly outlined their current research programmes. M. Suzuki

performed a phylogenetic study of bacterial strains isolated from a marine sponge by sequencing their 16S rRNA and GyrB genes. In both dendrograms, several isolates clustered with [*Flexibacter*] *maritimus* and [*Flexibacter*] *ovolyticus*, making it possible to describe two new species; the manuscript resulting from this study has subsequently been published [*Int J Syst Evol Microbiol* **51** (2001), 1639–1652]. J. Bowman carried on his extensive study of Antarctic sea ice isolates. A paper describing three new *Flavobacterium* species and proposing a new genus to accommodate [*Flavobacterium*] *salegens* was in press at the time of the meeting, and has subsequently been published [McCammon & Bowman, *Int J Syst Evol Microbiol* **50** (2000), 1055–1063]. Another paper describing a new species related to [*Cytophaga*] *uliginosa* that was ready to be submitted for publication had to be emended following the recent publication of the genus *Cellulophaga* [Johansen *et al.*, *Int J Syst Bacteriol* **49** (1999), 1231–1240]. This manuscript has sub-

sequently been published [*Int J Syst Evol Microbiol* **50** (2000), 1861–1868]. As he could not find any phenotypic or chemotaxonomic features to characterize the different taxa with menaquinone 7, Y. Nakagawa is sequencing the 16S rRNA gene of several strains of each branch in order to determine their signature sequences; he is also characterizing *Flavobacterium*–*Cytophaga* strains retrieved from mangrove environments. B. Holmes summarized a study of the carbapenemases of *Chryseobacterium meningosepticum*, such organisms being a reservoir of various metallo- β -lactamases which could facilitate the transfer of antibiotic resistance to other, more pathogenic, bacteria in hospital environments; a manuscript presenting the results of this study has subsequently been published [*Antimicrob Agents Chemother* **44** (2000), 1448–1452].

Minute 19. Adjournment. The open meeting was adjourned at 16:10.

Jean-François Bernardet, *Secretary*