Phylogenetic relationships amongst the saltwater members of the genus *Bacteriovorax* using *rpoB* sequences and reclassification of *Bacteriovorax stolpii* as *Bacteriolyticum stolpii* gen. nov., comb. nov.

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Members of the saltwater genus *Bacteriovorax*, formerly known as the marine *Bdellovibrio*, are obligate predatory bacteria that prey selectively on other Gram-negative bacteria. Previous phylogenetic analysis based on the 16S rRNA genes of saltwater *Bacteriovorax* isolates from environmental samples revealed 11 distinct phylogenetic clusters based on $\geq 96.5\%$ gene sequence similarity. In other micro-organisms, the gene coding for the β -subunit of RNA polymerase (*rpoB*) has been shown to be more discriminating than 16S rRNA genes. In this study, *rpoB* sequences from *Bacteriovorax* isolates were analysed to determine whether the results would be consistent with those based on 16S rRNA gene sequences. A 1242 bp region of the *rpoB* gene from 74 saltwater *Bacteriovorax* strains and two freshwater isolates, *Bacteriovorax stolpii* Uki2^T and *Peredibacter starrii* A3.12^T, was amplified by PCR and analysed. The sequences were aligned and phylogenetic trees were constructed using a neighbour-joining algorithm. The resulting tree showed that the *rpoB* sequences produced smaller subdivisions of isolates, but were nevertheless consistent with the clusters determined using 16S rRNA gene sequences. Thus, the highly conserved 16S rRNA gene sequences provided good phylogenetic information and the *rpoB* gene sequences permitted greater differentiation in order to further subdivide phylogenetically distinct groups within the genus *Bacteriovorax*. Also, on the basis of the extensive diversity and large distance between the saltwater members of the genus *Bacteriovorax* and the freshwater/soil *Bacteriovorax*, a reclassification of *Bacteriovorax stolpii* as *Bacteriolyticum stolpii* gen. nov., comb. nov. is proposed. A new family, *Peredibacteraceae* fam. nov., is also described.

INTRODUCTION

Members of the genera *Bdellovibrio* and *Bacteriovorax* and similar organisms are obligate predatory bacteria that prey on other Gram-negative bacteria and can be isolated from many environmental sources (Stolp, 1968). They have a unique life cycle in which they have an extracellular attack phase followed by penetration into the periplasm of a host cell. In the periplasm, they undergo filamentous growth

and divide into several daughter cells that lyse the host cell to enter the extracellular attack phase (Ruby, 1991).

The genus *Bacteriovorax* was proposed with the reclassification of *Bdellovibrio stolpii* and *Bdellovibrio starrii* as members of the genus (Baer *et al.*, 2000). Subsequently, the saltwater isolates *Bacteriovorax marinus* and *Bacteriovorax litoralis* were placed into the genus based on 16S rRNA gene sequence analysis, DNA G + C ratio, DNA–DNA hybridization and salinity experiments that differentiated them from *Bdellovibrio* spp. (Baer *et al.*, 2004) and the freshwater *Bacteriovorax* strains, *Bacteriovorax stolpii* and *Bacteriovorax starrii* (subsequently renamed *Peredibacter starrii* by Davidov & Jurkevitch, 2004). Delineation of

Abbreviation: *rpoB*, β -subunit of RNA polymerase.

The GenBank/EMBL/DDBJ accession numbers for the *rpoB* gene sequences determined in this study are EF536750–EF536823 and EU240892–EU240893.

species within the saltwater members of the genus *Bacteriovorax* is not yet resolved. Identification and classification of these micro-organisms are difficult because of the general absence of morphological or physiological features that can be tested in the presence of the prey cells. A number of attempts to set up reliable identification schemes based on phenotypic characterization or prey susceptibility, as well as initial molecular characterization, have been reported in previous studies (Seidler *et al.*, 1969; Taylor *et al.*, 1974; Marbach *et al.*, 1975; Sutton & Besant, 1994; Pineiro *et al.*, 2004).

The *rpoB* gene, which encodes the β -subunit of RNA polymerase, has been shown to be a useful and more discriminating alternative to the 16S rRNA gene for inferring phylogenetic relationships (Case *et al.*, 2007). It has previously been demonstrated to be a suitable target on which to base species identification for the genera *Staphylococcus* (Drancourt & Raoult, 2002), *Mycoplasma* (Kim *et al.*, 2003), *Bacillus* (De Clerck & De Vos, 2004) and *Acinetobacter* (La Scola *et al.*, 2006).

In the case of the saltwater members of the family *Bacteriovoracaceae*, previous phylogenetic analysis based on 16S rRNA genes of isolates from various locations worldwide has revealed eight distinct phylogenetic clusters, along with three outlier isolates, based on $\geq 96.5\%$ gene sequence similarity (Pineiro *et al.*, 2007). In order to better define the phylogeny of the genus *Bacteriovorax*, 74 saltwater isolates, representatives of the 16S rRNA gene clusters and three outlier isolates, were selected, their *rpoB* genes were sequenced and a phylogenetic tree was constructed. The *rpoB* gene sequences from two freshwater isolates, *Bacteriovorax stolpii* and *Peredibacter starrii* (Baer *et al.*, 2000), were included. The objective of this study was to demonstrate that phylogenetic analysis based on the *rpoB* gene from *Bacteriovorax* environmental isolates was consistent with and more discriminatory than the phylogenetic clustering obtained using 16S rRNA genes. Finally, the reclassification of *Bacteriovorax stolpii* as *Bacteriolyticum stolpii* gen. nov., comb. nov. is proposed based on the extensive diversity between the saltwater members of the genus *Bacteriovorax* and the isolates derived from freshwater and soil.

METHODS

Bacterial strains and DNA processing. Seventy-four saltwater *Bacteriovorax* isolates were obtained from environmental samples collected in various countries as previously described (Pineiro *et al.*, 2007). The freshwater/soil strains *Bacteriovorax stolpii* and *Peredibacter starrii*, donated by Dr E. Jurkevitch, were also included in the study. Lytic plaques of each isolate were obtained using a double overlay method with prey *Vibrio parahaemolyticus*, as described previously (Schoeffield & Williams, 1990). *Bacteriovorax stolpii* and *P. starrii* were maintained as described by Baer *et al.* (2000). Prey cells were removed by filtration and DNA was extracted from the *Bacteriovorax* cells using a Qiagen kit according to the manufacturer's specifications. The DNA samples utilized were the same as those previously used for sequencing the 16S rRNA genes from these isolates (Pineiro *et al.*, 2007).

Selection of *rpoB* primers. Primers were designed based on aligned *rpoB* sequences from a spirochaete, *Leptospira interrogans* (GenBank accession no. AE010300); two gammaproteobacteria, *Yersinia pestis* (AL590842) and *Salmonella enterica* (AL627279); one alphaproteobacterium, *Silicibacter pomeroyi* (CP000031), and four deltaproteobacteria, *Geobacter sulfurreducens* (AE017180), *Desulfovibrio vulgaris* (AE017285), *Bdellovibrio bacteriovorus* HD100 (BX842654) and the saltwater strain *Bacteriovorax marinus* SJ^T (http://www.sanger.ac.uk/Projects/B_marinus/). The primers were selected in two highly conserved regions flanking a variable region of the *rpoB* gene based on an analysis of variable regions using BioEdit (Hall, 2001). The primers *rpoBF*, 5'-CCTAATGGGATCGAACATG-3', and *rpoBR*, 5'-GATAA CACCTTTGT TACCGTG-3', resulted in a 1242 bp fragment and were used for both amplification and sequencing.

DNA sequencing and phylogenetic analysis. The *rpoB* gene fragments were obtained using Taq Ready to Go PCR beads (Amersham Biosciences) according to manufacturer's specifications. All the PCR mixtures were subjected to an initial denaturation at 94 °C for 2 min and to 35 cycles of denaturation for 30 s at 94 °C, 30 s at 50 °C for annealing and 10 s at 72 °C for extension, followed by 7 min at 72 °C for a final extension. PCR products were purified with Multiscreen PCR plates (Millipore) and sequenced using the BigDye Terminator v3.1 cycle sequencing kit (Applied Biosystems). The resulting fragments were separated and recorded in an ABI 3730xl automatic sequencer (Applied Biosystems). All PCR products were sequenced in both directions. Raw sequence data were compiled using the Phred/Phrap/Consed package (Ewing & Green, 1998; Ewing *et al.*, 1998; Gordon *et al.*, 1998). The low quality bases from the ends were removed and the resulting sequences were aligned using CLUSTAL_X v1.8 (Jeanmougin *et al.*, 1998). Chimeras were tested for by using the sequences aligned against those of *Vibrio parahaemolyticus*. Analysis of the aligned sequences was performed using the distance matrix, neighbour-joining and bootstrapping algorithms as implemented in PAUP (Swofford, 2002; PAUP version 3.1, Sinauer Associates). Trees were constructed using maximum-parsimony and maximum-distance methods and the uncorrected 'p' model for substitution (Fig. 1). Branch supporting values were evaluated with 1000 bootstrap replications.

RESULTS AND DISCUSSION

The primers chosen for *rpoB* gene amplification from the *Bacteriovorax* isolates included a region that began and ended at bases 2073 and 3315, respectively, of the *Bacteriovorax marinus* SJ^T *rpoB* gene (http://www.sanger.ac.uk/Projects/B_marinus/). The *rpoB* gene sequences were obtained from 74 saltwater and one freshwater *Bacteriovorax* isolate, aligned and analysed using a neighbour-joining algorithm. The phylogenetic tree was constructed using *G. sulfurreducens*, another member of the class *Deltaproteobacteria*, as the outgroup as previously described for 16S rRNA gene analysis (Pineiro *et al.*, 2007). The phylogenetic tree analysis based on *rpoB* gene sequences showed 15 groups, labelled A to O, and numerous singletons (Fig. 1). The groups were defined by bootstrapping of 1000 replicates. If groups were defined as those isolates that grouped together with bootstrap value of $\geq 96\%$, then 15 groups were identified. Alternatively, if the groups were assigned by sequence similarity of $\geq 96\%$ between members of the same group, then only 9 groups could be found. Groups C, D and E, G and H, and K and L coalesced when classified by similarity, since the isolates had

Fig. 1. Phylogenetic tree for *Bacteriovorax* isolates based on *rpoB* gene sequences. Percentages with cluster designations are *rpoB* gene sequence similarity. The numbers on the branches correspond to bootstrap values based on 1000 replicates. Groups that bootstrap together $\geq 96\%$ of the time based on their *rpoB* gene sequence are represented by letters. Clusters based on 16S rRNA gene sequence analysis are numbered consistently with previously published works (Davidov & Jurkevitch, 2004; Pineiro *et al.*, 2007). GenBank accession numbers are given in parentheses. Sanger*: http://www.sanger.ac.uk/Projects/B_marinus/. Bar, 10 nucleotide changes.

gene sequence similarities of $\geq 96\%$. Less stringent conditions will produce larger groups, but the groups need to be based on data from additional loci.

The *rpoB* gene sequences of the saltwater *Bacteriovorax* isolates were compared with those of the freshwater isolates *Bacteriovorax stolpii* and *Peredibacter starrii*. The genetic distances to *Bacteriovorax stolpii* ranged from 19 to 24% nucleotide differences and for *P. starrii* from 24 to 27%. These distances were comparable to those within the saltwater *Bacteriovorax* isolates (21 to 24%) and were less than to *Bdellovibrio bacteriovorus* (37 to 41%) and to other bacteria (43 to 51%). While these distances clearly indicate that the various taxa are distinct, the distance metric is unlikely to be accurate because of the high likelihood of multiple substitutions at a single site.

A comparison of the *rpoB* sequences of the saltwater *Bacteriovorax* isolates with those from genera of other classes of the phylum *Proteobacteria* revealed insertion/deletion mutations (Fig. 2). In this paper, insertion/deletions are referred to relative to *Bacteriovorax marinus* SJ^T and no inference is intended regarding the phylogeny of these events. When other bacterial species (belonging to various classes of the phylum *Proteobacteria*) were examined, a 6 bp insertion was found starting at base 2363 of *Bacteriovorax marinus* SJ^T in the species *G. sulfurreducens*, *Salmonella enterica* and *Y. pestis*, while a 3 base insertion was found in *Silicibacter pomeroyi*, *Bdellovibrio bacteriovorus* and *D. vulgaris* (Fig. 2a). A second insertion/deletion occurrence started at nucleotide position 3052 of the *Bacteriovorax marinus* SJ^T sequence. *Silicibacter pomeroyi* was found to have a 15 bp deletion in

Fig. 2. CLUSTAL_X v1.81 multiple sequence alignment for *rpoB* numbering based on *Bacteriovorax marinus* SJ^T (http://www.sanger.ac.uk/Projects/B_marinus/). Asterisks identify invariant nucleotides.

this region and the two gammaproteobacteria had 54 bp deletions (Fig. 2b). A third region where insertion/deletion events were detected was around bases 3135 to 3239 of the *Bacteriovorax marinus* SJ^T sequence. Through this region *Bacteriovorax marinus*, *Bdellovibrio bacteriovorus* and *D. vulgaris* had the same number of nucleotides, *G. sulfurreducens* had 3 more nucleotides, while *Silicibacter pomeroyi*, *Salmonella enterica* and *Y. pestis* had 3 fewer nucleotides (Fig. 2c). It is worth noting that all of the insertions/deletion events were multiples of three, consistent with the conservation of the reading frame so that a proper enzyme can be formed. These insertions/deletions did not separate phylogenetic groups within the *Bacteriovorax* isolates. The first separated *Bacteriovorax* isolates from the other members of the class *Deltaproteobacteria*, the second separated the *deltaproteobacteria* from the other subdivisions and the third separated out *G. sulfurreducens* from the other members of the class *Deltaproteobacteria*.

The phylogenetic groups based on *rpoB* gene sequences were compared with the clusters based on 16S rRNA gene sequences (Fig. 1). The 16S rRNA gene clusters III, IV, V, IX, X, XII, XIII were identified previously (Pineiro *et al.*, 2007) and were composed of isolates with $\geq 96.5\%$ gene sequence similarity. In general, there was very good agreement between the groups identified by *rpoB* gene sequencing and the clusters identified by 16S rRNA gene sequencing. Groups C, D and E, based on *rpoB* gene sequences, and cluster V, based on 16S rRNA gene

sequences, had identical sets of isolates and contained only isolates from estuarine environments. Similarly, groups K and L and cluster XI had an identical set of isolates and were from tropical locations (Miami, FL, USA, and Trinidad, Cuba). The groups A and B, G, H and I, K and L, and M and N, were most closely related to the other groups from the same 16S rRNA gene cluster (X, III, XI and XII). These groups may well represent smaller phylogenetic taxa within the clusters defined by 16S rRNA gene sequence analysis as is the case for the genus *Prochlorococcus* which has six ecotypes with 16S rRNA gene sequences that diverge by less than 3% (Johnson *et al.*, 2006). The genus *Salinispora* has three species with 99% or greater similarity at the 16S rRNA gene locus (Jensen & Mafnas, 2006) and *Yersinia pestis* has species defined both by biochemical tests and by multi-locus sequencing typing that cannot be distinguished by 16S rRNA gene sequences (Kotetishvili *et al.*, 2005). These observations and the groups found within the identified clusters may indicate that the family *Bacteriovoracaceae* is even more diverse than shown in this study. However, before these taxonomic groups are elevated to recognized species, additional information will be required to meet the criteria set forth by the ad hoc Committee for the Re-evaluation of the Species Definition in Bacteriology (Stackebrandt *et al.*, 2002) and others (Gurtler & Mayall, 2001; Rosselló-Mora & Amann, 2001).

Although insertion/deletion events are generally rare in 16S rRNA genes, there were four locations where these events have occurred in the 16S rRNA gene of the family

Bacteriovoracaceae. Two events distinguish the predatory deltaproteobacteria. One, a single base insertion relative to the *Escherichia coli* sequence (AF233451) near *E. coli* nucleotide position 1113 (Fig. 3a), was found in all isolates from the families *Bdellovibrionaceae* and *Bacteriovoracaceae*. A second single base insertion was found in the family *Bacteriovoracaceae* near *E. coli* nucleotide position 436. In contrast, members of the family *Bdellovibrionaceae* had a 23 base insertion at this location (Fig. 3b). This genetic variance unambiguously separated these two families of predatory bacteria. The other two events subdivided the family *Bacteriovoracaceae*. The third location was near *E. coli* nucleotide position 164 where most saltwater members of the family *Bacteriovoracaceae* had a single base deletion, in contrast to the freshwater *Bacteriovorax*/*Peredibacter* lineage that had a 10 base deletion. The saltwater cluster XIII, including strain MED2, also had the larger deletion, consistent with the presence of strain MED2 as an outlier at the base of the *rpoB* phylogenetic tree. The fourth location was between nucleotide positions 181 and 199 in the *E. coli* sequence. There were six distinct patterns within the family *Bacteriovoracaceae* and two more within the family *Bdellovibrionaceae*. The family *Bdellovibrionaceae* had three

or five more bases than *E. coli* (Fig. 3d), consistent with the insertion in the 16S rRNA gene near nucleotide position 436 and the *rpoB* difference near to nucleotide position 2363 of *Bacteriovorax marinus* SJ^T. These three observations all supported the separation of these two families. There were five distinct patterns within the family *Bacteriovoracaceae* (Fig. 3d). Three patterns were found in the saltwater clusters. 16S rRNA gene clusters V, IX and X had 16 more bases than the *E. coli* sequence, clusters III, IV, XI and XII had 17 bases more than *E. coli*, and cluster XIII containing MED2 had only four more bases. The freshwater isolates also showed three patterns, cluster VII containing *P. starrii* had nine more bases than the *E. coli* sequence, while cluster I containing *Bacteriovorax stolpii* and strain PNEc1 had 14 and 15 more bases, respectively. These data confirm the division of cluster XIII away from the other saltwater isolates and the separation of the saltwater and freshwater isolates. The partitions are also consistent with the groupings seen using the *rpoB* gene sequence. Clusters V, IX and X corresponded to *rpoB* groups A–F; clusters III, IV, XI and XII to groups G–O; and cluster XIII, containing MED2, was distinct. The distinctiveness of cluster XIII may explain why, despite repeated attempts, we were unable to obtain an *rpoB*

(a)	
<i>Bacteriovoracaceae</i>	CAACGAGCGCAACCCCTATCCTTAGTTGCCAGCA-TTAAGTTGGGCACT
	*
<i>E. coli</i>	CAACGAGCGCAACCCCTATCCTTTGTTGCCAGCGGTCCGGCCGGAACT
	1113
(b)	
<i>Bacteriovoracaceae</i>	CTCTGTGAGAAGGAATAATGGTGTAGGGTCCAATAGGCCTTACATTTG
	+
<i>Bdellovibrio</i> spp.	CTCTGTGCGAGGGGAATAACACAA-----TG

<i>E. coli</i>	TACTTTACGCGGGAGGAAGGA-GTAAAGTTAATACCTTGTCTCATTG
	436
(c)	
Freshwater <i>Bacteriovorax</i> VI, VII (<i>P. starrii</i>), VIII	
Saltwater XIII (MED2)	TAACCAGGAGAAATCCTGGCTAATACCGCATAAG-----TAAG

Other saltwater <i>Bacteriovorax</i>	TAACATAATCGAAAGATTAGCTAATACCGCATAAG-ACCACGGGAAGTG
	*
<i>E. coli</i>	TAACATAATCGAAACGGTAGCTAATACCGCATAACGTCGCAAGACCAAA
	164
(d)	
Clusters IX, X & V	AAAAGT--GCTAGAAAGATGCCTCTCCTTGAAGCATTACTATGAGAT
	* + +++ ++++++
XI, XII, IV & III	AAATATC-ATTAGAAAGGATGCCTCTCCTTGAAGCATTCACTGTAAGAT
	+ +++ ++++++
Cluster XIII (MED2)	TAAT-----TAGAAAGGATGCTCCGGCA-----TTCGCCGAAAGAT
	*** ** + +++ ++
<i>P. starrii</i> (VII)	TAAT-----AAGAAAGTGGGCG-CCGCAAGG--GCTCATGCTGAAAGAA
	*** ** + +++ * +++++ +++++ -9
<i>B. stolpii</i> (I)	GACTGT--GCTTGAAAGATGCCTCTGCATAT--GCATTGCTATTAGAT
	* + +++ +++++ +++++ -4
PNEc1 (I)	TAGTAAT-GCTTGAAAGGATGCCTCTGTATAT--GCATCCACTAAAAGAT
	+ +++ +++++ +++++
Most <i>Bdellovibrio</i> isolates	CGGTTTCTGAGGTCAAAGGT-----TT-----TTCGCTTCAAGAT
	+ +
<i>Bdellovibrio</i> R4, tra & oral	AGGTTTCATGAGGTCAAAGGA-----GCAA-----TTCGCTCGAAGAT
	+ + ++ ***** +++++ ** -13
<i>E. coli</i>	GAGGGGG-AC-CTTCGGG--CCTCT-----TGCATCCGGAT
	181 199 -18

Fig. 3. CLUSTAL_X v1.81 multiple sequence alignment for 16S rRNA genes. All asterisks represent deleted bases and all plus signs represent inserted bases relative to the *E. coli* 16S rRNA gene (GenBank accession no. AF233451). The underlined nucleotides in the *E. coli* sequence are associated with the number underneath.

sequence from the other isolate in the cluster, GSL4B. The isolate GSL37 was an outlier located near the base of the *rpoB* gene tree and was ambiguously placed on the basis of 16S rRNA gene sequence alone. The pattern for insertions/deletions in the 16S rRNA gene for this isolate was the same as that of isolates in clusters V, IX and X. Finally, the outlier isolates previously revealed by 16S rRNA gene sequence analysis, strains IP and NZ7, were consistent with the phylogenetic analysis using the *rpoB* gene.

In conclusion, we found that the *rpoB* nucleotide sequences were more variable than the 16S rRNA gene sequences. The variation was consistent with that observed in sequence and insertions/deletions in the 16S rRNA locus. Some of the 16S rRNA gene clusters were subdivided by the greater nucleotide variation in the *rpoB* gene, possibly indicating that more narrowly delineated phylogenetic groups may be defined among the saltwater isolates of the genus *Bacteriovorax*. The formal establishment of defined taxonomic groups at the species level will require information from additional loci.

The wide dissimilarities in 16S rRNA gene sequence analysis between the family *Bdellovibrionaceae* and the newly described family *Bacteriovoracaceae* served as a basis for the construction of the genus *Bacteriovorax* (Baer *et al.*, 2000) and, even more recently, the reclassification of *Peredibacter starrii* was proposed (Davidov & Jurkevitch, 2004). The differences in 16S rRNA and *rpoB* gene sequences between the genera *Bdellovibrio*, *Bacteriovorax* and *Peredibacter* are large enough to place each in its own family. Our additional data give rise to additional taxonomic categories. The reclassification of *Bacteriovorax stolpii* as *Bacteriolyticum stolpii* gen. nov., comb. nov. is proposed based on the extensive diversity between the saltwater members of the genus *Bacteriovorax* and the freshwater/soil isolates.

Description of *Bacteriolyticum* gen. nov.

Bacteriolyticum (Bac.ter'io.lyt.i.cum. Gr. n. *bakterion* staff, cane and in biology, a bacterium; Gr. adj. *lutikos* able to loosen, able to dissolve; N.L. neut. n. *Bacteriolyticum* a dissolver of bacteria).

This genus consists of Gram-negative bacteria that prey upon other Gram-negative bacteria to complete a biphasic life cycle. The morphological description of the genus is the same as that of the type and only described species, *Bacteriolyticum stolpii*.

Description of *Bacteriolyticum stolpii* comb. nov.

Bacteriolyticum stolpii (stol'pi.i. N.L. masc. gen. n. *stolpii* of Stolp, named after the American microbiologist Stolp).

Basonym: *Bdellovibrio stolpii* Seidler *et al.* 1972 (Approved Lists 1980).

Other homotypic synonym: *Bacteriovorax stolpii* Baer *et al.* 2000.

The description of the species is based on those of Baer *et al.* (2000) and Seidler *et al.* (1972). The species is separate from the genera *Peredibacter* and *Bacteriovorax* as *Bacteriovorax stolpii* has very low levels of DNA–DNA relatedness, 4 % and 7.7 % to *P. starrii* and *Bacteriovorax marinus* SJ^T, respectively (Baer *et al.*, 2004). 16S rRNA gene sequence analysis shows the similarity between the freshwater/soil isolate *Bacteriovorax stolpii* and the saltwater species *Bacteriovorax marinus* SJ^T to be 89 %. The maximum 16S rRNA gene sequence similarity to any saltwater member of the genus *Bacteriovorax* is 90.8 % and ranges down to 81 % (Pineiro *et al.*, 2007).

The type strain is Uki2^T (=ATCC 27052^T=DSM 12778^T=DSM 50722^T).

Description of *Peredibacteraceae* fam. nov.

Peredibacteraceae (Pe.re.di.bac'ter.a.ce.ae. N.L. fem. n. *Peredibacter* the genus of Gram-negative, aerobic, predatory bacteria is the type genus of the family; suffix *aceae* denoting family; N.L. fem. pl. n. *Peredibacteraceae* the *Peredibacter* family).

The description of the family *Peredibacteraceae* is based on the description of the genus *Peredibacter* (Davidov & Jurkevitch, 2004) and the data from this study. This family is composed of Gram-negative, vibrioid shaped bacteria about 0.5 µm in length. Obligate predators of other Gram-negative bacteria. They exhibit a biphasic life cycle consisting of a motile attack phase and a phase that dwells in the periplasm of prey bacteria. They are a monophyletic offshoot of the family *Bacteriovoracaceae*. The only recognized species are found in freshwater and soil environments. The DNA G + C contents of the saltwater *Bacteriovorax* strains SJ^T, AQ and JS5 are in the range of 37.7–38.3 mol% (Baer *et al.*, 2004) compared with the higher values of 43.5 mol% for *Peredibacter starrii* and 41.8 mol% for *Bacteriolyticum stolpii* [previously *Bacteriovorax stolpii* (Seidler *et al.*, 1972)]. These species grow on prey in freshwater. The two species have distinctive numbers of nucleotides between *E. coli* bases 181 and 199 that differ from both the families *Bacteriovoracaceae* and *Bdellovibrionaceae*. The type genus is *Peredibacter* (Davidov & Jurkevitch, 2004 who revised Seidler *et al.*, 1972).

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