43 The Family Cyclobacteriaceae

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Abstract

Cyclobacteriaceae, a family belonging to the order Cytophagales, comprises the genera Cyclobacterium, Algoriphagus, Aquiflexum, Belliella, Cecembia, Echinicola, Fontibacter. Indibacter, Litoribacter, Mongoliicoccus, Mongoliitalea, Nitritalea, Rhodonellum, and Shivajiella. Phylogenetic analysis based on 16S rRNA gene sequences clearly clustered all the genera into the family Cyclobacteriaceae. Members of the family are known to have a wide range of morphological properties like cell shape, size, motility, colony color and shape; physiological properties like growth temperature range, salinity range, and nitrate reduction; and chemotaxonomic properties like polar lipids, quinones, and fatty acid profiles. Members of the family are found in diverse habitats ranging from cold marine regions like Antarctica, freshwater bodies, algal/microbial mats, marine waters, marine sediments, mangroves, haloalkaline soda lakes, hot springs, mud volcanoes, and ikaite tufa columns. Some of the species of the family are known to degrade a number of polysaccharides and other macromolecules like casein and lipids. Whole genome sequencing and annotation of five members of Cyclobacteriaceae family were done so far. A number of genes related to

carbohydrate metabolism, carotenoid biosynthesis, antibiotic resistance, and quorum-sensing regulation were found. Pathogenicity is not reported among the *Cyclobacteriaceae* members.

This contribution is a modified and updated version of previous family descriptions (Nedashkovskaya OI, Ludwig W, Family II. *Cyclobacteriaceae* fam. nov. In: Krieg NR, Staley JT, Brown DR, Hedlund BP, Paster BJ, Ward NL, Ludwig W, Whitman WB (eds) Bergey's manual of systematic bacteriology, The *Bacteroidetes*, *Spirochaetes*, *Tenericutes* (*Mollicutes*), *Acidobacteria*, *Fibrobacteres*, *Fusobacteria*, *Dictyoglomi*, *Gemmatimonadetes*, *Lentisphaerae*, *Verrucomicrobia*, *Chlamydiae*, and *Planctomycetes*, vol 4, 2nd edn. Springer, New York, pp 423–441, 2011).

Taxonomy, Historical and Current

Short Description of the Family

Cyc.lo.bac.te.ri.a'ce.ae. N.L.neut.n. *Cyclobacterium* type genus of the family; suff. -aceae ending to denote a family; N.L. fem.pl.n. *Cyclobacteriaceae* the *Cyclobacterium* family. (Modified from Bergey's manual) The description is an emended version of the description given in *Bergey's Manual*, 2nd edition. Vol. 4. (Nedashkovskaya and Ludwig 2011).

The family Cyclobacteriaceae is a member of the order Cytophagales (Leadbetter 1974), phylum Bacteroidetes. The family contains the type genus Cyclobacterium (Raj and Maloy 1990), Algoriphagus (Bowman et al. 2003), Aquiflexum (Brettar et al. 2004b), Belliella (Brettar et al. 2004a), Cecembia (Anil Kumar et al. 2012b), Echinicola (Nedashkovskaya et al. 2006), Fontibacter (Kämpfer et al. 2010), Indibacter (Anil Kumar et al. 2010a), Litoribacter (Tian et al. 2010), Mongoliicoccus (Liu et al. 2012), Mongoliitalea (Yang et al. 2012), Nitritalea (Anil Kumar et al. 2010b), Rhodonellum (Schmidt et al. 2006a), and Shivajiella (Anil Kumar et al. 2012a). Litoribacter (Tian et al. 2010) and Rhodonellum (Schmidt et al., 2006a) are included in the family based on the phylogenetic studies, while genera Hongiella (Nedashkovskaya et al. 2004) and Chimaereicella (Nedashkovskava et al. 2007b) are combined into the genus Algoriphagus. Cell shape varies from rod, ring/ horseshoe to coccoid forms. Majority of them are nonmotile, and in very few gliding motility is seen. Gram stain negative. The predominant isoprenoid quinone is menaquinone MK7. Mostly the polar lipids are diphosphatidylglycerol, phosphatidylcholine, phosphatidylglycerol, phosphatidylethanolamine, and a number of unidentified aminolipids, aminophospholipids, phospholipids, and lipids. The DNA G+C values range from 35.0 to 62.8 mol%. Usually found in the following habitats: microbial/algal mats, soil, cold marine water, mud volcanoes, hot springs, marine solar salterns, freshwater, coral Palythoa species, and ikaite tufa columns.

Phylogenetic Structure of the Family and Its Genera

According to the phylogenetic branching of Cyclobacteriaceae type strains in the RaxML 16S rRNA gene tree of the All-Species Living Tree Project (Munoz et al. 2011), the family is moderately related to the families Cytophagaceae, Flammeovirgaceae, Chitinophagaceae, and Rhodothermaceae. Among these, Cytophagaceae and Cyclobacteriaceae were broad groups clustered together, and others are less species-rich families, which were clustered outside this group. Apart from this, some of the species related to the genera Flexibacter and Thermonema of the family Cytophagaceae were clustered separately (Fig. 43.1). All members of the Cyclobacteriaceae family were clustered together. Members of the genera Algoriphagus, Cyclobacterium, Echinicola, and Fontibacter were clustered with their respective members.

Molecular Analyses

DNA-DNA Hybridization Studies

The family *Cyclobacteriaceae* consists of a total of 14 genera. DNA-DNA hybridization (DDH) studies have been conducted for many of them. The available DDH results show that all of the tested species have similarity values below 40 %, indicating low relatedness to each other. The relatedness results were 37.5 % between *Algoriphagus aquaeductus* and *Algoriphagus aquaetilis* (Rau et al. 2012), 36 % between *Algoriphagus aquimarinus* and *Algoriphagus ratkowskyi* (Nedashkovskaya et al. 2004), 9–35 % between *Algoriphagus locisalis* and the type strains, 28.3 % between *Chimaereicella boritolerans* and *Chimaereicella alkaliphilia* (Ahmed et al. 2007), 24.5 % between *Cyclobacterium qasimii and Cyclobacterium amurskyense* (Shivaji et al. 2012), and 9.5 % between *Rhodonellum psychrophilum* and *Belliella baltica* (Schmidt et al. 2006a).

DNA-DNA relatedness experiments were not carried out between a number of novel taxa with their respective nearest neighbor, as the level of 16S rRNA gene sequence similarity between the novel taxa was less than 96.9 %, which is below the cutoff value (97 %) suggested by Stackebrandt and Goebel (1994) for genomic distinction of species.

The list of taxa for which DNA-DNA relatedness experiments includes Algoriphagus were not carried aquatilis (95.2 % sequence similarity with Algoriphagus mannitolivorans); Algoriphagus faecimaris (95.8 % with Algoriphagus hitonicola); Algoriphagus hitonicola (93.8 % with Hongiella ornithinivorans); Algoriphagus namhaensis (96.1 % with Algoriphagus halophilus); Algoriphagus terrigena (93.8-95.7 % with Algoriphagus species); Hongiella mannitolivorans, Hongiella halophila, and Hongiella ornithinivorans (88.7-91.2 % with Cyclobacterium sp.); and Cecembia calidifontis (96.4 % with Cecembia lonarensis).



Fig. 43.1

Phylogenetic reconstruction of the family *Cyclobacteriaceae* based on 16S rRNA and created using the neighbor-joining algorithm with the Jukes-Cantor correction. The sequence dataset and alignment were used according to the All-Species Living Tree Project (LTP) database (Yarza et al. 2010; http://www.arb-silva.de/projects/living-tree). The tree topology was stabilized with the use of a representative set of nearly 750 high-quality type strain sequences proportionally distributed among the different bacterial and archaeal phyla. In addition, a 40 % maximum frequency filter was applied in order to remove hypervariable positions and potentially misplaced bases from the alignment. Scale bar indicates estimated sequence divergence

The *Rhodonellum psychrophilum* showed a very low relatedness of only 9.5 % with its nearest phylogenetic neighbor *Belliella baltica*, and also its physiological and chemotaxonomic properties were found to be distinct (Schmidt et al. 2006a); hence, a new genus, *Rhodonellum*, was made under *Cyclobacteriaceae* family to accommodate this new species. The 16S rRNA gene study of several novel genera, namely, *Aquiflexum, Belliella, Cecembia, Fontibacter, Indibacter, Litoribacter, Mongoliicoccus, Mongoliitalea, Nitritalea, and Shivajiella* showed very less sequence similarities (<95 %), with the nearest taxa: hence, no DDH experiments were conducted by the authors.



Fig. 43.2

Score-oriented dendrogram generated by the BioTyper software (version 3.0, Bruker Daltonics) showing the similarity of MALDI-TOF mass spectra of cell extracts of selected type strains and other related strains of the family *Cyclobacteriaceae*

MALDI-TOF

MALDI-TOF (matrix-assisted laser desorption/ionization timeof-flight) mass spectrometric approach is a high throughput, reliable, and simple molecular tool for the classification and identification of microorganisms (Holland et al. 1996; Fenselau and Demirev 2001). Minimal sample preparation efforts, life cycle costs, and open, expandable library were more advantageous features. The tool compares the mass spectra acquired between 2,000 and 20,000 Da of biomolecules (ribosomal proteins) extracted from the microbial cells (Tóth et al. 2008). By comparison of generated mass spectrometric fingerprints with reference databases, several species of bacteria, yeasts, and fungi will be reliably identified. The ribosomal proteins were identified by searching the database (http://au.expasy.org/ srs5bin/cgi-bin/wgetz). Dendrograms based on the similarity of MALDI-TOF mass spectra provide an insight into the phylogenetic relationship of species in good agreement with results obtained by sequence comparison of 16S rRNA gene (Anil Kumar and Srinivas, unpublished).

Similar to the 16S rRNA gene sequence, dendrogram type strains of *Cecembia* as well as *Algoriphagus* are closely related (**Fig. 43.2**). The species of the respective taxa were clustered-together. And most of the clusters agree in the 16S rRNA gene sequence-based trees and MALDI-TOF MS-based

dendrograms. There is no report till now about the use of MALDI-TOF approach for characterization of the *Cyclobac-teriaceae* members.

Genome Comparison

As of now, whole genome sequences of only five taxa have been reported. The reported whole genome sequence of Algoriphagus sp. PR1 (Alegado et al. 2011) (GOLD ID Gi09039) is 4,776,234 bp long, contains 3,954 genes including three 16S rRNA genes, and has a G+C mol% of 38.69 %. The G+C value is within the range of 35-42 mol% deduced for the species by HPLC and thermal denaturation methods. The annotation of the genome revealed presence of several proteins, which account for the diversity of carbohydrate-active enzymes in Algoriphagus sp. PR1 (Alegado et al. 2011). Enzymes like glycosyltransferases, glycoside hydrolases, carbohydrate esterases, and polysaccharide lyases constitute for the high capacity of polysaccharide degradation. The genome of the bacterium is reported to have a repertoire, which is more similar to Bacteroidetes found in gut; this may be related to their interaction with choanoflagellates (Alegado et al. 2011).

Another reported whole genome is of *Belliella baltica* DSM 15883^T (GOLD ID Gc02256); it is 4,196,595 bp long, has 3,879

genes including five 16S rRNA genes and 39 tRNA genes, and has a G+C mol% of 36.81 % which is closer to value generated through spectrophotometric method (Brettar et al. 2004a). On the study of the genome annotation, enzymes like penicillin amidase and isopenicillin N synthase, which are a part of penicillin synthesis pathway, were found. Penicillin-binding protein 2, penicillin repressor, and beta-lactamase class penicillin-binding protein, which impart resistance against the antibiotic penicillin, other beta-lactam antibiotics, and cellulase (glycosyl hydrolase family 5), were also found in the genome (GOLD ID Gc02256).

The third reported whole genome is of Cyclobacterium marinus DSM 745^T (GOLD ID Gi04013); it is 6,451,537 bp long, has 5,149 genes including three 16S rRNA genes and 39 tRNA genes, and has a G+C mol% of 38.1 % which is slightly lesser than the value generated through spectrophotometric method (Raj and Maloy 1990). The annotation contains cellulose-degrading enzymes, cellulase M, and cellulase (glycosyl hydrolase 5 family). The genome reports the presence of a beta-lactamase enzyme, which imparts resistance against beta-lactam antibiotics like penicillins, cephamycins, and carbapenems (ertapenem) (carbapenems are relatively resistant to beta-lactamase) (Papp-Wallace et al. 2011). Aldehyde dehydrogenase enzyme, which catalyzes dehydrogenation of aldehydes (Crabb et al. 2004); sulfatase enzyme, which catalyzes the hydrolysis of sulfate esters; fatty acid desaturase enzyme, which removes two hydrogen atoms from a fatty acid, creating a carbon/carbon double bond; and arsenate reductase enzyme, responsible for arsenate reduction, which can prove to be very useful in arsenic detoxification (Mukhopadhyay and Rosen 2002) were detected in the genome (GOLD ID Gi04013).

The fourth reported whole genome is of Nitritalea halalkaliphila strain LW7^T (Anil Kumar et al., 2010b); the reported genome is 3,633,701 bp long, has 3,145 genes including four rRNA genes and 38 tRNA genes, and has a G+C mol% of 48.58 % (Jangir et al. 2012) which is very close to the value generated through spectrophotometric method (Anil Kumar et al. 2010b). The annotation reports beta-lactamase class C and other penicillin-binding proteins, which provide resistance against beta-lactam antibiotics like penicillins, cephamycins, and, to some extent, carbapenems, which are relatively resistant to beta-lactamase (Papp-Wallace et al. 2011). Also, the annotation has cellulase M and related proteins, which may have cellulose-degrading properties. DNA photolyase enzyme was also present in the genome annotation, which indicates UV protection features in the bacterium. Interestingly, we found acyl-homoserine lactone acylase enzyme in the annotation of the genome. This acyl-homoserine lactone acylase enzyme is involved in quenching of quorum sensing in bacteria (Lin et al. 2003). This can have a very promising antibacterial application as quorum sensing is very important for proper bacterial colonization, and hence if the enzyme can quench the quorum sensing, it can provide a newer approach in bacterial disease control (Dong et al. 2001).

The fifth reported genome is of *Cecembia lonarensis* strain LW9^T (Shivaji et al. 2012), which is 4,842,844 bp long, has 4,261

genes including 39 tRNA genes and two 16S rRNA genes, and has a G+C mol% of 40.96 % which is very close to the value generated through spectrophotometric method (Anil Kumar et al. 2012b). The genome carries methicillin resistance regulatory protein, which provides resistance against methicillin, a narrow-spectrum antibiotic (Garcia-Castellanos et al. 2004). Isopenicillin N epimerase, penicillin amidase, and various penicillin-binding proteins were also found in the genome of the bacterium, suggesting probable production of penicillin antibiotic. Catalase-peroxidase enzyme was also present in the genome of the bacterium. This enzyme has a catalase and a broad-spectrum peroxidase activity. Alpha-amylase enzyme is also present in the genome, which suggests amylase enzyme production by the bacterium. Lycopene cyclase, an enzyme of lycopene synthesis pathway, was also present in the genome of the bacterium. Lycopene is a very potent biological oxygen quencher (Di Mascio et al. 1989), and it has several health benefits (Böhm et al. 2012).

Phenotypic Analyses

The Family Cyclobacteriaceae

The main features of members of *Cyclobacteriaceae* are listed in **●** *Tables* 43.1–43.7.

Cyclobacterium Raj and Maloy 1990, 345^{VP} Emend. Ying, Wang, Yang, and Liu 2006, 2929^{VP}

Cy.clo.bac.ter'i.um. Gr. n. *cyclos* a circle L. neut. n. *bacterium* a small rod; N.L. neut. n. *Cyclobacterium* a circle-shaped bacterium.

Cells are aerobic, Gram stain negative, psychrotolerant, moderately halotolerant, nonflagellated, nonmotile, and nonspore forming. Morphological and chemotaxonomic properties are indicated in **•** *Table 43.1*.

At present the genus comprises four species, *Cyclobacterium* marinus Raj^{T} , *Cyclobacterium amurskyense* KMM 6143^T, *Cyclobacterium lianum* HY9^T, and *Cyclobacterium qasimii* M12-11B^T. The type species of the genus is *Cyclobacterium* marinus, type strain is Raj^{T} .

Cells are curved, ringlike, or horseshoe shaped with outer diameter ranging between 0.6 μ m and 1.8 μ m and width ranging between 0.2 μ m and 0.7 μ m (Ying et al. 2006; Shivaji et al. 2012). Colonies on marine agar (MA) are circular, convex, and pink pigmented with entire edge. Colonies of *C. marinum* are opaque (Raj and Maloy 1990) and of *C. qasimii* are translucent (Shivaji et al. 2012). Being a marine microorganism, *Cyclobacterium* strains have definite NaCl requirement for optimal growth, but absolute requirement is not observed except for *C. lianum* which requires 0.1 % w/v NaCl. Tolerance varies from 6 % to 12 % among species (Ying et al. 2006; Shivaji et al. 2012). Temperature range for growth is between 4 °C and 42 °C (with optimum grows within a temperature range between 4 °C and 42 °C and 42 °C and

Table 43.1

Phenotypic characteristics of genera of Cyclobacteriaceae. Taxa: 1. Algoriphagus; 2. Aquiflexum; 3. Belliella; 4. Cecembia; 5. Cyclobacterium; 6. Echinicola; 7. Fontibacter; 8. Indibacter; 9. Litoribacter; 10. Mongoliicoccus; 11. Mongoliitalea; 12. Nitritalea; 13. Rhodonellum; and 14. Shivaiiella

	1	2	3	4	5	6	7	8	6	10	11	12	13	14
Cell morphology:														
Regular rods	+	+	+	+	-	+	+	+	+	-	+	+	+	+
Ringlike/horseshoe	Ι	Ι	I	Ι	+	Ι	I	I	Ι	I	Ι	Ι	I	1
Coccus	I	I		I	I				I	+	I	1		
Cell size (µm)	0.3-0.7 ×	0.3-0.6 ×	0.3–0.5 ×	0.5–0.6 ×	0.3-0.7 ×	0.3–0.5 ×	0.3-0.6	0.5-0.7	0.3-0.5 ×	0.7-1.2	$0.5 - 1 \times 3 - 8$	0.7-0.8	0.7-1	0.5-0.8
	0.3-10	1.1–4.8	0.9–3	1.5–3	0.8-1.5	1.1–2.3	×	imes 2–3	1.2-1.5			\times 2–3	×	× 2–5
							1.5–3.2						0.8–3	
Gliding motility	Ι	Ι		Ι	Ι	+	I	Ι	Ι	Ι	Ι	Ι	I	1
Colony color	Pink	Red	Pink/orange red	Red/orange	Pink	Pink/yellow orange	Orange	Red/ orange	Bright red	Pink	Orange red	Red/ orange	Bright red	Drange
Oxidase	+	+	+		+	+	+	+	+	+	+	+	1	+
Catalase	+	I	1	+	+	+	+	+	+	+	+	1	+	+
Nitrate reduction	D	+	+	Ι	Ι		I	+	+	+	+	I	1	+
Salinity range (%)	0-10	0-6	0–6	0-4	0-12	0–15	1–3	0-8	6-0	0-10	0-5.5	1–22	0-3)–3
Growth at:														
5 °C	D	+	+	Ι	+	D		-	-	+	+	-	+	
25 °C	D	+	+	+	+	+	+	+	+	+	+	+	-	+
40 °C	D	+		+	+	+	+	+		+	+	+	-	+
Hydrolysis of starch	D	+	+	Ι	Ι	+	Ι	+	+	+	+	Ι	+	1
Major polar lipids	PC, PE, L, APL, AL, PL	PE, L	PE, PL, L	PE, APL, L	PC, PE, AL, L	PE, APL, AL, L	PE, AL, L	PE, AL, L	pu	PC, PE	PC, PE, PG	PE, AL, PL, L	pu	DPG, PG, PE
Major fatty acids	i-C _{15:0} , i-C _{17:1}	i-C _{15:0} , ai-	i-C _{15:0} , i-C _{17:1}	i-C _{15:0} , i-	i-C _{15:0} , ai-	C _{15:0} , C _{16:1}	i-C _{15:0} , i-	i-C _{15:0} ,	i-C _{17:1} @9c,	i-C _{15:0} , ai-	i-C _{15:0} , i-	i-C _{15:0} ,	i-C _{17:1}	-C _{15:0} ,
	<i>0</i> 9с, i-С _{17:0}	C _{15:0} , i-C _{15:1}	ω9c, C _{16:1} ω5c,	C _{16:0} , ai-	C _{15:0} , i-C _{17:1}	<i>ω5</i> c, i-C _{17:1}	C _{17:1}	ai-C _{15:0} ,	С _{15:0} 3-ОН,	C _{15:0} , i-C _{16:0} ,	C _{17:0} 3-OH,	i-C _{17:0}	ω9c,	лі.
	3-OH, SF3	G, i-C _{16:1} H, . C 2011	i-C _{15:1} G, i-C _{17:0}	C _{15:0} , i-C _{16:1}	ω9c, i-C _{17:0}	ω9c, C _{17:1}	ω9c, . C	i-C _{17:1}	i-C _{17:0} 3-OH,	C _{16:0} , i-C _{17:0}	C _{15:1} @6c, i-	3-OH	i-C _{17:0}	C _{15:0} ,
		Н-L _{17:0} 30П, Н-С _{17:1} 009С, сега	3011, C _{16:0} , SF3	П, 213	5-UH, SF3	000, I-U170 3-OH, SF3	3-OH	09с, i-С _{17:0} 3-Он	ors, L _{17:1} ø6c	3-UH, SF3, SF9	с _{16:0} 3-UП, SF4, SF9		SF3	30H,
Major respiratory quinone	MK7	MK7	MK7	MK7	MK7	MK7	MK7	MK7	MK7	MK7	MK7	MK5	pu	MK7
DNA G+C content (mol%)	35–49	38.4	35.4	40.5	41–45	44-46	53.2	43	45.1	62.8	39.8-41.2	49	44.2	41.2- 41.3
)ata are taken from Ahn 2006); Yoon et al. (2004,	ned et al. (2007); 2005a, b, 2006);	Bowman et al. (2 Shivaji et al. (20	2003); Brettar et al. (2)12); Nedashkovskay	2004a, b); Nedasł 'a and Ludwig, (2	nkovskaya et al. (111); Anil Kuma	(2004, 2006, 2007; ir et al. (2010a, b, .	a, b); Schmid [;] 2012a, b); an	t et al. (<mark>2006</mark> . d Srinivas et	a); Tiago et al. (2 al. (2012)	:006a); Van Trap	pen et al. (2004)	; Yi and Chi	un (2004); '	/ing et al.

Summed features represent groups of two or three fatty acids that cannot be separated by GLC with the MIDI system. Summed feature 3 contains C₁₆₁ 0/7 and/or C₁₆₁ 0/7 and/or C₁₆₁ 0/7 and/or C₁₆₁ 0/2.

Symbols: + >85 % positive, - 0-15 % positive, D different reactions occur in different taxa (species of a genus). Sf summed feature, DPG diphosphatidylglycerol, PC phosphatidylcholine, PE phosphatidylethanolamine, PG phosphatidy@ycerol, AL unidentified aminolipid, APL unidentified aminophospholipid, PL unidentified phospholipid, L unidentified lipid, i iso, ai anteiso, MK menaquinone, nd no data available Summed feature 9 contains i-C $_{17:1}$ ω 9c and/or 10-methyl-C $_{16:0}$

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Table 43.2 Differentiating phenotypic properties of type strains of Cyclobacterium

	Cvclobacterium	Cvclobacterium	Cvclobacterium lianum	Cvclobacterium aasimii
Characteristic	marinus Raj ^T	amurskyense KMM 6143 ^T	HY9 ^T	M12-11B ^T
Cell size (outer diameter \times cell width) in μm	0.8–1.5 × 0.3–0.7	0.9–1.2 × 0.3–0.4	1.5–1.8 × 0.4–0.5	0.6–0.7 × 0.2
Salinity range (% w/v) NaCl	0–10	0–10	0.1–12	0–6
Temperature range for growth (°C)	4–42	4–40	15–42	4–30
Optimum temperature (°C)	20–25	25	33	18
Ornithine decarboxylase	+	_	nd	—
Oxidase	w	+	+	w
β -galactosidase	+	+	+	_
Hydrolysis of:				
Aesculin	+	+	+	_
DNA	-	-	-	+
Gelatin	+	-	-	—
Tween 20	-	-	+	—
Tween 40	+	-	w	—
Tween 80	-	-	w	nd
Acid production from:	•			
D-Glucose	_	_	+	+
Ribose	+	+	w	+
Methyl α-D-glucoside	_	_	+	+
Glycerol	_	_	w	_
Utilization of:				
Sorbose	+	+	_	_
Salicin	_	+	nd	_
Mannitol	+	_	w	+
Methyl α-D-glucoside	-	-	+	+
Melezitose	+	+	+	w
Gluconate	-	+	+	_
Methionine	+	-	nd	w
Cysteine	_	+	nd	+
Aspartic acid	+	w	v	+
Isoleucine	+	_	nd	+
Alanine	-	-	_	+
Histidine	-	+	nd	+
Susceptibility to:		I		
Penicillin	_	+	+	+
Kanamycin	_	+	+	_
Streptomycin	+	_	_	_
Tetracycline	+	_	_	+
Maior fatty acids	i-C15:0, ai-C15:0,	i-C15:0, ai-C15:0, i-C15:1 G,	i-C15:0, ai-C15:0, i- C17:1 (09c)	i-C15:0, ai-C15:0, i-C15:1 G, C15:1 06(
	i-C _{15:1} G, SF3, SF9	SF3, SF9	C _{17:0} 3-OH, SF3	C _{17:1 ω6c} , SF3, SF9
Polar lipids	PE, PC, AL2, L1 to L3	PE, L1 to L5	nd	PE, PC, AL1, L1 to L3
Menaquinones	MK7	MK7	nd	MK7
DNA G+C content (mol%)	41.9	41.3	45.2	40.5

Data taken from Ying et al. 2006; Shivaji et al. 2012; and Nedashkovskaya and Ludwig 2011

Summed features represent groups of two or three fatty acids that cannot be separated by GLC with the MIDI system. Summed feature 3 contains $C_{16:1} \omega 7c$ and/or $C_{16:1} \omega 6c$. Summed feature 9 contains i- $C_{17:1} \omega 9c$ and/or 10-methyl- $C_{16:0}$.

+ Positive, – negative, w weakly positive, v variable, nd no data available, i iso, ai anteiso, SF summed feature, PC phosphatidylcholine, PE phosphatidylethanolamine, AL unidentified aminolipid, L unidentified lipid, MK menaquinone

🖸 Table 43.3

Differentiating phenotypic properties of type strains of Algoriphagus. Taxa: 1, Algoriphagus alkaliphilus; 2, Algoriphagus antarcticus; 3, Algoriphagus aquaeductus; 4, Algoriphagus aquatilis; 5, Algoriphagus aquimarinus; 6, Algoriphagus boritolerans; 7, Algoriphagus chordae; 8, Algoriphagus faccimaris; 9, Algoriphagus halophilus; 10, Algoriphagus hitonicola; 17, Algoriphagus olei; 18, Algoriphagus ornithinivorans; 19, Algoriphagus ratkowskyi; 20, Algoriphagus terrigena; 21, Algoriphagus vanfongensis; 22, Algoriphagus winogradskyi; 11, Algoriphagus jejuensis; 12, Algoriphagus locisalis; 13, Algoriphagus lutimaris; 14, Algoriphagus mannitolivorans; 15, Algoriphagus marincola; 16, Algoriphagus namhaensis; 23. Alaoriphaqus yeomjeoni

	1	2	3	4	5	9	7	8	6	10	11	12 1	1	14	15	16	17	18	19	20	21	22	23
Nitrate reduction	+	I	+	I	I	I		+		+	1		+	-				I	v	+	I	+	
Salinity range for growth (%)	0–3	0-5	0-4	0–3	0-10	0-3	1–10	0-8 (8-0	1.5-5	0-5	1-9 0)-8 0		1-9	0.5-6	0-3	0-10	0-6	1-7	0-8	0-6	1–9
Temperature range for growth (°C)	11–39	5-25	15-42	20-37	4-34	17–37	4–32	0-42	10-41	20-40	4-30	4-35 2	1-40 1	10-42	10-45	10-40	10–37	10-40	-2-25	10–36	12–35	4–39	4-35
Catalase	+	+	+	+	+	+	+	*	+	+	+	+	+		+	+	+	+	+	+	+	+	+
Oxidase	+	+	+	+	+	+	+	+	+		+	+	+		+	+	+	+	+	+	+	+	+
β -galactosidase	+	+	+	+	+	+	+	-	+		+	+	+		+	1	+	+	+	+	+	+	+
H ₂ S production	I	I		+	I	I	I				1						I	I	I	I	I	I	I
Hydrolysis of:																1							
Agar	I	I		pu	+	I	+	1			+	-				I	I	I	I	Ι	I	I	I
Casein	Ι	1	+	w	+	pu	-	-	_	pu		+	-	-	+	+	-	Ι	+	+	-	I	+
Gelatin	+	Ι	+		+	-	1	+	+	pu	+	+	+			+	-	+	v	-	+	+	~
Starch	+	Ι	+	+	Ι	+	I	+	+	+	-	-	+		+	+	I	+	^	-	I	+	I
DNA	+	Ι	+	Ι	+	pu	I	+	1	+	n bn	+ pu	+	, ,	+	I		+	^	pu	Ι	Ι	Ι
Tween 20	pu	pu	+	+	+	pu	+	+	+	pu	+	+	- pt		+	+	Ι	Ι	+	+	Ι	+	+
Tween 40	pu	pu		+	+	pu	+	+ M	+	pu	+	+	- pı		+	+	Ι	+	+	+	Ι	+	+
Tween 80	pu	pu		+	+	pu		T N	+		+	+	- pı		+	+		+		+	I		+
Acid production fr	:mo																						
L-arabinose	+	Ι			Ι		I	1			+	+	1		+	I	Ι	Ι	+	+	Ι	I	Ι
Cellobiose	+	Ι		N	+	+	+	+	,	+	+	+	+	بر	+	I			+	+	+	+	+
D-fructose	+			8	pu	I	pu	+	- pu	+	N	+		-		8	Ι	I	+	+	I	pu	+
D-galactose	+	Ι		pu	Ι	+	+	+	-	pu	+	+	1		+	1			+	+	Ι	+	+
D-glucose	+	I	+	N	+	+	+	+	+	+	+	+	1	-	+	+		+	+	+	+	+	+
D-lactose	+	Ι	+	N	+	+	+	-	-	+	M	+	-	-	+			Ι	Ι	+		+	+
Maltose	+	Ι	+	w	Ι	+	+	+	+	+	+	+	-		+	+	Ι	Ι	+	+	+	+	+
D-mannose	+	I	+	v	+	+	+	w	- pu	+		+	1		+	I	Ι		+	+	+	+	Ι
Melibiose	+	I	+	w	+		+	+	-	+	+	+	-	-	+	1			+	+	-	+	+
Raffinose		Ι	+	nd	I	+	+	- pu		+	+	+	1		+	I	Ι	Ι	+	+	Ι	+	~
L-rhamnose	Ι	I	+	I	+	+	+	- pu			M	+	1	,		I	Ι	I	+	I	+	+	+
Sucrose	+	I	+	>	+	I	+	+	+	+	+	+	+	,	+			I		+	I	+	+

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Trehalose	+		+	>	bu	-	pu	pu	pu	+	+	>	+	I	+	Ι	I	I	+	+	I	pu	+
DL-xylose	I				+	I	+	I	+	I	+	+	I	I	+	>	I	I	+	+	+	+	+
	1	2	3	4	5	9	7	8	6	10	11	12	13	14	15	16	17	18	19	20	21	22	23
<i>N</i> -acetyl- _D - glucosamine	I	I	+	I	+	+	I	I	+	+	+	pu	pu	I		pu	I	I	+	pu	+	+	Ι
Utilization of:																							
L-arabinose	+	I	I	I	+	I	+	I	+	I	+	+	I	+	+	pu	I	I	+	+	+	+	I
D-galactose	+	pu	+	Ι	pu	+	+	I	+	pu	N	1		Ι	+	pu	+	I	+	+	pu	+	+
p-glucose	+	Ι	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-lactose	+	pu	+	+	+	+	+	I	+	+	M	+	+	+	+	pu	+	+	+	+	+	+	+
Maltose	+	Ι	+	+	+	+	+	I	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
D-mannose	+	I	+	+	+	+	+	I	+	+	+	+	+	+	+	pu	+	+	+	+	+	+	+
D-xylose	Ι	pu	pu	Ι	+	Ι	+	Ι	+	Ι	+	1	+	+	+	M	+	+	+	+	+	+	+
D-mannitol	Ι	Ι	I	pu	+	+	Ι	I	Ι	Ι	+	I	pu	+	I	pu	Ι	Ι	+	Ι	Ι	I	Ι
Sorbitol	+	pu	I	pu	Ι	Ι	I	I	Ι	I	+	pu	pu	I	I	pu	Ι	Ι	+	pu	Ι	I	pu
myo-Inositol	I	pu	+	Ι	I	I	Ι	+	Ι	Ι	pu	pu	pu	Ι	I	pu	Ι	Ι	+	Ι	I	I	Ι
Glycerol	Ι	pu	+	Ι	Ι	pu	Ι	I	-	Ι	pu	pu	pu	Ι	Ι	pu	Ι	Ι	Ι	pu	-	Ι	Ι
N-acetyl- ^{D-} glucosamine	I	I	+	+	+	I	+	+	+	+	+	pu	pu	+		pu	+	+	+	pu	+	+	+
Susceptibility	to:																				_	_	
Gentamicin	pu	pu	pu	pu	I	+	I	I	I	pu	I	1	+	I	I	+	+		I	I	+	I	I
Tetracycline	pu	pu	+	pu	+	+	I	I	+	pu	+	I	I	+	+	Ι	Ι	+	+	Ι	+	>	Ι
Major fatty acids (>5 %)	i- C _{15:0} , i-C _{17:1} <i>w9c</i> , i- C _{17:0} 3-OH, SF3	i: C _{15:0} , G, i- C _{17:0} 3-OH, SF3	i-C _{15:0} , C _{16:1} <i>ω5c</i> , i- C _{17:1} <i>ω9c</i> , i- C _{17:0} 3- OH, SF3	i-C _{15:0} , i-C _{17:1} <i>ω9c</i> , SF3	i-C _{15:0} , i C _{17:1} 	- ai-C ₁₅₀ , i i-C ₁₅₀ i C ₁₆₀ , i- C ₁₆₁ , i- C ₁₇₁ w <i>9</i> C, SF3	i- i- i- C _{15:0} , i- C _{15:0} , 3- OH, SF3	i- C _{15:0} , C _{16:0} , C _{18:0} i- C _{17:0} 3-OH	i-C _{15:0} , i-C _{16:0} , i-C _{17:1} <i>ω9</i> C, i- C _{17:0} 3- OH, SF3	i- C _{15:0} , i-C _{17:1} <i>w9c</i> , i- C _{17:0} 3-OH, SF3	г. С _{15:0} , SF3	гі С _{15:0} , С 1 _{7:0} 3- ОН, SF3	і- С15:0/ İ-С17:1 0.9 <i>с</i> , İ- С17:0 3-ОН, SF3	i-C _{15:0} , i- C _{16:0} , i- C _{17:1} <i>w9c</i> , C _{17:1} <i>w6c</i> , i-C _{17:0} 3- OH, SF3	і С1 _{5:0} , С1 _{6:0} , і і С1 _{7:0} 3- ОН, SF3	i-C _{15:0} , i-C _{16:0} , i-C _{16:1} , i-C _{17:0} 3-OH, SF3	i-C _{15:0} , i- C _{15:1} G, i-C _{17:1} <i>w9</i> c, i- C _{17:0} 3- OH, SF3	ai-C _{15:0} , C _{15:0} , i- C _{16:0} , i- C _{17:1} <i>m9</i> C _{17:1} <i>m9</i> SF3	i- i- C _{15t} i-C _{15t} <i>w5c</i> , c,i- C _{17t} 3-O7t SF3	i- i- i- i- i- i- i- i- i- i-	i :C ₁₅₀ i C ₁₅₁ G, C ₁₅₀ 3 - OH, i C ₁₇₀ 3- OH, SF3	- i- C-15:0, C-17:0 3- 3- OH, SF3	i C1 _{5:0} , i C1 _{7:0} 3 SF3
Major polar lipids	ри	pu	PE, PL, APL, L	pu	pu	pu	pu	pu	pu	pu	DPG, PE, AL, APL, L	hd	pu	ри	pu	PC, PE, L	PE, PL, AL, APL, L	pu	PC, PE, PL, L	DPG, PE, - AL, APL,	pu	hd	pu
			1 2		3	4 5	9	7	ø	6	10	11	1	2 13	14	15	16	17 1	8 19	20	21	22	23
DNA G+C con	tent (mo	1%)	43.5 3	9-41	41.8	43 41	42.5	37-	40 42	.5 37	, 43.	4 48	3.5 42	2 41.4	42	43	44.8	43 3	8 35	49	43.8	39-42	41
Data taken from, B Summed features _i + Positive, – negë	bowman et represent ative, w w	al. (2003) groups of eakly poo); Copa-Patiŕ f two or thre sitive, v variċ	io et al. (<mark>2</mark> C e fatty acio able, <i>nd</i> no	008); Lee et ds that canr o data avail	al. (2012); Li ot be separ. lable, <i>SF</i> sur	et al. (2011 ated by GL nmed feat), (2009); N C with the ure, <i>P</i> C pł	Jedashkovs MIDI syste Nosphatidyl	kaya et al. m. Summé choline, <i>P</i> I	(2004, 200 ed feature E phosphi	<mark>07b);</mark> Oh et 3 contain: atidylethar	t al. (2012) 15 C _{16:1} <i>ω7</i> 10 nolamine,	; Park et al. (c and/or C _{16:} <i>DPG</i> diphos	2010); Rau 1 <i>യ6c</i> iphatidylgl	i et al. (2013 lycerol, <i>AP</i> L	l); Van Trapț unidentifie	pen et al. (2 :d aminopf	004); Yoon ospholipid,	et al. (2005a , AL unider	ı, b, 2006); an tified aminol	id Young e	t al. (2009) identifiec
phospholipid, L un	videntified	lipid, <i>i</i> isc	o, ai anteiso																				

Table 43.4 Differentiating phenotypic properties of type strains of Belliella

Characteristic	Belliella baltica BA134 ^T	Belliella pelovolcani CC-SAL-25 ^T
Cell size (width $ imes$ length) in μ m	0.3–0.7 × 1.5–3.0	0.3 × 2.0–5.0
Colony color	Pink/orange	Pink/red
Temperature range for growth (°C)	4–37	25–40
Optimum temperature for growth (°C)	25	37
pH range for growth	6–10	6–9
Salinity range for growth (%)	0–6	0.5–6
Nitrate reduction to nitrite	+	_
H ₂ S production	_	+
Hydrolysis of gelatin	-	+
Esterase lipase	w	+
API 50 CH:		
D-Fructose	w	-
D-Galactose	+	-
D-Glucose	+	-
Methyl α-D-mannopyranoside	_	+
Methyl α-⊃-glucopyranoside	_	+
N-acetylglucosamine	-	+
Amygdalin	_	+
Arbutin	_	+
Salicin	w	+
Inulin	_	+
Raffinose	w	+
Glycogen	w	+
Xylitol	_	+
Gentiobiose	_	+
Turanose	_	+
d- Lyxose	_	+
D-Tagatose	_	+
D-Fucose	-	+
∟-Fucose	_	+
D-Arabitol	_	+
Potassium gluconate	-	+
Potassium 2-ketogluconate	_	+

Table 43.4 (continued)

Characteristic	Belliella baltica BA134 ^T	Belliella pelovolcani CC-SAL-25 [⊤]
Potassium	_	+
5-ketogluconate		
Utilization of:		
Citrate	-	+
Adonitol	-	+
Rhamnose	_	+
Major fatty acids	i-C _{15:1} G, i-C _{15:0} , ai- C _{15:0} , C _{17:1} ω6c, i-C _{17:1} ω9c, i-C _{17:0} 3-OH, SF3	i-C _{15:0} , C _{15:1} ω6c, C _{17:1} ω6c, i-C _{17:1} ω9c, i-C _{17:0} 3-OH, SF3
DNA G+C content (mol%)	35.3	40

Data taken from Brettar et al. (2004a); Arun et al. (2009)

Summed features represent groups of two or three fatty acids that cannot be separated by GLC with the MIDI system. Summed feature 3 contains C_{16:1} ω 7c and/or C_{16:1} ω 6c

+ Positive, - negative, w weakly positive, SF summed feature, i iso, ai anteiso

optimally at 20–25 °C (Shivaji et al. 2012) compared to *C. lianum*, which grows between 15 °C and 42 °C with optimum growth at 33 °C (Ying et al. 2006). Lowest optimum temperature was observed in *C. qasimii* with 18 °C and grows between 4 °C and 30 °C (Shivaji et al. 2012). And being chemoorganoheterotroph with strictly respiratory type of metabolism, strains of *Cyclobacterium* grow in peptone seawater medium and also in modified Zobell medium, containing peptone, yeast extract, and seawater, and utilize a number of organic compounds (including tricarboxylic acid [TCA] cycle intermediates also) in the presence of NaCl, as single carbon sources (Raj and Paveglio 1983). Differentiating phenotypic properties is listed in \bigcirc *Table 43.2*.

Cyclobacterium strains are non-fermentative aerobes, do not produce gas, but produce acid oxidatively from different carbohydrates and not from alcohols (Raj and Paveglio 1983). Strains do not hydrolyze agar, cellulose, chitin, starch, and urea, but aesculin is hydrolyzed by all except *C. qasimii* (Ying et al. 2006; Shivaji et al. 2012). Catalase and oxidase are positive in all strains, but *C. qasimii* is weakly positive for oxidase activity (Shivaji et al. 2012). *C. qasimii* hydrolyzes DNA, *C. marinum* can hydrolyze Tween 40, and *C. lianum* can hydrolyze Tween 20 strongly and Tween 40 and Tween 80 weekly (Ying et al. 2006) (**•** *Table 43.2*).

Because of relatively restricted biochemical potential (oxidation of few mono-, di-, tri-, and polysaccharides and some TCA cycle intermediates), strains of *Cyclobacterium* seem to survive on small biomolecules released due to the continuous

Table 43.5 Differentiating phenotypic properties of type strains of *Cecembia*

Characteristic	Cecembia Ionarensis LW9 ^T	Cecembia calidifontis TU-13 ^T
Cell size (width $ imes$ length) in μ m	0.5-0.6 × 1.5-3.0	0.5-0.7 × 1.0- 3.0
Colony color	Reddish orange	Red
Temperature range for growth (°C)	10–40	20–50
Optimum growth temperature (°C)	30–37	45
pH range for growth	7.5–10	6.5–10
Salinity range for growth (%)	0-4	0–7
NaCl optimum (%)	2	1
Nitrate reduction	_	+
Oxidase	_	+
β -Galactosidase	_	+
Hydrolysis of:		
Aesculin	-	+
Casein	_	+
Gelatin	-	+
Starch	_	+
Acid production from:		
D-Ribose	_	+
L-Xylose	-	+
Methyl	-	+
β -D-xylopyranoside		
∟-Sorbose	-	+
Inositol	_	+
D-Fucose	_	+
L-Fucose	_	+
D-Galactose	_	+
Methyl α-D-mannopyranoside	_	+
D-Melibiose	-	+
Inulin	_	+
D-Melezitose	_	+
Starch	_	+
Gentiobiose	_	+
D-Tagatose	_	+
Potassium	_	+
5-ketogluconate		
Major fatty acids	i-C _{15:0} , ai-C _{15:0} , i-C _{16:0} , i-C _{16:1} H, i-C _{17:1} <i>@9c</i> , i-C _{17:0} 3-OH, SF3	C _{15:0} , i-C _{15:0} , ai-C _{15:0} , i-C _{17:0} 30H, SF9

Table 43.5 (continued)

Characteristic	Cecembia Ionarensis LW9 ^T	Cecembia calidifontis TU-13 ^T
Major polar lipids	PE, AL1, AL2, L1-L7	PE, APL, AL, PL1, PL2, L1, L2
DNA G+C content (mol%)	40.5	40.5 ± 0.5

Data taken from Anil Kumar et al. (2012b); Albuquerque et al. (2012)

Summed features represent groups of two or three fatty acids that cannot be separated by GLC with the MIDI system. Summed feature 3 contains $C_{16:1}$ $\omega 7c$ and/or $C_{16:1}$ $\omega 6c$. Summed feature 9 contains i- $C_{17:1}$ $\omega 9c$ and/or 10-methyl- $C_{16:0}$

+ Positive, – negative, SF summed feature, PE phosphatidylethanolamine, AL unidentified aminolipid, APL unidentified aminophospholipid, PL unidentified phospholipid, L unidentified lipid, i iso, ai anteiso

biochemical turnover of large macromolecules by other life forms in the marine environment (Raj and Paveglio 1983). Glucose oxidized mainly via the Embden-Meyerhof pathway (glycolysis), while gluconate is catabolized mainly via the Entner-Doudoroff pathway, both in conjunction with the TCA cycle as a secondary pathway and with some participation of the pentose phosphate pathway (Raj and Paveglio 1983). All strains utilized L-arabinose, D-arabinose, xylose, rhamnose, cellobiose, melibiose, melezitose, raffinose, trehalose, glucose, lactose, maltose, fructose, galactose, sucrose, mannose, inulin, ribose, methyl α-D-mannoside, and glutamic acid (Nedashkovskaya and Ludwig 2011; Shivaji et al. 2012). Nitrate is not reduced; H₂S, indole, and acetoin (Voges-Proskauer reaction) are not produced (Nedashkovskaya and Ludwig 2011; Shivaji et al. 2012). Positive for β -galactosidase in all strains except C. *qasimii* (Ying et al. 2006; Shivaji et al. 2012). Adonitol, dulcitol, and malonate were not utilized in any of the strains (Nedashkovskaya and Ludwig 2011; Shivaji et al. 2012). Hydrolysis of aesculin DNA, gelatin, and Tween 20, 40, 80; acid production from D-glucose, methyl α -D-glucoside, and glycerol; and utilization of sorbose, salicin, mannitol, methyl α -D-glucoside, gluconate, methionine, cysteine, alanine, histidine; susceptibility to penicillin, kanamycin, streptomycin, and tetracycline act as distinguishing phenotypic characteristics (Table 43.2).

The predominant cellular fatty acids are iso- $C_{15:0}$, summed feature 3 (iso- $C_{15:0}$ 2-OH and/or $C_{16:1}$ $\omega7c$), iso- $C_{17:1}$ $\omega9c$, iso- $C_{17:0}$ 3-OH, and anteiso- $C_{15:0}$ (Nedashkovskaya and Ludwig 2011). The predominant cellular fatty acids of the type strains of the genus *Cyclobacterium* are straight-chain unsaturated, branched-chain unsaturated and saturated, explicitly iso- $C_{15:0}$, iso- $C_{15:1}$ G, $C_{15:1}$ $\omega6c$, $C_{17:1}$ $\omega6c$, iso- $C_{17:1}$ $\omega9c$, iso- $C_{17:1}$ $\omega9c$ and 10-methyl $C_{16:0}$) (**)** *Table 43.2*). The species of the genus differs among each other with respect to the

Table 43.6

Differentiating phenotypic properties of type strains of Echinicola

Characteristic	Echinicola pacifica KMM 6172 ^T	Echinicola jeungdonensis HMD3054 ^T	Echinicola shivajiensis AK12 ^T	Echinicola vietnamensis KMM6221 ^T
Colony color	Pink	Yellow orange	Orange	Light pink
Cell size (width \times length)	0.3–0.5 × 1.0–2.0	0.7–0.8 × 1.4–1.5	0.5–1.0 × 1.5–2.5	0.4–0.5 × 1.0–2.0
in μm				
Salinity growth range (%)	0–12	1–15	0.5–12	0–15
Temperature range for growth (°C)	6–41	15–42	5–40	6–44
Temperature optimum (°C)	25–28	20	30–37	30–32
Citrate utilization	+	nd	-	-
Nitrate reduction	-	+	+	-
H ₂ S production	+	nd	-	-
Arginine dihydrolase	-	-	+	-
L-Pyrrolidonyl arylamidase	_	nd	+	-
L-Proline arylamidase	-	nd	+	+
β -N-acetyl galactosaminidase	-	nd	+	-
ELLMAN	+	nd	+	_
Hydrolysis of:	I			
Aesculin	+	+	+	_
Agar	+	_	_	-
Gelatin	+	_	_	-
Starch	+	-	-	+
Tween 20/40/60/80	-/+/-/-	nd	-	-/+/-/-
Utilization of:				
Fructose	+	+	+	-
Arabinose	+	+	-	_
Cellobiose	+	+	+	-
Glucose	+	+	+	_
Rhamnose	+	+	-	-
Mannose	+	+	-	-
Melibiose	-	+	+	_
Sucrose	+	+	+	-
Galactose	+	+	+	-
Xylose	+	nd	-	-
Sorbitol	—	+	+	-
Lactose	+	+	_	-
Trehalose	+	+	+	-
Raffinose	-	+	+	_
Maltose	+	+	-	-

Characteristic	Echinicola pacifica KMM 6172 ^T	Echinicola jeungdonensis HMD3054 ^T	Echinicola shivajiensis AK12 ^T	Echinicola vietnamensis KMM6221 ^T
Major fatty acids (>5 %)	i-C _{15:0} , C _{16:1} <i>ω5c</i> , i-C _{17:0} 3OH, C _{17:1} <i>ω6c</i> , SF3, SF9	i-C _{15:0} , ai-C _{15:0} , i-C _{17:0} 3- OH, SF3, SF9	i-C _{15:0} , i-C _{17:0} 3OH, SF3, SF4, SF9	i-C _{15:0} , C _{16:1} <i>ω5c</i> , i-C _{17:0} 3OH, SF3, SF4, SF9
Polar lipids	PE, AL, L	nd	PE, APL, L	PE, AL, L
DNA G+C content (mol%)	44–45	46.9	46.2	45.9

Table 43.6 (continued)

Data taken from Nedashkovskaya and Ludwig (2011); Kim et al. (2011); and Srinivas et al. (2012)

Summed features represent groups of two or three fatty acids that cannot be separated by GLC with the MIDI system. Summed feature 3 contains $C_{16:1} \omega 7c$ and/or $C_{16:1} \omega 6c$. Summed feature 4 contains $C_{14:0}$ 2-OH and/or i- $C_{15:0}$ 2-OH. Summed feature 9 contains one or more of the fatty acids i- $C_{17:1} \omega 9c$ and 10-methyl- $C_{16:0}$ + Positive, – negative, *wnd* no data available, *SF* summed feature, *PE* phosphatidylethanolamine, *AL* unidentified aminolipid, *APL* unidentified aminophospholipid, *L* unidentified lipid, *i* iso, *ai* anteiso

presence/absence/abundance of listed predominant fatty acids except iso- $C_{15:0}$, anteiso- $C_{15:0}$, and summed feature 3 which are common predominant fatty acids among all taxa. In contrast with the other ring-forming genera (*Runella*, *Spirosoma*, and *Flectobacillus*) which comprise spermidine as the cellular polyamine, *C. marinum* contains homospermidine as the cellular polyamine (Hamana and Nakagawa 2001). MK7, phosphatidylethanolamine, and unidentified lipids were present in all species of the genus *Cyclobacterium* (Shivaji et al. 2012). Phosphatidylcholine and unidentified aminolipids were present in *C. marinum* and *C. qasimii* but absent in *C. amurskyense* (Shivaji et al. 2012). There is no data available for the presence of menaquinone and polar lipids in type strain *C. lianum*. The DNA G+C content (mol%) ranges between 40.5 and 45.2 among the type strains (**•** *Table 43.2*).

Algoriphagus Bowman, Nichols, and Gibson 2003, 1351^{VP}, Emend. Nedashkovskaya, Vancanneyt, Van Trappen, Vandemeulebroecke, Lysenko, Rohde, Falsen, Frolova, Mikhailov, and Swings 2004, 1762^{VP}, Emend. Nedashkovskaya, Kim, Kwon, Shin, Luo, Kim, and Mikhailov 2007b, 1993^{VP}

Al.go.ri.pha'gus. L. masc. n. *algor* cold; Gr. masc. n. *phagos* glutton; N.L. masc. n. *Algoriphagus* the cold eater.

Cells are aerobic rods, Gram stain negative, psychrotolerant, moderately halotolerant, non-flagellated, nonmotile, and nonspore forming. Morphological and chemotaxonomic properties are indicated in **O** *Table 43.1*.

At present the genus comprises twenty-three species, the type species of the genus is *Algoriphagus ratkowskyi*, and type strain is IC025^T (http://www.bacterio.cict.fr/a/algoriphagus.html).

Cells are slender rod shaped usually measuring between 0.3 and 0.7×0.3 and $10.0 \,\mu\text{m}$ (Nedashkovskaya and Ludwig 2011). No gliding motility produces nondiffusible carotenoid pigments but not flexirubin type of pigments. Colonies on MA are regular, circular, convex, shiny, and smooth, with entire edges

and pink/orange pigmented with diameter of 0.5-3 mm after cultivation for 48 h (Nedashkovskaya and Ludwig 2011). Type strains of the genus Algoriphagus have been isolated from diverse environments, namely, algal mats, athalassohaline lagoon, corals, freshwater, marine sediments, marine solar saltern, microbial mats in Antarctic lakes, nonsaline alkaline groundwater, oil-contaminated soil, sea ice, seawater, and soil (Nedashkovskaya and Ludwig 2011). Being a marine microorganism Algoriphagus strains require nearly seawater concentration of NaCl for optimal growth. Although absolute requirement of NaCl is not observed in many species, NaCl is required for the growth for seven species with concentrations ranging from 0.5 % to 1 % (w/v) NaCl. Tolerance varies from 3 % to 10 % among species (**D** *Table 43.3*). Temperature range for growth is between -2 °C and 42 °C. The type strains A. ratkowskyi and also A. antarcticus are psychrophilic and can able to grow between -2 °C and 25 °C with an optimum temperature for growth that is between 16 °C and 19 °C (Bowman et al. 2003; Van Trappen et al. 2004). A. faecimaris grows within a wide range of temperature of 0-42 °C (Li et al. 2011). Eight type strains are psychrotolerant and rest are mesophilic (Table 43.3). All strains do not form acid from inositol/mannitol and positive for oxidase, catalase, alkaline phosphatase, and β -galactosidase activities. Arginine dihydrolase and tryptophan deaminase are not produced. Aesculin is hydrolyzed. Cellulose (CM-cellulose and filter paper) and urea are not degraded, but agar, casein (9 strains), gelatin (14 strains), starch (13 strains), DNA (10 strains), Tween 20 (14 strains), Tween 40 (14 strains), Tween 80 (11 strains), and chitin may be decomposed (Table 43.3). Three strains A. aquimarinus, A. chordae, and A. jejuensis were able to hydrolyze agar (Nedashkovskaya et al. 2004; Lee et al. 2012). H₂S and indole are not produced. The species of the genus Algoriphagus have many common phenotypic features. However, they can be differentiated from each other by several phenotypic traits as shown in **S** Table 43.3.

Acid is produced from a number of carbon sources, namely, L-arabinose (7 strains), cellobiose (16 strains), D-fructose (11 strains), D-galactose (12 strains), D-glucose (20 strains), D-lactose (14 strains), maltose (18 strains), D-mannose

Table 43.7 Differentiating phenotypic properties of type strains of *Fontibacter*

Characteristic	Fontibacter flavus CC- GZM-130 [⊤]	Fontibacter ferrireducens SgZ-2 ^T
Cell size (width \times length) in μm	0.3–0.6 × 1.5–3.2	0.2–0.4 × 1.5–3
Colony color	Bright orange	Orange
Salinity growth range (%)	0–3.5	0–5
Temperature growth range (°C)	25–40	15–37
Temperature optimum (°C)	30	25–30
pH growth range	6–9.5	6.5–8.5
Oxidase	+	_
β -Galactosidase	-	+
Lysine decarboxylase	+	_
Ornithine decarboxylase	+	-
Voges-Proskauer reaction	_	+
Tryptophane deaminase	_	+
Esterase lipase (C8)	+	-
Acid phosphatase	+	-
β -Glucosidase	+	-
Trypsin	-	+
Acid production from:		
D-glucose	_	w
Maltose	w	+
Mannitol	-	-
Melibiose	-	+
Rhamnose	+	-
D-sucrose	-	+
Utilization of:		
D-Glucose	_	+
Salicoside	-	+
D-Melibiose	-	+
L-Fucose	_	+
D-Sorbitol	_	+
D-Sucrose	_	+
D-Maltose	_	+
Citrate	_	+
Major fatty acids	i-C _{15:0} , ai-C _{15:0} , i-C _{15:1} G, i-C _{17:0} 3OH, i-C _{17:1} ω9c	i-C _{15:0} , i-C _{17:0} 3OH, i-C _{17:1} ω9c

Table 43.7 (continued)

Characteristic	Fontibacter flavus CC- GZM-130 ^T	Fontibacter ferrireducens SgZ-2 [⊤]
Phospholipids	PE, AL, PL, L	PE, AL, L
DNA G+C content (mol%)	53.2	55.3

Data taken from Kämpfer et al. (2010) and Zhang et al. (2012)

+ Positive, – negative, w weakly positive, PE phosphatidylethanolamine, AL unidentified aminolipid, PL unidentified phospholipid, L unidentified lipid, *i* iso, *ai* anteiso

(15 strains), melibiose (14 strains), raffinose (12 strains), L-rhamnose (11 strains), sucrose (15 strains), trehalose (11 strains), DL-xylose (12 strains), and N-acetyl-D-glucosamine (9 strains). A. antarcticus does not produce acid from any of the substrates used and also cannot able to utilize those carbon sources (Van Trappen et al. 2004; Nedashkovskaya and Ludwig 2011). Similarly A. olei was not able to produce acid from the tested substates but able to utilize a number of substrates for growth (Young et al. 2009). A. ornithinivorans produces acid from only D-glucose (Nedashkovskaya et al. 2007b; Nedashkovskaya and Ludwig 2011), and A. lutimaris produces acid from only two substrates (cellobiose and sucrose) (Park et al. 2010). A. locisalis produces acid from all the tested substrates (Yoon et al. 2005b; Nedashkovskaya and Ludwig 2011). Rest of the type strains varies among each other in acid production from diverse carbon sources ranging between 29 % and 93 %. Strains can utilize a number of carbon substrates L-arabinose, D-galactose, D-glucose, D-lactose, maltose, D-mannose, D-xylose, D-mannitol, sorbitol, myo-inositol, glycerol, and N-acetyl-D-glucosamine but vary among each other (Young et al. 2009; Nedashkovskaya and Ludwig 2011) (Table 43.3). A. ratkowskyi was able to utilize all the tested carbon sources except glycerol (Young et al. 2009; Nedashkovskaya and Ludwig 2011). A. faecimaris utilizes only three substrates (D-glucose, myo-inositol, and N-acetyl-D-glucosamine) (Li et al. 2011). Algoriphagus strains vary among each other with respect to the susceptibility to gentamicin and tetracycline (**)** Table 43.3). The complete genome sequence of Algoriphagus sp. strain PR1 revealed the presence of required genes for a complete TCA cycle and complete glycolysis and pentose phosphate pathways. The genome encodes the presence of 62 glycoside hydrolases, 71 glycosyltransferases, 2 polysaccharide lyases, and 10 carbohydrate esterases, comprising a lofty aptitude for polysaccharide degradation (Alegado et al. 2011) which is evident from their phenotypic properties. Algoriphagus sp. PR1 forms pink-pigmented colonies. The genome analysis also reveals the presence of genes which encodes numerous carotenoid biosynthetic enzymes which are required for the biosynthesis of carotenoids which impart color to the cells (Alegado et al. 2011). In *Algoriphagus* sp. strain KK10202C (synthesize Zeaxanthin), seven genes were transcribed in the same direction, among which five of them were involved in carotenoid synthesis. This cluster had a unique gene organization, with an isoprenoid gene, *ispH* (previously named *lytB*), being present among the carotenoid synthesis genes. The lycopene β -cyclase encoded by the *crtYcd* gene appeared to be a fusion of bacterial heterodimeric lycopene cyclase CrtYc and CrtYd (Tao et al. 2006).

The major cellular fatty acids are unsaturated, and branched-chain saturated fatty acids iso-C_{15:0}, iso-C_{17:0} 3-OH, and summed feature 3 comprising iso-C_{15:0} 2-OH and/or C_{16:1} ω7c (Nedashkovskaya and Ludwig 2011) (**)** Table 43.3) present in all type strains except in A. jejuensis (Lee et al. 2012). A. jejuensis, A. aquatilis, and A. boritolerans comprise iso- $C_{15:0}$ and summed feature 3 as major fatty acids (>5 %) and also contain the third dominant fatty acid iso-C_{17:0} 3-OH in slightly lesser (4.7, 4.0, and 4.9%, respectively) quantity (Liu et al. 2009; Nedashkovskaya et al. 2007b). A. faecimaris comprises iso-C_{15:0} and iso-C_{17:0} 3-OH as major fatty acids but contains less amount of summed feature 3 (3.2 %) (Li et al. 2011). Algoriphagus species were distinguished based on the presence/absence/concentration of other major fatty acids, namely, iso-C_{15:1} G, anteiso-C15:0, iso-C15:0 3-OH, C16:0, iso-C16:0, iso-C16:1, C16:1 ω5c, C_{17:1} ω6c, iso-C_{17:1} ω9c, and C_{18:0} (**)** Table 43.3). The major respiratory quinone is MK7 (Nedashkovskaya and Ludwig 2011). Phosphatidylethanolamine (PE) and unidentified polar lipid (L) were commonly present among the tested Algoriphagus species (Table 43.3). Apart from the common polar lipids, A. aquaeductus comprises unidentified aminophospholipid (APL) and unidentified phospholipid (PL) (Rau et al. 2012); A. jejuensis and A. terrigena comprise diphosphatidylglycerol (DPG), APL, and unidentified aminolipid (AL) (Lee et al. 2012); A. namhaensis comprises phosphatidylcholine (PC) (Oh et al. 2012); A. olei comprises AL, APL, and PL (Young et al. 2009); and A. ratkowskyi comprises PC and PL (Bowman et al. 2003) (Table 43.3). The DNA G+C content varies between 35 and 49 mol% among the type strains of the genus Algoriphagus (Table 43.3).

Belliella Brettar, Christen and Höfle 2004a, 69^{VP}, Emend. Anil Kumar, Srinivas, Madhu, Sravan, Shashi Singh, Naqvi, Mayilraj and Shivaji 2012, 2256^{VP}

Bel.li.el'la. N.L. fem. dim. n. *Belliella* named in honor of the aquatic microbiologist Russell Bell of the University of Uppsala.

Cells are aerobic rods, Gram stain negative, psychrotolerant/ mesophilic, moderately halotolerant, nonmotile (nonflagellated and no gliding motility), and nonspore forming. Morphological and chemotaxonomic properties are listed in **O** *Table 43.1*. At present the genus comprises two species, *Belliella baltica* $BA134^{T}$ and *Belliella pelovolcani* CC-SAL-25^T. The type species of the genus is *Belliella baltica*; type strain is $BA134^{T}$ (Brettar et al. 2004a). Differentiating phenotypic properties is listed in \bigcirc *Table 43.4*.

Cells are slender rods and occur as single or chains of up to five cells usually measuring between 0.3 and 0.7 \times 1.5 and 5.0 µm (Arun et al. 2009). Produce carotenoid pigments but not flexirubin type of pigments. Colonies on MA are regular, circular, smooth, and convex with entire edges, pink pigmented and transparent when young, but become orange and opaque with long incubation (>2 weeks at 20 °C) (Brettar et al. 2004a). Colony edges are spreading type in case of B. pelovolcani (Arun et al. 2009). Type strains of the genus Belliella have been isolated from marine or estuarine or mud volcano (Brettar et al. 2004a; Arun et al. 2009). B. baltica does not require NaCl for growth, but B. pelovolcani requires 0.5 % (w/v) NaCl; both tolerate up to 6 % (w/v) NaCl and grow optimally at 3 % (w/v) NaCl (Table 43.4). B. baltica grows in between 4 °C and 37 °C with optimum at 25 °C, but B. pelovolcani grows in between 25 °C and 40 °C with optimum at 37 °C (Brettar et al. 2004a; Arun et al. 2009). Oxidase and catalase positive. Nitrate is reduced to nitrite by B. baltica (Nedashkovskaya and Ludwig 2011); aesculin, starch, and DNA are hydrolyzed but not gelatin. Chemoorganoheterotrophic. Strain B. baltica was positive for acid production from L-arabinose, D-xylose, galactose, D-glucose, D-fructose, aesculin, salicin, cellobiose, maltose, lactose, melibiose, sucrose, trehalose, D-raffinose, and starch; enzymic activities: α - and β -glucosidases, β -galactosidase, acidic and alkaline phosphatases, lipase (C8), leucine, valine and cystine arylamidases, chymotrypsin, trypsin, and naphthol phosphohydrolase; assimilation of glucose, arabinose, and maltose; and utilization of D-galactose, gentiobiose, a-D-glucose, α -D-lactose, lactulose, maltose, D-trehalose, acetic acid, α -ketobutyric/glutaric/valeric acids, and L-glutamic acid as substrates (Brettar et al. 2004a). B. pelovolcani oxidized a number of carbon sources, namely (positive with the Biolog GN2 system), α -cyclodextrin, dextrin, glycogen, Tween 40, Tween 80, L-arabinose, cellobiose, i-erythritol, D-fructose, D-galactose, gentiobiose, α -D-glucose, α -D-lactose, lactulose, maltose, D-mannose, melibiose, methyl β -D-glucoside, D-psicose, raffinose, sucrose, trehalose, turanose, succinic acid monomethyl ester, acetic acid, L-alaninamide, L-alanine, L-alanyl glycine, L-glutamic acid, L-ornithine, L-threonine, inosine, uridine, thymidine, α -D-glucose 1-phosphate, and D-glucose 6-phosphate; carbon sources not oxidized include (negative with the Biolog GP2 system) N-acetyl-D-galactosamine, N-acetyl-D-glucosamine, adonitol, D-arabitol, L-fucose, myo-inositol, D-mannitol, L-rhamnose, D-sorbitol, xylitol, pyruvic acid methyl ester, cis-aconitic acid, citric acid, formic acid, D-galactonic acid lactone, D-gluconic acid, D-galacturonic acid, D-glucosaminic acid, D-glucuronic acid, α-hydroxybutyric β -hydroxybutyric acid, γ-hydroxybutyric acid. acid. p-hydroxyphenylacetic acid, itaconic acid, α-ketobutyric acid,

 α -ketoglutaric acid, α -ketovaleric acid, DL-lactic acid, malonic acid, propionic acid, quinic acid, D-saccharic acid, sebacic acid, succinic acid, bromosuccinic acid, succinamic acid. glucuronamide, D-alanine, L-asparagine, L-aspartic acid, glycyl L-aspartic acid, glycyl L-glutamic acid, L-histidine, hydroxy-Lproline, L-leucine, L-phenylalanine, L-proline, L-pyroglutamic acid, D-serine, L-serine, DL-carnitine, y-aminobutyric acid, urocanic acid, phenylethylamine, putrescine, 2-aminoethanol, 2,3-butanediol, glycerol, and $DL-\alpha$ -glycerol phosphate; positive for β -galactosidase, H₂S production, citrate utilization, acetoin production, and gelatinase (in API 20E system); and aesculin, gelatinase and β -galactosidase (in API 20NE) and alkaline and acid phosphatase, butyrate esterase, caprylate esterase (C8), leucine arylamidase, valine arylamidase, cystine arylamidase, trypsin, α-chymotrypsin, naphthol-AS-B-1-phosphohydrolase, β -galactosidase, and α -glucosidase (in API ZYM) (Arun et al. 2009).

Both strains contain unsaturated, branched saturated/unsaturated fatty acids, namely, iso- $C_{15:0}$, $C_{17:1} \omega 6c$, iso- $C_{17:1} \omega 9c$, iso- $C_{17:0}$ 3-OH, and summed feature 3 comprising iso- $C_{15:0}$ 2-OH and/or $C_{16:1} \omega 7c$ as predominant fatty acids (Arun et al. 2009). Other dominant fatty acids are iso- $C_{15:1}$ G, anteiso- $C_{15:0}$, and $C_{15:1} \omega 6c$ (**)** *Table 43.4*). The major respiratory quinone is MK7 (Nedashkovskaya and Ludwig 2011; Anil Kumar et al. 2012b). *B. baltica* comprises phosphatidylethanolamine (PE), two unidentified phospholipids (PL), and nine unidentified lipids (L) (Anil Kumar et al., 2012b). The DNA G+C content varies between 35.3 and 40 mol% (**)** *Table 43.4*).

Cecembia Anil Kumar, Srinivas, Madhu, Sravan, Shashi Singh, Naqvi, Mayilraj, and Shivaji 2012, 2256^{VP}

Ce.cem'bi.a. N.L. fem. n. *Cecembia*, arbitrary name derived from the abbreviation CCMB Centre for Cellular and Molecular Biology.

Cells are aerobic rods, Gram stain negative, mesophilic/ slightly thermophilic, moderately halotolerant, and nonmotile (nonflagellated and no gliding motility). Morphological and chemotaxonomic properties are indicated in **2** *Table 43.1*.

At present the genus comprises two species, *Cecembia lonarensis* LW9^T and *Cecembia calidifontis* TU-13^T. The type species of the genus is *Cecembia lonarensis*, type strain is LW9^T (Anil Kumar et al. 2012b). Differentiating phenotypic properties is listed in Table 43.5.

Cells are slender rods occur singly measuring between 0.5 and 0.7×1.0 and $3.0 \,\mu\text{m}$ (Anil Kumar et al. 2012b; Albuquerque et al. 2012b). Colonies on half-strength MA are regular, circular, grow up to 2–3 mm in diameter, smooth texture, reddish orange pigmented, translucent, and raised with entire margins (Anil Kumar et al. 2012b). Produce carotenoid pigments but not flexirubin type of pigments. Type strains of

the genus Cecembia have been isolated from haloalkaline water or hot spring runoff (Anil Kumar et al. 2012b; Albuquerque et al. 2012). Both taxa do not require NaCl for growth but differed with respect to optimum and tolerance (Table 43.5). C. lonarensis grows between temperature range of 10 and 40 °C, optimally between 30 °C and 37 °C but C. calidifontis between 20 °C and 50 °C, optimally at 45 °C (Table 43.5). Catalase activity is positive. Oxidase and β -galactosidase activities and nitrate reduction vary among species (Table 43.5). Aesculin, casein, gelatin, and starch are hydrolyzed by C. calidifontis but not C. lonarensis (Table 43.5). C. lonarensis cannot produce acid from D-ribose, L-xylose, methyl β -D-xylopyranoside, L-sorbose, inositol, D-fucose, L-fucose, D-galactose, methyl α -D-mannopyranoside, D-melibiose, inulin, D-melezitose, starch, gentiobiose, D-tagatose, and potassium 5-ketogluconate, but C. calidifontis produces acid from the above substrates (Albuquerque et al. 2012). C. lonarensis can utilize adonitol, cellobiose, citrate, D-arabinose, fructose, glucose, lactose, L-arabinose, maltose, mannose, malonate, melibiose, raffinose, rhamnose, sucrose, and xylose but cannot utilize amygdalin, dulcitol, galactose, glucosamine, glycerol, inositol, inulin, mannitol, melezitose, methyl α -D-glucoside, methyl α -D-mannoside, ribose, salicin, sodium gluconate, sorbitol, sorbose, trehalose, and xylitol. C. calidifontis requires yeast extract for growth. C. calidifontis is positive for alkaline phosphatase, esterase (C4), esterase lipase (C 8), leucine arylamidase, valine arylamidase, cystine arylamidase, trypsin, *a*-chymotrypsin, acid phosphatase, naphthol-AS-BI-phosphohydrolase, α -galactosidase, β -galactosidase, α -glucosidase, β -glucosidase, N-acetyl- β -glucosaminidase, and α-mannosidase (in API ZYM); utilizes D-glucose, D-fructose, D-galactose, D-mannose, L-rhamnose, L-fucose, D-ribose, D-xylose, D-arabinose, L-arabinose, sucrose, maltose, lactose, D-cellobiose, D-trehalose, D-raffinose, D-melibiose, glycerol, α -ketoglutarate, DL-lactate, acetate, pyruvate, succinate, malate, citrate, fumarate, D-gluconate, D-glucuronate, aspartate, glutamate, L-alanine, L-asparagine, L-histidine, L-lysine, proline, L-glutamine, L-arginine, L-serine, valine, L-phenylalanine, L-leucine, L-isoleucine, and L-ornithine; but not L-sorbose, ribitol, xylitol, sorbitol, mannitol, myo-inositol, erythritol, D-arabitol, L-arabitol, benzoate, formate, glycine, and methionine (Albuquerque et al. 2012).

The predominant fatty acids present in *C. lonarensis* are iso- $C_{15:0}$, anteiso- $C_{15:0}$, iso- $C_{16:1}$, iso- $C_{16:1}$ H, summed feature 3 (comprising $C_{16:1} \omega 7c/C_{16:1} \omega 6c$), iso- $C_{17:1} \omega 9c$, and iso- $C_{17:0}$ $_{3-OH}$ (Anil Kumar et al. 2012b). The predominant fatty acids present in *C. calidifontis* are $C_{15:0}$, i- $C_{15:0}$, ai- $C_{15:0}$, i- $C_{17:0}$ 3OH, and summed feature 9 comprising iso- $C_{17:1} \omega 9c$ and/or $C_{16:0}$ 10-methyl. Menaquinone MK7 is the only respiratory quinone. The polar lipids consist of phosphatidylethanolamine, unidentified aminolipids, and unidentified lipids (**D** *Table* 43.5). *C. calidifontis* also comprises unidentified aminophospholipids and unidentified phospholipids (**D** *Table* 43.5). The DNA G+C content is 40.5 for both taxa, but one was determined by spectrophotometrically and other by HPLC (Anil Kumar et al. 2012b; Albuquerque et al. 2012 respectively).

Echinicola Nedashkovskaya, Kim, Vancanneyt, Lysenko, Shin, Park, Lee, Jung, Kalinovskaya, Mikhailov, Bae, and Swings 2006, 955^{VP}

E.chi.ni.co'la. L. masc. n. *echinus -i* a sea urchin; L. suff. *-cola* derived from L. masc. or fem. n. *incola* a dweller; N.L. fem. n. *Echinicola* a sea urchin dweller.

Cells are aerobic, Gram stain negative, mesophilic, halotolerant, and nonflagellated but motile by gliding. Morphological and chemotaxonomic properties are listed in • *Table 43.1*.

At present the genus comprises four species, *Echinicola pacifica* KMM 6172^{T} , *Echinicola jeungdonensis* HMD3054^T, *Echinicola shivajiensis* AK12^T, and *Echinicola vietnamensis* KMM6221^T. The type species of the genus is *Echinicola pacifica*, type strain is KMM 6172^{T} . Differentiating phenotypic properties is listed in \bigcirc *Table 43.6*.

Cells are rod shaped measuring 0.3–1.0 \times 1.0–2.5 μ m. Colonies on MA are circular, 2-3 mm in diameter, convex, shiny, pink pigmented, and sunken smooth. into agar (Nedashkovskaya et al. 2006). Colonies of E. jeungdonensis on MA are convex, circular, smooth, and yellow orange pigmented with entire margin (Kim et al. 2011). Colonies of E. shivajiensis on MA are circular, 1-3 mm in diameter, smooth, shiny, orange, translucent, and raised with entire margin (Srinivas et al. 2012). Color of the colonies is due to nondiffusible carotenoid pigments, and flexirubin-type pigments are not present. Type strains were isolated from different marine environments, namely, sea urchin Strongylocentrotus intermedius, solar saltern, brackish water pond, and seawater from mussel farm (Nedashkovskaya and Ludwig 2011; Kim et al. 2011; Srinivas et al. 2012). Being a marine microorganism Echinicola strains have definite NaCl requirement for optimal growth, but NaCl requirement is not observed in E. pacifica and E. vietnamensis. Tolerance varies from 12 % to 15 % among species (Nedashkovskaya and Ludwig 2011; Kim et al. 2011; Srinivas et al. 2012). Temperature range for growth is between 5 °C and 44 °C (with optimum growth temperature range is 20–37 °C). E. jeungdonensis grows within a narrow temperature range from 15 °C to 42 °C but optimally at 20 °C (Kim et al. 2011), and others grow at wider growth temperatures () Table 43.6). Aerobic, chemoorganoheterotrophic, and can ferment D-glucose. Differentiating phenotypic properties displayed in

Positive for catalase, oxidase, and β -galactosidase activities. Aesculin is hydrolyzed by all type strains except *E. vietnamensis*. Agar and gelatin are hydrolyzed by *E. pacifica*. Starch and Tween 40 are hydrolyzed by *E. pacifica* and *E. vietnamensis* (**Table 43.6**). Nitrate reduction; citrate utilization; H₂S production, arginine dihydrolase, L-pyrrolidonyl arylamidase, L-proline arylamidase, and β -N-acetylgalactosaminidase activities; and ELLMAN reaction vary among the type strains. *E. vietnamensis* does not able to utilize arabinose, cellobiose, fructose, galactose, glucose, lactose, maltose, mannose, melibiose, raffinose, rhamnose, sorbitol, sucrose, trehalose, and xylose, but the three type strains utilize most of the substrates but vary with each other (**•** *Table 43.6*). Acid is produced by a number of substrates L-arabinose, D-cellobiose, D-glucose, D-lactose, D-maltose, D-mannose, L-rhamnose, and DL-xylose, but it varies within the species (Nedashkovskaya and Ludwig 2011; Kim et al. 2011; Srinivas et al. 2012).

The main cellular fatty acids are straight-chain unsaturated and branched-chain unsaturated fatty acids, namely, iso-C15-02 iso-C_{17:0} 3-OH, summed feature 3 (iso-C_{15:0} 2-OH and/or C_{16:1} ω 7c), and summed feature 9 (iso-C_{17:1} ω 9c and/or C_{16:0} 10-methyl) (Kim et al. 2011; Srinivas et al. 2012). The species of the genus differs among each other with respect to the presence/absence/abundance of listed predominant fatty acids except anteiso-C15:0, C16:1 w5c, C17:1 w6c, and summed feature 4 (C_{14:0} 2-OH and/or iso-C_{15:0} 2-OH) (**)** *Table 43.6*). The major respiratory quinone is MK7; phosphatidylethanolamine and unidentified lipids were present in all tested species of the genus Echinicola (Table 43.6). Unidentified aminolipids were present in E. pacifica and E. vietnamensis, and unidentified aminophospholipid was present in E. shivajiensis. The DNA G+C content (mol%) ranges between 44 and 46.9 among the type strains (**S** Table 43.6).

Fontibacter Kämpfer, Young, Chen, Rekha, Fallschissel, Lodders, Chou, Shen, Frischmann, Busse, and Arun 2010, 2067^{VP}

Fon.ti.bac'ter. L. n. *fons* fontis a spring; N.L. masc. n. *bacter* a rod; N.L. masc. n. *Fontibacter* a rod isolated from a spring sample.

Cells are aerobic rods, Gram stain negative, mesophilic, nonmotile, nonflagellated, and nonspore forming. Morphological and chemotaxonomic properties are indicated in **S** *Table 43.1*.

At present the genus comprises two species, *Fontibacter flavus* CC-GZM-130^T and *Fontibacter ferrireducens* SgZ-2^T. The type species of the genus is *Fontibacter flavus*, type strain is CC-GZM-130^T (Kämpfer et al. 2010).

Cells are slender rods, with rounded ends, occur singly measuring between 0.2 and 0.5×1.5 and 3.0 µm (Kämpfer et al. 2010; Zhang et al. 2012). Differentiating phenotypic properties is listed in \bigcirc *Table 43.7*.

Colonies on nutrient agar or tryptic soy agar (TSA) are circular, smooth, orange pigmented, and convex with entire edges, growing up to 4 mm (Kämpfer et al. 2010; Zhang et al. 2012). Type strains of the genus *Fontibacter* have been isolated from hot spring and from anode biofilm of a wastewater-fed microbial fuel cell that was continuously operated for 3 months

(Kämpfer et al. 2010; Zhang et al. 2012). F. flavus requires NaCl for growth and tolerates up to 3 % w/v, but F. ferrireducens does not require NaCl for growth but optimal growth at 1 % and tolerates up to 5 % w/v (Table 43.7). Mesophilic, grows between 15 °C and 40 °C, and optimally between 25 °C and 30 °C (O Table 43.7). Catalase activity is positive. Oxidase, β -galactosidase, lysine decarboxylase, ornithine decarboxylase, tryptophan deaminase, esterase lipase, acid phosphatase, and β -glucosidase activities vary among both species (Table 43.5). Aesculin hydrolysis is negative (Kämpfer et al. 2010; Zhang et al. 2012). F. ferrireducens is negative for methyl red test; indole and H₂S production; and hydrolysis of gelatin, starch, and urea (Zhang et al. 2012). F. flavus hydrolyzes bis-pNP-phosphate, L-glutamate-y-3-carboxy-p-nitroanilide (NA), o-nitrophenyl (NP)- β -D-galactopyranoside, pNP- α -Dglucopyranoside, $pNP-\beta$ -D-glucopyranoside, and pNP-phosphoryl-choline; cellobiose, D-fructose, D-galactose, D-glucose, maltose, D-mannose, melibiose, sucrose, and trehalose utilized (weekly) as sole carbon sources; but not acetate, N-acetylgalactosamine. adipate, D-adonitol, L-alanine, β -alanine, 4-aminobutyrate, L-aspartate, azelate, cis-aconitate, citrate, fumarate, gluconate, glutarate, L-histidine, 3-hydroxybenzoate, 4-hydroxybenzoate DL-3-hydroxybutyrate, itaconate, DL-lactate, L-leucine, L-malate, D-mannitol, mesaconate, L-ornithine, oxoglutarate, L-phenylacetate, L-phenylalanine, L-proline, propionate, pyruvate, D-ribose, L-serine, suberate, trans-aconitate, and L-tryptophan (Kämpfer et al. 2010). F. ferrireducens produces acid from glucose, maltose, melibiose, and sucrose but F. flavus from rhamnose and weekly from maltose (Table 43.7). F. ferrireducens utilizes L-fucose, D-glucose, D-melibiose, salicoside, D-sorbitol, D-sucrose, and D-maltose, but does not utilize N-acetylglucosamine, L-alanine, L-arabinose, capric acid, citrate, glycogen, 1-histidine, 3-hydroxybenzoic acid, 4-hydroxybenzoic acid, 3-hydroxybutyric acid, inositol, itaconic acid, D-mannitol, potassium 2-ketogluconate, potassium 5-ketogluconate, L-proline, propionic acid, L-rhamnose, D-ribose, L-serine, sodium acetate, sodium lactic acid, suberic acid, and valeric acid; positive for activities of N-acetyl- β glucosaminidase, alkaline phosphatase, α -chymotrypsin, cystine arylamidase, esterase C4, α -glucosidase, leucine arylamidase, valine arylamidase, and β -galactosidase (in API ZYM) (Zhang et al. 2012). The other distinguishing characteristics of F. ferrireducens are it reduces Fe (III) to Fe (II) and can grow both aerobically and also anaerobically (Zhang et al. 2012).

The common predominant fatty acids (>5 %) present in *Fontibacter* are branched saturated and unsaturated, namely, iso- $C_{15:0}$, iso- $C_{17:0}$ 3OH, iso- $C_{17:1} \omega 9c$, but *F. flavus* also comprises anteiso- $C_{15:0}$ and iso- $C_{15:1}$ G as predominant fatty acids (**Table 43.7**). Menaquinone MK7 is the major respiratory quinone system. The common polar lipids present are phosphatidylethanolamine, unidentified aminolipids, and unidentified lipids (**Table 43.7**). *F. flavus* also comprises unidentified phospholipid (**Table 43.7**). The DNA G+C content is between 53.2 and 55.3 mol% (**Table 43.7**).

Aquiflexum, Indibacter, Litoribacter, Mongoliicoccus, Mongoliitalea, Nitritalea, Rhodonellum, and Shivajiella are monospecific genera. Some of their salient properties have been covered under "short description of genera" and in • *Table 43.1*.

Aquiflexum Brettar, Christen, and Höfle 2004b, 2339^{VP}

A.qui.fle'xum. L. fem. n. *aqua* water; L. part. adj. *flexus -a -um* bent, winding; N.L. neut. n. *Aquiflexum* to indicate the bacterium's aquatic origin and its long flexible rods.

The type strain of *Aquiflexum balticum* BA160^T was isolated from surface water of the central Baltic Sea (Brettar et al. 2004b) and is oxidase and catalase positive. Cells are long, straight rods measuring 0.3–0.6 × 1.1–4.8 µm. Colonies on half-strength MA after 2–3 days of incubation at 30 °C are circular, smooth, convex, red pigmented and transparent and entire, but after 1 week incubation under similar conditions colonies become opaque (Brettar et al. 2004b). Cells contain carotenoids but not flexirubin type of pigments. NaCl is not required for growth, but optimal growth obtained with 1.5 % w/v NaCl and tolerates up to 6 % w/v NaCl (Brettar et al. 2004b; Nedashkovskaya and Ludwig 2011). Growth occurs between 4 °C and 40 °C (optimum around 30 °C) and pH 7.0–9.0 (optimum pH at 7.0) (Nedashkovskaya and Ludwig 2011).

Hydrolyzes aesculin, gelatin, and starch but not cellulose and tyrosine. Indole is not produced. Does not grow on media containing 0.5 % yeast extract, casein, DNA, chitin, or pectin. Aminopeptidase was produced (Brettar et al. 2004b).

In API 50CHE system, acid is produced from *N*-acetylglucosamine, amygdalin, arbutin, cellobiose, esculin, D-fructose, L-fucose, galactose, β -gentiobiose, D-glucose, glycogen, inulin, 5-ketogluconate, lactose, maltose, D-mannose, melezitose, melibiose, α -methyl-D-glucoside, α -methyl-D-mannoside, D-raffinose, rhamnose, salicin, sucrose, starch, trehalose, D-turanose, and xylitol (Brettar et al. 2004b).

In API 20NE and API ZYM systems, positive for *N*-acetyl- β -glucosaminase acid and alkaline phosphatase; chymotrypsin; β -galactosidase; α - and β -glucosidase; leucine-, valine-, and cystine arylamidase; trypsin; and naphthol phosphohydrolase (Brettar et al. 2004b).

In API 20NE system, assimilated glucose, arabinose, mannose, *N*-acetylglucosamine, maltose, and gluconate (Brettar et al. 2004b).

In Biolog GN2 system, utilized acetic acid, L-arabinose, cellobiose, L-fructose, D-galactose, gentiobiose, α -D-glucose, α -ketoglutaric acid, lactic acid, α -D-lactose, lactulose, maltose, D-mannose, β -methyl-D-glucoside, monomethyl-succinate, propionic acid, D-psicose, D-sorbitol, sucrose, D-trehalose, and turanose (Brettar et al. 2004b).

Cells contains high percentage of branched-chain fatty acid with predominant fatty acids (>5 %) being iso- $C_{15:0}$, anteiso- $C_{15:0}$, iso- $C_{15:1}$ G, and iso- $C_{16:1}$ H. The polar lipids comprise phosphatidylethanolamine, unidentified aminolipid, unidentified phospholipid, and unidentified lipids. The genomic DNA G+C content is 38.4 mol% (Brettar et al. 2004b). Indibacter Anil Kumar, Srinivas, Madhu, Manorama and Shivaji 2010, 725^{VP}, Emend. Anil Kumar, Srinivas, Madhu, Sravan, Shashl Singh, Naqvi, Mayilraj, and Shivaji 2012, 2256^{VP}

In.di.bac'ter. L. n. *India* India; N.L. masc. n. *bacter* a rod; N.L. masc. n. *Indibacter* a rod from India, referring to the isolation of the bacterial strain from India.

The type strain of *Indibacter alkaliphilus* LW1^T was isolated from a water sample collected at a depth of 3.5 m from Lonar Lake, is catalase and oxidase positive. Cells are Gram stain negative, nonmotile, aerobic, slender rods occur singly, measuring $0.5-0.7 \times 2.0-3.0 \mu$ m. Colonies on half-strength MA after 2–3 days, incubated at 30 °C are circular, smooth, convex, red orange pigmented and opaque with entire margin, and grows up to 2–3 mm in diameter. Cells contain carotenoids but no flexirubin type of pigments. NaCl is not required for growth, but optimal growth is between 0.5 % and 6 % w/v NaCl and tolerates up to 8 % w/v NaCl. Growth occurs between 15 °C and 40 °C (optimum between 30 °C and 37 °C) and pH 7.5–12.0 (optimum pH at 10.0) (Anil Kumar et al. 2010a).

Positive for ornithine and lysine decarboxylase activities; hydrolysis of aesculin and starch; nitrate is reduced to nitrite; production of acid from D-arabinose, cellobiose, D-fructose, D-glucose, lactose maltose, melezitose, melibiose, L-rhamnose and xylose; assimilation of adonitol, aesculin, D-arabinose, L-arabinose, fructose, galactose, glucosamine, glucose, lactose, mannose, maltose, melibiose, raffinose, rhamnose, sucrose, trehalose and xylose; but negative for arginine dihydrolase, β -galactosidase, tryptophan deaminase and urease activities; methyl red and Voges-Proskauer reactions; hydrolysis of Tween 60, Tween 85 and gelatin; H₂S gas production; production of acid from adonitol, aesculin, dulcitol, erythritol, D-galactose, inositol, inulin, mannitol, D-mannose, raffinose, salicin, D-sorbitol sucrose, trehalose and xylitol; assimilation of amygdalin, citrate, dulcitol, glycerol, inositol, inulin, malonate, mannitol, melezitose, methyl a-D-glucoside, methyl a-Dmannoside, ONPG, ribose, salicin, sodium gluconate, sorbitol, sorbose, and xylitol (Anil Kumar et al. 2010a).

Cells comprise high percentage of branched-chain fatty acid with predominant fatty acids (>5 %) being iso- $C_{15:0}$, iso- $C_{17:1}$ ω 9*c*, iso- $C_{17:0}$ 3-OH, and anteiso- $C_{15:0}$. MK7 is the major respiratory quinone. The polar lipids consist of phosphatidylethanolamine, unidentified aminolipid, and unidentified lipids. The genomic DNA G+C content is 42.7 \pm 1 mol% (Anil Kumar et al. 2010a, 2012b).

Litoribacter Tian, Wang, Hu, Zhang, Xiao, Chen, Lai, Wen, and Cui 2010, 3000^{VP}

Li.to.ri.bac'ter. L. n. *litus* -oris shore, beach, strand; N.L. masc. n. *bacter* rod; N.L. masc. n. *Litoribacter* rod from the shore.

The type strain of *Litoribacter ruber* YIM CH208^T was isolated from a sediment sample collected from the shore of a soda lake in Yunnan, southwest China, is oxidase and catalase

positive. Cells are aerobic, straight rods measuring 0.3–0.5 \times 1.2–1.5 µm. Colonies are circular, smooth, convex, bright redpigmented and grows up to 2–3 mm diameter on marine agar. Cells contain carotenoids. NaCl is not required for growth, but optimal growth obtained between 0.5 % and 2.5 % w/v NaCl and tolerates up to 9 % w/v NaCl. Growth occurs between 20 °C and 37 °C (optimum at 28 °C) and pH 7.5–10.5 (optimum pH at 8.5) (Tian et al. 2010).

Hydrolyzes aesculin, starch, and Tween 40 but not cellulose, gelatin, Tween 20, and Tween 80 (Tian et al. 2010).

In API 50CH system, acid is produced from amygdalin, aesculin, cellobiose, L-fucose, gentiobiose, D-glucose, lactose, maltose, D-mannose, methyl α -D-glucopyranoside, raffinose, salicin, starch, and sucrose (Tian et al. 2010).

In API 20NE system, positive for nitrate reduction, aesculin hydrolysis, arginine dihydrolase, urease, and β -galactosidase; utilization of acetate, benzoate, citrate, fructose, galactose, glycerol, glucose, lactose, malate, D-mannitol, D-mannose, raffinose, ribose, starch, sucrose, D-sorbitol, and trehalose as sole carbon sources (Tian et al. 2010).

In API ZYM system, positive for α -galactosidase, alkaline and acid phosphatases, esterase (C4), esterase lipase (C8), lipase (C14), leucine arylamidase, valine arylamidase, cystine arylamidase, trypsin, α -chymotrypsin, naphthol-AS-BI-phosphohydrolase, α - and β -glucosidases, and *N*-acetyl- β -glucosaminidase, but negative for β -galactosidase, α -mannosidase, α -fucosidase, and β -glucuronidase (Tian et al. 2010).

Major fatty acids are iso- $C_{17:1} \omega 9c$, $C_{15:0}$ 3-OH, iso- $C_{17:0}$ 3-OH, summed feature 3 (iso- $C_{15:0}$ 2-OH and/or $C_{16:1} \omega 7c$), and $C_{17:1} \omega 6c$. Predominant isoprenoid quinone is MK7, and the genomic DNA G+C content is 45.1 mol% (Tian et al. 2010).

Mongoliicoccus Liu, Wang, Li, Feng, Liu, and Wang 2012, 2210^{VP}

Mon.go.li.i.coc'cus. N.L. n. *Mongolia* Mongolia; N.L. masc. n. *coccus* (from Gr. masc. n. Kokkos grain) coccus; N.L. masc. n. *Mongoliicoccus* a coccus from Mongolia, referring to the isolation of the type strain from the Mongolia Plateau.

The type strain of *Mongoliicoccus roseus* MIM28^T was isolated from Lake Horsemeno, a soda lake located in Hunsandake Desert, China, on the Mongolia Plateau, is oxidase and catalase positive. Cells are Gram-stain-negative cocci measuring 0.7–1.2 μ m in diameter. Cells are aerobic, chemoheterotrophic, moderately halotolerant, and mesophilic. Colonies on MA after 3 days at 30 °C are circular, smooth, convex, and pink pigmented. NaCl is not required for growth, but optimal growth occurs between 3 % and 4 % and tolerates up to 10 % w/v NaCl. Growth occurs between 4 °C and 42 °C (optimum between 30 °C and 33 °C) and pH 6–10 (optimum pH between 8 and 9) (Liu et al. 2012).

Hydrolyzes aesculin and starch but not Tweens 20, 40, and 80. Nitrate is reduced to nitrite. Hydrogen sulfide is not produced from L-cysteine (Liu et al. 2012).

Acid production is positive from aesculin, amygdalin, arbutin, cellobiose, galactose, β -gentiobiose, D-glucose, D-fructose, inulin, 2-ketogluconate lactose, maltose, D-mannose, raffinose, salicin, starch, sucrose, turanose, and D-xylose (Liu et al. 2012).

Growth occurs in presence of acetic acid, *N*-acetyl-D-galactosamine, *N*-acetyl-D-glucosamine, adonitol, L-alaninamide, L-alanine, L-alanylglycine, L-arabinose, L-asparagine, L-aspartic acid, 2,3-butanediol, DL-carnitine, cellobiose, citric acid, α -cyclodextrin, dextrin, i-erythritol, D-galactose, D-galacturonic acid, gentiobiose, D-gluconic acid, D-glucosaminic acid, α -Dglucose, α -D-glucose 1-phosphate, D-glucose 6-phosphate, D-glucuronic acid, L-glutamic acid, DL- α -glycerol phosphate, glycogen, L-histidine, inosine, α -ketoglutaric acid, DL-lactic acid, α -lactose, lactulose, L-leucine, maltose, D-mannitol, D-mannose, D-melibiose, myo-inositol, L-ornithine, D-psicose, L-pyroglutamic acid, pyruvic acid methyl ester, quinic acid, raffinose, D-rhamnose, D-serine, L-serine, D-sorbitol, sucrose, thymidine, trehalose, turanose, and uridine (Liu et al. 2012).

Growth does not occur in presence of D-alanine, γ -hydroxybutyric acid, α -ketovaleric acid, malonic acid, phenylehylamine, D-saccharic acid, and xylitol (Liu et al. 2012).

Positive for enzymatic activities like acid phosphatase, *N*-acetyl- β -glucosaminidase, alkaline phosphatase, α -chymotrypsin, cystine arylamidase, esterase (C4), esterase lipase (C8), α -fucosidase, α -galactosidase, β -galactosidase, α -glucosidase, β -glucosidase, leucine arylamidase, naphthol-AS-BIphosphohydrolase, trypsin, and valine arylamidase (Liu et al. 2012).

The major fatty acids are branched saturated to unsaturated, namely, iso- $C_{15:0}$, anteiso- $C_{15:0}$, iso- $C_{16:0}$, $C_{16:0}$, iso- $C_{17:0}$ 3-OH, summed feature 3 ($C_{16:1} \ \omega 7c$ and/or $C_{16:1} \ \omega 6c$), and summed feature 9 (iso- $C_{17:1} \ \omega 9c$ and/or $C_{16:0}$ 10-methyl). The major respiratory quinone is menaquinone MK7. The polar lipids comprise phosphatidylcholine and phosphatidylethanolamine, and the genomic DNA G+C content is 62.8 mol% (Liu et al. 2012).

Mongoliitalea Yang, Liu, Bao, Feng, Liu, Zhang, and Zhao 2012, 650^{VP}

Mon.go.li.i.ta'le.a. N.L. n. *Mongolia* Mongolia; L. fem. n. *talea* rod, stick; N.L. fem. n. *Mongoliitalea* a rod from Mongolia, referring to the isolation of the bacterial strain from Mongolia Plateau.

The type strain of *Mongoliitalea lutea* MIM18^T was isolated from an alkaline and saline lake on the Mongolia Plateau and is oxidase and catalase positive. Cells are Gram-stain-negative rods measuring $0.5-1.0 \times 3.0-8.0 \mu$ m. Cells are aerobic, chemoheterotrophic, alkaliphilic, halotolerant, and mesophilic. Colonies on MA after 3 days at 30 °C are circular, smooth, convex, and pink pigmented. NaCl is not required for growth, but optimal growth occurs between 3 % and 4 % and tolerates up to 10 % w/v NaCl. Growth occurs between 4 °C and 42 °C (optimum at 30–33 °C) and pH 6–10 (optimum pH at 8–9) (Yang et al. 2012). Hydrolyzes gelatin but not Tweens 20, 40, and 80. Nitrate is reduced to nitrite. Hydrogen sulfide is produced. Phenylalanine deaminase is positive (Yang et al. 2012).

Acid production is positive from aesculin, amygdalin, L-arabinose, arbutin, cellobiose, D-fructose, β -gentiobiose, D-glucose, 2-ketogluconate, lactose, maltose, D-mannose, melibiose, methyl α -D-mannoside, raffinose, salicin, starch, sucrose, and turanose (Yang et al. 2012).

Utilizes following substrates: y-aminobutyric acid, L-alaninamide, L-alanine, L-alanyl glycine, arabinose, L-asparagine, cellobiose, α-cyclodextrin, dextrin, D-fructose, D-galactose, α -D-glucose, glucuronamide, L-glutamic acid, α -ketoglutaric acid, *α*-lactose, lactulose, maltose, L-ornithine, raffinose, succinic acid, sucrose, L-threonine, trehalose, and turanose, but 2-aminoethanol, D-arabitol, bromosuccinic not acid. 2,3-butanediol, L-carnitine, cis-aconitic acid, erythritol, formic acid, L-fucose, D-galactonic acid, glycerol, glycogen, glycyl-Laspartic acid, L-histidine, β -hydroxybutyric acid, hydroxyphenylacetic acid, inosine, itaconic acid, lactone, malonic acid, phenylehylamine, propionic acid, D-psicose, putrescine, L-rhamnose, sebacic acid, succinamic acid, uridine, urocanic acid, and xylitol (Yang et al. 2012).

Positive for the enzyme activities acid phosphatase, *N*-acetyl- β -glucosaminidase, alkaline phosphatase, α -chymotrypsin, cystine arylamidase, esterase (C4), esterase lipase (C8), β -galactosidase, α -glucosidase, β -glucosidase, leucine arylamidase, α -mannosidase, naphthol phosphohydrolase, trypsin, and valine arylamidase (Yang et al. 2012).

The major fatty acids are branched saturated to unsaturated, namely, iso- $C_{15:0}$, iso- $C_{17:0}$ 3-OH, $C_{15:1}$ $\omega 6c$, iso- $C_{16:0}$ 3-OH, summed feature 4 ($C_{14:0}$ 2-OH and/or iso- $C_{15:0}$ 2-OH), and summed feature 9 (iso- $C_{17:1}$ $\omega 9c$ and/or $C_{16:0}$ 10-methyl). The major respiratory quinone is menaquinone MK7. The polar lipids comprise phosphatidylcholine, phosphatidylglycerol, and phosphatidylethanolamine, and the genomic DNA G+C content is 39.8 mol% (Yang et al. 2012).

Nitritalea Anil Kumar, Srinivas, Pavan Kumar, Madhu, and Shivaji 2010, 2324^{VP}, Emend. Anil Kumar, Srinivas, Madhu, Sravan, Shashi Singh, Naqvi, Mayilraj, and Shivaji 2012, 2256^{VP}

Ni.tri.ta'le.a. L. n. *nitrum* natural soda; L. fem. n. *talea* a rod, a stick; N.L. fem. n. *Nitritalea* a rod or stick from natural soda, referring to the isolation of the first strain from a soda lake.

The type strain of *Nitritalea halalkaliphila* LW7^T was isolated from a water sample collected at a depth of 4.5 m from Lonar Lake and is oxidase and catalase positive. Cells are Gram stain negative, straight rods, measuring 0.7–0.8 × 2.0–3.0 μ m. Cells are aerobic, chemoheterotrophic, haloalkaliphilic, and mesophilic. Colonies on half-strength MA after 3 days at 30 °C are circular, smooth, convex and dark reddish orange pigmented, and opaque with entire edges. NaCl is required for growth; growth ranges between 1 % and 22 %

with optimal growth occurs between 2 % and 5 % (w/v) NaCl. Growth occurs between 25 $^{\circ}$ C and 40 $^{\circ}$ C (optimum pH at 37 $^{\circ}$ C) and pH 7.5–12 (optimum at 10) (Anil Kumar et al. 2010b).

Hydrolyzes aesculin and starch but not Tweens 20, 40, and 80. Nitrate is reduced to nitrite. Hydrogen sulfide is not produced from L-cysteine (Anil Kumar et al. 2010b).

Positive for ornithine and lysine decarboxylase activities; hydrolysis of gelatin; but negative for arginine dihydrolase, β -galactosidase, and phenylalanine deaminase activities; methyl red and Voges-Proskauer reactions; hydrolysis of aesculin, starch, and Tween 60; nitrate reduction; and H₂S gas production (Anil Kumar et al. 2010b).

Acid production is positive from the following substrates: cellobiose, dulcitol, D-fructose, galactose, glucose, glycogen, inositol, inulin, maltose, mannitol, mannose, melibiose, sorbitol, and sucrose and negative from aesculin, amygdalin, arabinose, erythritol, inositol, mannoside, methyl α -D-glucoside, raffinose, ribose, sorbose, and xylose (Anil Kumar et al. 2010b).

Utilization is positive from DL-aspartic acid, cellobiose, citrate, dulcitol, fructose, glucose, glutamine, L-histidine, inositol, inulin, isoleucine, lactose, malonate, mannose, melibiose, *o*-nitrophenyl β -galactoside, pyruvic acid, raffinose, succinic acid, sucrose, trehalose, L-valine, and glutamic acid and negative for acetate, adonitol, aesculin, D-arabinose, L-arabinose, L-arginine, galactose, D-gluconic acid, glucosamine, glycerol, maltose, D-glucoside, leucine, mannitol, melezitose, methyl α -Dmannoside, L-methionine, methyl α -ribose, phenylalanine, L-proline, rhamnose, salicin, L-serine, sodium gluconate, sorbose, sorbitol, threonine, L-tyrosine, xylitol, and xylose (Anil Kumar et al. 2010b).

The major fatty acids are branched saturated to unsaturated, namely, iso- $C_{15:0}$ and iso- $C_{17:0}$ 3-OH. The polar lipids comprise phosphatidylethanolamine, unidentified aminolipid, unidentified phospholipids, and unidentified lipids, and the genomic DNA G+C content is 49 mol% (Anil Kumar et al. 2010b).

Rhodonellum Schmidt, Priemé, and Stougaard 2006, 2891^{VP}

Rho.do.nell.um. Gr. neut. n. *rhodon* a rose; L. neut. dim. ending - *ellum*; N.L. dim. neut. n. *Rhodonellum* a small rose, referring to the red color of the colonies.

The type strain of *Rhodonellum psychrophilum* GCM71^T was isolated from the cold and alkaline ikaite tufa columns of the Ikka Fjord in southwest Greenland and is catalase positive and oxidase negative. Cells are Gram-stain-negative rods, measuring $0.7-1.0 \times 0.8-3.0 \mu$ m. Cells are aerobic, chemoheterotrophic, alkaliphilic, and psychrophilic. When grown under low light intensities, colonies are circular, smooth, and red pigmented due to the presence of carotenoids, and when grown at light intensities of 20–40 μ E m⁻² s⁻¹, colonies are white to light red (Schmidt et al. 2006a).

NaCl is not required for growth; optimal growth occurs at 0.6 %, tolerates up to 3 % (w/v) NaCl. Growth occurs

between 0 °C and 22 °C (optimum at 5 °C) and pH 7.5–10.7 (optimum pH between 9.2 and 10.0) (Schmidt et al. 2006a).

Positive for utilization of the following carbon sources: fructose, galactose, glucose, glycerol, lactose, maltose, mannose, sodium acetate, sorbitol, starch, sucrose, and xylose (Schmidt et al. 2006a).

In API 20NE system positive for *N*-acetylglucosamine, glucose, maltose and mannose assimilation, β -galactosidase, and β -glucosidase activities (Schmidt et al. 2006a).

In API ZYM system positive for acid phosphatase, *N*-acetyl- β -glucosaminidase, alkaline phosphatase, α -chymotrypsin, cystine arylamidase, esterase, esterase lipase (C8), α -galactosidase, β -galactosidase, α -glucosidase, β -glucosidase, leucine arylamidase, α -mannosidase, naphthol-AS-BI-phosphohydrolase, trypsin, and valine arylamidase (Schmidt et al., 2006a).

The major fatty acids are branched saturated to unsaturated, namely, iso- $C_{15:1}$ G, iso- $C_{15:0}$, iso- $C_{16:1}$ H, iso- $C_{17:1}\omega 9c$, $C_{17:1}\omega 6c$, iso- $C_{17:0}$ 3-OH, summed feature 3 (iso- $C_{15:0}$ 2-OH and/or $C_{16:1} \omega 7c$), and summed feature 4 ($C_{14:0}$ 2-OH and/or iso- $C_{15:0}$ 2-OH). The genomic DNA G+C content is 44.2 mol% (Schmidt et al. 2006a).

Shivajiella Anil Kumar, Aravind, Francis, Bhumika, Ritika, Priyashanth, and Srinivas 2012, 323^{VP}

Shi.va.ji'.el'la. N.L. fem. dim. n. *Shivajiella*, named after Dr. Shivaji, an eminent Indian microbiologist who has made a significant contribution to our knowledge of heterotrophic bacteria from different habitats worldwide.

The type strain of *Shivajiella indica* NIO-S1^T was isolated from a water sample of a pond adjacent to the coast, Kakinada, Bay of Bengal, India, is oxidase and catalase positive. Cells are Gram stain negative, straight long rods, measuring 0.5–0.8 × 2.0–5.0 µm. Cells are aerobic, chemoheterotrophic, mesophilic, and alkalitolerant. Colonies on MA after 2 days of incubation at 30 °C are circular, smooth, raised and orange pigmented, and translucent with entire margin. NaCl is not required for growth; optimal growth occurs between 1 % and 2 % and tolerates up to 3 % (w/v) NaCl. Growth occurs between 25 °C and 37 °C (optimum at 30 °C) and pH 7–11 (optimum pH at 11) (Anil Kumar et al. 2012a). β -galactosidase activity is present.

In Hi25TM Enterobacteriaceae Identification Kit, positive for nitrate is reduction; negative for lysine and ornithine decarboxylase and phenylalanine deaminase activities; H_2S and indole production; methyl red and Voges-Proskauer reactions; hydrolysis of aesculin, casein, cellulose, starch, Tween 40, and urea (Anil Kumar et al. 2012a).

In the VITEK GN ID card system, positive for β -N-acetylglucosaminidase, α-galactosidase, β -galactosidase, glu-glyarg-arylamidase, α -glucosidase, y-glutamyl transferase, arylamidase, glycine arylamidase phosphatase, L-proline tyrosine arylamidase, and β -xylosidase activities; utilization of D-cellobiose and D-trehalose; variable for

ala-phe-pro-arylamidase, glutamyl arylamidase, and β -glucosidase activities; and negative for β -*N*-acetylgalactosaminidase, β -alanine arylamidase, courmarate, β -glucuronidase, lipase, and L-pyrrolidonyl arylamidase activities; utilization of L-arabitol, 5-keto-D-gluconate, L-histidine, L-lactate, L-malate, D-maltose, D-mannitol, D-mannose, PalatinoseTM (isomaltulose), D-sorbitol, and D-tagatose; L-lactate and succinate alkalinization; fermentation of glucose; and 0/129 resistance (Anil Kumar et al. 2012a).

In Hi25TM Enterobacteriaceae Identification Kit, produces acid from cellobiose, galactose, lactose, malonate, maltose, mannose, melibiose, methyl α -D-glucoside, sucrose, trehalose, and xylitol (Anil Kumar et al. 2012a).

In HiCarbohydrateTM Kit, ferment the cellobiose, citrate, galactose, lactose, malonate, mannose, melibiose, methyl α -D-glucoside, sucrose, and trehalose (Anil Kumar et al. 2012a).

The major fatty acids are branched saturated to unsaturated, namely, iso- $C_{15:0}$, anteiso- $C_{15:0}$, iso- $C_{17:0}$ 3OH, and summed feature 3 ($C_{16:1} \ \omega 7c$ and/or $C_{16:1} \ \omega 6c$). The major respiratory quinone is menaquinone MK7. The polar lipids include diphosphatidylglycerol, phosphatidylglycerol, phosphatidylglycerol, phosphatidylglycerol, DNA G+C content is 41.2–41.3 mol% (Anil Kumar et al. 2012a).

Isolation, Enrichment and Maintenance Procedures

Isolation

The members of *Cyclobacteriaceae* grow on a wide range of media. The selection of a particular medium depends on the source of sample. Most of the members were reported from marine habitats, which were isolated by plating the sample on marine agar or by growing on medium with artificial or natural seawater composition following the dilution technique.

Species of the genus *Cyclobacterium* can be isolated by plating the serially diluted sample on low-organic marine agar plates containing (w/v) 0.05 % peptone, 0.01 % yeast extract; the salt concentration was same as in marine agar and incubating at 30 °C for 10 days (Ying et al. 2006) or by using low-nutrient medium containing (w/v) 0.005 % casein hydrolysate, 0.005 % peptone, and 5.0 ml of Wolfe mineral elixir (g/liter of distilled water: nitrilotriacetic acid, 1.5: MgS0₄.3.0; MnS0₄, 0.5; NaCl, 1.0; FeS0₄, 0.1; CaCl₂, 0.1; CoCl₂, 0.1; ZnS0₄, 0.1; CuS0₄, 0.01; AlK(SO₄)₂, 0.01; H₃BO₃, 0.01; Na₂Mo0₄, 0.01) (Raj and Maloy 1990; Wolin et al. 1963).

Psychrophilic species of the genus *Algoriphagus* were isolated from microbial mats of Antarctica. For isolation, mats homogenized aseptically in normal saline (0.86 % w/v, NaCl) water were placed on to MA by standard dilution-plating technique using filtered seawater as a solvent and incubated aerobically at 4 °C or 20 °C (Van Trappen et al. 2002). Species from sea ice cores were isolated by plating the melted ice core samples after serial dilutions in sterile seawater, on marine agar, and incubating at 4 and 25 °C for 2 months (Bowman et al. 1997). For the isolation of marine strains of Algoriphagus, the samples were plated directly or after standard dilution series on to marine agar plates and incubated at 25-28 °C (Yoon et al. 2005a, b). Isolation of the strains from the soil was performed by dilution-plating technique using ten times diluted nutrient agar and incubating at 25 °C (Yoon et al. 2006). Strains were isolated from freshwater pipe by directly streaking the freshwater sample on half-strength nutrient agar medium (Rau et al. 2012). Some species were isolated from coral Palythoa sp., collected from sea, by directly plating the tissue homogenates on medium containing (L^{-1}) Bacto peptone (Difco), 5.0 g; sucrose, 5.0 g; glucose 1.0 g; yeast extract (Difco), 2.5 g; KH₂PO₄, 0.1 g; MgSO₄, 0.1 g; and Bacto agar (Difco), 15.0 g in 30 % (v/v) natural seawater and 70 % (v/v) distilled water (Nedashkovskaya et al. 2007b).

Members of the genera *Aquiflexum*, *Belliella*, and *Shivajiella* can be isolated from seawater, algal mat, or mud volcano by plating the serially diluted sample on marine agar and incubating at 25–30 °C (Brettar et al. 2004a, b; Anil Kumar et al. 2012a).

Members of the genera *Indibacter*, *Nitritalea*, *Cecembia*, *Mongoliicoccus*, and *Litoribacter* can be isolated from soda lakes by plating the sample on marine agar medium with or without adjusting to pH 10.0 (with 20 % w/v Na₂CO₃) and incubating at 25–30 °C for 15 days (Anil Kumar et al. 2010a, b, 2012b; Tian et al. 2010; Yang et al. 2012).

The species of the genus *Echinicola* were isolated by using dilution-plating technique on MA (Difco) after incubation for 3 days at 25 °C (Kim et al. 2011; Nedashkovskaya et al. 2006) or by direct plating of the seawater sample on the medium containing 0.5 % (w/v) peptone, 0.2 % (w/v) yeast extract, 0.2 % (w/v) casein hydrolysate, 0.1 % (w/v) glucose, 0.02 % (w/v) KH₂PO₄, 0.005 % (w/v) MgSO₄, and 1.5 % (w/v) agar (Difco) in 50 % (v/v) natural seawater and 50 % (v/v) distilled water (Nedashkovskaya et al. 2007a).

Species of the genus *Fontibacter* can be isolated from hot spring water by plating the sample on nutrient agar and incubating at 30-40 °C (Kämpfer et al. 2010).

Members of the genus *Rhodonellum* were isolated from a permanently cold (4 $^{\circ}$ C) and alkaline (pH 10.4) ikaite tufa columns. They were isolated by plating small grains of ikaite material on a tenfold diluted R2A medium agar plates and incubating at 5 $^{\circ}$ C for 1–3 months (Schmidt et al. 2006a, b).

Enrichment

The members of the family do not require any special enrichment procedures for their isolation from the samples. The samples can be directly plated or by using the standard dilution-plating technique on the suitable medium, viz., mostly MA for the samples collected from marine habitats or marine agar with the pH adjusted to 9–10 by 20 % (w/v) Na_2CO_3 for the samples from haloalkaline soda lakes or nutrient agar medium or R2A medium can be used for freshwater strains.

Maintenance

The members of the family are preserved using the routine techniques like freeze drying, cryopreservation. For short-term preservation, the log phase cultures on agar plates can be preserved at 4 °C for 4–6 weeks, and for long-term preservation, they can be freeze dried using 10 % skim milk as a cryoprotectant or cryopreserved as 20 % v/v glycerol stocks prepared with the appropriate medium, in liquid nitrogen at -196 °C or at -20 or -80 °C freezers.

Ecology

Habitat

The habitats of *Cyclobacteriaceae* members mostly include the marine salterns, seawater, sea sediment, hot springs, soda lakes, mud volcanoes, algal mats, soil, sea ice cores, coral species, ikaite tufa columns, and freshwater lakes.

Cyclobacterium marinus (Raj and Maloy 1990) and *Cyclobacterium amurskyense* (Nedashkovskaya et al. 2005) were isolated from seawater, while *Cyclobacterium lianum* (Ying et al. 2006) was isolated from a sediment sample collected at a depth of 100 m from the sea.

Algoriphagus ratkowskyi (Bowman et al. 2003) was isolated from sea ice and cyanobacterial mat from saline lake. Algoriphagus alkaliphilus (Tiago et al. 2006) from an alkaline groundwater (pH 11.4). Algoriphagus antarcticus (Van Trappen et al. 2004) was isolated from microbial mats in Antarctic lakes. Algoriphagus aquaeductus (Rau et al. 2012) was isolated from a freshwater pipe. Algoriphagus aquatilis (Liu et al. 2009) was isolated from a freshwater lake. Algoriphagus aquimarinus, Algoriphagus chordate, Algoriphagus winogradskyi (Nedashkovskaya et al. 2004), Algoriphagus jejuensis (Lee et al. Algoriphagus marincola (Yoon et al. 2012), 2004; Nedashkovskaya et al., 2007b), and Algoriphagus namhaensis (Oh et al. 2012) were isolated from seawater. Algoriphagus boritolerans (Ahmed et al. 2007; Nedashkovskava et al., 2007b) was isolated from soil; Algoriphagus faecimaris (Li et al. 2011) was isolated from a sediment sample of the sea. Algoriphagus halophilus, Algoriphagus mannitolivorans, Algoriphagus ornithinivorans (Yi and Chun 2004; Nedashkovskaya et al. 2004), and Algoriphagus lutimaris (Park et al. 2010) were isolated from a tidal flat. Algoriphagus hitonicola (Copa-Patiño et al. 2008) was isolated from an athalassohaline lagoon. Algoriphagus olei (Young et al. 2009) was isolated from a oil-contaminated soil; Algoriphagus terrigena (Yoon et al. 2006) soil; Algoriphagus was isolated from vanfongensis (Nedashkovskaya et al. 2007b) was isolated from a coral Palythoa sp.; Algoriphagus yeomjeoni (Yoon et al. 2005a) and Algoriphagus *locisalis* (Yoon et al. 2005b) were isolated from a marine solar saltern.

Species of *Belliella baltica* (Brettar et al. 2004a) were isolated from seawater, while species of *Belliella pelovolcani* (Arun et al. 2009) from a mud volcano.

Members of the genus *Cecembia* were isolated from soda lake (Anil Kumar et al., 2012b) and hot spring runoff (Albuquerque et al. 2012).

Echinicola pacifica, the type species of the genus, was isolated from the sea urchin *Strongylocentrotus intermedius* (Nedashkovskaya et al. 2006). *Echinicola jeungdonensis* was isolated from a solar saltern (Kim et al. 2011), *Echinicola vietnamensis* was isolated from seawater (Nedashkovskaya et al. 2007a), and *Echinicola shivajiensis* (Srinivas et al. 2012) was isolated from a brackish water pond.

Fontibacter species were isolated from hot spring (Kämpfer et al. 2010) and microbial fuel cell (Zhang et al. 2012).

Aquiflexum balticum, the only species of the genus, was isolated from the seawater (Brettar et al. 2004b).

Indibacter alkaliphilus (Anil Kumar et al. 2010a), Nitritalea halalkaliphila (Anil Kumar et al. 2010b), Litoribacter ruber (Tian et al. 2010), Mongoliicoccus roseus (Liu et al. 2012), and Mongoliitalea lutea (Yang et al. 2012) were isolated from haloalkaline soda lakes.

Rhodonellum psychrophilum, the only species of the genus, was isolated from cold and alkaline ikaite tufa columns (Schmidt et al. 2006a, b).

Shivajiella indica, the only species of the genus, was isolated from an algal mat collected from marine fish pond (Anil Kumar et al. 2012a).

Pathogenicity and Clinical Relevance

The family Cyclobacteriaceae consists mostly of marine species; none of which are reported to be pathogenic. Nitritalea halalkaliphila strain LW7^T, as discussed earlier, has a promising new approach towards the antibacterial activity. It can quench quorum sensing by inactivating acyl-homoserine-lactone (AHL) by its acyl-homoserine-lactone acylase enzyme, and hence the bacterial intercellular signaling, required for regulation of growth, is hampered. This has many adverse effects on the growth of the affected bacteria including inability to swarm (Lin et al. 2003). Another clinically important bacteria in Cyclobacteriaceae family is Cecembia lonarensis strain LW9^T. As discussed earlier, genome of this bacterium has a lycopene-producing gene. Lycopene is a carotenoid generally found in tomato and some microorganisms too. Lycopene is the most active antioxidant. It is reported to have several health benefits including prevention from cancer, especially in case of lungs, stomach, and prostate gland. It is also debated to have antimutagenic effects, but there is not enough evidence to support this theory (Polívková et al. 2010). Chimaereicella boritolerans strain T-22^T has a very strong alpha-chymotrypsin enzyme activity (Ahmed et al. 2007). Chymotrypsin is an enzyme with many clinical applications, for

example, it is used as an anti-inflammatory agent; it is used to reduce liver damage in burn patients and to assist in wound repair. It helps in loosening of phlegm in case of asthma, bronchitis, and sinus and lung diseases. Chymotrypsin can be taken by mouth, as a shot or even applied directly over the skin (topically) for inflammation cases. Chymotrypsin is also sometimes used to reduce eye damage during cataract surgery.

Application

Cyclobacteriaceae family has a total of 14 genera, and these genera include 45 species. *Belliella baltica* DSM 15883^T, *Cyclobacterium marinus* DSM 745^T, and *Nitritalea halalkaliphila* strain LW7^T report the presence of cellulose-degrading enzymes in their genome annotation and can be screened for cellulase production. Cellulase finds its uses in pulp and paper industry, textile industry, bioethanol industry, wine and brewery industry, food processing industry, animal feed industry, detergent industry, olive oil extraction, carotenoid extraction, and waste management (Kuhad et al. 2011).

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