

**RHODOCISTA CENTENARIA GEN. NOV., SP. NOV.,
A CYST-FORMING ANOXYGENIC PHOTOSYNTHETIC
BACTERIUM AND ITS PHYLOGENETIC POSITION
IN THE PROTEOBACTERIA ALPHA GROUP**

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To elucidate the taxonomic position of “*Rhodospirillum centenum*” IAM 14193^T (=ATCC 43720^T) (T=type strain), a cyst-forming anoxygenic phototrophic bacterium, the small rRNA sequences (about 1,300 bases) were determined for the organism, two cyst-forming isolates (MT-SP-2 and MT-SP-3), *Rhodopseudomonas palustris* ATCC 17001^T and *Erythrobacter longus* JCM 6170^T by using the reverse transcription-dideoxy sequencing method. “*Rhodospirillum centenum*” IAM 14193^T and the two isolates had high phylogenetic affinity (98.8% homology). The sequences of the two isolates were identical. Comparative analysis of the presently determined sequences and the reference sequences (1,107 bases) revealed that the three cyst-forming organisms belonged to the α group of the class *Proteobacteria*, and were distantly related to *Rhodospirillum rubrum* (the type species, α -1 subgroup) (89.9% homology) and the representative members of other subgroups (α -2, -3 and -4) (87.6–89.8% homology). They constituted a new phylogenetic branch in the α group. “*Rhodospirillum centenum*” IAM 14193^T possessed Q-9 as the major quinone but no rhodoquinones or menaquinones, that was a quinone system distinct from that of other anoxygenic phototrophs. The phylogenetic evidence and the phenotypic characteristics indicated that “*Rhodospirillum centenum*” IAM 14193^T did not belong to the genus *Rhodo-*

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spirillum. We proposed *Rhodocista centenaria* gen. nov., sp. nov. for the organism.

"*Rhodospirillum centenum*" IAM 14193^T (= ATCC 43720^T) (T = type strain) was isolated from a water sample (temperature, ca. 55°C) of the source pool at Thermopolis Hot Springs, Wyoming, U.S.A. (4). The bacterium is unusual among anoxygenic phototrophic bacteria in possessing cytoplasmic 'R-body' and forming desiccation- and heat-resistant cysts when grown under aerobic conditions in the dark (4). Favinger et al. (4) reported the bacterium to be a member of the genus *Rhodospirillum* on the bases of vegetative cell morphology and biochemical properties. However, the cyst-formation is a unique character and implies the possibility that the bacterium is not a member of the genus *Rhodospirillum*. In 1987, one of the authors (Y. H.) and T. Satoh isolated two anoxygenic phototrophic bacteria from waste water of paste-producing plant in Japan that formed cyst-cells microscopically similar to those of "*Rhodospirillum centenum*."

With the purpose to establish the taxonomic position of "*Rhodospirillum centenum*" IAM 14193^T, we determined the 16S rRNA primary structures of the organism and the two cyst-forming isolates, and compared these structures with those of *Rhodospirillum rubrum* (the type species), the other representative members of *Rhodospirillaceae* and non-phototrophic members of the *Proteobacteria* α group. In this paper the evidences are presented that "*Rhodospirillum centenum*" is phylogenetically and phenotypically distinct from *Rhodospirillum rubrum* and other proteobacteria and the name *Rhodocista centenaria* gen. nov., sp. nov. is proposed for the organism.

MATERIALS AND METHODS

Bacterial strains and cultivation. The strains which we examined for 16S rRNA sequencing were "*Rhodospirillum centenum*" IAM 14193^T (= ATCC 43720^T), two isolated strains, MT-SP-2 and MT-SP-3, *Rhodopseudomonas palustris* ATCC 17001^T and *Erythrobacter longus* JCM 6170^T.

"*Rhodospirillum centenum*" IAM 14193^T and the two isolates were grown on SA medium (8, 9) that contained (per liter) 1.0 g of sodium acetate, 1.0 g of sodium succinate, 0.5 g of KH₂PO₄, 0.6 g of K₂HPO₄, 1.0 g of (NH₄)₂SO₄, 0.2 g of MgSO₄·7H₂O, 0.2 g of NaCl, 0.05 g of CaCl₂·2H₂O, 0.1 g of Na₂S₂O₃·5H₂O, 0.1 g of yeast extract (Difco Laboratories, Detroit, Mich., U.S.A.), 1 ml of vitamin mixture and 1 ml of trace element solution, pH 6.8. The vitamin mixture was composed of 50 mg of thiamine hydrochloride, 50 mg of niacin, 30 mg of *p*-aminobenzoic acid, 10 mg of pyridoxine hydrochloride, 5 mg of biotin and 5 mg of vitamin B₁₂ per 100 ml. The trace element solution was composed of 1.0 g of disodium EDTA, 2.0 g of FeCl₃·6H₂O, 0.1 g of ZnCl₂, 0.1 g of MnCl₂·4H₂O, 0.1 g of H₃BO₃, 0.1 g of CoCl₂·H₂O, 20 mg of Na₂MoO₄·2H₂O, 10 mg of CuCl₂·2H₂O, 10 mg of NiCl₂·6H₂O and

5 mg of Na_2SeO_3 per liter. "*Rhodospirillum centenum*" IAM 14913^T was grown at 40°C. The two isolates and *Rhodopseudomonas palustris* ATCC 17001^T were grown at 30°C. For the determination of 16S rRNA sequences these were cultivated using screw-capped test tubes (160 mm long, 15 mm wide) half-filled with culture broth. For the determination of the quinone system and the guanine-plus-cytosine (G + C) content of DNA, "*Rhodospirillum centenum*" IAM 14193^T and the two isolates were cultivated in 300 ml broth in 500 ml conical flask with cotton plug. Cultures were illuminated with tungsten lamps. *Erythrobacter longus* JCM 6170^T was grown aerobically in Marine broth 2216 (Difco) at 30°C. For the maintenance, cultures were grown on agar medium of the above composition under the illumination and kept at 10°C in the dark. For cyst formation, cultures were grown aerobically on the agar medium containing 0.3% yeast extract (Difco) and 0.2% Casamino Acids (Difco), pH 6.8, in the dark.

Preparation and analysis of 16S rRNA. Total cellular ribosomal RNAs were extracted by the modified method of Marmur (16), Woese et al. (28), and Yamada and Kawasaki (31). The cells (ca. 0.1 g, wet weight) harvested and washed once with TMK buffer (50 mM Tris-HCl, pH 7.6, containing 10 mM magnesium acetate and 25 mM KCl) were suspended in 500 μl of TMK buffer and lysed on 0.04% (w/v) chicken egg white lysozyme at 37°C for 10 min. The suspension was added with 50 μl of 10% (w/v) sodium dodecyl sulfate and 100 μl of macaloid suspension (15) and shaken. The mixture was added with 200 μl of phenol solution (14) and 200 μl of chloroform-isoamyl alcohol (24:1, v/v), vigorously shaken for 2 min on a Vortex and centrifuged at 12,000 rpm for 1 min. This deproteinization procedure was repeated again. The nucleic acids were precipitated by adding 150 μl of 10 M ammonium acetate and 700 μl of isopropyl alcohol. The precipitates by centrifugation were rinsed twice with 700 μl of 70% ethanol, dried and dissolved in 400 μl of TMK buffer. The DNA was decomposed by incubation with 1 μl of DNase I (60–80 U/ μl , Takara Shuzo Co., Ltd., Kyoto, Japan) at 30°C for 15 min. The enzyme was removed by treating with 200 μl of phenol solution and 200 μl of chloroform-isoamyl alcohol (24:1, v/v) and centrifuged at 12,000 rpm for 1 min. The rRNAs in the supernatant were precipitated with 100 μl of ammonium acetate and 500 μl of isopropyl alcohol and rinsed twice with 70% ethanol. The precipitates were dried and dissolved in ca. 25 μl of 10 mM Tris-HCl (pH 8.5). One μl of this suspension was confirmed for the purity of 16S rRNA by the 1% agarose gel electrophoresis.

Nucleotide sequences were determined by the method of Lane et al. (13), using reverse transcriptase (Takara Shuzo Co., Ltd.). The oligonucleotide primers used in this experiment were 1400R 5'-CGGTGTGTACAAGGCCC-3' (positions 1,401 to 1,385 on *Escherichia coli* numbering system (1,2)), 1240R 5'-CATTGTAGC-ACGTGTGTA-3' (1,241 to 1,224), 1110R 5'-AGGGTTGCGCTCGTTG-3' (1,115 to 1,100), 920R 5'-CCGTCAATTCATTTGAGTTTC-3' (926 to 907), 800R 5'-CTACCAGGGTATCTAATC-3' (803 to 786), 700R 5'-TCTACGCATTTCACCGCTAC-3' (704 to 688), 520R 5'-GTATTACCGCGGCTGCTG-3' (536 to 519) and 350R 5'-CTGCTGCCTCCCGTA-3' (357 to 343).

Data analysis. The sequences were aligned, and the homology values and the phylogenetic distances (K_{nuc} values (12)) were determined. An unrooted phylogenetic tree was reconstructed by applying the algorithm of the neighbor-joining method (22) to K_{nuc} values. The statistical significance of some group positions in the tree was reexamined using the bootstrap method of Felsenstein (5) with 100 replicates.

Quinone system. Quinone system was determined according to the procedures of Ohara et al. (19) and Nakase and Suzuki (18). Quinone fractions were extracted with chloroform-methanol (2:1, v/v) from the lyophilized cells. The fractions were separated with preparative thin-layer chromatography (TLC) developed with petroleum benzine-diethyl ether (9:1, v/v) into two fractions, the ubiquinone fraction and the fraction which showed the R_f value corresponding to rhodoquinones. For the latter fraction, TLC was carried out again with benzene as a developer. The both fractions were then subjected to chromatography with a high performance liquid chromatograph (Millipore Corp., Waters Chromatography Division, Milford, Mass., U.S.A.) equipped with a Crest-Pak C 18S column (4.6 by 150 mm) (Japan Spectrophotometric Co., Ltd., Hachioji, Japan). Quinones were detected with the spectrophotometer Lambda-Max Model 481 LC (Millipore Corp., Waters Chromatography Division) at 275 nm. Methanol-isopropanol (2:1, v/v) was used as an eluent.

The G + C content of DNA. DNA was extracted and purified by using the method of Saito and Miura (21). Phenol-chloroform mixture (1:1, v/v) was used at the later steps of deproteinization. The G + C content of the DNA was estimated by using the thermal denaturation method (17,32). DNA from *Escherichia coli* IAM 1264 (strain K-12) (G + C content, 51.6 mol% (26)) was used as the reference.

RESULTS AND DISCUSSION

The 16S rRNA sequences of "*Rhodospirillum centenum*" IAM 14193^T, two phototrophic isolates, MT-SP-2 and MT-SP-3, *Rhodopseudomonas palustris* ATCC 17001^T and *Erythrobacter longus* JCM 6170^T were determined. The nucleotide sequences have been deposited at the DDBJ (Mishima, Japan) sequences data base under accession numbers D12699-D12703. These sequences were comprised of about 1,300 bases ranging between the positions 2 and 1,380 (*E. coli* numbering system) together with the reference sequences of *Rhodospirillum rubrum* ATH 1.1.1^T (EMBL accession number, M32020) and *Escherichia coli* (1,2,30).

The sequences of these bacteria were aligned with those of other representative phototrophic and non-phototrophic members of proteobacteria which were from the data libraries. For analyzing a phylogenetic relationships more precisely, the homology values (percentage similarities) and the K_{nuc} values were calculated by computing 1,107 bases ranging between positions 2 and 1,380 with the exclusion of the following regions: positions 74-96, 179-220, 454-478, 841-845, 1,106-1,035,

Table 1. Similarity and K_{nuc} values for 1,107 nucleotide regions of 16S rRNA sequences of anoxygenic phototrophic bacteria and other proteobacteria.^a

Strain	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
1. "Rsp. centenaria" IAM 14193 ^T		98.8	98.8	89.9	89.8	87.6	88.5	88.6	87.9	87.6	87.6	85.8	84.3	84.4	83.7	83.2	83.4
2. Isolate MT-SP-2	0.0118		100.0	90.0	89.7	87.9	88.3	88.6	87.5	88.0	87.5	85.4	84.2	84.1	83.7	83.0	83.5
3. Isolate MT-SP-3	0.0118	0.0000		90.0	89.7	87.9	88.3	88.6	87.5	88.0	87.5	85.4	84.2	84.1	83.7	83.0	83.5
4. Rsp. rubrum ATH 1.1.1 ^T	0.1090	0.1079	0.1079		89.3	87.9	86.8	88.3	87.8	87.2	87.3	83.1	82.7	84.6	83.5	84.6	84.2
5. Mag. gryphiswaldense MSR-1 ^T	0.1099	0.1110	0.1110	0.1152		87.9	87.9	88.5	88.3	87.0	87.3	84.3	83.9	84.1	83.8	82.6	82.5
6. Rps. palustris ATCC 17001 ^T	0.1364	0.1331	0.1331	0.1326	0.1327		92.4	90.5	89.5	87.4	88.3	82.8	82.7	83.5	81.8	81.2	81.4
7. Rps. acidophila strain 7050	0.1248	0.1272	0.1272	0.1460	0.1324	0.0804		90.0	90.3	86.9	88.2	82.7	81.6	83.2	81.9	82.2	81.8
8. Rmi. vannielii EY33	0.1235	0.1235	0.1235	0.1287	0.1254	0.1016	0.1078		90.2	87.9	87.2	84.3	82.9	84.3	83.7	83.3	81.8
9. A. tumefaciens DSM 30150	0.1322	0.1365	0.1365	0.1337	0.1274	0.1133	0.1037	0.1051		88.4	89.1	82.9	80.8	84.5	82.7	83.2	82.3
10. Rba. capsulatus ATCC 33303	0.1354	0.1311	0.1311	0.1411	0.1441	0.1385	0.1445	0.1326	0.1261		86.2	81.6	80.5	82.7	80.8	82.0	81.2
11. Ery. longus JCM 6170 ^T	0.1574	0.1630	0.1630	0.1916	0.1764	0.1954	0.1962	0.1764	0.1941	0.2117	0.1929	83.0	81.8	83.6	82.3	81.8	83.6
12. Rcy. purpureus "Ames" 6770 ^T	0.1769	0.1781	0.1781	0.1964	0.1813	0.1977	0.2120	0.1940	0.2236	0.2273	0.2087	0.1037	90.3	86.9	84.9	82.1	81.9
13. P. testosteroni ATCC 11996 ^T	0.1756	0.1791	0.1791	0.1733	0.1789	0.1874	0.1908	0.1768	0.1746	0.1976	0.1862	0.1440	0.1688	84.9	84.3	80.6	80.7
14. C. vinosum	0.1834	0.1834	0.1834	0.1870	0.1822	0.2095	0.2069	0.1833	0.1966	0.2215	0.2026	0.1685	0.1767	87.2	82.8	83.7	83.2
15. Esc. coli	0.1908	0.1932	0.1932	0.1737	0.1990	0.2179	0.2035	0.1896	0.1913	0.2059	0.2087	0.2045	0.2253	0.1953	0.1871	83.5	84.4
16. D. desulfuricans ATCC 27774	0.1884	0.1873	0.1873	0.1779	0.2000	0.2149	0.2094	0.2082	0.2025	0.2166	0.1859	0.2073	0.2250	0.1853	0.1910	0.1760	
17. Mpx. xanthus MD207																	

^a The values on the upper right triangle are homology values (percentage similarities), and the values on the lower left triangle are K_{nuc} values.

Abbreviations: Rsp., Rhodospirillum; Mag., Magnetospirillum; Rps., Rhodospseudomonas; Rmi., Rhodomicrobium; A., Agrobacterium; Rba., Rhodobacter; Ery., Erythrobacter; Rcy., Rhodocyclops; P., Pseudomonas; C., Chromatium; Esc., Escherichia; D., Desulfovibrio; Myx., Myxococcus; ATCC, American Type Culture Collection (Rockville, Maryland); DSM, Deutsche Sammlung von Mikroorganismen und Zellkulturen (Braunschweig); IAM, Institute of Applied Microbiology, The University of Tokyo (Bunkyo-ku, Tokyo); JCM, Japan Collection of Microorganisms, RIKEN (Wako, Saitama).

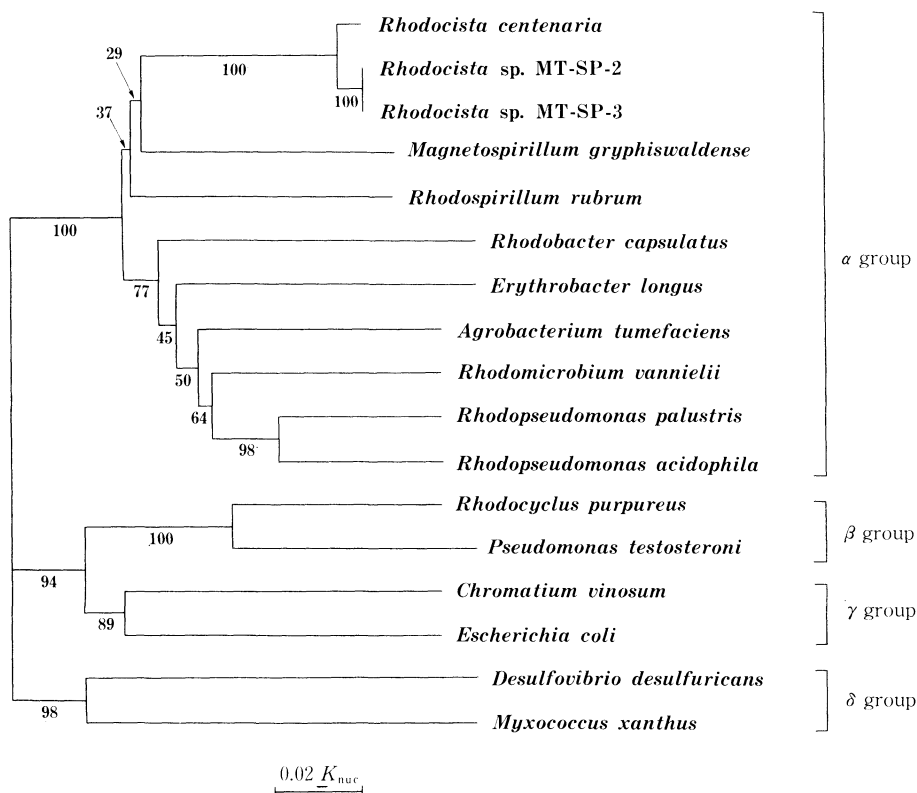


Fig. 1. Phylogenetic relationship of the cyst-forming bacteria, *Rhodocista centenaria* ("Rhodospirillum centenum") IAM 14193^T, *Rhodocista* sp. MT-SP-2 and MT-SP-3, derived from a distance matrix on 16 S rRNA sequences of the representatives of proteobacteria. Numbers indicate percentage bootstrap values of 100 replicates. For abbreviations and strain numbers see footnote of Table 1.

1,133–1,141 and 1,258–1,278 (Table 1). These regions were excluded because the variations among the sequences were so large due to the many deletions and insertions that exact alignment could not be expected, and because the inclusion of too variable regions in the phylogenetic analysis would give unreliable relationships at higher taxonomic ranks.

A phylogenetic tree reconstructed by applying the neighbor-joining method is shown in Fig. 1. "*Rhodospirillum centenum*" IAM 14193^T and the two phototropic isolates exhibited high levels of sequence relatedness (98.8% homology), and the two isolates were completely homologous (Table 1). The isolates were phylogenetically close to "*Rhodospirillum centenum*" IAM 14193^T.

"*Rhodospirillum centenum*" IAM 14193^T was proved to belong to the α group of the class *Proteobacteria* (24). However, the bacterium exhibited considerably lower homology with *Rhodospirillum rubrum* ATH 1.1.1^T (89.9%) and other

genera of the α group (87.6–89.8%) (Table 1). Based on homology values of 16S rRNA sequences “*Rhodospirillum centenum*” IAM 14193^T was distinct at least at generic level from *Rhodospirillum rubrum* ATH 1.1.1^T which was the type strain of the type species of the genus *Rhodospirillum*.

The presence of four subgroups in the *Proteobacteria* α group was proposed based on oligonucleotide cataloging by Woese et al. (29). For analyzing the phylogenetic place of “*Rhodospirillum centenum*” IAM 14193^T in the *Proteobacteria*, the 16S rRNA sequences of the following strains of proteobacteria were computed: *Rhodospirillum rubrum* ATH 1.1.1^T (M32020) of α -1 subgroup, *Rhodopseudomonas palustris* ATCC 17001^T (present result), *Rhodopseudomonas acidophila* strain 7050^T (M34128), *Rhodomicrobium vannielii* strain EY33 (M34127) and *Agrobacterium tumefaciens* DMS 30150 (M11223) (33) of α -2 subgroup, *Rhodobacter capsulatus* ATCC 33303 (M34129) of α -3 subgroup, *Erythrobacter longus* JCM 6170^T (present result) of α -4 subgroup, *Rhodocyclus purpureus* “Ames” 6770^T (M34132) and *Pseudomonas testosteroni* ATCC 11996^T (M11224) (32) of β group, *Chromatium vinosum* (M26629) and *Escherichia coli* (1,2,30) of γ group, *Desulfovibrio desulfuricans* ATCC 27774 (20) and *Myxococcus xanthus* MD207 (20) of δ group, and *Magnetospirillum gryphiswaldense* MSR-1^T (23). The homology values among the genera of α group ranged continuously from 86.2 to 90.5%. The homology values of 87.6 to 89.9% between each of the three cyst-forming bacteria and the other members of α *Proteobacteria* were within this range. However, the cyst-formers constituted a rather deep branch that was distinct from any other lineages and could not be assigned to any of subgroups of the *Proteobacteria* α group.

The quinone systems of the species of the genus *Rhodospirillum* are compared in Table 2. “*Rhodospirillum centenum*” IAM 14193^T had Q-9 but no rhodoquinones or menaquinones, and its quinone system was different from that of the other species (6, 7, 10, 11). Furthermore, the organism had G + C content of 69.9 mol% (68.3 mol% (3)) that differed from the G + C content of 60.5–65.8% (Bd) or 62.1–63.5 mol% (T_m) of the other *Rhodospirillum* species (27) except for *Rhodospirillum salinarum* (68.3%). The organism can be clearly differentiated chemotaxonomically from other members of *Rhodospirillaceae* by the combination of quinone system and G + C content of the DNA.

The two cyst-forming isolates were confirmed to share the following characters with “*Rhodospirillum centenum*” IAM 14193^T; cyst formation, vibrioid to spiral cell shape, bacteriochlorophyll *a*, lamellae membrane system, Q-9 and G + C content of DNA (69.2 and 69.8 mol% for MT-SP-2 and MT-SP-3, respectively). On the bases of phylogenetic relatedness and phenotypic characteristics the two isolates are considered to belong to the same taxon of “*Rhodospirillum centenum*” IAM 14193^T at least at the generic level.

“*Rhodospirillum centenum*” IAM 14193^T has a morphologically unique characteristic that the cells of the strain form cysts when grown under aerobic conditions in the dark. The cyst formation had not been known in the anoxygenic phototro-

Table 2. Characteristics of "*Rhodospirillum centenum*" and the species of the genus *Rhodospirillum*.

Species	Cell shape width/length (μm) ^a	Intracyto- plasmic membrane ^a	DNA base ratio (G + C mol%) ^a	Quinone system ^c
" <i>R. centenum</i> "	vibrio, spiral ^b 1-2/3	Lamellae ^b	69.9 (68.3 ^b)	Q-9
<i>R. rubrum</i>	vibrio, spiral 0.8-1.0/7-10	Vesicles	63.8-65.8	Q-10, RQ-10
<i>R. fulvum</i>	vibrio, spiral 0.5-0.7/3.5	Lamellar stacks	64.3-65.3	Q-9, MK-9
<i>R. molischianum</i>	vibrio, spiral 0.7-1.0/4-9	Lamellar stacks	61.7-64.8	Q-9, MK-9
<i>R. photometricum</i>	spiral 1.2-1.5/7-30	Lamellar stacks	65.8	Q-8, RQ-8
<i>R. salexigens</i>	vibrio, spiral 0.6-0.7/1-6	Lamellae	64	Q-10, MK-10
<i>R. salinarum</i>	rod, spiral 0.3/1-3	Vesicles	68.1	Q-10, MK-10

^a Referred from Trüper and Imhoff (27) except for "*R. centenum*."

^b Referred from Favinger et al. (4).

^c Referred from Hiraishi et al. (6), Hiraishi and Hoshino (7), Imhoff (10) and Kato et al. (11) except for *R. centenaria*.

phic bacteria until "*Rhodospirillum centenum*" was isolated (4), and this is an important morphological characteristic to distinguish it from other genera of the anoxygenic phototrophic bacteria.

The distinctness in phylogenetic relationships and chemotaxonomic and morphological characteristics shown above supports an idea that "*Rhodospirillum centenum*" IAM 14193^T should be placed in a genus apart from the genus *Rhodospirillum*. "*Rhodospirillum centenum*" was described and effectively published by Favinger et al. in 1989 (4). The term "centenum" means "a hundred" in Latin and was used as a specific epithet to commemorate "a century" that had passed since 1887 when the first description of a phototrophic bacterium was published by Esmarch (3). However, it has not been validated and has no standing in bacterial nomenclature. Therefore, we propose the establishment of a new genus and a new species *Rhodocista centenaria* for "*Rhodospirillum centenum*" IAM 14193^T. The strain IAM 14193 (= ATCC 43720) is designated as the type strain of the species. The two isolates, MT-SP-2 and MT-SP-3, are included in the genus *Rhodocista*. However, the characterization of the two strains is not sufficient to establish their taxonomic status in this paper. Therefore, we will report the species of these two isolates in following paper.

Description of *Rhodocista* gen. nov.

Rhodocista (Rho.do.ci'sta. L. fem. n. rhodos the rose; L. fem. n. cista a basket; M.L. fem. n. *Rhodocista* red basket).

The description is based on the previous publications by Favinger et al. (1989) (4) and Stadtwald-Demchick et al. (1990) (25) for the strain ATCC 43720^T, and our present results.

Cells are vibrioid to spiral in shape, and 3 μm long and 1 to 2 μm wide. Motile by means of long polar flagellum. Gram-negative. Internal photosynthetic membrane is present as lamellae lying parallel to the cytoplasmic membrane. Growth is possible phototrophically in the light and microaerophilically or aerobically in the dark. Photosynthetic pigments are bacteriochlorophyll *a* and carotenoids of the spirilloxanthin series. The cells are converted to thick-walled cysts which are resistant to desiccation and heat, when aerobically grown on agar media. Major quinone is Q-9. The G + C content of the DNA is 69–70 mol%.

The type species is *Rhodocista centenaria*.

Strains examined: IAM 14193, MT-SP-2, and MT-SP-3.

Description of *Rhodocista centenaria* sp. nov.

Rhodocista centenaria (ce'n.te.naria. L. f. adj. centenaria relating to a hundred, to commemorate a century after the publication of the first description of phototrophic bacteria in 1887. *Rhodocista centenaria* red basket (isolated) a century (after the first publication of phototrophic bacterium)).

Cells are vibrioid to spiral in shape, and 3 μm long and 1 to 2 μm wide. The cells are actively motile by means of a single, long polar flagellum. Internal phototrophic membrane is lamellae and lies parallel to the cytoplasmic membrane. Color of anaerobically grown cultures is pink. Living cells show absorption maxima at 800, 875 and 587 nm. Photosynthetic pigments are bacteriochlorophyll *a* and carotenoids of the spirilloxanthin series.

Malate or other C4 dicarboxylic acids is not utilized for phototrophic growth as the carbon sources. The carbon sources utilized are pyruvate, lactate and acetate. Butyrate, caproate and caprylate also support phototrophic growth when supplemented with bicarbonate (9 mM). Ammonia, dinitrogen, glutamate, glutamine and alanine are utilized as nitrogen sources. Biotin and vitamin B₁₂ are required as growth factors. Growth occurs at pH 6.8. Optimum temperature is between 39 and 45°C with maximum of ca. 47°C. The cells grow aerobically as a chemoheterotroph in darkness. With butyrate as the sole carbon source, the spiral-shaped cells are converted to cysts which are desiccation- and heat-resistant; the filter-fixed cysts kept at the temperature of 55°C for 2 days are viable. Major quinone is Q-9. The G + C content of the DNA is 69.9 mol%. It was isolated from a water sample of source pool of hot spring.

The type strain is IAM 14193 (= ATCC 43720).

This species is monotypic.

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