Allochromatium humboldtianum sp. nov., isolated from soft coastal sediments

Wilbert Serrano,[1,2] Jan Schrübbers,[1]† Rudolf Amann[2] and Ulrich Fischer[1,2]

Correspondence
Wilbert Serrano
wserrano@uni-bremen.de
Ulrich Fischer
ufischer@mpi-bremen.de

[1]Zentrum für Umweltforschung und nachhaltige Technologien, Fachbereich 2 Biologie/Chemie, Abteilung Marine Mikrobiologie, Universität Bremen, D-28359 Bremen, Germany

[2]Max Planck Institute for Marine Microbiology, Celsiusstraße 1, D-28359 Bremen, Germany

A novel purple sulfur bacterium, strain AX1YPE^T, was isolated from marine sediments sampled at 47 m depth in Callao Bay, Peru. Strain AX1YPE grew anaerobically, synthesizing bacteriochlorophyll *a* and carotenoid pigments of the spirilloxanthin series. Cells were Gram-stain-negative rods and actively motile by a polar flagellum. Strain AX1YPE was able to grow photolithoautotrophically with sulfide and thiosulfate as electron donors. This new phototrophic organism utilized ammonium salt, N_2, urea and glutamate as nitrogen sources. Strain AX1YPE had a DNA base composition of 63.9 mol% G+C. Analysis of the 16S rRNA gene sequence indicated that strain AX1YPE clusters in a separate branch within the genus *Allochromatium* of the family *Chromatiaceae.* Strain AX1YPE showed 16S rRNA gene sequence similarities of 98.2 % with *Allochromatium vinosum* DSM 180^T and *Allochromatium minutissimum* DSM 1376^T, 98.1 % with *Allochromatium phaeobacterium* JA144^T, 97.3 % with *Allochromatium renukae* DSM 18713^T and 96.8 % with *Allochromatium warmingii* DSM 173^T. DNA–DNA hybridization values to the type strains of its closest relatives, *A. vinosum* and *A. minutissimum*, were 59 and 64 %, respectively. The predominant fatty acid of strain AX1YPE^T was $C_{18:1}\omega$;7*c* and it notably possessed $C_{20:1}$ as a minor component. PCR-based molecular typing (Box A1R and randomly amplified polymorphic DNA) produced a unique banding pattern for strain AX1YPE^T in comparison with the type strains of *A. vinosum* and *A. minutissimum*. Based on data from this polyphasic taxonomic study, which also includes average nucleotide identity comparison of five concatenated housekeeping genes, strain AX1YPE^T is considered to represent a novel species of the genus *Allochromatium* for which the name *Allochromatium humboldtianum* sp. nov. is proposed. The type strain is AX1YPE^T (=DSM 21881^T=KCTC 15448^T).

Purple sulfur bacteria (PSB) are anoxygenic photosynthetic prokaryotes which grow anaerobically in the presence of light and use H_2S as an electron donor (Pfennig & Trüper, 1992). The class *Gammaproteobacteria* includes the two families *Chromatiaceae* and *Ectothiorhodospiraceae* as representative phylogenetic branches of PSB (Imhoff *et al.*, 1998). A common feature of the *Chromatiaceae* is the intracellular storage of sulfur globules while the members of the other family deposit them outside the cell.

Current phylogenetic reconstructions within the family *Chromatiaceae* are based on comparative 16S rRNA gene sequence analysis, DNA–DNA hybridization and the presence of discriminatory phenotypic properties (Anil Kumar *et al.*, 2008; Imhoff *et al.*, 1998;). The genus *Allochromatium* comprises, at the time of writing, five recognized species, namely

†Present address: Bregau GmbH & Co.KG, Mary-Astell-Str. 10, D-28359 Bremen, Germany.

This paper is dedicated to the memory of the famous German microbiologist Hans G. Schlegel, who passed away in March 2013 and who has influenced significantly the development of microbiology in Germany.

Abbreviations: ANI, average nucleotide identity; DDH, DNA–DNA hybridization; PSB, purple sulfur bacteria; RAPD, randomly amplified polymorphic DNA.

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of Allochromatium *humboldtianum* AX1YPE^T is EU442182.

Two supplementary figures and two supplementary tables are available with the online Supplementary Material.

Allochromatium vinosum, *Allochromatium minutissimum*, *Allochromatium warmingii* (see Imhoff *et al.*, 1998), *Allochromatium renukae* (Anil Kumar *et al.*, 2008) and *Allochromatium phaeobacterium* (Srinivas *et al.*, 2009).

In this report, a new PSB, strain AX1YPE^T, isolated from soft marine sediments retrieved from shallow depth in Callao Bay, Peru, is described using a polyphasic taxonomic study, which also includes DNA–DNA hybridization (DDH) comparison and multilocus sequence analysis. To our knowledge, strain AX1YPE^T is the first marine representative of the genus *Allochromatium*.

Sediment samples were collected during a cruise of IMARPE R/V *Jose Olaya* along the central coast of Peru in April 2005 from the nearshore station 1 (12° 02′ S 72° 13′ W) at 47 m depth. Undisturbed sediment samples were obtained with a plexiglas multicorer sampler. Aliquots of the top surface sediments, which also contained *Thioploca* mats, were used for first enrichment cultures. Aliquots were inoculated in 50 ml screw capped bottles filled with anoxic basal medium as described by Pfennig & Trüper (1981) containing 3 % NaCl. The medium was supplemented with sulfide (4 mM, final concentration), thiosulfate (2 mM) and acetate (2 mM). Initial incubation on board was done at room temperature under continuous illumination of a 40 W tungsten lamp. Subsequently, a pure culture was obtained by successive repurification steps using both liquid cultures and deep agar shake dilution series. The agar shake method was also used to test oxygen requirements. Optimal salinity, carbon substrate assimilation and utilization of different electron donors were tested using anoxic basal medium prepared as mentioned above. Tests for assimilation of several carbon sources were carried out in the presence or absence of $NaHCO_3$. Nitrogen source utilization was tested by replacing ammonium chloride from the basal medium by N_2, urea or glutamate. Diazotrophy was tested by cultivating the organism under N_2 atmosphere and was confirmed by repeated subculturing (three times). All experiments were performed using completely filled 15 ml Hungate tubes and screw cap bottles under continuous light of 100 μmol photons m^{-2} s^{-1} at 30 °C. Growth was measured turbidometrically at 650 nm in a Novaspec II (Pharmacia Biotech) UV spectrophotometer.

Cell morphology and motility were observed under a Zeiss phase-contrast microscope. Intra-cytoplasmic structures and flagella were documented by transmission electron microscopy (Zeiss EM 902A). The *in vivo* absorption spectra of cells in a sucrose suspension were measured with a Beckman DU 640 model UV spectrophotometer according to Pfennig & Trüper (1992).

For analysis of cellular fatty acids, strain AX1YPE^T was grown in liquid medium according to Pfennig & Trüper (1981) with 3 % NaCl, while its closest relatives *A. vinosum* DSM 180^T and *A. minutissimum* DSM 1376^T were grown in the same medium but without NaCl. The strains were incubated at 30 °C in all cases. Cells were harvested at the beginning of the stationary growth phase and stored lyophilized for further analysis. Analysis was carried out by the identification service of the Deutsche Sammlung von Mikroorganismen und Zellkulturen, Braunschweig, Germany.

Genomic DNA was extracted according to Marmur (1961). Purity of the extracted genomic DNA was checked spectrophotometrically and by gel electrophoresis. Determination of the DNA G+C content was made by the thermal denaturation method according to Marmur & Doty (1962). DNA–DNA hybridization was performed following the renaturation kinetic method according to De Ley *et al.* (1970) and as reported by Serrano *et al.* (2009).

PCR amplification of the 16S rRNA gene of strain AX1YPE^T was done according to Muyzer *et al.* (1993). Sequence alignment and phylogenetic tree reconstruction were carried out as described by Serrano *et al.* (2009). The final phylogentic tree was reconstructed by the maximum-likelihood method using the program Phy-ML (Guindon & Gascuel, 2003). Primers and conditions for amplification and sequencing of five protein-coding genes, *soxB* (507 bp), *cbbL* (492 bp), *groES* (729 bp), *fusA* (555 bp), *mfd* (222 bp), and the intergenic-transcriber spacer (ITS, 420 bp) sequences have been reported previously (Serrano *et al.*, 2011). Average nucleotide identity (ANI) analyses on the concatenated sequences (in total 2925 bp) were calculated according to Goris *et al.* (2007), using the ANI calculator (http://enve-omics.ce.gatech.edu/ani/index). In addition, PCR-based fingerprinting methods were also employed to study the banding pattern differences between strain AX1YPE^T and *A. vinosum* DSM 180^T and *A. minutissimum* DSM 1376^T. Randomly amplified polymorphic DNA (RAPD) and BOX A1R analyses were performed according to Serrano *et al.* (2009).

Morphological features are shown in Fig. 1. Cells of strain AX1YPE^T were rod-shaped, of 2 × 3 μm in size with one polar flagellum (Fig. 1a). Analysis of the ultrastructure

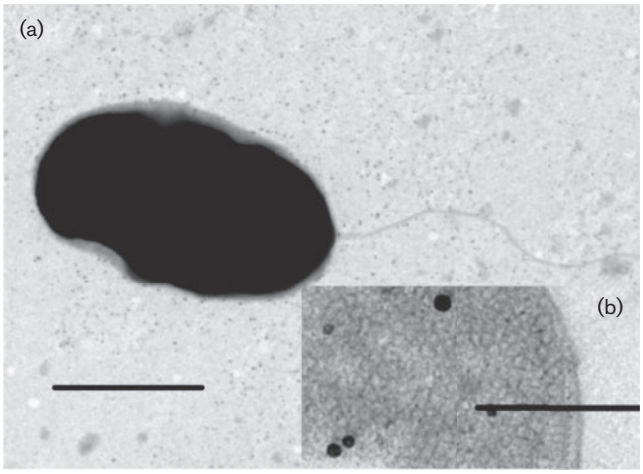

Fig. 1. Electron micrographs of a negative stained cell of strain AX1YPE^T showing a polar flagellum (a) and an ultrathin-section exhibiting the vesicular type photosynthetic membrane system (b). Bars, 2 μm (a), 0.5 μm (b).

revealed the presence of an internal membrane system of the vesicular type (Fig. 1b). The colour of the cell suspension was brownish red and the presence of bacteriochlorophyll *a* was evidenced by peaks of absorption maxima at 372, 592, 799 and 862 nm, while carotenoid pigments seem to be of the spirilloxanthin series with absorption peaks at 472 and 504 nm (Fig. S1, available in the online Supplementary Material).

Strain AX1YPE^T was strictly anaerobic and was able to grow photolithoautotrophically with sulfide as an electron donor and CO_2. However, growth was faster when the organism grew photolithoheterotrophically with sulfide or thiosulfate as electron donors and pyruvate or acetate as carbon sources. The complete morphological, physiological and molecular characteristics are summarized in Table 1. Sulfide was essential for growth of strain AX1YPE^T under all growth conditions tested, while sulfite, cysteine and hydrogen did not support growth. Although strain AX1YPE^T was isolated from a marine environment, the organism was able to grow without NaCl, but tolerated up to 5 % NaCl (optimum 3 %). The pH for growth ranged from 6.5 to 8.5, with an optimum at pH 6.9–8.0. Good growth was observed at 25–35 °C (optimum 30 °C). Strain AX1YPE^T did not require vitamin B_{12} for growth. It is remarkable that strain AX1YPE^T was able to use different nitrogen sources such as ammonium, glutamate, N_2 or urea. The use of glutamate and urea has not previously been reported for other *Allochromatium* strains (Table 1).

The fatty acid profile of strain AX1YPE^T was different from those of the closest relatives *A. vinosum* DSM 180^T and *A. minutissimum* DSM 1376^T (Table S1). The predominant component was the unsaturated fatty acid $C_{18:1}\omega 7c$ (44.6 % of the total). This fatty acid was also found in the type strains of both *A. vinosum* and *A. minutissimum* but in lower amounts (28.8 and 27.9 %, respectively). $C_{15:0}$ 3-OH and iso-$C_{16:0}$ 3-OH were found in the type strains of *A. vinosum* and *A. minutissimum* but not in strain AX1YPE^T, whereas the unsaturated eicosanoic long-chain fatty acid $C_{20:1}$ was characteristic of strain AX1YPE^T (0.6 %).

The DNA base composition of strain AX1YPE^T was 63.9 mol% G+C, which is within the range reported for the genus *Allochromatium* (Table 1). As strain AX1YPE^T shows 16S rRNA gene sequence similarity of 98.2 % to its closest relatives, the type strains of *A. vinosum* and *A. minutissimum* (Fig. 2), species differentiation was ensured additionally by using DDH studies as recommended by Tindall *et al.* (2010) and Imhoff & Caumette (2004).

DDH comparison gave values of 59 and 64 % relatedness between strain AX1YPE^T and the type strains of *A. vinosum* and *A. minutissimum*, respectively. DDH studies clearly revealed that strain AX1YPE^T showed values below 70 % to the type strains of all species compared (Table 1). DDH values were lower yet close to the accepted threshold for species delineation of 70 % suggested by Wayne *et al.* (1987).

The final phylogenetic tree was reconstructed using high-quality sequences as recommended by Tindall *et al.* (2010). In the present work we incorporated into the reconstructed tree the full 16S rRNA gene sequence from the complete genome sequence of *A. vinosum* DSM 180^T (Weissgerber et al., 2011). Strain AX1YPE^T clustered with the type strains of *A. phaeobacterium* and *A. warmingii* (99 % bootstrap support), and with that of *A. renukae*, well separated from *A. vinosum* DSM 180^T and *A. minutissimum* DSM 1376^T (Fig. 2). In a previous study, multilocus sequence analysis of all available *Allochromatium* strains including strain AX1YPE^T showed a comparable tree topology (Serrano *et al.*, 2011). In both cases, *A. vinosum* strains and *A. minutissimum* DSM 1376^T showed a trend of forming an independent cluster in the phylogenetic trees, suggesting a close relationship among them (see Serrano *et al.*, 2011). In fact, an ANI-predicting study based on the concatenation of five housekeeping protein-coding genes plus intergenic-transcribed spacer (ITS) sequences (2925 bp) performed in this study confirmed this close relationship between *A. vinosum* DSM 180^T and *A. minutissimum* DSM 1376^T, with an ANI value of 97.37 % (Table S2). This value fell into the ANI category of $\geqslant$96 % according to Kim *et al.* (2014), which is above the accepted threshold for species determination based on ANI (Konstantinidis *et al.*, 2006; Richter & Rosselló-Móra, 2009). By contrast, when strain AX1YPE^T was compared with *A. vinosum* DSM 180^T, *A. minutissimum* DSM 1376^T and *A. renukae* DSM 18713^T it exhibited ANI values of 94.95, 94.63 and 93.52 %, respectively. All these values are below 95 %, an accepted boundary limit of species demarcation using ANI (Kim *et al.*, 2014; Konstantinidis *et al.*, 2006; Richter & Rosselló-Móra, 2009). Besides, PCR fingerprinting patterns obtained by the use of the primer sets BOX A1R and RAPD (SMO3) have shown that the banding profile of strain AX1YPE^T clearly differentiates it from *A. vinosum* DSM180^T and *A. minutissimum* DSM 1376^T (Fig. S2).

Strain AX1YPE^T could clearly be differentiated from other species of the genus *Allochromatium* based on its ability to use a variety of nitrogen sources. We conclude that strain AX1YPE is sufficiently distinct in its phenotype, genotype and phylogeny to represent a novel species in the genus *Allochromatium*, for which the name *Allochromatium humboldtianum* sp. nov. is proposed.

Description of *Allochromatium humboldtianum* sp. nov.

Allochromatium humboldtianum (hum.boldt.i.a′num. N.L. neut. adj. *humboldtianum* named after the famous German naturalist Alexander von Humboldt).

Cells are Gram-stain-negative rods, measuring about 2 × 3 μm in size and are motile by a polar flagellum. Culture suspension is brownish red. Pigments include bacteriochorophyll *a* and

Table 1. Characteristics of strain AX1YPE^T and species of the genus *Allochromatium*

Strains: 1, AX1YPE^T; 2, *A. vinosum* DSM 180^T; 3, *A. minutissimum* DSM 1376^T; 4, *A. renukae* JA136^T; 5, *A. phaeobacterium* JA 144^T; 6, *A. warmingii* DSM 173^T. Data for taxa 1–3 were obtained in this study; data for taxa 4–6 were obtained from Pfennig & Trüper (1992), Anil Kumar *et al.* (2008) and Srinivas *et al.* (2009), except where indicated. All organic substrates and nitrogen sources were tested during photolithoheterotrophic growth with $Na_2S.9H_2O$ (4 mM). NA, Not applicable; NR, not reported

Characteristic	1	2	3	4	5	6
Cell shape	Ovoid to rod	Rod	Rod	Avoid to rod	Rod	Rod
Cell size (width × length; μm)	2.0 × 3.0	2.0 × 2.5–6.0	1.0–1.2 × 2.0	2.0–2.5 × 3.0–5.0	1.0–1.5 × 2.0–4.0	3.5–4.0 × 5.0–11.0
Flagellation	Single polar	Single polar	Single polar	NR	NR	NR
Colour of cell suspension	Brown–red	Brown–red	Brown–red	Purple–violet	Brown	Purple–violet
Carotenoids	Spirilloxanthin	Spirilloxanthin	Spirilloxanthin	Lycopene	Rhodopinal	Rhodopinal
NaCl optimum (%)	3	0–1	0–1	None	None	NR
Vitamin B_{12} requirement	−	−	−	−	−	+
Optimum temperature (°C)	25–35	25–35	25–35	28–32	30	25–30
Optimum pH (range)	6.9–8.0 (6.5–8.5)	7.0–7.3 (6.5–7.6)	7.0–7.3 (6.5–7.6)	7.2–8.0 (6.8–8.5)	7.5 (7.0–8.0)	7.0 (6.5–7.3)
Photolithoautotrophy	+	+	+	+	+	+
Photoheterotrophy	+	+	+	+	+	+
Nitrogen source(s) utilized	Ammonium salts, N_2 urea, glutamate	Ammonium salts	Ammonium salts	Armonium salts, N_2	Ammonium salts	Ammonium salts
Organic substrates utilized:						
Formate	+	+	+	−	NR	−
Acetate	+	+	+	+	−	+
Fumarate	+	+	+	+	+	−
Malate	+	+	+	+	+	−
Pyruvate	+	+	+	+	NR	NR
Glycolate	+	+	+	−	−	−
Fructose	−	−	−	NR	+	NR
Glucose	+	−	−	+	−	−
Molecular features						
DDH relatedness (%)*	100	59 ± 5.44	64 ± 2.38	61 ± 3.59	NA	NA
16S rRNA similarity (%)*	100	98.2	98.2	97.3	98.1	96.8
DNA G+C content (mol %)	63.9 (T_m)	64.3 (T_m)	63.7 (T_m)	63.3 (HPLC)	59.8 (HPLC)	55.1–60.2 (T_m)

*Data were obtained in this study.

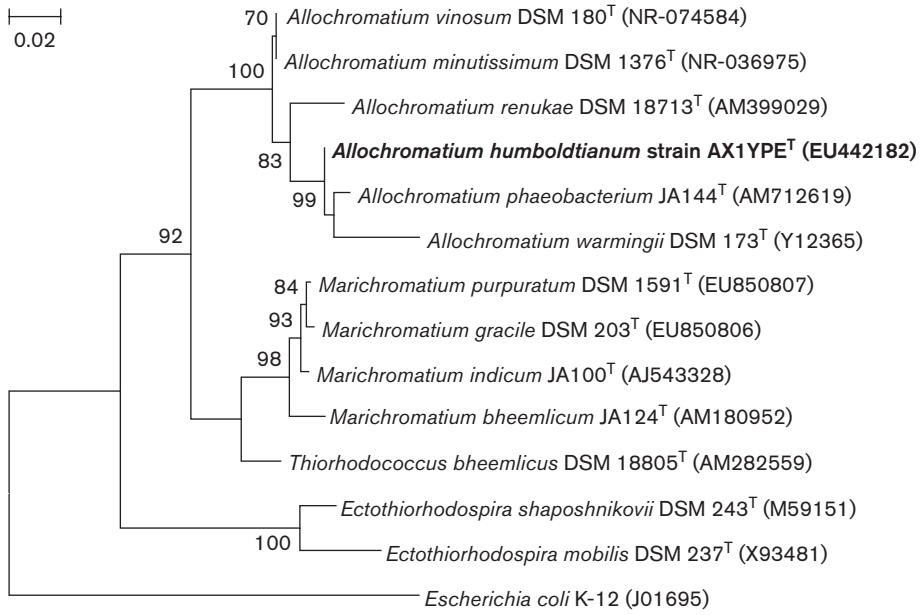

Fig. 2. 16S rRNA-based tree reflecting the phylogenetic relationship between strain AX1YPE^T and reference species in the family *Chromatiaceae*. The tree was reconstructed with the maximum-likelihood method using the program PhyML of nearly full-length 16S rRNA gene sequences. Accession numbers of the reference sequences are given in parentheses. *Escherichia coli* K-12 was used as outgroup. Bar, 2 % estimated sequence divergence.

carotenoids of the spirilloxanthin series, and the photosynthetic membrane system is of the vesicular type. No addition of vitamin B_{12} is required for growth. Growth modes of this strictly anaerobic bacterium are photolithoautotrophy, photolithoheterotrophy and chemomixotrophy. Sulfide and thiosulfate serve as electron donors and elemental sulfur formed as an intermediate oxidation product is stored inside the cells. Organic substrates such as formate, lactate, fumarate, malate, pyruvate, glycolate and glucose are photoassimilated in the presence of sulfide and absence of carbonate. The organism is able to use the nitrogen compounds ammonium, urea and glutamate. Diazotrophic growth is also possible. Grows with 0–5 % NaCl (optimum 3 %), at 25–35 °C (optimum 30 °C) and at pH 6.5–8.5 (optimum pH 6.9–8.0). Tolerates up to 8 mM sulfide.

The type strain is AX1YPE^T (=DSM 21881^T=KCTC 15448), which was isolated from marine coastal sediments at 47 m depth in Callao Bay, Peru. The DNA G+C content of the type strain is 63.9 mol% (T_m).

Acknowledgements

We thank the captain and the crew of the R/V *Jose Olaya*, our scientific colleagues at Instituto del Mar del Peru (IMARPE), and all the scientific team on board the April 2005 cruise. We acknowledge Dr K. Palinska, University of Oldenburg, Germany, for her technical assistance with electron microscopy and Professor Dr M. Friedrich, Department of Microbial Ecophysiology, University of Bremen, for the use of his laboratories to finish the experiments (W. S.). Funding was provided by the Max Planck Society; W. S. was also supported by a fellowship from the 'Zentrale Kommission für Forschung und wissenschaftlichen Nachwuchs', University of Bremen. We are grateful to Professor Dr Hans G.Trüper, Universität Bonn, Germany, for his suggestion regarding the specific epithet. W. S. was a member of the International Max Planck Research School of Marine Microbiology (MarMic). Financial support by a DAAD stipend and through the MarMic Alumni Programme to W. S. is gratefully acknowledged.

References

Anil Kumar, P., Srinivas, T. N. R., Sasikala, Ch. & Ramana, ChV. (2008). *Allochromatium renukae* sp. nov. *Int J Syst Evol Microbiol* **58**, 404–407.

De Ley, J., Cattoir, H. & Reynaerts, A. (1970). The quantitative measurement of DNA hybridization from renaturation rates. *Eur J Biochem* **12**, 133–142.

Goris, J., Konstantinidis, K. T., Klappenbach, J. A., Coenye, T., Vandamme, P. & Tiedje, J. M. (2007). DNA-DNA hybridization values and their relationship to whole-genome sequence similarities. *Int J Syst Evol Microbiol* **57**, 81–91.

Guindon, S. & Gascuel, O. (2003). A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. *Syst Biol* **52**, 696–704.

Imhoff, J. F. & Caumette, P. (2004). Recommended standards for the description of new species of anoxygenic phototrophic bacteria. *Int J Syst Evol Microbiol* **54**, 1415–1421.

Imhoff, J. F., Süling, J. & Petri, R. (1998). Phylogenetic relationships among the *Chromatiaceae*, their taxonomic reclassification and description of the new genera *Allochromatium*, *Halochromatium*, *Isochromatium*, *Marichromatium*, *Thiococcus*, *Thiohalocapsa* and *Thermochromatium*. *Int J Syst Bacteriol* **48**, 1129–1143.

Kim, M., Oh, H. S., Park, S. C. & Chun, J. (2014). Towards a taxonomic coherence between average nucleotide identity and 16S rRNA gene sequence similarity for species demarcation of prokaryotes. *Int J Syst Evol Microbiol* **64**, 346–351.

Konstantinidis, K. T., Ramette, A. & Tiedje, J. M. (2006). The bacterial species definition in the genomic era. *Phil Trans R Soc B* **361**, 1929–1940.

Marmur, J. (1961). A procedure for the isolation of deoxyribonucleic acid from micro-organisms. *J Mol Biol* **3**, 208–218.

Marmur, J. & Doty, P. (1962). Determination of the base composition of deoxyribonucleic acid from its thermal denaturation temperature. *J Mol Biol* **5**, 109–118.

Muyzer, G., de Waal, E. C. & Uitterlinden, A. G. (1993). Profiling of complex microbial populations by denaturing gradient gel electrophoresis analysis of polymerase chain reaction-amplified genes coding for 16S rRNA. *Appl Environ Microbiol* **59**, 695–700.

Pfennig, N. & Trüper, H. G. (1981). Isolation of members of the families Chromatiaceae and Chlorobiaceae. In *The Prokaryotes*, pp. 279–289. Edited by M. P. Starr, H. Stolp, H. G. Trüper, A. Balows & H. G. Schlegel. Berlin: Springer.

Pfennig, N. & Trüper, H. G. (1992). The family Chromatiaceae. In *The Prokaryotes*, pp. 3200–3221. Edited by A. Balows, H. G. Trüper, M. Dworkin, W. Harder & K. H. Schleifer. Berlin: Springer.

Richter, M. & Rosselló-Móra, R. (2009). Shifting the genomic gold standard for the prokaryotic species definition. *Proc Natl Acad Sci U S A* **106**, 19126–19131.

Serrano, W., Amann, R. & Fischer, U. (2009). A new moderately thermophilic and high sulfide tolerant biotype of *Marichromatium gracile*, isolated from tidal sediments of the German Wadden Sea: *Marichromatium gracile* biotype *thermosulfidiphilum*. *Syst Appl Microbiol* **32**, 1–7.

Serrano, W., Amann, R., Rosselló-Móra, R., Herbert, R. A. & Fischer, U. (2011). The genus *Allochromatium* (Chromatiales *Chromatiaceae*) revisited: a study on its intragenic structure based on multilocus sequence analysis (MLSA) and DNA-DNA hybridization (DDH). *Syst Appl Microbiol* **34**, 590–594.

Srinivas, T. N. R., Anil Kumar, P., Sucharitha, K., Sasikala, Ch. & Ramana, ChV. (2009). *Allochromatium phaeobacterium* sp. nov. *Int J Syst Evol Microbiol* **59**, 750–753.

Tindall, B. J., Rosselló-Móra, R., Busse, H. -J., Ludwig, W. & Kämpfer, P. (2010). Notes on the characterization of prokaryote strains for taxonomic purposes. *Int J Syst Evol Microbiol* **60**, 249–266.

Wayne, L. G., Brenner, D. J., Colwell, R. R., Grimont, P. A. D., Kandler, O., Krichevsky, M. I., Moore, L. H., Moore, W. E. C., Murray, R. G. E. & other authors (1987). International Committee on Systematic Bacteriology. Report of the ad hoc committee on reconciliation of approaches to bacterial systematics. *Int J Syst Bacteriol* **37**, 463–464.

Weissgerber, T., Zigann, R., Bruce, D., Chang, Y. -J., Detter, J. C., Han, C., Hauser, L., Jeffries, C. D., Land, M. & other authors (2011). Complete genome sequence of *Allochromatium vinosum* DSM 180^{T}. *Stand Genomic Sci* **5**, 311–330.