**NOTE**

**Pelczaria aurantia ATCC 49321** (= DSM 12801) is a strain of Kocuria rosea (Flügge 1886) Stackebrandt et al. 1995

Peter Schumann, Brian J. Tindall, Ulrike Mendrock, Ina Kramer and Erko Stackebrandt

Author for correspondence: Erko Stackebrandt. Tel: +49 531 2616352. Fax: +49 531 2616418. E-mail: erko@dsmz.de

Phylogenetic and chemotaxonomic analyses of Pelczaria aurantia ATCC 49321 (= DSM 12801) indicate that this species is very closely related to Kocuria rosea. The DNA–DNA reassociation value of 87.1% determined for the type strains of the two species supports this finding. The results of phylogenetic analysis of the 16S rDNA of a subculture of the original strain of Pelczaria aurantia, deposited at the National Institutes of Health, Bethesda, MD, USA, as ‘Neisseria aurantia’, are identical to those for strain ATCC 49321T and indicate that Pelczaria aurantia ATCC 49321T is an authentic subculture of the original culture described by Poston (1993). On the basis of these findings it is concluded that P. aurantia ATCC 49321T and K. rosea DSM 20447T are members of the same taxon. The taxonomic consequences of this union are discussed.

Keywords: Pelczaria aurantia, Kocuria rosea, Actinobacteria, Micrococcineae

**Pelczaria aurantia** ATCC 49321T was proposed as a new species within a new genus (Poston, 1993) on the basis of the fact that it was a Gram-positive coccus, often appearing as a doublet or tetrad, exhibiting histidine in the cell wall and a DNA G+C content of 59 mol%. Polar lipid analysis indicated that the phospholipids phosphatidylglycerol, phosphatidylethanolamine and phosphatidylethanolamine were present and the major carotenoid pigment was tentatively identified as rhodopin. On the basis of the fact that it shared only very low DNA similarities with Gram-positive and Gram-negative cocci as determined by DNA reassociation studies (<20%), in addition to the fact that there was also no significant RNA–DNA hybridization (<5%) to the same range of taxa, the taxonomic distinctness of this organism was indicated. Subsequently, this organism has been included in a chemotaxonomic survey which showed that the type strain (the only strain included in the species) contained l-lysine as the diamino acid of the peptidoglycan (type A3z, according to Schleifer & Kandler, 1972) and menaquinones of the MK-8(Hα) type (Miyadoh, 1997). Because this combination is typical for some Gram-positive cocci, *P. aurantia* was included in the *Atlas of Actinomycetes* (Miyadoh, 1997) in the list of chemotaxonomic characteristics of actinobacteria and related taxa. However, neither a strain designation nor the phylogenetic analysis was included in the *Atlas*.

During the course of the extension of the 16S rDNA database of type strains of actinobacterial species we analysed the 16S rDNA of *P. aurantia* DSM 12801T (= ATCC 49321T, kindly provided by the American Type Culture Collection). Extraction of DNA, PCR amplification and cycle sequencing analysis of the PCR product were performed according to described methods (Rainey et al., 1996). The phylogenetic analysis of the 16S rDNA, based upon 1502 nucleotides, revealed that strain DSM 12801T was 100% identical to the type strain of *Kocuria rosea* (DSM 20447T), which agrees with the chemotaxonomic characterization of these strains as indicated in the *Atlas of Actinomycetes* (Miyadoh, 1997). However, this contradicts the data published by Poston (1993) and in order to confirm the presence of these properties in strain DSM 12801T (= ATCC 49321T) the most widely used chemotaxonomic characteristics were investigated for *P. aurantia* DSM 12801T and *K. rosea* DSM 20447T by using previously published methods (Groth et al., 1996). The results of this chemotaxonomic investigation were compared to the data of Poston (1993) given in the original description for strain ATCC 49321T and the data given in the *Atlas of Actinomycetes* (Miyadoh, 1997) (Table 1). Examination of the results obtained in this study and the results published by Poston (1993) needs further comment.
In our study on *P. aurantia* DSM 12801T and *K. rosea* DSM 20447T, using published methods for DNA isolation (Cashion et al., 1977; Escara & Hutton, 1980) and the spectrophotometric measurement of DNA–DNA reassociation (Huß et al., 1983; Jahnke, 1992), these two strains show 87.1% DNA–DNA similarity, which would indicate that they are members of the same species. This is also supported by the fact that the two strains share 100% 16S rDNA sequence similarity. This is in contrast to data published by Poston (1993), which indicated that there was no significant DNA–DNA hybridization or RNA–DNA hybridization between these two strains (Poston, 1993). They did, however, find canthaxanthin in a number of the taxa within the *Kocuria* group (see Ratledge & Wilkinson, 1988; B. J. Tindall, unpublished results).

In our study, the cellular fatty acid profiles of *P. aurantia* DSM 12801T and *K. rosea* DSM 20447T were found to be very similar and contained both 12-methyl tetradecanoic (ai-C15:0) and 13-methyl tetradecanoic acid (i-C15:0) as the predominating components (Table 2). The polar lipid composition reported for *P. aurantia* (Poston, 1993) is phosphatidylglycerol, phosphatidylethanolamine and phosphatidylcholine, which, like the DNA–DNA and RNA–DNA data, indicates that this organism is neither a member of the species *K. rosea*, nor a member of the genus *Kocuria*. In fact, this polar lipid pattern is not found in any of the taxa within the *Arthrobacter–Micrococcus* line of descent, but is not uncommon in members of the α subclass of the Proteobacteria (see Ratledge & Wilkinson, 1988; B. J. Tindall, unpublished results).

Although we did not examine the pigment composition of *P. aurantia*, it should be noted that the methods used by Poston (1993), HPLC and photodiode array detection, were sufficient to tentatively identify the major pigment as rhodopin. Despite the reports that *K. rosea* (ATCC 516) produces canthaxanthin, together with a range of other bicyclic carotene derivatives (Cooney & Berry, 1981; Cooney et al., 1966; Schwartzel & Cooney, 1970, 1972; Ungers & Cooney, 1968), Nelis & De Leenheer (1989) were unable to find these pigments in *K. rosea* CCM 839 or ATCC 516 (which are identical strains). They did, however, find canthaxanthin in a *Brevibacterium* species. It is significant that Poston (1993) used the methods of Nelis & De Leenheer (1989), which separate and spectroscopically detect

### Table 1. Chemotaxonomic characteristics of *P. aurantia* (as reported in different studies) and of *K. rosea*

<table>
<thead>
<tr>
<th>Property</th>
<th><em>P. aurantia</em> ATCC 49321T</th>
<th><em>P. aurantia</em> 1</th>
<th><em>P. aurantia</em> DSM 12801T</th>
<th><em>K. rosea</em> DSM 20447T</th>
</tr>
</thead>
<tbody>
<tr>
<td>Petidoglycan: amino acid ratio</td>
<td>Glu: Ser: His: Lys: Ala</td>
<td>ND</td>
<td>Glu: Lys: Ala (1:1:5:3:4)</td>
<td>ND</td>
</tr>
<tr>
<td>Peptidoglycan type</td>
<td>ND</td>
<td>A3x, Lys-Ala</td>
<td>A3x, Lys-Ala</td>
<td>A3x, Lys-Ala</td>
</tr>
<tr>
<td>Polar lipids</td>
<td>PC, PE, PG</td>
<td>ND</td>
<td>PG, DPG, PL, GL</td>
<td>PG, DPG, (PI, PL, GL)</td>
</tr>
<tr>
<td>Menaquiones</td>
<td>ND</td>
<td>MK-8(H4)</td>
<td>MK-8(H4), MK-9(H4), MK-10(H4)</td>
<td>MK-8(H4), MK-9(H4), MK-10(H4)</td>
</tr>
<tr>
<td>DNA G+C content (mol %)</td>
<td>59 (HPLC), 60 (Tm)</td>
<td>59</td>
<td>72 (HPLC)</td>
<td>66–75 (Tm)</td>
</tr>
</tbody>
</table>

* Poston (1993).
† Miyadoh (1997).
‡ This study.
§ Stackebrandt et al. (1995).

### Table 2. Percentage cellular fatty acid composition of the type strains of *P. aurantia* and of *K. rosea*

<table>
<thead>
<tr>
<th>Organism</th>
<th>C14:0</th>
<th>C16:0</th>
<th>C16:1</th>
<th>i-C14:0</th>
<th>i-C15:0</th>
<th>i-C16:0</th>
<th>i-C17:0</th>
<th>ai-C15:0</th>
<th>ai-C17:0</th>
<th>ai-C17:1</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>P. aurantia</em></td>
<td>3.4</td>
<td>6.4</td>
<td>2.1</td>
<td>2.0</td>
<td>11.6</td>
<td>2.5</td>
<td>1.1</td>
<td>6.5</td>
<td>4.5</td>
<td>3.0</td>
<td>98.6</td>
</tr>
<tr>
<td><em>K. rosea</em></td>
<td>1.5</td>
<td>1.6</td>
<td>5.9</td>
<td>1.8</td>
<td>7.6</td>
<td>1.6</td>
<td>7.0</td>
<td>4.3</td>
<td>4.3</td>
<td>98.2</td>
<td></td>
</tr>
</tbody>
</table>

* Stackebrandt et al. (1995).
rhodopin, provides details of the separation and detection of canthaxanthin and showed that *K. rosea* CCM 839 or ATCC 516 had a distinctive pigment pattern that was not identical to *P. aurantia* (Poston, 1993). It should be noted that rhodopin is not uncommon in members of the anoxygenic phototrophic bacteria within the α subclass of the *Proteobacteria* (Schmidt, 1978).

The obvious discrepancy between the chemotaxonomic data given for strain ATCC 49321T of Poston (1993), originally deposited in the American Type Culture Collection as *Neisseria aurantia*, and the data for strain DSM 12801T (= ATCC 49321T) obtained from the ATCC in 1999 could be explained by the failure to maintain the original culture in any of the collections involved. To verify the authenticity of strain ATCC 49321T (= DSM 12801T), Michael Poston, formerly of the Laboratory of Biochemistry, National Institutes of Health, Bethesda, MD, USA, arranged to ship a subculture of his original culture (labelled 'N. aurantia') to the DSMZ, where this strain was analysed with respect to the almost complete 16S rDNA sequence. The sequence thus obtained was 100% identical to those of *P. aurantia* DSM 12801T (= ATCC 49321T) and *K. rosea* DSM 20447T, which confirms the authenticity of ATCC 49321T and DSM 12801T subsequently subcultured from the ATCC strain.

At the time of description of *P. aurantia* Poston 1994 (see Poston, 1993), members of the genus *Kocuria* were classified as members of the genus *Micrococcus*, i.e. *K. rosea* was classified as *Micrococcus roseus*. *M. roseus* ATCC 186T was included in the DNA–DNA and RNA–DNA reassociation experiments between *P. aurantia* ATCC 49321T and a variety of Gram-positive and Gram-negative bacteria (Poston, 1993), but no significant DNA–DNA reassociation was reported with any organism (12% for low stringency; < 5% for high stringency), nor could any close evolutionary relationship be detected by RNA–DNA hybridization. The lack of reassociation between any of the reference species was, among others, the main rationale for describing strain ATCC 49321T as the type strain of a new species within a new genus. Histidine, the presence of which in cell wall preparations is the unique feature of *P. aurantia*, is probably not a constituent of the peptidoglycan, but a contaminating cellular protein.

On the basis of the 16S rDNA sequence data, DNA–DNA hybridization data, chemotaxonomic data and morphological data, we consider that DSM 12801T (= ATCC 49321T) (the type strain of *P. aurantia*) and *K. rosea* DSM 20447T are strains of the same species. However, we consider that the data that we have collected does not allow us to apply Rule 42 of the International Code of Nomenclature of Bacteria (Lapage et al., 1992) and simply unify the genera *Pelczaria* Poston 1994 and *Kocuria* Stackebrandt et al. 1995. In an accompanying Request for an Opinion (Tindall et al., 2000), we propose that ATCC 49321T (= DSM 12801T) be placed in the species *K. rosea* (Flügge 1886) Stackebrandt et al. 1995 and that the generic name *Kocuria* Stackebrandt et al. 1995 be retained on the basis of the fact that there are significant differences between the circumscription of the taxon *P. aurantia* (Poston, 1994) and the properties of the strains currently in circulation, which are supposed to represent the type strain of the species.

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**References**


P. Schumann and others


