The genus *Marinomonas* is composed of Gram-negative bacterial strains that have been found distributed across many different marine environments (Sanchez-Amat & Solano, 2005). The number of species in this genus has increased during recent years, after the initial description of the genus by Van Landschoot & De Ley (1983). At the time of writing, the list of species with validly published names in the genus *Marinomonas* included *M. communis* and *M. vaga* (formerly *Alteromonas communis* and *A. vaga*; Van Landschoot & De Ley, 1983; Baumann et al., 1972), *M. mediterranea* (Solano & Sanchez-Amat, 1999), *M. primoryensis* (Espinosa et al., 2003), *M. pontica* (Ivanova et al., 2005), *M. dokdonensis* (Yoon et al., 2005), *M. aquimarina* (Macian et al., 2005), *M. ushuaiensis* (Prabagaran et al., 2005), *M. polaris* (Gupta et al., 2006), *M. ostreistagni* (Lau et al., 2006), *M. arctica* (Zhang et al., 2008), *M. basaltis* (Chang et al., 2008), *M. arenicola* (Romanenko et al., 2009), *M. balearica* and *M. pollencensis* (Espinosa et al., 2010), *M. brasiliensis* has since been added to the genus and *M. communis* and *M. basaltis* have been recognized as heterotypic synonyms (Chimetto et al., 2011).

It has been reported that the microbiota of the marine seagrass *Posidonia oceanica* contains a diverse range of species of the genus *Marinomonas*, namely, *M. balearica*, *M. pollencensis* and *M. mediterranea* (Espinosa et al., 2010). Here, we report the characterization of five novel strains of the genus *Marinomonas* isolated from *Posidonia oceanica*.
Samples of seagrass were collected from different meadows on the coast of the Balearic Islands, Spain, and processed as described previously (Espinosa et al., 2010). After plating on Marine Agar 2216 and incubating for 24–72 h at 25 °C, several hundreds of strains were isolated. In a preliminary step, partial 16S rRNA gene sequences, corresponding approximately to the fragment between position 70 and 660 of the 16S rRNA gene of Escherichia coli, were obtained from all the isolated strains by the Molecular Diagnosis Center (MDC, Orihuela, Spain) following the method of Martínez-Murcia et al. (1999). DNA sequences were determined by direct sequencing of the PCR products using an ABI 3100 Avant sequencer (Applied Biosystems).

Searches using BLAST (Altschul et al., 1997) were performed with the sequences obtained and those showing higher identity to 16S rRNA sequences of species of the genus Marinomonas. The highest DNA–DNA relatedness value was 31% for M. pontica 46-16T, the next lowest value being 29% for M. mediiterranea, which was also highly related to M. alcarazii, M. foliarum and M. pontica 46-16T (Table 1). Taking into consideration that M. mediterranea, M. alcarazii and M. pollencensis were also detected in the culturable microbiota of P. oceanica, Fig. 1 shows that this plant harbours a great phylogenetic diversity of strains of the genus Marinomonas, including strains from most of the branches within the genus.

It was found that, in all cases, these values were much lower than the value of 70%, recommended as the threshold for the delineation of bacterial species (Wayne et al., 1987). The highest DNA–DNA relatedness value was 31% between M. pontica 46-16T and strain IVIA-Po-155T. The DNA–DNA relatedness between M. pontica 46-16T and strain IVIA-Po-14b T was 26.8% in spite of a 99.5% 16S rRNA gene sequence similarity. M. pontica 46-16T differed from the strains studied in that it was oxidase-positive, able to use m-hydroxybenzoate as carbon source (Table 1) and did not contain the unknown fatty acid of ECL 11.799 that was detected in other strains (Table 2). It is important to point out that, in spite of the differences indicated, the major fatty acid profiles of all the strains analysed in this study agreed with those reported for other members of the genus Marinomonas (Espinosa et al., 2010). Strain IVIA-Po-14b T was very similar to M. pontica 46-16T, generously gifted by E. Ivanova, and M. dokonensis DSW10-10T, obtained from the DSMZ, were used as reference strains since they were the species that proved to be most closely related to the novel strains.

The phylogenetic analysis confirmed the results of the BLAST searches, indicating that the novel strains characterized in this study belonged to the genus Marinomonas (Fig. 1). Taking into consideration that M. mediterranea, M. alcarazii and M. pollencensis were also detected in the culturable microbiota of P. oceanica, Fig. 1 shows that this plant harbours a great phylogenetic diversity of strains of the genus Marinomonas, including strains from most of the branches within the genus.

Strains IVIA-Po-14b T, IVIA-Po-145 T and IVIA-Po-155 T were most closely related to M. pontica 46-16T (Fig. 1); the 16S rRNA gene sequence similarities between these strains and M. pontica 46-16T were 99.5 ± 0.2, 98.2 ± 0.2 and 99.0 ± 0.2%, respectively. In order to determine whether they belonged to the same species, DNA–DNA hybridization studies were performed comparing these four strains. It was found that, in all cases, these values were much lower than the value of 70%, recommended as the threshold for the delineation of bacterial species (Wayne et al., 1987). The highest DNA–DNA relatedness value was 31% between M. pontica 46-16T and strain IVIA-Po-155T. The DNA–DNA relatedness between M. pontica 46-16T and strain IVIA-Po-14b T was 26.8% in spite of a 99.5% 16S rRNA gene sequence similarity.

M. pontica 46-16T differed from the strains studied in that it was oxidase-positive, able to use m-hydroxybenzoate as carbon source (Table 1) and did not contain the unknown fatty acid of ECL 11.799 that was detected in other strains (Table 2). It is important to point out that, in spite of the differences indicated, the major fatty acid profiles of all the strains analysed in this study agreed with those reported for other members of the genus Marinomonas (Espinosa et al., 2010). Strain IVIA-Po-14b T was very similar to M. pontica 46-16T in many respects but differed from it, in addition to the characteristics mentioned above, in its capacity to use galactose but not m-hydroxybenzoate as sole carbon source (Tables 1 and 2). Strains IVIA-Po-145 T and IVIA-Po-155 T differed from strains IVIA-Po-14b T and M. pontica 46-16T in their cell shape, being curved rods. The capacity to use L-tyrosine as sole carbon and energy source was a distinctive character of strain IVIA-Po-14b T (Table 1).

These results indicate that strains IVIA-Po-14b T, IVIA-Po-145 T and IVIA-Po-155 T can be considered as novel species of the genus Marinomonas, for which the names M. alcarazii sp. nov., M. rhizomae sp. nov. and M. foliarum sp. nov. are proposed.
Strains IVIA-Po-181T and IVIA-Po-159T were phylogenetically related to *M. dokdonensis* DSW10-10T (Fig. 1). However, these two strains formed a well-defined separate cluster. This cluster represents the most abundant cultivable group of species of the genus *Marinomonas* isolated from the microbiota of *P. oceanica* since 35 out of the 43 strains that were initially isolated belong to it. Strains IVIA-Po-181T and IVIA-Po-159T showed 16S rRNA gene sequence similarities in the range of 98.3–98.5% with *M. dokdonensis* DSW10-10T and although the similarity value between the two strains was higher than 99.4%, they showed a DNA–DNA relatedness of only 42.4% to each other, suggesting that they represent different species. This assumption was supported by the fact that strains IVIA-Po-181T and IVIA-Po-159T showed DNA–DNA relatedness values of 10.5 and 25.9%, respectively, to their closest relative, *M. dokdonensis* DSW10-10T.

In terms of their phenotypic characteristics, cells of strains IVIA-Po-181T and IVIA-Po-159T differed from *M. dokdonensis* DSW10-10T, by being helical in shape and oxidase-negative (Table 1) and by containing the fatty acid C12:1 3-OH but not C12:0 (Table 2). The detection of an unknown fatty acid of ECL 11.799 in strain IVIA-Po-181T was the main differential characteristic between these two novel strains (Table 2). Accordingly, strains IVIA-Po-181T and

**Table 1.** Phenotypic characteristics of strains IVIA-Po-14bT, IVIA-Po-145T, IVIA-Po-155T, IVIA-Po-181T, IVIA-Po-159T and the type strains of closely related species of the genus *Marinomonas*

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
</tr>
</thead>
<tbody>
<tr>
<td>DNA G+C content (mol%)</td>
<td>46</td>
<td>46.2</td>
<td>45.5</td>
<td>46.2</td>
<td>45.5</td>
<td>44.7</td>
<td>47.8</td>
</tr>
<tr>
<td>Cell shape</td>
<td>Helical*</td>
<td>Helical</td>
<td>Curved</td>
<td>Curved</td>
<td>Straight</td>
<td>Helical</td>
<td>Helical</td>
</tr>
<tr>
<td>Oxidase</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>NaCl tolerance (%)</td>
<td>0.5–10</td>
<td>0.5–10</td>
<td>0.5–10</td>
<td>0.5–10</td>
<td>2–10</td>
<td>0.5–10</td>
<td>0.5–10</td>
</tr>
<tr>
<td>Temperature range (°C)</td>
<td>4–33</td>
<td>5–37</td>
<td>5–25</td>
<td>5–25</td>
<td>4–37</td>
<td>5–25</td>
<td>5–37</td>
</tr>
<tr>
<td>Lipase</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>w</td>
</tr>
<tr>
<td>Carbon source utilization</td>
<td>Galactose</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Sucrose</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Glycine</td>
<td>+</td>
<td>+</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>L-Tyrosine</td>
<td>–</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td></td>
<td>m-Hydroxybenzoate</td>
<td>+</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

*Data from Espinosa et al. (2010).*
†Data from the present study.
Table 2. Cellular fatty acid composition of strains IVIA-Po-14bT, IVIA-Po-145T, IVIA-Po-155T, IVIA-Po-181T, IVIA-Po-159T and type strains of closely related species of the genus Marinomonas

<table>
<thead>
<tr>
<th>Fatty acid</th>
<th>1*</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
</tr>
</thead>
<tbody>
<tr>
<td>C10:0 3-OH</td>
<td>6.54</td>
<td>9.72</td>
<td>7.84</td>
<td>8.25</td>
<td>7.61</td>
<td>12.64</td>
<td>15.70</td>
</tr>
<tr>
<td>Unknown</td>
<td>4.01</td>
<td>0.64</td>
<td>0.39</td>
<td>8.43</td>
<td>5.01</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>11.799</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C12:0</td>
<td>1.36</td>
<td>-</td>
<td>1.21</td>
<td>3.65</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>C12:1 3-OH</td>
<td>8.40</td>
<td>3.65</td>
<td>5.92</td>
<td>7.70</td>
<td>-</td>
<td>6.71</td>
<td>14.26</td>
</tr>
<tr>
<td>C14:0</td>
<td>1.60</td>
<td>1.02</td>
<td>1.02</td>
<td>1.12</td>
<td>0.69</td>
<td>1.05</td>
<td>1.17</td>
</tr>
<tr>
<td>C16:0</td>
<td>11.73</td>
<td>11.81</td>
<td>13.30</td>
<td>22.20</td>
<td>8.92</td>
<td>15.15</td>
<td>12.95</td>
</tr>
<tr>
<td>C16:1ω7c</td>
<td>34.26</td>
<td>22.73</td>
<td>24.05</td>
<td>27.25</td>
<td>20.48</td>
<td>13.10</td>
<td>15.96</td>
</tr>
<tr>
<td>C17:0 Cyclo</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>C18:0</td>
<td>2.27</td>
<td>4.35</td>
<td>4.07</td>
<td>3.08</td>
<td>2.46</td>
<td>3.69</td>
<td>3.92</td>
</tr>
<tr>
<td>C18:1ω7c</td>
<td>22.90</td>
<td>43.52</td>
<td>43.60</td>
<td>37.50</td>
<td>40.05</td>
<td>32.83</td>
<td>33.16</td>
</tr>
</tbody>
</table>

*Data from Espinosa et al. (2010).

IVIA-Po-159T, represent two novel species, for which the names M. posidonica sp. nov. and M. aquiplantarum sp. nov. are proposed.

This study has revealed that the seagrass Posidonia oceanica harbours a diverse group of bacterial species belonging to the genus Marinomonas, within which the creation of five novel species is proposed.

**Description of Marinomonas alcarazii sp. nov.**

*Marinomonas alcarazii* (a1.ca.ra’zì.i. N.L. masc. gen. n. alcarazii of Alcaraz, named after Miquel Alcaraz, Spanish scientist).

Cells are helical, motile with a single polar flagellum, Gram-reaction-negative, strictly aerobic, catalase-positive and oxidase-negative. Organic growth factors are not required. Requires Na\(^+\) for growth. Grows in 0.5–10 % (w/v) NaCl and at 5–25 °C. Negative for amylase, agarase, lipase and protease (gelatin) activities. Glucose, maltose, sucrose, sorbitol, L-tyrosine, glutamate, α-ketoglutarate, fructose, malate and acetate are used as carbon and energy sources but lactose, butyrate, valerate, tyrosine, glycine and m-hydroxybenzoate are not. The major fatty acids (>10 % of total fatty acids) are C16:0, C16:1ω7c and C18:1ω7c.

The type strain is IVIA-Po-145T (=CECT 7731T =NCIMB 14672T). The DNA G+C content of the type strain is 46.2 mol%.

**Description of Marinomonas foliarum sp. nov.**

*Marinomonas foliarum* (fo.li.a’rum. L. gen. pl. n. foliarum of leaves, the part of the plant Posidonia oceanica from which the type strain was isolated).

Cells are curved rods, motile with a single polar flagellum, Gram-reaction-negative, strictly aerobic, catalase-positive and oxidase-negative. Organic growth factors are not required. Requires Na\(^+\) for growth. Grows in 0.5–10 % (w/v) NaCl and at 5–25 °C. Negative for amylase, agarase, lipase and protease (gelatin) activities. Glucose, maltose, sucrose, sorbitol, L-tyrosine and m-hydroxybenzoate are not. The major fatty acids (>10 % of total fatty acids) are C16:0, C16:1ω7c and C18:1ω7c.

The type strain is IVIA-Po-155T (=CECT 7730T =NCIMB 14671T). The DNA G+C content of the type strain is 45.5 mol%.

**Description of Marinomonas posidonica sp. nov.**

*Marinomonas posidonica* (po.si.do’ni.ca. N.L. fem. adj. posidonica relating to the genus name Posidonia, of Posidonia oceanica, the plant from which the type strain was isolated).

Cells are curved rods, motile with a single polar flagellum, Gram-reaction-negative, strictly aerobic, catalase-positive and oxidase-negative. Organic growth factors are not required. Requires Na\(^+\) for growth. Grows in 0.5–10 % NaCl and at 5–25 °C. Negative for amylase, agarase, lipase and protease (gelatin) activities. Glucose, maltose, galactose, sucrose, sorbitol, glutamate, α-ketoglutarate, fructose, malate and acetate are used as carbon and energy sources but lactose, butyrate, valerate, tyrosine, glycine and m-hydroxybenzoate are not. The major fatty acids (>10 % of total fatty acids) are C16:0, C16:1ω7c and C18:1ω7c.

The type strain is IVIA-Po-151T (=CECT 7731T =NCIMB 14672T). The DNA G+C content of the type strain is 46.2 mol%.
The type strain is IVIA-Po-181T (=CECT 7376T =NCIMB 14433T). The DNA G+C content of the type strain is 44.7 mol%.

**Description of *Marinomonas aquiplantarum* sp. nov.**

*Marinomonas aquiplantarum* (a.qui.plan.ta’rum. L. fem. n. *aqua* water; L. fem. n. *planta* plant; N.L. gen. pl. n. *aquiplantarum* of aquatic plants, referring to *Posidonia oceanica*).

Cells are helical, motile with polar flagella at one or both poles of the cells, Gram-reaction-negative, strictly aerobic, catalase-positive and oxidase-negative. Organic growth factors are not required. Requires Na+ for growth. Grows in 0.5–10 % (w/v) NaCl and at 5–37 °C. Negative for amylase, agarase and protease (gelatin) activities. Lipase (TWEEN 80) activity is variable with some strains showing a low activity. Glucose, maltose, galactose, sucrose, sorbitol, glutamate, α-ketoglutarate, fructose, malate and acetate are used as carbon and energy sources but lactose, butyrate, valerate, tyrosine and m-hydroxybenzoate are not. The major fatty acids (>10 % of total fatty acids) are C10:0 3-OH, C12:1 3-OH, C16:0, C16:1ω7c and C18:1ω7c.

The type strain is IVIA-Po-159T (=CECT 7732T =NCIMB 14673T). The DNA G+C content of the type strain is 47.8 mol%.

**Acknowledgements**

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**References**


