Morphology and small-subunit rRNA gene sequences of two novel marine ciliates, Metanophrys orientalis spec. nov. and Uronemella sinensis spec. nov. (Protista, Ciliophora, Scuticociliatia), with an improved diagnosis of the genus Uronemella

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The morphology and infraciliature of two novel marine scuticociliates, Metanophrys orientalis spec. nov. and Uronemella sinensis spec. nov., collected from sandy beaches at Qingdao, China, were investigated using live observation and protargol-staining methods. Metanophrys orientalis spec. nov. is distinguished by the following characteristics: marine habitat and a slender to elongate oval body with pointed anterior end and rounded caudal end, in vivo about 25–50 μm long; buccal field about a quarter to a third of body length; nine or ten somatic kineties with dikinetids approximately in anterior half of body, monokinetids in posterior half; membranelles 1 and 2 almost equal in length and composed of two and three longitudinal rows of kinetids respectively; paroral membrane with zigzag structure extending anteriorly to middle portion of membranelle 2; contractile vacuole pore located at posterior end of somatic kinety 1. The genus Uronemella is redefined as follows: marine form with an elongate-elliptical or inverted pear-shaped body; apical plate conspicuous; buccal field about two-thirds of body length, cytostome subequatorially located; oral apparatus Uronema-like; somatic kineties comprising a mixture of dikinetids and monokinetids. Uronemella sinensis spec. nov. is recognized by having an elongate-elliptical body with truncated apical frontal plate, size in vivo about 25–35×15–20 μm, nine or ten somatic kineties, membranelle 1 consisting of two or three basal bodies, contractile vacuole pore at posterior end of somatic kinety 1. This study also compared the small-subunit rRNA gene sequences of these two species with other closely related species to show the sequence divergence, which ranged from 3.53 to 9.60 %. Phylogenetic analyses support the contention that the genus Uronemella is monophyletic, while Metanophrys is non-monophyletic.

INTRODUCTION

As common members of ecosystems in habitats worldwide, the ciliates in the subclass Scuticociliatia exhibit great species richness and biological diversity and often act as symbionts or pathogens of aquatic animals (Fan et al., 2011a; Gao et al., 2012a, b; Lobban et al., 2011; Pan et al., 2010; Song & Wilbert, 2002; Song et al., 2002; Whang et al., 2013). Investigations of scuticociliates in intertidal sediments have demonstrated that this group is much more diverse than previously assumed (Fan et al., 2011a, b; Foissner et al., 1994; Foissner & Wilbert, 1981; Pan et al., 2011; Song, 2000). Moreover, with the application of molecular techniques in taxonomy, species need to be compared not only at the morphological level but also at the molecular level, and there is a particular need for further descriptions and comparisons at the molecular level in this group (Gao et al., 2010, 2013; Grolière et al., 1978; Medlin et al., 1988; Miao et al., 2011; de Puytorac et al., 2011a; Gao et al., 2012a, b; Lobban et al., 2011; Pan et al., 2010; Song & Wilbert, 2002; Song et al., 2002; Whang et al., 2013).
The genus *Uronemella* de Puytorac, 1974 was established by de Puytorac et al. (1974) with *Metanophrys duchoni* as the type species, and four species have since been added to the genus, namely *Metanophrys elongata* (Biggar & Wenrich, 1932) Groliére et al., 1978, *Metanophrys chini* Small & Lynn, 1985, *Metanophrys sinensis* Song & Wilbert, 2000 and *Metanophrys similis* Song et al., 2002. Among them, *Metanophrys sinensis* and *Metanophrys similis* have been found in Chinese seas and described several times (Song & Wilbert, 2000b; Song et al., 2009). This genus is distinguished by the following features: body with pointed anterior end and no apical plate, cytosome above mid-body, membranelle 1 composed of two rows of kinetids, each with six kinetosomes; membranelle 2 equal to membranelle 1 in length, three-rowed, paroral membrane with zigzag structure extending anteriorly to middle portion of membranelle 2, single caudal cilium (Strüder & Wilbert, 1982).

The genus *Uronemella* Song & Wilbert, 2002, meanwhile, comprises three nominal species from marine habitats, *Uronemella filificum* Kahl, 1931, *U. binucleata* (Song, 1993) Song & Wilbert, 2002 and *U. parafilificum* Gong et al., 2007 (see Fig. 4), and is generally recognized by having a prominent buccal field (which accounts for more than 50% of body length), a dominant apical plate and a typical rotatory movement with the help of a sticky thread associated with the caudal cilium (Borror, 1963; Gong et al., 2007; Pérez-Uz et al., 1996; Pérez-Uz & Hope, 1997; Song & Wilbert, 2002; Thompson & Kaneshiro, 1968; Wilbert & Kahan, 1981).

In this paper, two novel species are described and the diagnosis of the genus *Uronemella* is emended based on current observations. Additionally, the paper contributes to the currently very limited molecular data relating to these two genera by comparing their small-subunit (SSU) rRNA gene sequences with those of closely related species.

### METHODS

*Metanophrys orientalis* spec. nov. was collected on 13 October 2010 from Yangkou bathing beach (36°14′N 120°40′E) in Laoshan district, Qingdao, China (water temperature about 23 °C, pH 7.6 and salinity 30%). *Uronemella sinensis* spec. nov. was isolated on 4 October 2010 from the Shilaoren bathing beach (36°5′N 120°27′E) in Laoshan district, Qingdao, China (water temperature 17.2 °C, pH 7.4 and salinity 35%). In each case, the upper 15 cm layer of sand was collected together with some water from the site. Ciliates were maintained in glass Petri dishes (9–10 cm across) as raw cultures for 1 week at room temperature and then isolated by using a glass micropipette.

Isolated cells were observed and photographed *in vivo* using differential interference contrast microscopy. Protargol (Wilbert, 1975) and Chatton–Lwoff (Wilbert & Song, 2008) methods were used to reveal the infraciliature and argyrome, respectively. Counts and measurements of stained specimens were performed at magnifications of 100× to 1250×. Drawings were carried out with the help of a camera lucida (Foissner, 2006). Systematics and terminology are mainly according to Lynn (2008) and Small & Lynn (1985).

The SSU rRNA gene sequences of *Metanophrys orientalis* spec. nov. and *Uronemella sinensis* spec. nov. were deposited in the GenBank database with the accession numbers JN885084 and JN885083, respectively (Gao et al., 2012a), but they were considered as unidentified forms at that time. In this study, these two sequences were compared with those of another 14 morphologically similar species as follows: *Uronema marinum* (GenBank accession no. GQ465466), *Uronema elegans* (AY103090), *Uronema heteromarinum* (FJ870100), *Uronema filificum* (EF486866), *Uronema parafilificum* (HM236337), *Metanophrys sinensis* (HM236336), *Metanophrys similis* (AY314803), *Paranophrys magna* (JN885089), *Mesanophrys carini* (JN885085), *Parauronema venezuelanum* (JN885087), *Glaucocena trihyneme* (GQ214552), *Miaminiopsis avidus* (JN885091), *Anoplyroides haenophila* (AF107779) and *Homalogastra setosa* (EF158844). Sequences were aligned using CLUSTAL W implemented in BioEdit 7.0 (Hall 1999) using pairwise analysis.

Bayesian inference (BI) analyses were performed with MrBayes version 3.1.2 (Ronquist & Huelsenbeck, 2003) using the GTR + I + G model selected by MrModeltest version 2.2 (Nylander, 2004) according to the AIC criterion. Markov chain Monte Carlo simulations were run with two sets of four chains using the default settings: chain length 1 000 000–3 000 000 generations, with trees sampled every 100 generations. The first 25% of sampled trees were discarded as burn-in. All remaining trees were used to calculate posterior probabilities using a majority rule consensus. Maximum-likelihood (ML) trees were reconstructed with PhyML version 2.4.4 (Guindon & Gascuel, 2003) using the best model according to the AIC criterion selected by Modeltest version 3.4 (Posada & Crandall, 1998). The reliability of internal branches was assessed using non-parametric bootstrapping with 1000 replicates. Phylogenetic trees were visualized with TreeView version 1.6.6 (Page, 1996) and MEGA version 4 (Tamura et al., 2007).

### RESULTS AND DISCUSSION

*Metanophrys orientalis* spec. nov. (Figs 1, 2 and S1; Tables 1 and 2) (subclass Scuticociliata Small, 1967; order Philasterida Small, 1967; genus *Metanophrys* de Puytorac et al., 1974)

**Diagnosis.** Medium-sized, slender to elongated oval, *in vivo* about 25–50 µm with pointed anterior end; buccal field about a quarter to a third of body length; nine or ten somatic kineties with dikinetids approximately in anterior half of body length; membranelle 1 (M1) composed of two rows of kinetids, each with six kinetosomes; membranelle 2 (M2) the same length as membranelle 1, three-rowed; contractile vacuole pore located at posterior end of somatic kinety 1; single caudal cilium present; marine habitat.

**Type locality.** Yangkou bathing beach, Laoshan district of Qingdao, northern China (36°14′N 120°40′E).

**Type slides.** The holotype slide (registration no. PXM-2010101301) and one paratype slide (registration no. NHMUK 2013.7.4.1) with protargol-stained specimens are deposited in the Laboratory of Protozoology, Ocean University of China, and the Natural History Museum, London, UK, respectively.
**Etymology.** The species-group name *orientalis* (eastern, of the Orient) refers to the fact that this species was first isolated from Chinese coastal waters.

**Description.** Cell size in vivo 25–50 × 12–20 μm. Body shape usually elongated oval to slender with anterior end distinctly pointed, posterior rounded (Figs 1a–c and 2a and Fig. S1a, available in IJSEM Online). Body asymmetrical in outline when viewed from ventral side with anterior end slightly curved sideways (Figs 2a–c and S1a–c). Ventral side slightly straightened, while dorsal side convex (Figs 1a, 2a, b, S1a and S2a). Buccal field a quarter to a third of body length, with buccal cilia about 8–10 μm long (Figs 2c, f and S1c, f). Somatic cilia densely arranged and about 7–8 μm

**Fig. 1.** *Metanophrys orientalis* spec. nov. from life (a–c) and after staining with protargol (e–g) and silver nitrate (d). (a) Ventral view of a representative individual. (b, c) To show different body shapes. (d) Caudal view to show silverline system. (e, f) Ventral (e) and dorsal (f) views of the same specimen, showing infraciliature and nuclear apparatus. (g) Detailed structure of the buccal area. M1–3, Membranelles 1, 2 and 3; Ma, macronucleus; PM, paroral membrane; Sc, scutica. Bars, 20 μm (a, b, e, f).

**Fig. 2.** *Metanophrys orientalis* spec. nov. from life (a–h, m) and after staining with protargol (k, l, m) and silver nitrate (i, j). (a–c) Different individuals; arrow in (c) indicates crystals. (d) Ventral view; arrow marks the single prolonged caudal cilium. (e) Individual undergoing binary fission. (f) Anterior region of cell; arrowheads mark buccal cilia. (g) Ventral view showing crystals (arrowhead). (h) To show contractile vacuole (arrow). (i) Ventral view, to show M1 (arrowhead). (j) Ventral view, showing contractile vacuole pore (arrowhead). (k, l) Ventral (k) and dorsal (l) views of the same specimen, showing infraciliature and nuclear apparatus. (m) Detailed infraciliature of buccal area. (n) Detailed view of cortex; arrowheads mark extrusomes. M1–3, Membranelles 1, 2 and 3; Ma, macronucleus; PM, paroral membrane; Sc, scutica. Bars, 20 μm (a–c, e) and 15 μm (k, l).
Table 1. Morphometric characterization of Metanophrys orientalis spec. nov. (upper rows) and Uronemella sinensis spec. nov. (lower rows)

Data are based on protargol-stained specimens.

<table>
<thead>
<tr>
<th>Character</th>
<th>Min.</th>
<th>Max.</th>
<th>Mean</th>
<th>SD</th>
<th>CV (%)</th>
<th>n</th>
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</thead>
<tbody>
<tr>
<td>Body length (µm)</td>
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<td>58</td>
<td>49.2</td>
<td>4.9</td>
<td>9.9</td>
<td>20</td>
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<tr>
<td>Body width (µm)</td>
<td>34</td>
<td>46</td>
<td>59.1</td>
<td>3.7</td>
<td>9.2</td>
<td>19</td>
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<tr>
<td>Length of buccal field (µm)</td>
<td>27</td>
<td>40</td>
<td>33.3</td>
<td>4.1</td>
<td>12.3</td>
<td>20</td>
</tr>
<tr>
<td>Number of somatic kineties</td>
<td>20</td>
<td>31</td>
<td>25.2</td>
<td>2.8</td>
<td>11.0</td>
<td>19</td>
</tr>
<tr>
<td>Number of rows in membranelle 1</td>
<td>11</td>
<td>16</td>
<td>13.4</td>
<td>1.7</td>
<td>12.6</td>
<td>15</td>
</tr>
<tr>
<td>Number of rows in membranelle 2</td>
<td>22</td>
<td>30</td>
<td>25.1</td>
<td>2.1</td>
<td>10.1</td>
<td>15</td>
</tr>
<tr>
<td>Number of rows in membranelle 3</td>
<td>9</td>
<td>10</td>
<td>9.3</td>
<td>0.5</td>
<td>1.8</td>
<td>18</td>
</tr>
<tr>
<td>Width of macronucleus (µm)</td>
<td>9</td>
<td>10</td>
<td>9.6</td>
<td>1.0</td>
<td>4.8</td>
<td>19</td>
</tr>
</tbody>
</table>

X. Pan and others

Table 2. SSU rRNA gene sequence dissimilarities among 16 scuticociliates

Values below the diagonal are pairwise distances (%); those above the diagonal are numbers of different sites. Species described in the present study are marked in bold.

<table>
<thead>
<tr>
<th>Species</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
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<th>13</th>
<th>14</th>
<th>15</th>
<th>16</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Uronemella sinensis spec. nov.</td>
<td>152</td>
<td>150</td>
<td>156</td>
<td>76</td>
<td>83</td>
<td>141</td>
<td>163</td>
<td>143</td>
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<td>164</td>
<td>125</td>
<td>107</td>
<td>111</td>
<td>105</td>
<td>97</td>
<td></td>
</tr>
<tr>
<td>2. Uronema mariniuin</td>
<td>170</td>
<td>191</td>
<td>139</td>
<td>150</td>
<td>169</td>
<td>165</td>
<td>181</td>
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<td>105</td>
<td>107</td>
<td>111</td>
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<td>3. Uronema elegans</td>
<td>106</td>
<td>124</td>
<td>133</td>
<td>130</td>
<td>135</td>
<td>168</td>
<td>133</td>
<td>176</td>
<td>154</td>
<td>83</td>
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<td>122</td>
<td>122</td>
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<td></td>
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<tr>
<td>4. Uronema heteromarinum</td>
<td>147</td>
<td>155</td>
<td>159</td>
<td>171</td>
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<td>174</td>
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<td>134</td>
<td>136</td>
<td>134</td>
<td>138</td>
<td></td>
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<tr>
<td>5. Uronemella filificum</td>
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<td>119</td>
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<td>139</td>
<td>123</td>
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<td>116</td>
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<td>114</td>
<td>112</td>
<td>112</td>
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<td></td>
</tr>
<tr>
<td>6. Uronema parafilificum</td>
<td>8</td>
<td>12</td>
<td>10.4</td>
<td>7.6</td>
<td>10.2</td>
<td>20</td>
<td>20</td>
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<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>7. Metanophrys orientalis spec. nov.</td>
<td>7</td>
<td>16</td>
<td>13.6</td>
<td>7.2</td>
<td>13.2</td>
<td>19</td>
<td>19</td>
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<td>19</td>
<td>19</td>
<td>19</td>
<td>19</td>
<td>19</td>
<td>19</td>
<td></td>
</tr>
</tbody>
</table>

Nine or ten somatic kineties with dikinetids arranged in approximately in anterior half of each row and monokinetids positioned posteriorly (Figs 1e, f, 2k, l and S1k, l). M1 slightly away from apex and comprising two rows of kinetids with six basal bodies each (Figs 1g and 2i). M2, as long as M1, also composed of about six basal bodies in each row. Membranelle 3 (M3) located close to M2, and normally comprised three short arranged rows of basal bodies (Figs 1g, 2m and S1m). Paroral membrane (PM) extended to about anterior third of body. Contractile vacuole pore located at posterior end of kinety 2 (Figs 1d, 2j and S1j).

SSU rRNA gene sequence (Table 2). The SSU rRNA gene of Metanophrys orientalis is 1735 bp long with a G+C content of 44.78 mol%. It differs from those of morphologically similar species at 62–169 positions (Table 2).

Remarks and comparison. The novel form is similar to three nominal species, Metanophrys elongata (Biggar & Wenrich, 1932) Groliére et al., 1978, Metanophrys similis Song et al., 2002 and Metanophrys sinensis Song & Wilbert, 2000 (Fig. 4).

Compared with Metanophrys orientalis spec. nov., Metanophrys elongata has a larger body (100–120 µm vs
25–50 \( \mu m \), a larger number of somatic kineties (15–20 vs nine or ten) and highly developed, extremely long M1 and M2 (Grolière et al., 1978, 1980).

Metanophrys similis can be separated easily from Metanophrys orientalis by the different arrangement of the scutica (basal bodies solitary and sparsely distributed in a long row in Metanophrys similis vs in pairs and closely packed in Metanophrys orientalis), the appearance of the pellicle (notched in the former vs smooth in the latter) and the different number of rows in M1 (two in Metanophrys similis vs one in Metanophrys orientalis) (Song et al. 2002) (Fig. 4g, h).

The novel species most closely resembles Metanophrys sinensis (Song & Wilbert, 2000b). The latter, however, can be distinguished from Metanophrys orientalis in (i) the presence or absence of extrusomes (absent in Metanophrys sinensis vs arranged in rows between somatic kineties in Metanophrys orientalis); (ii) distinctly longer M1 than M2 in Metanophrys sinensis (vs they are equal in length in Metanophrys orientalis); (iii) structure of M2 (two-rowed vs three-rowed); (iv) the length of the buccal field relative to the body length (half in Metanophrys sinensis vs a quarter to a third in Metanophrys orientalis); (v) the location of the contractile vacuole pore (at posterior end of kinety 2 in Metanophrys sinensis vs at posterior end of kinety 1 in Metanophrys orientalis) (Song & Wilbert, 2000b); (vi) dikinetids approximately in anterior two-thirds of each somatic kinety in Metanophrys sinensis (vs half in Metanophrys orientalis) (Fig. 4n).

Comparison of SSU rRNA sequences shows that the sequence of the novel species has differences in 62–132 gene sites compared with those of its congeners, but it differs from that of Paranophrys magna in 73 sites, indicating its closer relationship compared with Metanophrys similis, which is consistent with the phylogenetic analyses (Gao et al., 2012a). The sequence difference of 88 sites between the novel form and Anophyroides haemophila indicates a possible close relationship between Metanophrys and Anophyroides (Table 2).

The topologies of the phylogenetic trees reconstructed using BI and ML analyses were similar; therefore, only the
BI tree is shown (Fig. 5). Our phylogenetic trees show that the three species of the genus *Metanophrys* included in the analyses are divided into two clades: *Metanophrys similis* clusters with members of the genus *Mesanophrys*. *Metanophrys sinensis* branches as a sister group with the *Paranophrys magna* clade (1.00 BI, 100 ML) and *Metanophrys orientalis* spec. nov. forms a sister taxon to the branch comprising the above three species (1.00 BI, 100 ML). *Metanophrys similis* is well characterized and, based on its morphology, its generic placement is not in doubt (Song et al., 2002). Why *Metanophrys similis* clusters in a separate clade from its congeners is unclear, but may reflect the fact that phylogenetic relationships at this level cannot be resolved completely using SSU rRNA gene sequence data alone.

*Uronemella sinensis* spec. nov. (Figs 3 and S2; Tables 1 and 2; subclass Scuticociliatia Small, 1967; order Philasterida Small, 1967; genus *Uronemella* Song & Wilbert, 2002)

Some novel characters were found in the novel species; hence, an improved diagnosis of the genus *Uronemella* is supplied here, based on both previous studies and the present study.

**Improved diagnosis of genus Uronemella**

Body generally elongate-elliptical or pear-shaped, with an apical plate; buccal field about two-thirds of the total body length; membranelle 1 (M1) consisting of one row of basal bodies, membranelles 2 and 3 (M2 and M3) with two or more longitudinal rows of basal bodies; paroral membrane (PM) extending anteriorly to about the mid-level of M2; somatic kineties comprising a mixture of dikinetids and monokinetids; marine habitat.

**Uronemella sinensis** spec. nov.

**Diagnosis.** Body elongate-elliptical with truncated apical frontal plate, *in vivo* about 25–35 × 15–20 μm, buccal field about 65% of body length; nine or ten somatic kineties; membranelle 1 one-rowed with two or three basal bodies; contractile vacuole caudally positioned near ventral margin with its opening pore at posterior end of somatic kinety 1; marine habitat.

**Type locality.** A sandy beach named Shilaoren (salinity: 35%, in Laoshan district of Qingdao (36° 5' 30" N 120° 27' 54" E), northern China.

**Type slides.** The holotype slide (registration no. PXM-20101004) and one paratype slide (registration no. NHMUK 2013.7.4.2) with protargol-stained specimens were deposited in the Laboratory of Protozoology, Ocean University of China, and the Natural History Museum, London, UK, respectively.
**Etimology.** The species gets its name *sinesis* due to the locality where it was isolated (China).

**Description.** Size *in vivo* about 25–35 × 15–20 μm, elongate-elliptical in outline becoming wider toward posterior end (Figs 3a, e and S2a, e). Anterior end truncated, with a conspicuous apical plate, dorsal area broadly rounded (Figs 3a, h and S2a, h). Buccal field about 65 % of body length (Figs 3a and S2a). Swimming moderately fast while rotating about main body axis, sometimes quiet on the bottom.

Nine or ten somatic kineties (SK) arranged longitudinally, which usually have dikinetids in anterior quarter to a third of each row and monokinetids positioned posteriorly (Figs 3c, d, k and S2c, d, k). Buccal apparatus as shown in Figs 3b, i and S2b, i: M1 distinct subapically positioned, separated from other membranelles and consisting of two or three basal bodies; M2 relatively large and composed of two longitudinal rows of basal bodies; M3 comprising three longitudinal rows (Figs 3b, c, k and S2b, c, k, l). PM positioned on right of buccal cavity, terminating anteriorly to M2 (Figs 3b, c). Scutica consists of two or three basal bodies. Scuticum large and spherical, located mostly at anterior region. Contractile vacuole about 5 μm in diameter, positioned caudally near ventral side (Figs 3f and S2f). Swimming moderately fast while rotating about main body axis, sometimes quiet on the bottom.

**SSU rRNA gene sequence** (Table 2). The SSU rRNA gene is 1753 bp long with a G+C content of 43.18 mol%. The sequence differs from those of morphologically similar species at 76–164 positions (Table 2).

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**Fig. 5.** Phylogenetic tree inferred from SSU rRNA gene sequences, showing the positions of *Metanophrys orientalis* spec. nov. and *Uronemella sinensis* spec. nov. (bold). Nodal support for branches in the BI and ML trees are marked in order. *Wilbertia typica*, *Eurytomatella sinica*, *Histobalan tum natans*, *Pleurocroma coronatum* and *Cyclidium porcatum* are the outgroup taxa. Bar, 2 substitutions per 100 nucleotide positions.
Remarks and comparison. In terms of live morphology, infraciliature and habitat, two morphologically similar species should be compared with the novel species, Uronemella filificum Kahl, 1931 and U. parafilificum Gong et al., 2007 (Fig. 4).

U. filificum can be separated from the novel species through its body shape (pear-shaped vs elongate-ellipsoid), more somatic kineties (21–23 vs nine or ten), larger apical plate and behaviour (thigmotactic vs non-thigmotactic) (Song & Wilbert, 2002) (Fig. 4i, j).

U. parafilificum also differs from U. sinensis in having more somatic kineties (21–23 vs nine or ten) and more basal bodies in its membranelles (six vs two or three) and in its behaviour (thigmotactic vs non-thigmotactic) (Gong et al., 2007) (Fig. 4f).

Results from the comparison of SSU rRNA sequences support the morphological identification of the novel form, which is also consistent with the phylogenetic analyses (Gao et al., 2012a); the sequence of the novel species differs from those of U. filificum and U. parafilificum in 76 and 83 sites, respectively (Table 2).

In addition to U. sinensis spec. nov., SSU rRNA gene sequences of two congener, U. parafilificum and U. filificum, are available. All of these sequences were included in the phylogenetic analyses. As shown in Figs 5 and 53, the three Uronemella species form a monophyletic assemblage, with full support (1.00 BI, 100 ML). Noticeably, U. sinensis branches as a sister group with the U. parafilificum–U. filificum clade. The above analyses support the contention that the genus Uronemella is monophyletic.

In conclusion, the morphological and molecular data consistently support the validity of the species Uronemella sinensis spec. nov.

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