



Surgical site infection after hip replacement due to a novel *Peptoniphilus* species, provisionally named ‘*Peptoniphilus nemausus*’ sp. nov.

Cécilia Enault, Fabien Aujoulat, Alix Pantel, Nicolas Cellier, Catherine Lechiche, Bernard Mégy, Jean-Philippe Lavigne, Hélène Marchandin

► To cite this version:

Cécilia Enault, Fabien Aujoulat, Alix Pantel, Nicolas Cellier, Catherine Lechiche, et al.. Surgical site infection after hip replacement due to a novel *Peptoniphilus* species, provisionally named ‘*Peptoniphilus nemausus*’ sp. nov.. *Anaerobe*, 2020, 61, pp.102071. 10.1016/j.anaerobe.2019.102071 . hal-02860916

HAL Id: hal-02860916

<https://hal.umontpellier.fr/hal-02860916>

Submitted on 22 Aug 2022

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L’archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d’enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution - NonCommercial 4.0 International License

**Surgical site infection after hip replacement due to a novel *Peptoniphilus* species,
provisionally named '*Peptoniphilus nemausus*' sp. nov.**

Cécilia Enault¹, Fabien Aujoulat², Alix Pantel³, Nicolas Cellier⁴, Catherine Lechiche⁵,
Bernard Mégy⁴, Jean-Philippe Lavigne³, Hélène Marchandin^{6*}

¹ Department of Microbiology, Nîmes University Hospital, Nîmes, France

² HydroSciences Montpellier, CNRS, IRD, Univ Montpellier, Montpellier, France

³ National Institute of Health and Medical Research, INSERM U1047, University of
Montpellier, Department of Microbiology, CHU Nîmes, Nîmes, France

⁴ Department of Orthopaedic Surgery, Nîmes University Hospital, Nîmes, France

⁵ Department of Infectious Diseases, Nîmes University Hospital, Nîmes, France

⁶ HydroSciences Montpellier, CNRS, IRD, University of Montpellier, Department of
Microbiology, CHU Nîmes, Nîmes, France

* Corresponding author: Prof. Hélène Marchandin, UMR 5569 HydroSciences Montpellier,
Equipe Pathogènes Hydriques Santé Environnements, U.F.R. des Sciences Pharmaceutiques
et Biologiques, Université de Montpellier, 15, avenue Charles Flahault, BP 14491, 34093
Montpellier Cedex 5, France

helene.marchandin@umontpellier.fr

Declarations of interest: none.

24 **Abstract**

25 We report a case of surgical site infection after total hip prosthesis replacement due to an
26 ofloxacin-resistant *Peptoniphilus* isolate belonging to an unknown species for which the name
27 ‘*Peptoniphilus nemausus*’ sp. nov. is proposed. Follow-up was favourable under clindamycin
28 and rifampin for 3 months in this patient whom had a *Proteus mirabilis* infection treated by
29 fluoroquinolone.

30

31 **Key words:** *Peptoniphilus*, infection, anaerobe, resistance, surgical site infection, prosthetic
32 joint infection.

33

Text

Gram-positive anaerobic cocci (GPAC) are important members of the human microbiota, that can also act as opportunistic pathogens in humans. GPAC were shown to be the more frequently isolated anaerobes in microbiological laboratories (24-31% of the total number of isolated anaerobes) [1,2]. While *Finegoldia magna* and *Parvimonas micra* represent about half of the isolated GPAC [3,4] and are the most studied, several less known genera of GPAC like *Anaerococcus* and *Peptoniphilus*, are involved in various opportunistic human infections, mainly as part of polymicrobial infections [2].

A 66-year old woman was admitted to the rehabilitation unit of the University Hospital of Nîmes on March 8, 2018, after revision of her total hip prosthesis on March 1. The patient presented with morbid obesity (body mass index 52 defining grade 3 obesity). She had no hormone replacement treatment since menopause that occurred at the age of 55. Her history includes arterial hypertension, breast cancer in remission after surgery and radiotherapy, and under current hormonal therapy by letrozole, an aromatase inhibitor with bone loss side effects. Initial arthroplasty was performed on February 8, for painful hip and functional impotence revealing extensive osteolysis of the femoral head with previously undiagnosed osteoporosis. At the same time, a Vitamin D deficiency of 10 nmol/L (normal range: 30-100 nmol/L) was found requiring supplementation. Early periprosthetic fracture occurred at weightbearing initiation and hip prosthesis replacement, including removal of the failed implant, lavage and implantation of a femoral component that has a long stem, was performed on March 1. Microbiological investigations showed an early prosthetic joint infection (PJI), as the 3 samples taken during hip prosthesis revision were positive for *Proteus mirabilis*. PJI was treated by intravenous ofloxacin (600 mg per day) and the introduction of bisphosphonates to correct the osteoporosis and the vitamin D deficiency.

In the rehabilitation unit, a surgical site infection was suspected at the beginning of April and confirmed by CT-scan on April 10. Surgical lavage and debridement were performed on April, 12th as part of the management of the infection with Debridement, Antibiotics, Irrigation and Retention (DAIR). Eight surgical samples were obtained (1 periprosthetic fluid, 4 periprosthetic tissue and 3 bone samples). Direct examination showed either rare or rather numerous polymorphonuclear depending on the sample and Gram-positive cocci were visualized after Gram stain of a periprosthetic tissue sample leading to the instauration of an intravenous antimicrobial therapy by cefotaxime plus vancomycin. Samples were analyzed according to national recommendations [5]. Anaerobic cultures were positive after 7 days of incubation of the periprosthetic fluid and the 4 tissue specimens and grew a strictly anaerobic Gram-positive coccus. The three bone samples remained negative. Identification by MALDI-TOF mass spectrometry (Vitek® MS, bioMérieux, Marcy-l'Etoile, France) was unsuccessful. Antimicrobial susceptibility testing was performed using Etest strips (bioMérieux) according to the recommendations of the Antibigram committee of the French Society for Microbiology for anaerobes [6]. The isolate was susceptible to all antibiotics tested (MICs of imipenem and rifampin < 0.02 mg/L, MICs of amoxicillin and coamoxiclav. < 0.016 mg/L, MIC of metronidazole 0.016 mg/L, MIC of linezolid 0.125 mg/L and MIC of clindamycin 0.75 mg/L) except ofloxacin (MIC > 32 mg/L). The multidisciplinary team for the management of PJI of our hospital decided an antimicrobial treatment switch to clindamycin (2400 mg per day) and rifampin (1200 mg per day) for 3 months, on April 23th. A favourable outcome was noted after the end of the treatment and a one-year period of clinical follow-up after a novel total hip prosthesis has been implanted in July 2018.

For the identification of the GPAC isolated in pure culture from a deep-tissue infection, we tested the isolate with another commercially available MALDI-TOF MS system, (Maldi Biotyper Microflex®, Bruker Daltonics, Bremen, Germany), as differences in identification

performances between MALDI-TOF systems have been previously reported for identification of anaerobes [7]; however, no identification was obtained for the clinical isolate using this alternative MALDI-TOF MS system. We also performed 16S rRNA gene sequencing as previously described [8]. Sequence analysis (1388 nt) showed the isolate, belonged to the genus *Peptoniphilus*, but to an as yet unknown species. Indeed, a similarity table constructed using utilities implemented in Biological sequence alignment editor (BioEdit) software (<http://www.mbio.ncsu.edu/bioedit/bioedit.html>) revealed that the type strains of *Peptoniphilus coxii* (97.9% of 16S rRNA gene sequence identity) and *Peptoniphilus ivorii* (94.6%), as well as the type strains of the two non validated species '*Peptoniphilus urinimassiliensis*' (96.6%) and '*Peptoniphilus pacaensis*' (96.1%) were the most closely related species of the clinical isolate [9-12]. However, the highest 16S rRNA gene sequence identity observed between the clinical isolate (strain 1804121828, GenBank accession number: MK945758) and the type strain of *Peptoniphilus coxii* was below the threshold for species identification, *i.e.*, less than 98.7% of 16S rRNA gene identity [13], suggesting the clinical isolate to belong to a novel species in the genus *Peptoniphilus* [14]. The 16S rRNA gene sequence of the clinical isolate was also compared with those of the type strains of species of the genus *Peptoniphilus* through phylogenetic analysis. Evolutionary distances were analysed using the neighbour-joining (NJ) method (Kimura two-parameter substitution model) using phylogenetic analyses available at <http://www.phylogeny.fr> [14]. Phylogenetic analysis supported the inclusion of the isolate in a new species based on a clearly individualized branching within the genus *Peptoniphilus* and the cluster *P. coxii* / '*P. pacaensis*' / '*P. urinimassiliensis*' / *P. ivorii* (Figure 1). A formal characterization of the novel species is ongoing and the name '*Peptoniphilus nemausus*' sp. nov. is proposed for this novel species pertaining to the Nîmes town in the south of France, where the strain supporting the description of the species was isolated.

The genus *Peptoniphilus* was individualized in 2001 to accommodate strictly anaerobic Gram-positive cocci previously classified in the genus *Peptostreptococcus* that were butyrate-producers, non-saccharolytic and that used peptone and amino acids as major energy sources [15]. Since then, a growing number of species has been described and currently 17 species are validly published (<http://www.bacterio.net/peptoniphilus.html>) [16] and 9 others have been proposed without current valid publication (May 20, 2019) (Figure 1). Among the genus, species can be distinguished by phenotypic assays (allowing the determination of a metabolic profile) that are not routinely performed in clinical microbiology laboratories, particularly since the development of MALDI-TOF MS; therefore, species identification is currently based on mass spectrometry and, when unsuccessful, on molecular tools [17]. MALDI-TOF MS is a powerful and rapid identification tool; however, databases are currently incomplete and optimization of current databases for anaerobes is ongoing [18-20]. MS was unable to identify our clinical isolate and 16S rRNA gene sequence analysis was required revealing the clinical isolate to belong to an unknown species and showing a still underestimated diversity in this genus. A formal description of this new species based on a polyphasic taxonomy approach has been undertaken.

Members of the different human microbiota, *Peptoniphilus* spp. have been reported in a large variety of human endogeneous polymicrobial infections due to the pathogenesis process of such infections, *i.e.*, polymicrobial infections involving members of the contiguous microbiota through contamination of initially sterile anatomical sites. Clinical relevance of *Peptoniphilus* spp. has been mainly demonstrated after isolation from skin and soft tissues, chronic wounds (pressure ulcer, diabetic foot wounds), osteoarticular samples, genitourinary (vaginal infections) and respiratory tract (pleural empyema, chronic rhinosinusitis) [2].

Anaerobic infections remain rare in patients with prosthetic joints and mostly involved species originating from the cutaneous microbiota like *Cutibacterium* (formerly

Propionibacterium) acnes and *Finegoldia magna* [21-24]. Despite *Peptoniphilus* spp. have been previously identified during osteoarticular [21,25,26] and soft tissue infections [27,28], we were unable to find a case similar to that described herein, *i.e.*, surgical site infection following PJI, among the 122 publications retrieved in the PubMed database using the “*Peptoniphilus*” search term (July 8, 2019). In the present case, despite anaerobes were not reported during initial infection and the portal of entry or origin of the *Peptoniphilus* isolate remained unidentified, it is likely that it has been selected by ofloxacin therapy towards initial *P. mirabilis* infection, as the isolate displayed high level resistance to ofloxacin. Fluoroquinolones are one of the therapeutic options in the management of osteoarticular infections in case of susceptibility of the causative microorganism, as they displayed good penetration profiles into bone tissues and synovial fluid [29]. If the antimicrobial susceptibility patterns of the main encountered anaerobic pathogens in bone and joint infections, *C. acnes* and *F. magna*, is documented, antimicrobial resistance patterns of the overall GPAC have received less interest being for long considered as microorganisms susceptible to antibiotics with anti-anaerobic activity. However, studies including or focused on GPAC revealed high rate of resistance towards some antibiotics used in the management of osteoarticular anaerobic infections, 25% of GPAC displayed resistance to clindamycin in most recent studies for example [22,29]; reported some multidrug resistant clinical isolates [30] while revealing heterogeneity in antibiotic susceptibility patterns between species [4,29,31,32]. Regarding ofloxacin, a large study conducted in France, *i.e.*, 170 GPAC isolated from diverse anatomical sites including 16.5% of *Peptoniphilus* spp. all identified by 16S rRNA gene sequencing, showed a global rate of resistance of 63% but revealed that all *Peptoniphilus* - but also all *Anaerococcus* - clinical isolates studied displayed resistance to ofloxacin [3]. In case of deep monomicrobial infection of a normally sterile body site as in the present case, antimicrobial susceptibility testing is recommended to guide the treatment [33];

however, in case of polymicrobial infection involving both aerobes and anaerobes or several anaerobes, antimicrobial susceptibility testing is usually less systematically performed on all isolated anaerobes and one should then consider the presence of potentially resistant microorganisms, not only members of the *Bacteroides fragilis* group but also some GPAC, among the cultivable microbiota in the choice of the best therapeutic option.

Acknowledgments

The authors thank Nathalie Jeanney for excellent technical assistance.

Authors belong to the Centre Regional des Infections Ostéo-Articulaires complexes (CRIOAc) Sud Méditerranée (AP, NC, CL, BM, and JPL), to the Clinique du Pied Diabétique Gard Occitanie (NC, CL, BM, and JPL) and/or to the FHU INCh (Fédération Hospitalo-Universitaire Infections Chroniques, Aviesan) (all authors).

We thank the Nîmes University Hospital for its structural, human and financial support through the award obtained by our team during the internal call for tenders « Thématiques phares ».

References

- [1] Murdoch DA. Gram-positive anaerobic cocci. Clin Microbiol Rev. 1998; 11:81-120.
- [2] Murphy EC, Frick IM. Gram-positive anaerobic cocci--commensals and opportunistic pathogens. FEMS Microbiol Rev. 2013; 37:520-53. doi: 10.1111/1574-6976.12005.
- [3] Jean-Pierre H, Ribot J, Esquerre L, Guzzi-Domanico A, Michon A-L, Jumas-Bilak E. Les cocci à Gram positif anaérobies, identification et sensibilité aux antibiotiques : à propos de 170 souches isolées au CHRU de Montpellier. 30ème Réunion Interdisciplinaire de Chimiothérapie Anti -Infectieuse, 2-3 décembre 2010, Paris.
- [4] Veloo AC, Welling GW, Degener JE. Antimicrobial susceptibility of clinically relevant Gram-positive anaerobic cocci collected over a three-year period in the Netherlands. Antimicrob Agents Chemother. 2011; 55:1199-203. doi: 10.1128/AAC.01771-09.
- [5] Société Française de Microbiologie. Chapter 30. Infections osseuses et articulaires. Référentiel en microbiologie médicale (REMIC): Société Française de Microbiologie Ed; 2018; p. 313-20.
- [6] Société Française de Microbiologie. Bactéries anaérobies à Gram positif. In: CASFM / EUCAST. Société Française de Microbiologie Ed. 2019; p.122-3. Available at: <http://www.sfm-microbiologie.org>.
- [7] Veloo AC, Knoester M, Degener JE, Kuijper EJ. Comparison of two matrix-assisted laser desorption ionisation-time of flight mass spectrometry methods for the identification of clinically relevant anaerobic bacteria. Clin Microbiol Infect. 2011; 17:1501-6. doi: 10.1111/j.1469-0691.2011.03467.x.
- [8] Carlier JP, Marchandin H, Jumas-Bilak E, Lorin V, Henry C, Carrière C, Jean-Pierre H. *Anaeroglobus geminatus* gen. nov., sp. nov., a novel member of the family Veillonellaceae. Int J Syst Evol Microbiol. 2002; 52:983-86.

200 [9] Citron DM, Tyrrell KL, Goldstein EJ. *Peptoniphilus coxii* sp. nov. and *Peptoniphilus*
201 *tyrrelliae* sp. nov. isolated from human clinical infections. *Anaerobe*. 2012; 18: 244-8. doi:
202 10.1016/j.anaerobe.2011.11.008.

203 [10] Diop K, Diop A, Michelle C, Richez M, Rathored J, Bretelle F, Fournier PE, Fenollar
204 F. Description of three new *Peptoniphilus* species cultured in the vaginal fluid of a woman
205 diagnosed with bacterial vaginosis: *Peptoniphilus pacaensis* sp. nov., *Peptoniphilus raoultii*
206 sp. nov., and *Peptoniphilus vaginalis* sp. nov. *Microbiologyopen*. 2019; 8:e00661. doi:
207 10.1002/mbo3.661.

208 [11] Brahimi S, Cadoret F, Founier PE, Moal V, Raoult D. '*Peptoniphilus urinimassiliensis*'
209 sp. nov., a new bacterial species isolated from a human urine sample after de novo kidney
210 transplantation. *New Microbes New Infect*. 2017; 16:49–50.

211 [12] Song Y, Liu C, Finegold SM. *Peptoniphilus gorbachii* sp. nov., *Peptoniphilus olsenii*
212 sp. nov., and *Anaerococcus murdochii* sp. nov. isolated from clinical specimens of human
213 origin. *J Clin Microbiol*. 2007; 45:1746-52.

214 [13] Stackebrandt E, Ebers J. Taxonomic parameters revisited: tarnished gold standards.
215 *Microbiology Today*. 2016; 33:152-5.

216 [14] Dereeper A, Guignon V, Blanc G, Audic S, Buffet S, Chevenet F, Dufayard JF,
217 Guindon S, Lefort V, Lescot M, et al. Phylogeny.fr: robust phylogenetic analysis for the non-
218 specialist. *Nucleic Acids Res*. 2008; 36:W465-9.

219 [15] Ezaki T, Kawamura Y, Li N, Li ZY, Zhao L, Shu S. Proposal of the genera
220 *Anaerococcus* gen. nov., *Peptoniphilus* gen. nov. and *Gallicola* gen. nov. for members of the
221 genus *Peptostreptococcus*. *Int J Syst Evol Microbiol*. 2001; 51:1521-8.

222 [16] Parte AC. LPSN – List of Prokaryotic names with Standing in Nomenclature
 223 (bacterio.net), 20 years on. *International Journal of Systematic and Evolutionary*
 224 *Microbiology*. 2018; 68:1825-9; doi: 10.1099/ijsem.0.002786.

225 [17] Nagy E, Boyanova L, Justesen US; ESCMID Study Group of Anaerobic Infections.
 226 How to isolate, identify and determine antimicrobial susceptibility of anaerobic bacteria in
 227 routine laboratories. *Clin Microbiol Infect*. 2018; 24: 1139-48. doi:
 228 10.1016/j.cmi.2018.02.008.

229 [18] Veloo AC, Erhard M, Welker M, Welling GW, Degener JE. Identification of Gram-
 230 positive anaerobic cocci by MALDI-TOF mass spectrometry. *Syst Appl Microbiol*. 2011;
 231 34:58-62. doi: 10.1016/j.syapm.2010.11.005.

232 [19] Veloo AC, de Vries ED, Jean-Pierre H, Justesen US, Morris T, Urban E, Wybo I, van
 233 Winkelhoff AJ; ENRIA workgroup. The optimization and validation of the Biotyper MALDI-
 234 TOF MS database for the identification of Gram-positive anaerobic cocci. *Clin Microbiol*
 235 *Infect*. 2016; 22: 793-8. doi: 10.1016/j.cmi.2016.06.016.

236 [20] Veloo ACM, Jean-Pierre H, Justesen US, Morris T, Urban E, Wybo I, Kostrzewa M,
 237 Friedrich AW; ENRIA workgroup. Validation of MALDI-TOF MS Biotyper database
 238 optimized for anaerobic bacteria: The ENRIA project. *Anaerobe*. 2018; 54: 224-30. doi:
 239 10.1016/j.anaerobe.2018.03.007.

240 [21] Walter G, Vernier M, Pinelli PO, Million M, Coulange M, Seng P, Stein A. Bone and
 241 joint infections due to anaerobic bacteria: an analysis of 61 cases and review of the literature.
 242 *Eur J Clin Microbiol Infect Dis*. 2014; 33: 1355-64. doi: 10.1007/s10096-014-2073-3.

243 [22] Shah NB, Tande AJ, Patel R, Berbari EF. Anaerobic prosthetic joint infection.
 244 *Anaerobe*. 2015; 36:1-8. doi: 10.1016/j.anaerobe.2015.08.003.

245 [23] Lebowitz D, Kressmann B, Gjoni S, Zenelaj B, Groscurin O, Marti C, Zingg M,
 246 Uçkay I. Clinical features of anaerobic orthopaedic infections. *Infect Dis (Lond)*. 2017;
 247 49:137-40.

248 [24] Rieber H, Frontzek A, Jerosch J, Alefeld M, Strohecker T, Ulatowski M, Morawietz
 249 T, Hinsenkamp S, Bell A, Kücüköylü D, Frommelt L. Periprosthetic joint infection caused
 250 by anaerobes. Retrospective analysis reveals no need for prolonged cultivation time if
 251 sensitive supplemented growth media are used. *Anaerobe*. 2018; 50:12-8. doi:
 252 10.1016/j.anaerobe.2018.01.009.

253 [25] La Scola B, Fournier PE, Raoult D. Burden of emerging anaerobes in the MALDI-
 254 TOF and 16S rRNA gene sequencing era. *Anaerobe*. 2011; 17:106-12. doi:
 255 10.1016/j.anaerobe.2011.05.010.

256 [26] Verma R, Morrad S, Wirtz JJ. *Peptoniphilus asaccharolyticus*-associated septic
 257 arthritis and osteomyelitis in a woman with osteoarthritis and diabetes mellitus. *BMJ Case*
 258 *Rep*. 2017; pii: bcr-2017-219969. doi: 10.1136/bcr-2017-219969.

259 [27] Brazier J, Chmelar D, Dubreuil L, Feierl G, Hedberg M, Kalenic S, Könönen E,
 260 Lundgren B, Malamou-Ladas H, Nagy E, Sullivan A, Nord CE; ESCMID Study Group on
 261 Antimicrobial Resistance in Anaerobic Bacteria. European surveillance study on
 262 antimicrobial susceptibility of Gram-positive anaerobic cocci. *Int J Antimicrob Agents*. 2008;
 263 31:316-20. doi: 10.1016/j.ijantimicag.2007.11.006.

264 [28] Thabit AK, Fatani DF, Bamakhrama MS, Barnawi OA, Basudan LO, Alhejaili SF.
 265 Antibiotic penetration into bone and joints: An updated review. *Int J Infect Dis*. 2019; 81:128-
 266 36. doi: 10.1016/j.ijid.2019.02.005.

267 [29] Jeverica S, Kolenc U, Mueller-Premru M, Papst L. Evaluation of the routine
 268 antimicrobial susceptibility testing results of clinically significant anaerobic bacteria in a

269 Slovenian tertiary-care hospital in 2015. *Anaerobe*. 2017; 47:64-9. doi:
 270 10.1016/j.anaerobe.2017.04.007.

271 [30] Shilnikova II, Dmitrieva NV. Evaluation of antibiotic susceptibility of Gram-positive
 272 anaerobic cocci isolated from cancer patients of the N.N.Blokhin Russian cancer research
 273 center. *J Pathog*. 2015; 648134.

274 [31] Brazier JS, Hall V, Morris TE, Gal M, Duerden BI. Antibiotic susceptibilities of
 275 Gram-positive anaerobic cocci: results of a sentinel study in England and Wales. *J Antimicrob*
 276 *Chemother*. 2003; 52:224-8.

277 [32] Veloo AC, van Winkelhoff AJ. Antibiotic susceptibility profiles of anaerobic
 278 pathogens in The Netherlands. *Anaerobe*. 2015; 31:19-24. doi:
 279 10.1016/j.anaerobe.2014.08.011.

280 [33] Gajdács M, Spengler G, Urbán E. Identification and antimicrobial susceptibility
 281 testing of anaerobic bacteria: Rubik's cube of clinical microbiology? *Antibiotics (Basel)*.
 282 2017; 6(4). pii: E25. doi: 10.3390/antibiotics6040025.

283

284 **Legend to figure**

285 **Fig. 1.** Neighbor-joining phylogenetic tree showing the relationship between the 16S rRNA
286 gene sequences of *Peptoniphilus* strain 1804121828^T (type strain of the proposed novel
287 species '*Peptoniphilus nemausus*') and of species, either validated or not, in the genus
288 *Peptoniphilus*. Alignment length was 1166 nt. Names for effectively published but non-
289 validated species are indicated between quotes. GenBank accession numbers are indicated in
290 parentheses. Bootstrap support was computed after 1000 reiterations. Bootstrap values are
291 indicated at the corresponding nodes when >70%. *Ezakiella peruensis* was used as the
292 outgroup microorganism.

293 * indicates species with uncertain taxonomic status, as *P. senegalensis* and '*P. rhinitis*' might
294 be synonym species of *P. tyrrelliae* and *P. lacydoensis*, respectively.

