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Corynebacterium phoceense, resident member of the urogenital microbiota?

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Abstract

Corynebacterium phoceense is a Gram-positive species previously isolated from human urine. Although other species from the same genus have been associated with urinary tract infections, *C. phoceense* is currently believed to be a non-pathogenic member of the urogenital microbiota. Prior to our study, only two isolates were described in the literature, and very little is known about the species. Here, we describe *C. phoceense* UFMG-H7, the first strain of this species isolated from the urine of healthy cattle. The genome for this isolate was produced and compared to the two other publicly available *C. phoceense* as well as other *Corynebacterium* genome assemblies. Our in-depth genomic analysis identified four additional publicly available genome assemblies that are representatives of the species, also isolated from the human urogenital tract. Although none of the strains have been associated with symptoms or disease, numerous genes associated with virulence factors are encoded. In contrast to related *Corynebacterium* species and *Corynebacterium* species from the bovine vaginal tract, all *C. phoceense* strains examined code for the SpaD-type pili suggesting adherence is essential for its persistence within the urinary tract. As the other *C. phoceense* strains analysed were isolated from the human urogenital tract, our results suggest that this species may be specific to this niche.

DATA SUMMARY

Raw reads as well as the genome assembly for *C. phoceense* UFMG-H7 have been deposited under accession numbers SRR13080711 and GCF_015686525.1, respectively. Accession numbers for all publicly genome sequences examined in this work are listed in Table S1 (available in the online version of this article).

INTRODUCTION

Corynebacterium species have been detected in the healthy urinary microbiomes of both human males and females [1–4]. In fact, the male urinary microbiota is enriched with *Corynebacterium* spp. [1]. *Corynebacterium* species, however, also have been associated with urinary tract infections (UTIs) and symptoms. For instance, *C. coyleae* and *C. riegelii* have been associated with urgency urinary incontinence in women [2] and *C. urealyticum* has been associated with UTIs (see review [5]). *C. urealyticum* also has been found as the causative agent of UTIs in cats [6, 7] and dogs [6, 8], while other species, including *C. renale*, *C. cystidis* and *C. pilosum*, have been associated with UTIs in cattle [9–14].

Recently new species of *Corynebacterium* have been identified from human urine samples including *C. urinapleomorphum* Marseille-P2799^T [15] and *C. phoceense* MC1^T [16]. Neither has been found to be the causative agent of infection. Subsequent to the description of *C. phoceense* in 2016 [16], two other strains were described. The first by Intanoo and colleagues [17] classified an isolate from cattle rumen as *C. phoceense* based upon 16S rRNA gene sequence homology to the type strain, although neither the 16S rRNA gene sequence or genome was published. The second *C. phoceense* strain was collected from

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Keywords: bovine microbiome; *Corynebacterium phoceense*; urinary microbiome; urogenital microbiome.

Abbreviations: ANI, average nucleotide identity; LB, Lysogeny Broth; TEM, transmission electron microscopy; UTI, urinary tract infection.

Raw reads as well as the genome assembly for *C. phoceense* UFMG-H7 have been deposited under accession numbers SRR13080711 and GCF_015686525.1, respectively.

Five supplementary tables are available with the online version of this article.

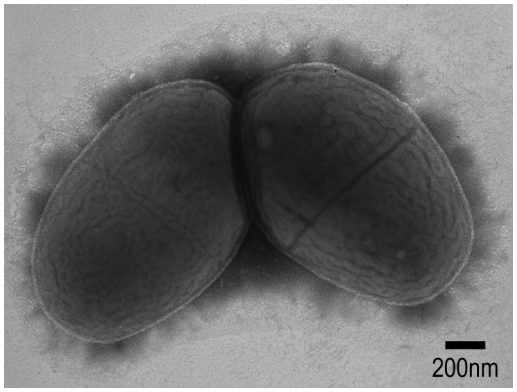

Fig. 1. TEM image of *C. phoceense* UFMG-H7

a human urine sample and its genome assembly is publicly available (strain 272, Accession no. GCF_006547305). The 16S rRNA gene sequences of *C. phoceense* MC1^T and 272 are identical. Recently we discovered a third representative of this species, *C. phoceense* UFMG-H7, which was isolated from urine from a healthy Gyr heifer.

METHODS

C. phoceense UFMG-H7 was collected in May 2019, from a purebred Gyr heifer at the Agricultural Research Company of Minas Gerais State (EPAMIG) (approved by the Ethics Committee in Animal Experimentation of the Universidade Federal de Minas Gerais, Brazil [CEUA/UFMG - 40/2019]). Prior to collection, the animal's vulva was washed with soap and distilled water. Mid-stream urine was collected using a sterile 50 ml conical tube and stored at −20 °C for 48 h until processing in the lab. Then 2 ml aliquots were centrifuged, and the supernatant was spread onto Lysogeny Broth (LB) agar plates. Plates were incubated overnight at 37 °C. Individual colonies were picked and regrown in LB under the same conditions, and this process was repeated at least three times to obtain pure colonies. One of these isolates was confirmed as *C. phoceense* via 16S rRNA gene sequencing using the 63F/1387R primer pair; the 16S rRNA gene sequence is identical to that of MC1^T and 272. Transmission Electron Microscopy (TEM) imaging of the isolate revealed a morphology consistent with the species (Fig. 1). LB plates were streaked with the isolate and found to grow aerobically with and without 5% CO_2 as well as anaerobically. This is consistent with the observations of the type strain [16].

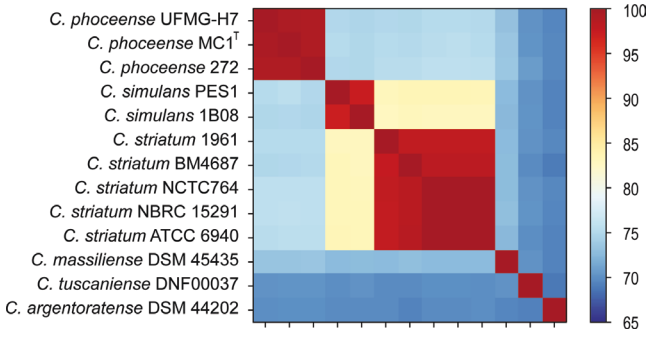

Fig. 2. Heatmap of ANI values for *C. phoceense* strains and representatives of other related *Corynebacterium* species

DNA was extracted (Qiagen DNeasy UltraClean microbial kit (Qiagen, Hilden, Germany)) and sent to the Microbial Genome Sequencing Centre (Pittsburg, PA) for library preparation and whole genome sequencing using the Illumina NextSeq 550 platform. Raw reads were trimmed using sickle (https://github.com/najoshi/sickle) and assembled using SPAdes v3.13 [18] producing a 2.7 Mbp genome in 41 contigs (GC%=63.4%; N50=165 055; coverage=138×). The raw reads and genome assembly for *C. phoceense* UFMG-H7 have been deposited in GenBank under accession numbers SRR13080711 and GCF_015686525.1, respectively. The publicly available genome was annotated using PGAP v4.13 [19].

RESULTS

With three genome assemblies now available for *C. phoceense*, we conducted a comparative genomic analysis for the species. Average Nucleotide Identity (ANI) values were calculated using JSpeciesWS [20] for *C. phoceense* UFMG-H7, other *C. phoceense* strains, and species previously identified [16] as closely related to *C. phoceense* (Fig. 2). *C. phoceense* is distinct from strains of *C. simulans* (ANI values range: 74.73–75.59) and *C. striatum* (ANI values range: 74.96–75.83). Additionally, this examination confirms that UFMG-H7 belongs to the *C. phoceense* species. It has an ANI value of 99.41 to the species' type strain MC1^T and an ANI value of 99.2 to *C. phoceense* strain 272.

To further investigate the *C. phoceense* species, we retrieved all publicly available complete genome sequences or assemblies for *C. phoceense* (*n*=3) and close relatives, including strains belonging to *C. massiliense* (*n*=1), *C. simulans* (*n*=3), and *C. striatum* (*n*=33) (Table S1). The pangenome for these 40 genomes consists of 4640 genes. We identified 519 genes within the core genome; these genes are single copy-number genes present in all 40 genomes. Core and pangenome analyses were conducted using anvi'o v6.2 [21]. A phylogenomic tree was derived based upon the alignment of the concatenation of the core amino acid sequences using anvi'o [21], FastTree [22], and iTOL [23] (Fig. 3). The core genome phylogeny further confirms that *C. phoceense* is a distinct species and *C. phoceense* UFMG-H7 is a representative of this species.

Narrowing our focus to just *C. phoceense*, 1456 unique coding regions are contained within the core genome of the species. Of these 515 are unique to the three *C. phoceense* genomes, i.e. they are not found in any of the *C. massiliense*, *C. simulans*, or *C. striatum* genomes from our prior pangenome analysis. We next queried each of the 515 amino acid sequences against the complete nr database using the blastp algorithm. Examination of these blast results repeatedly found hits to *C. phoceense* as well as several *Corynebacterium* sp. strains,

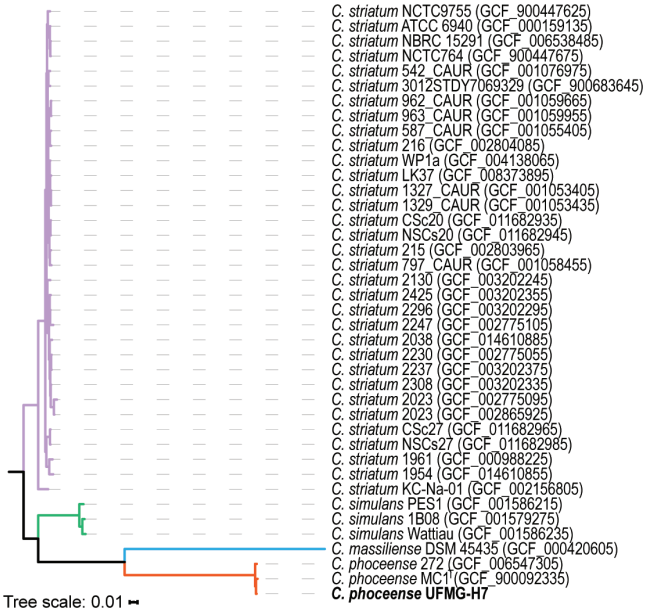

Fig. 3. Phylogenomic tree based upon the core genome for *C. phoceense* and related species. Purple branches indicate *C. striatum* genomes; green branches indicate *C. simulans* genomes; the blue branch is for the single representative of *C. massiliense*; the red branches indicate *C. phoceense* genomes, with the UFMG-H7 strain presented here shown in bold. The tree scale is shown, representative of the sequence divergence between the aligned, concatenated amino acid core sequences. The RefSeq accession number for each genome is indicated in parentheses

namely *Corynebacterium* sp. DNF00584 (GCF_001552965.1), *Corynebacterium* sp. CMW7794 (GCF_001563605.1), *Corynebacterium* sp. HMSC072G08 (GCF_001809185.1), and *Corynebacterium* sp. HMSC077B05 (GCF_001810505.1). We retrieved these four genome assemblies from NCBI and calculated their pair-wise ANI to the three *C. phoceense* genomes. We also compared the 16S rRNA and *rpoB* gene sequences. Fig. 4 shows the results of the ANI and *rpoB* analysis. The 16S rRNA gene sequences were identical among the sequences examined. These comparisons confirm that the four *Corynebacterium* species should in fact be classified as *C. phoceense*. Interestingly all *C. phoceense* genomes come from urogenital isolates suggesting that this is a bacterium specific to this niche. Furthermore, it is not human specific as our isolate *C. phoceense* UFMG-H7 comes from the bovine urinary tract.

Blast analysis also identified genes within the 515 gene set that were encoded by other *Corynebacterium* species not included in our pangenome analysis. Nevertheless, 436 genes are unique to the seven *C. phoceense* genomes and strictly conserved among the three named *C. phoceense* strains. It is important to note that these sequences were queried against the entire nr database, not just *Corynebacterium* species, and exhibited no recognisable homology to any annotated bacterial proteins in other species. The majority of these gene sequences encode for hypothetical proteins (Table S2). The genes were queried against the SEED database using the PATRIC server [24, 25]; only 19 of the 436 were assigned to a functional class. These genes represent genetic diversity of the species that has yet to be catalogued and whose function is unknown.

The seven *C. phoceense* genomes were next examined for antibiotic resistance genes using ResFinder with default parameters [26]. *C. phoceense* UFMG-H7, *C. phoceense* MC1^T, and *Corynebacterium* sp. HMSC077B05 were not predicted to contain any genes associated with antibiotic resistance. On the other hand, *C. phoceense* 272 carried the genes *erm*(X), *aph*(3′)-la, *aph*(3′)-lb, *aph*(6)-ld, and *cmx* which are predicted to confer resistance to steptogramin b, lincosamides, macrolides, aminoglycosides, and phenicols. *Corynebacterium* sp. DNF00584 and *Corynebacterium* sp. CMW7794 also encoded *erm*(X) and thus are predicted to be resistant to streptogramin b, lincosamides, and macrolides. *Corynebacterium* sp. CMW7794 and *Corynebacterium* sp. HMSC072G08 encoded *aph*(3′)-la, which confers resistance to aminoglycosides. All of the aforementioned antibiotic resistance genes were located within predicted genomic islands, per IslandViewer4 analysis [27], often flanked by IS6 family transposases. Therefore, there are no genes associated with antibiotic resistance that are carried by all strains of this species. Furthermore, prior genomic studies of other *Corynebacterium* species frequently find *erm*(X) [28, 29] and *aph* genes [30] among clinical isolates.

Because pathogenicity in other *Corynebacterium* species is often linked to prophage [31], we examined the seven *C. phoceense* genomes for prophages using PHASTER [32]. All of the strains were predicted to include at least one incomplete prophage (Table S3). Each prophage sequence

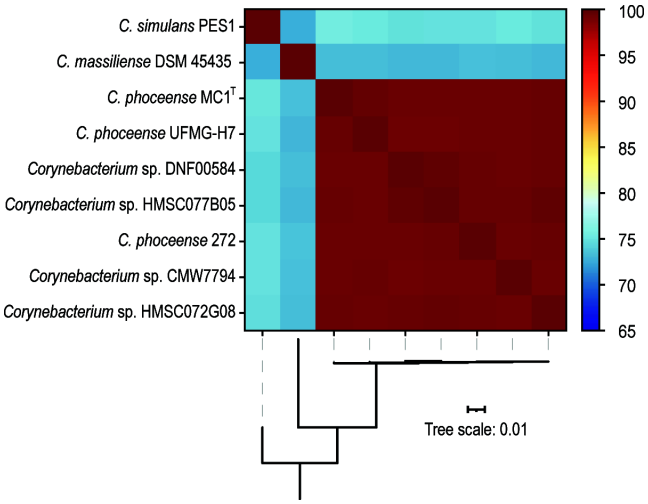

Fig. 4. Comparison of *Corynebacterium* strains identified as members of the *C. phoceense* species. The matrix shows that the ANI values of the *C. phoceense* strains and four *Corynebacterium* species are greater than 99%. The tree below the heatmap shows the *rpoB* gene tree. Isolates are ordered in the heatmap based upon the *rpoB* gene tree structure. The tree scale is shown, representative of the sequence divergence between the aligned *rpoB* gene nucleotide sequences

Table 1. Virulence factors conserved among *C. phoceense*, *C. simulans*, and *C. massiliense* genomes

Virulence factor class	Virulence factor	Related genes	*C. phoceense* (*n*=7)	*C. simulans* (*n*=3)	*C. massiliense* (*n*=1)
Adherence	SpaD-type pili	*spaD srtB*	+++++++ +++++	+	
Iron uptake	ABC-type haem transporter	*hmuT hmuU hmuV*	+++++++ +++++ +++++	+++	
	Siderophore-dependent iron uptake system	*irp6A irp6B irp6C*	+++++++ +++++ +++++	++	+
Regulation	Diphtheria toxin repressor	*dtxR*	+++++	++	+
	SenX3	*senX3*		+++	+
	MprA/B	*mprA*	+++++		
	Sigma A	*sigA/rpoV*	+++++	++	+
	Sigma D	*sigD*	+++++	++	
	WhiB3	*whiB3*	+++++	++	+
Amino acid and purine metabolism	Glutamine synthesis	*glnA1*		+++	+
	Lysine synthesis	*lysA*	+++++	+++	+
Copper uptake	Copper exporter	*ctpV*	+++++++	++	
Lipid and fatty acid metabolism	Pantothenate synthesis	*panC*	+++++++		
Phagosome arresting	Tyrosine phosphatase	*ptpA*	+++++		+
Protease	Proteasome-associated proteins	*mpa*	+++++	+++	+
		pafA	+++++++	+++	+
Secreted proteins	Protein kinase G	*pknG*	+++++++	+++	+
Secretion system	Accessory secretion factor	*secA2*	+++++++	++	+

*A "+" indicates the presence of related gene in a genome sequence.

was queried against the nr/nt database using discontiguous blast. Some exhibited similarity with genes in other characterized phages, frequently *Corynebacterium* phages infectious of other *Corynebacterium* species. The *C. phoceense* UFMG-H7 predicted prophage is 29.5 Kbp in length. We annotated this phage sequence using RAST [24] and blastp queries to the nr/nt database and were able to identify essential hallmark phage genes (Table S4). Nevertheless, the *C. phoceense* prophage sequences do not carry virulence factors or toxins.

The *C. phoceense* genomes encode for several recognized virulence factors, including genes related to ABC-type haem transporter, siderophore-dependent iron uptake system, lysine synthesis, copper export, pantothenate synthesis, proteasome-associate proteins, among others (Table S5). Genomes of *C. simulans* and *C. massiliense* also were examined for virulence factors, also using VFAnalyzer [33]. Table 1 lists the virulence factors conserved in genomes of one or more of these three species. *C. phoceense* strains include the *MprA/B* regulator, whereas the genomes of *C. simulans* and *C. massiliense* do not. However, *C. simulans* and *C. massiliense* encode for the *senX3* regulator and *C. phoceense* strains do not. Another distinct difference between these three species is the lack of the glutamine synthesis gene *glnA1* in *C. phoceense* and the presence of the pantothenate synthesis protein *panC* only in *C. phoceense*. Upon further inspection via tblastn queries, other genes encoding enzymes of the pantothenate biosynthesis pathway were identified in the *C. phoceense* genomes. Previous studies have shown that pantothenate biosynthesis plays an important role in membrane lipid synthesis and cell metabolism in bacteria (see review [34]). Furthermore panthothenate kinase was found to be enriched in bacterial species of the female urogenital tract [4]. It is important to note that representatives of these three species have been isolated from different body sites, *C. simulans* from skin and *C. massiliense* from hip joint fluid (Table S1). The differences between these environments may contribute to the different virulence factors identified.

DISCUSSION

Here we have presented the first isolation of a *C. phoceense* strain in cattle. Recently, two new *Corynebacterium*, *C. urogenitale* and *C. endometrii*, were described, both isolated from the bovine vaginal tract and nonpathogenic for uterine epithelial cells [35, 36]. Evidence suggests that many *Corynebacterium* species are commensal members of the uterine microbiome of cattle [37–40] and buffalo [41]. Evidence of the SpaD-type pili system (Table 1) conserved among the *C. phoceense* strains suggests that adhesion is central to its survival. We screened 18 publicly available *Corynebacterium* genomes from the human urinary microbiota (BioProject: PRJNA316969) for pili systems using VFAnalyzer [33]. While these genomes represent nine different species, only *C. amycolatum* and *C. aurimucosum* genomes include a pili system – the SpaD-type pili system. We found that neither the bovine *C. urogenitale* nor *C. endometrii* genomes code for a pili cluster. Both *C. amycolatum* and *C. aurimucosum* have recently been reported as opportunistic pathogens causing a variety of infections throughout the human body [42–44], including the urinary [44, 45] and vaginal [46] tracts. Furthermore, pili play an essential role in uropathogenic *E. coli* UTI infections in the human urinary tract [47]. Accordingly, the presence of the pili in the *C. phoceense* strains, coupled with the other virulence factors detected (Table 1), suggests that this species has pathogenic potential.

In contrast to the vaginal microbiota, very little is known about the urinary microbiota of healthy cattle. *C. phoceense* is phylogenetically distinct from the *Corynebacterium* species found within bovine vaginal samples. The fact that *C. phoceense* has been primarily detected within the urinary tract of humans and now cattle suggests that this species may be specific to this environment. Recent studies have found that some of the same bacterial species inhabit both the bovine and human urinary microbiota [48, 49]. Previous research found that the proteoglycan surface within the human and bovine bladder are similar [50]; in humans, this layer is a source for adhesion and nutrients by urinary bacteria [51]. Further isolation of *C. phoceense* is needed in order to ascertain if it is specific to the urinary or urogenital tract. While here we have contributed the third genome for the species as well as identified four strains that need to be reclassified as *C. phoceense*, the majority of the genes unique to the species have an unknown function. These may be key in ascertaining how and possibly the contributions of this species persists within the urinary environment.

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Author contributions
S.G.F., E.F.B.S. and C.P., conceptualized the study. S.G.F. and A.E., conducted formal analyses. All authors contributed to the investigation and writing review and editing. S.G.F., and C.P., wrote the original draft.

Conflicts of interest
The authors declare that there are no conflicts of interest.

Ethical statement
Sampling was approved by the Ethics Committee in Animal Experimentation of the Universidade Federal de Minas Gerais, Brazil (approval number: CEUA/UFMG - 40/2019).

References

1. Fouts DE, Pieper R, Szpakowski S, Pohl H, Knoblach S, *et al*. Integrated next-generation sequencing of 16S rDNA and metaproteomics differentiate the healthy urine microbiome from asymptomatic bacteriuria in neuropathic bladder associated with spinal cord injury. *J Transl Med* 2012;10:174.
2. Pearce MM, Hilt EE, Rosenfeld AB, Zilliox MJ, Thomas-White K, *et al*. The female urinary microbiome: A comparison of women with and without urgency urinary incontinence. *MBio* 2014;5:e01283-14.
3. Hilt EE, McKinley K, Pearce MM, Rosenfeld AB, Zilliox MJ, *et al*. Urine is not sterile: Use of enhanced urine culture techniques to detect resident bacterial flora in the adult female bladder. *J Clin Microbiol* 2014;52:871–876.
4. Thomas-White K, Forster SC, Kumar N, Van Kuiken M, Putonti C, *et al*. Culturing of female bladder bacteria reveals an interconnected urogenital microbiota. *Nat Commun* 2018;9:1557.
5. Kline KA, Lewis AL. Gram-positive uropathogens, polymicrobial urinary tract infection, and the emerging microbiota of the urinary tract. *Microbiol Spectr* 2016;4.
6. Bailiff NL, Westropp JL, Jang SS, Ling GV. *Corynebacterium urealyticum* urinary tract infection in dogs and cats: 7 cases (1996-2003). *J Am Vet Med Assoc* 2005;226:1676–1680.
7. Briscoe KA, Barrs VR, Lindsay S, Hoffmann KL, Cockwill KR, *et al*. Encrusting cystitis in a cat secondary to *Corynebacterium urealyticum* infection. *J Feline Med Surg* 2010;12:972–977.
8. Suarez ML, Espino L, Vila M, Santamarina G. Urinary tract infection caused by *Corynebacterium urealyticum* in a dog. *J Small Anim Pract* 2002;43:299–302.
9. Nicolet J, Fey H. Antibody coated bacteria in urine sediment from cattle infected with *Corynebacterium renale*. *Vet Rec* 1979;105:301–303.
10. Rebhun WC, Dill SG, Perdrizet JA, Hatfield CE. Pyelonephritis in cows: 15 cases (1982-1986). *J Am Vet Med Assoc* 1989;194:953–955.
11. Wallace LL, Bouchard G, Nicholson W, Turk J, Sweeney CL. Polypoid cystitis, pyelonephritis, and obstructive uropathy in a cow. *J Am Vet Med Assoc* 1990;197:1181–1183.
12. Yeruham I, Elad D, Avidar Y, Goshen T, Asis E. Four-year survey of urinary tract infections in calves in Israel. *Vet Rec* 2004;154:204–206.
13. Yeruham I, Elad D, Avidar Y, Goshen T. A herd level analysis of urinary tract infection in dairy cattle. *Vet J* 2006;171:172–176.
14. Braun U, Nuss K, Wehbrink D, Rauch S, Pospischil A. Clinical and ultrasonographic findings, diagnosis and treatment of pyelonephritis in 17 cows. *Vet J* 2008;175:240–248.
15. Niang EHA, Cl L, Morand A, Ndongo S, Raoult D, *et al*. *Corynebacterium urinapleomorphum* sp. nov., a new bacterial species isolated from human urine sample. *New Microbes New Infect* 2019;31:100576.
16. Cresci M, Ibrahima Lo C, Khelaifia S, Mouelhi D, Delerce J, *et al*. *Corynebacterium phoceense* sp. nov., strain MC1T a new bacterial species isolated from human urine. *New Microbes New Infect* 2016;14:73–82.

17. Intanoo M, Kongkeitkajorn MB, Pattarajinda V, Bernard JK, Callaway TR, *et al*. Isolation and screening of aflatoxin-detoxifying yeast and bacteria from ruminal fluids to reduce aflatoxin B 1 contamination in dairy cattle feed. *J Appl Microbiol* 2018;125:1603–1613.
18. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, *et al*. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. *J Comput Biol* 2012;19:455–477.
19. Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Nawrocki EP, *et al*. NCBI prokaryotic genome annotation pipeline. *Nucleic Acids Res* 2016;44:6614–6624.
20. Richter M, Rosselló-Móra R, Oliver Glöckner F, Peplies J. JSpeciesWS: a web server for prokaryotic species circumscription based on pairwise genome comparison. *Bioinformatics* 2016;32:929–931.
21. Eren AM, Esen ÖC, Quince C, Vineis JH, Morrison HG, *et al*. Anvi'o: an advanced analysis and visualization platform for' omics data. *PeerJ* 2015;3:e1319.
22. Price MN, Dehal PS, Arkin AP. FastTree 2--approximately maximum-likelihood trees for large alignments. *PLoS ONE* 2010;5:e9490.
23. Letunic I, Bork P. Interactive Tree Of Life (iTOL) v4: recent updates and new developments. *Nucleic Acids Res* 2019;47:W256–W259.
24. Brettin T, Davis JJ, Disz T, Edwards RA, Gerdes S, *et al*. RASTtk: a modular and extensible implementation of the RAST algorithm for building custom annotation pipelines and annotating batches of genomes. *Sci Rep* 2015;5:8365.
25. Davis JJ, Wattam AR, Aziz RK, Brettin T, Butler R, *et al*. The PATRIC Bioinformatics resource center: expanding data and analysis capabilities. *Nucleic Acids Res* 2020;48:D606–D612.
26. Bortolaia V, Kaas RS, Ruppe E, Roberts MC, Schwarz S, *et al*. ResFinder 4.0 for predictions of phenotypes from genotypes. *J Antimicrob Chemother* 2020;75:3491–3500.
27. Bertelli C, Laird MR, Williams KP, Simon Fraser University Research Computing Group, Lau BY, *et al*. Islandviewer 4: Expanded prediction of genomic islands for larger-scale datasets. *Nucleic Acids Res* 2017;45:W30–W35.
28. Ortiz-Pérez A, Martín-de-Hijas NZ, Esteban J, Fernández-Natal MI, García-Cía JI, *et al*. High frequency of macrolide resistance mechanisms in clinical isolates of *Corynebacterium* species. *Microb Drug Resist* 2010;16:273–277.
29. Salem N, Salem L, Saber S, Ismail G, Bluth MH. *Corynebacterium urealyticum*: a comprehensive review of an understated organism. *Infect Drug Resist* 2015;8:129–145.
30. Navas J, Fernández-Martínez M, Salas C, Cano ME, Martínez-Martínez L. Susceptibility to Aminoglycosides and distribution of aph and aac(3)-XI genes among *Corynebacterium striatum* clinical isolates. *PLoS One* 2016;11:e0167856.
31. Schroven K, Aertsen A, Lavigne R. Bacteriophages as drivers of bacterial virulence and their potential for biotechnological exploitation. *FEMS Microbiol Rev* 2021;45.
32. Arndt D, Grant JR, Marcu A, Sajed T, Pon A, *et al*. PHASTER: a better, faster version of the PHAST phage search tool. *Nucleic Acids Res* 2016;44:W16-21.
33. Liu B, Zheng D, Jin Q, Chen L, Yang J. VFDB 2019: A comparative pathogenomic platform with an interactive web interface. *Nucleic Acids Res* 2019;47:D687–D692.
34. Begley TP, Kinsland C, Strauss E. The biosynthesis of coenzyme a in bacteria. In: *Vitamins & Hormones*. Elsevier, . pp. 157–171.
35. Ballas P, Rückert C, Wagener K, Drillich M, Kämpfer P, *et al*. *Corynebacterium urogenitale* sp. nov. isolated from the genital tract of a cow. *Int J Syst Evol Microbiol* 2020;70:3625–3632.
36. Ballas P, Rückert C, Wagener K, Drillich M, Kämpfer P, *et al*. *Corynebacterium endometrii* sp. nov., isolated from the uterus of a cow with endometritis. *Int J Syst Evol Microbiol* 2020;70:146–152.
37. Laguardia-Nascimento M, Branco KMGR, Gasparini MR, Giannattasio-Ferraz S, Leite LR, *et al*. Vaginal microbiome characterization of nellore cattle using metagenomic analysis. *PLoS One* 2015;10:e0143294.
38. Wang M-L, Liu M-C, Xu J, An L-G, Wang J-F, *et al*. Uterine microbiota of dairy cows with clinical and subclinical endometritis. *Front Microbiol* 2018;9:2691.
39. Giannattasio-Ferraz S, Laguardia-Nascimento M, Gasparini MR, Leite LR, Araujo FMG, *et al*. A common vaginal microbiota composition among breeds of Bos taurus indicus (Gyr and Nellore). *Braz J Microbiol* 2019;50:1115–1124.
40. Ballas P, Reinländer U, Schlegl R, Ehling-Schulz M, Drillich M, *et al*. Characterization of intrauterine cultivable aerobic microbiota at the time of insemination in dairy cows with and without mild endometritis. *Theriogenology* 2021;159:28–34.
41. El-Jakee J, Ahmed W, El-Seedy F, Abd El-Moez S. Bacterial profile of the genital tract in femalebuffalo during the different reproductive stages. *Glob Vet* 2008;2:7–14.
42. Sugumaran R, Sistla S, Chavhan P, Deb AK. *Corynebacterium amycolatum*: An unusual cause of corneal ulcer. *BMJ Case Rep* 2020;13.
43. Borde K, Rao V, Shah M, Pavani N, Anand M. Not always a commensal: A case of mastitis by *Corynebacterium amycolatum*. *IDCases* 2020;20:e00728.
44. Lefèvre CR, Pelletier R, Le Monnier A, Corvec S, Bille E. Clinical relevance and antimicrobial susceptibility profile of the unknown human pathogen *corynebacterium aurimucosum*. *J Med Microbiol* 2021;70.
45. Lo S, Thiam I, Fall B, Ba-Diallo A, Diallo OF, *et al*. Urinary tract infection with *Corynebacterium aurimucosu*m after urethroplasty stricture of the urethra: a case report. *J Med Case Rep* 2015;9:156.
46. Chen X, Zhao X, Chen L, Zeng W, Xu H. Vaginitis caused by *corynebacterium amycolatum* in a prepubescent girl. *J Pediatr Adolesc Gynecol* 2015;28:e165-167.
47. Spaulding CN, Hultgren SJ. Adhesive PILI in UTI pathogenesis and drug development. *Pathogens* 2016;5.
48. Giannattasio-Ferraz S, Maskeri L, Oliveira AP, Barbosa-Stancioli EF, Putonti C. Draft genome sequence of *Citrobacter freundii* UFMG-H9, isolated from urine from a healthy bovine heifer (Gyr breed). *Microbiol Resour Announc* 2020;9.
49. Giannattasio-Ferraz S, Maskeri L, Oliveira AP, Barbosa-Stancioli EF, Putonti C. Three draft genome sequences of S*taphylococcus* species isolated from the urine of healthy bovine heifers (Gyr breed). *Microbiol Resour Announc* 2020;9.
50. Hurst RE, Zebrowski R. Identification of proteoglycans present at high density on bovine and human bladder luminal surface. *J Urol* 1994;152:1641–1645.
51. Neugent ML, Hulyalkar NV, Nguyen VH, Zimmern PE, De Nisco NJ. Advances in understanding the human urinary microbiome and its potential role in urinary tract infection. *MBio* 2020;11.