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Description of *Citrobacter cronae* sp. nov., isolated from human rectal swabs and stool samples

Philipp Oberhettinger¹,²,*, Leonard Schüle¹,³, Matthias Marschal¹,², Daniela Bezdan⁴, Stephan Ossowski⁵, Daniela Dörfel⁶,⁷, Wichard Vogel⁶, John W. Rossen⁴, Matthias Willmann¹,² and Silke Peter¹,²

**TAXONOMIC DESCRIPTION**

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**Abstract**

Nine independent Gram-negative bacterial strains were isolated from rectal swabs or stool samples of immunocompromised patients from two different wards of a university hospital. All isolates were phylogenetically analysed based on their 16S rRNA gene sequence, housekeeping gene *recN*, multilocus sequence analysis of concatenated partial *fusA*, *leuS*, *pyrG* and *rpoB* sequences, and by whole genome sequencing data. The analysed strains of the new species cluster together and form a separate branch with *Citrobacter werkmanii* NBRC105721ᵀ as the most closely related species. An average nucleotide identity value of 95.9–96% and computation of digital DNA–DNA hybridization values separate the new species from all other type strains of the genus *Citrobacter*. Biochemical characteristics further delimit the isolates from closely related *Citrobacter* type strains. As a result of the described data, a new *Citrobacter* species is introduced, for which the name *Citrobacter cronae* sp. nov. is proposed. The type strain is Tue2-1ᵀ with a G+C DNA content of 52.2 mol%.

**ISOLATION AND ECOLOGY**

*Citrobacter* species are Gram-negative microorganisms frequently encountered in the environment, but also from the intestinal tract of humans [1–3]. They were also reported as opportunistic pathogens causing wound infections, abscesses, severe forms of meningitis, endocarditis or bloodstream infections [4–8].

In clinical microbiology laboratories, *Citrobacter* species represent up to 6% of all isolated *Enterobacteriaceae* from clinical specimens [4]. As they can have chromosomal AmpC β-lactamases [9] as well as plasmid encoded carbapenemases [10], many antibiotics are ineffective increasing the intricacy of treatment [11]. To date, 15 *Citrobacter* species are published in the literature [12]. In the present study, nine independent clinical isolates (Tue2-1ᵀ, Tue2-3 and Tue2-5–Tue2-11) were investigated that originated from nine patients of two different wards with underlying haematological conditions. Isolates were collected and stored from rectal swabs or stool samples over a 3-year period (2012–2015), but even more isolates were obtained since 2016. Three of the strains (Tue2-1ᵀ, Tue2-3, Tue2-5) harbouring metallo-β-lactamase (MBL) enzymes were already characterized by comparative genomics using next generation sequencing, but could not be unambiguously identified to the species level by standard routine methods [13]. We first considered that the studied isolates belong to the species *Citrobacter werkmanii*. However, experimental evidence suggested that the three isolates belong to a new *Citrobacter* species, which will be characterized here. In order to gain further evidence for our new hypothesis we additionally characterized and sequenced six more isolates (Tue2-6–Tue2-11).

**METHODS**

MALDI-TOF AXIMA system assurance (bioMérieux; Saramis database version 4.09) and the Microflex LT instrument (Bruker Daltonics; MBT IVD Library.5627) were
performed on all isolates, but failed to unambiguously iden-
tify the strains. Additionally, biochemically based identifica-
tion using the API 20 E System (bioMérieux; apiweb) and the
vitek GN ID card (bioMérieux) was applied. Bacterial DNA
was extracted from cultures grown on Columbia agar with
5% sheep blood (Becton Dickinson) using the Genomic-
tip 100/G system (Qiagen) following the manufacturer’s instruc-
tions. For whole genome sequencing, libraries were prepared
using the TruSeq DNA LT Sample Prep Kit (Illumina) with
24 different barcodes using standard protocols as described
previously [13, 14]. Barcoded libraries were analysed by the
Agilent 2100 Bioanalyzer (Agilent Technologies) or QIAXcel
Advanced Instrument (Qiagen) and quantified by Real-Time
(RT)-PCR. Normalized libraries were pooled and sequenced
with v3 chemistry (2×300 bp) or with v2 chemistry (2×250 bp)
on the MiSeq platform (Illumina). Assembly of genome
sequences was performed using SPAdes (version 3.7.0) [15]
with default settings.

For phylogenetic analysis of the isolates, publically available
whole genome sequencing (WGS) data from Citrobacter type
strains were included in the analysis (Table S1). Progressive-
Mauve (version 2.3.1) [16] was run to conduct a full alignment
of 23 genomes using default settings and prophage regions
were investigated and excluded using phaster (phaster.ca)
[17]. Maximum-likelihood phylogenetic trees of 23 whole
genome-sequences were reconstructed by applying IQ-Tree
with 1000 bootstrap replicates. Alignments of 16S rRNA
gene sequences downloaded from EZ-Taxon [18], concat-
enated partial fusA (protein synthesis elongation factor-G),
leuS (leucine tRNA synthetase), pyrG (CTP synthetase) and
rpoB (β-subunit of RNA polymerase) [19] as well as recN

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**Fig. 1. Multilocus sequence analysis of concatenated partial fusA, leuS, pyrG and rpoB gene sequences extracted from whole genome
data of the study isolates (Citrobacter cronae Tue2-1\(^1\), Tue2-3, Tue2-5 – Tue2-11) and available genome data of Citrobacter type strains.
The scale bar represents the expected number of changes per site. Bootstrap values [%] are colour-coded for all nodes (based on 1000
replicates). The tree was rooted at midpoint.**
sequences (DNA repair) [2, 20] used for recent description of new *Citrobacter* species extracted from available WGS data were done by *clustal_w* (BioEdit version 7.2.5) followed by phylogenetic treeing with RAxML and the GTR model in conjunction with GAMMA rates [21]. Trees were visualized using FigTree (version 1.4.3). The average nucleotide identity (ANI) was assessed by *JSpecies* (version 1.2) [22, 23] based on BLAST +2.2.29 (ANib). The Genome-to-Genome Distance Calculator (GGDC 2.1) using the recommended Formula 2 was applied for *in silico* genome comparison and computation of digital DNA–DNA hybridization (dDDH) values [24]. Employing the online multi-locus sequence typing (MLST) service of the Center for Genomic Epidemiology (https://cge.cbs.dtu.dk/services/MLST/; version 2.0), MLST sequence types were obtained from assembled sequences based on the MLST scheme for *C. freundii* [25].

**PHYLOGENY**

MALDI-TOF using Bruker and bioMérieux systems as well as analysing the biochemical characteristics of these strains with API 20E and VITEK2 system (bioMérieux) did not allow for unambiguous identification of all nine study isolates on the species level. Dissecting the MLST type showed the same result for all nine strains isolated over the 3-year collection period. Their phylogenetic relationship to other *Citrobacter* type strains was assessed by analysing the 16S rRNA, the *recN* gene, the concatenated partial *fusA*, *leuS*, *pyrG* and *rpoB* genes as well as by WGS. 16S rRNA gene-based phylogeny represented a distinct branch of all isolates of the new *Citrobacter* species clustering together in group I including the formerly published species *Citrobacter freundii*, *Citrobacter youngae*, *Citrobacter braakii*, *Citrobacter werkmanii*, *Citrobacter gillenii* and *Citrobacter murliniae* [26] as well as *Citrobacter pasteuri* [19] and the recently described *C. europaeus* [2] and *C. portu- calensis* [20] (Fig. S1, available in the online version of this article). Regarding the limited resolution of 16S rRNA genes in discrimination of *Citrobacter* species [19, 27], the closest similarity in 16S rRNA gene comparison was found to *C. freundii* (99.73%). Phylogenetic analysis based on the *recN* gene (Fig. S2) as well as MLSA of concatenated partial *fusA*, *leuS*, *pyrG* and *rpoB* (Fig. 1) extracted from WGS data of type strains confirmed 16S rRNA gene-based clustering of all nine isolates in a separate branch. The maximum-likelihood tree generated using WGS data enabled further distinction of the new *Citrobacter* species from other type strains of the genus including the most closely related species *C. werkmanii* strain NBRC 105721T (Fig. 2).
MALDI-TOF using Bruker and bioMérieux systems as well as analysing the biochemical characteristics of these strains with API 20E and VITEK2 system (bioMérieux) did not allow for unambiguous identification of all nine study isolates on the species level. Dissecting the MLST type showed the same result for all nine strains isolated over the 3-year collection period. Their phylogenetic relationship to other Citrobacter type strains was assessed by analysing the 16S rRNA, the recN gene, the concatenated partial fusA, leuS, pyrG and rpoB genes as well as by WGS. 16S rRNA gene-based phylogeny represented a distinct branch of all isolates of the new Citrobacter species clustering together in group I including the formerly published species Citrobacter freundii, Citrobacter youngae, Citrobacter braakii, Citrobacter werkmanii, Citrobacter gillenii and Citrobacter murliniae [26] as well as the recently described C. europaeus [2] and C. portucalensis [20] (Fig. S1, available in the online version of this article). Regarding the limited resolution of 16S rRNA genes in discrimination of Citrobacter species [19, 27], the closest similarity in 16S rRNA gene comparison was found to C. freundii (99.73%). Phylogenetic analysis based on the recN gene (Fig. S2) as well as MLSA of concatenated partial fusA, leuS, pyrG and rpoB (Fig. 1) extracted from WGS data of type strains confirmed 16S rRNA gene-based clustering of all nine isolates in a separate branch. The maximum-likelihood tree generated using WGS data enabled further distinction of the new Citrobacter species from other type strains of the genus including the most closely related species C. werkmanii strain NBRC 105721T (Fig. 2).

### GENOME FEATURES

Species definition can also be based on ANI value [22, 28]. Therefore the new Citrobacter isolates were compared to all Citrobacter type strains with available WGS data (Table S2b). The closest relationship of the new Citrobacter species was found with C. werkmanii NBRC105721T (95.92%), slightly below the proposed cut-off value of 96% for the assignment of a new species [29]. In comparison, ANI values between all nine analysed isolates (Tue2-1, Tue2-3, Tue2-5–2–11) were above 99.5%, demonstrating their close relationship (Table S2a).

As described recently, dDDH can be used for delineation of a new bacterial species using WGS data [29]. As illustrated in Table S2b, dDDH values were calculated for all available Citrobacter type strains in relation to Tue2-1T. The lowest intergenomic distance of our nine analysed Citrobacter species isolates was found to C. werkmanii NBRC105721T with a dDDH value of 70%, exactly the cutoff proposed for bacterial species delineation [22, 28]. The dDDH values between any pair of the new Citrobacter species isolates were above 99.1%.

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**Table 1.** Biochemical characteristics of all Citrobacter cronae study isolates and closely related Citrobacter type strains

| Characteristics | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
|-----------------|---|---|---|---|---|---|---|---|---|
| Amygdalin       | + | – | – | – | – | – | – | – | – |
| Cellobiose      | + | – | v | v | v | + | + | v | v |
| Catalase        | + | v | v | v | v | – | – | + | + |
| Phosphatase     | – | + | NA | NA | + | NA | NA | NA | NA |
| α-Glucosidase   | – | – | + | v | – | NA | v | NA | v |
| Indole          | – | – | – | – | – | v | – | – | v |
| Melibiose       | – | – | – | v | – | + | + | + | v |
| β-Glucosidase   | v (66.6) | – | – | v | + | NA | v | NA | – |
| Adonitol        | v (44.4) | – | – | – | – | – | – | – | – |
| H₂S             | v (44.4) | + | + | + | + | + | + | v | v |
| Malonate        | v (88.8) | + | – | – | – | NA | – | NA | + |
| Ornithine       | v (44.4) | – | – | v | – | NA | – | NA | v |
| Sucrose         | v (77.7) | – | – | – | v | + | + | – | – |
| 5-Ketogluconate | v (66.6) | – | – | + | + | + | + | + | + |
| Data obtained from | This study | This study | [19, 30] | [19, 30] | [19] | [20] | [19, 30] | [2] | [19, 30] |
PHYSIOLOGY
Biochemical characteristics were analysed by the API 20E and VITEK2 systems and results are listed in Table 1 for all nine study isolates and closely related type strains of the genus Citrobacter [2, 19, 20, 30]. The data show that all nine C. cronae isolates are able to catabolize amygdalin distinguishing the novel species from all other closely related Citrobacter species tested. In addition all C. cronae isolates are able to catabolize cellobiose, which is not the case for C. werkmanii DSM17579T. No enzymatic activity for phosphatase could be found for C. cronae, whereas C. werkmanii DSM17579T was positive for phosphatase. Moreover due to some variable characteristics not found to be diverse in different C. werkmanii isolates [19, 30], C. cronae can be separated biochemically. Taken together, phylogenetic analysis based on 16S rRNA gene, recN, the concatenated partial genes ftsA, leuS, pyrG and rpoB and WGS data, calculation of genome relatedness by ANI and dDDH as well as biochemical properties classifies Citrobacter Tue2-1T, 2-3 and 2-5–2-11 as representing a new species within the genus Citrobacter for which we propose the name Citrobacter cronae sp. nov., with Tue2-1T as type strain.

DESCRIPTION OF CITROBACTER CRONAЕ SP. NOV.

Citrobacter cronae [cro‘nae. N.L. gen. n. cronae, pertaining to he CRONA (the landmark building of the university hospital; acronym for Surgery, Radiology, Orthopedics, Neurology, Anesthesiology) clinics, Tuebingen, Germany].

Citrobacter cronae is a Gram-stain-negative, oxidase-negative, catalase-positive (delayed), facultative anaerobic, rod-shaped bacterium. It is able to ferment the following carbohydrates: d- adonitol, citrate, potassium 5- keto- gluconate, sucrose, trehalose and amygdalin. The strains cannot utilize aesculin, and derivatives: d- mannitol, sorbitol, d- mannose, cellobiose, baebacterium. It is able to ferment the following carbohydrates:

C. cronae is Tue2-1 T, which β-glucosidase.

N-acetyl-β Glu- Gly- Arg- arylamidase, for α-glucosidase, phosphatase, lipase, lysine decarboxylase- galactosidase, but negative reactions

Semi-quantitative analysis of enzymatic activities of all strains demonstrate positive reactions for L- pyrrolidonaryl- amylidase and β- galactosidase, and negative reactions for α- glucosidase, phosphatase, lipase, lysine decarboxylase, tryptophan deaminase, gelatinase, α- galactosidase, Glu- Gly- Arg- aryiamidase, β- N- acetyl- galactosaminidase, β- xylosidase, β- alanin- aryiamidase- pNA and β- glucoronidase. Variable enzymatic reactions are seen for N- acetyl-β- glucosaminidase, ornithine decarboxylase, arginine dihydrolase, tyrosin arylamidase and β- glucosidase.

The type strain of Citrobacter cronae is Tue2-1T, which was isolated from a rectal swab of a patient hospitalized at University Hospital Tuebingen, Tuebingen, Germany. The G+C DNA content of the type strain is 52.2 mol%.

respectively. The culture certificate accession numbers are CCUG 73860 from the CCUG, Göteborg, Sweden, and DSM 110040 from the DSMZ, Braunschweig, Germany.

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

Ethical statement
The study was conducted in accordance with the local ethics committee from the medical faculty of the university clinics at Tübingen, Germany (407/2013R).

References
1. Arens S, Verbiest L. Differentiation and susceptibility of Citrobacter isolates from patients in a university hospital. Clin Microbiol Infect 1997;3:53–57.
2. Ribeiro TG, Clermont D, Branquinho R, Machado E, Peixe L et al. Citrobacter europeaus sp. nov., isolated from water and human faecal samples. Int J Syst Evol Microbiol 2017;67:170–173.
3. Borenstein D, Schauer DB. The Genus Citrobacter. In: Dworkin M, Falkow S, Rosenberg E, Schleifer K-H, Stackebrandt E (editors). The Prokaryotes: Volume 6: Proteobacteria: Gamma Subclass. New York, NY: Springer New York; 2006. pp. 90–98.
4. Lipsky BA, Hook EW, Smith AA, Plorde JJ. Citrobacter infections in humans: experience at the Seattle Veterans administration medical center and a review of the literature. Rev Infect Dis 1980;2:746–760.
5. Hodges GR, Degener CE, Barnes WG. Clinical significance of citrobacter isolates. Am J Clin Pathol 1978;70:37–40.
6. Vaz Marecos C, Ferreira M, Ferreira MM, Barroso MR, Sepsis BMR. Sepsis, meningitis and cerebral abscesses caused by Citrobacter koseri. BMJ Case Rep 2012;2012:bcr1020114941.
7. Samonis G, Karageorgopoulos DE, Kotteridis DP, Matthaiou DK, Sidiroplou V et al. Citrobacter infections in a general hospital: characteristics and outcomes. Eur J Clin Microbiol Infect Dis 2009;28:61–68.
8. Ribeiro CD, Davis P, Jones DM. Citrobacter koseri meningitis in a special care baby unit. J Clin Pathol 1976;29:1096–1098.
9. Porres- Osante N, Sænæ S, Somalo S, Torres C. Characterization of Beta-lactamases in Faecal Enterobacteraicae recovered from healthy humans in Spain: focusing on AmpC Polymorphisms. Microb Ecol 2015;70:132–140.
10. Peter S, Wolz C, Kaase M, Marschal M, Schultz B et al. Emergence of Citrobacter freundii carrying IMP-8 metallo-β-lactamase in Germany. New Microbes New Infect 2014;2:42–45.
11. Lavigne J-P, Defez C, Bouziges N, Mahamat A, Sotto A. Clinical and molecular epidemiology of multidrug-resistant Citrobacter spp. infections in a French university hospital. Eur J Clin Microbiol Infect Dis 2007;26:439–441.
12. Euzéby JP. List of bacterial names with standing in nomenclature: a folder available on the Internet. Int J Syst Bacteriol 1997;47:590–592.
13. Peter S, Bezdan D, Oberheftinger P, Vogel W, Dörfel D et al. Whole-Genome sequencing enabling the detection of a colistin-resistant hypermutating Citrobacter werkmanii strain harbouring a novel metallo-β-lactamase VIM-48. Int J Antimicrob Agents 2018;51:867–874.
14. Liese J, Schüle L, Oberhettinger P, Tschörner L, Nguyen T et al. Expansion of vancomycin-Resistant Enterococcus faecium in an academic tertiary Hospital in Southwest Germany: a large-scale whole-genome-based outbreak Investigation. Antimicrob Agents Chemother 2019:63.

15. Nurk S, Bankevich A, Antipov D, Gurevich AA, Korobeynikov A et al. Assembling single-cell genomes and mini-metagenomes from chimeric MDA products. J Comput Biol 2013:20:714–737.

16. Darling AE, Mau B, Perna NT. progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. PLoS One 2010;5:e11147.

17. 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–W21).

18. Yoon S-H, Ha S-M, Kwon S, Lim J, Kim Y et al. Introducing EzBioCloud: a taxonomically United database of 16S rRNA gene sequences and whole-genome assemblies. Int J Syst Evol Microbiol 2017:67:1613–1617.

19. Clermont D, Motreff L, Passet V, Fernandez J-C, Bizet C et al. Multilocus sequence analysis of the genus Citrobacter and description of Citrobacter pasteurii sp. nov. Int J Syst Evol Microbiol 2015:65:1486–1490.

20. Ribeiro TG, Gonçalves BR, da Silva MS, Novais Ângela, Machado E et al. Citrobacter portucalensis sp. nov., isolated from an aquatic sample. Int J Syst Evol Microbiol 2017:67:3513–3517.

21. Stamatakis A. RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. Bioinformatics 2014;30:1312–1313.

22. Richter M, Rosselló-Móra R. Shifting the genomic gold standard for the prokaryotic species definition. Proc Natl Acad Sci U S A 2009;106:19126–19131.

23. 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.

24. Meier-Kolthoff JP, Auch AF, Klenk H-P, Göker M. Genome sequence-based species delimitation with confidence intervals and improved distance functions. BMC Bioinformatics 2013:14:60.

25. Larsen MV, Cosentino S, Rasmussen S, Friis C, Hasman H et al. Multilocus sequence typing of total-genome-sequenced bacteria. J Clin Microbiol 2012:50:1355–1361.

26. Warren JR, Farmer JJ, Dewhirst FE, Birkhead K, Zembofer T et al. Outbreak of nosocomial infections due to extended-spectrum beta-lactamase-producing strains of enteric group 137, a new member of the family Enterobacteriaceae closely related to Citrobacter farmeri and Citrobacter amalonaticus. J Clin Microbiol 2000;38:3946–3952.

27. Naum M, Brown EW, Mason-Gamer RJ. Is 16S rDNA a reliable phylogenetic marker to characterize relationships below the family level in the enterobacteriaceae? J Mol Evol 2008;66:630–642.

28. Goris J, Konstantinidis KT, Klappenbach JA, Coenye T, Vandamme P et al. DNA-DNA hybridization values and their relationship to whole-genome sequence similarities. Int J Syst Evol Microbiol 2007:57:81–91.

29. Chun J, Oren A, Ventosa A, Christensen H, Arahal DR et al. Proposed minimal standards for the use of genome data for the taxonomy of prokaryotes. Int J Syst Evol Microbiol 2018;68:461–466.

30. Brenner DJ, Grimont PA, Steigerwalt AG, Fanning GR, Ageron E et al. Classification of citrobacteria by DNA hybridization: designation of Citrobacter farmeri sp. nov., Citrobacter youngae sp. nov., Citrobacter braakii sp. nov., Citrobacter werkmanii sp. nov., Citrobacter sedlakii sp. nov., and three unnamed Citrobacter genomospecies. Int J Syst Bacteriol 1993;43:645–658.

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