



## Draft Genome Sequences of Three Capnocytophaga canimorsus Strains Isolated from Healthy Canine Oral Cavities

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Here, we present the draft genome sequences of three strains of *Capnocytophaga canimorsus*, each isolated from a different dog's mouth. Genome analysis provided evidence that these organisms may belong to a different nonpathogenic subtype of *C. canimorsus*.

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apnocytophaga canimorsus is a capnophilic gliding Gramnegative bacterium from the Bacteroidetes phylum that is part of the normal flora of a dog's mouth (1, 2). It is not reported to cause infections in dogs, but it causes severe infections in humans who are in contact with dogs (3, 4). It has been estimated by culture-based methods that more than every other dog carries C. canimorsus in its normal oral flora (5). Because of the very specific culture conditions required by C. canimorsus strains, their prevalence has often been underestimated. A recent study using PCR-based methods reported that up to 74% of dogs carry C. canimorsus in their mouths (6). Interestingly, despite such a high prevalence, human infections remain very rare. Considering that in barely half of the reported cases, patients do not show any obvious risk factors (7), it is possible that different strains are not equally virulent and that only a tiny proportion is actually pathogenic for humans.

The three strains CcD38, CcD93, and CcD95 were isolated from canine oral swabs and partially characterized (5). The strains were selected as potential representatives of a subgroup of C. canimorsus, as suggested by 16S rRNA phylogenetics and various phenotyping assays (5, 8). Genomic DNA was extracted using the Genomic-tip 500/G DNA extraction kit (catalog no. 10262; Qiagen), according to the manufacturer's instructions, followed by an additional phenol-chloroform purification step. Sequencing was performed at LGC Genomics, Berlin, Germany, on one Illumina HiSeq 2000 channel and generated between 8.6 and 9.8 million 100-bp single reads per strain. Due to low mapping efficiency on the C. canimorsus 5 reference genome (9), de novo assembly was performed with Velvet with optimized k-mers (10). The sizes of the draft assemblies ranged from 2.56 to 2.74 Mb, with 191 (CcD95), 216 (CcD38), and 293 (CcD93) contigs. Genome annotation and preliminary analyses were performed by the LABGeM, France Génomique (11). The global G+C content (35.56%  $\pm$ 0.07%) differs slightly but significantly from that of clinical isolates of *C. canimorsus* (36.16%  $\pm$  0.08%; *P* < 0.0002). A total of 2,325 to 2,618 coding sequences (CDSs) were identified per genome, giving a core genome of 1,896 orthologous groups. When considering the whole Capnocytophaga pangenome, 290 genes

were conserved and exclusively found in the three genomes presented here. This is similar to the number of Capnocytophaga cynodegmi-specific genes (341) (12) and supports the idea of a speciation event within the C. canimorsus taxon. Besides 247 clusters of unknown function, 16 genes implicated in basal energy metabolism, 10 in polysaccharide synthesis, 11 in integral outer membrane proteins, and 5 in ABC transporters formed predominant functional classes of the species-specific core. In addition, a consensual phylogenetic tree based on 771 individual phylogenies of orthologous proteins conserved among all Capnocytophaga spp. exhibited a very clear discrimination between clinical (clustering together in 677 trees) and canine oral (clustering together in 729 trees) isolates of C. canimorsus, even compared to a different species, such as C. cynodegmi (clustering together in 686 trees). This is again consistent with a potential speciation event within the C. canimorsus taxon.

**Nucleotide sequence accession numbers.** These wholegenome shotgun projects have been deposited in ENA under the accession numbers CDOI00000000 (CcD38), CDOL00000000 (CcD93), and CDOH00000000 (CcD95). The versions described in this paper are the initial versions.

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## REFERENCES

- 1. Brenner DJ, Hollis DG, Fanning GR, Weaver RE. 1989. *Capnocytophaga canimorsus* sp. nov. (formerly CDC group DF-2), a cause of septicemia following dog bite, and *C. cynodegmi* sp. nov., a cause of localized wound infection following dog bite. J Clin Microbiol 27:231–235.
- Shin H, Mally M, Meyer S, Fiechter C, Paroz C, Zaehringer U, Cornelis GR. 2009. Resistance of *Capnocytophaga canimorsus* to killing by human complement and polymorphonuclear leukocytes. Infect Immun 77: 2262–2271. http://dx.doi.org/10.1128/IAI.01324-08.
- 3. Bobo RA, Newton EJ. 1976. A previously undescribed Gram-negative bacillus causing septicemia and meningitis. Am J Clin Pathol 65:564–569.

- 4. Butler T, Weaver RE, Ramani TK, Uyeda CT, Bobo RA, Ryu JS, Kohler RB. 1977. Unidentified Gram-negative rod infection. A new disease of man. Ann Intern Med 86:1–5. http://dx.doi.org/10.7326/0003-4819-86-1-1.
- Mally M, Paroz C, Shin H, Meyer S, Soussoula LV, Schmiediger U, Saillen-Paroz C, Cornelis GR. 2009. Prevalence of *Capnocytophaga canimorsus* in dogs and occurrence of potential virulence factors. Microbes Infect 11:509–514. http://dx.doi.org/10.1016/j.micinf.2009.02.005.
- 6. Suzuki M, Kimura M, Imaoka K, Yamada A. 2010. Prevalence of *Capnocytophaga canimorsus* and *Capnocytophaga cynodegmi* in dogs and cats determined by using a newly established species-specific PCR. Vet Microbiol 144:172–176. http://dx.doi.org/10.1016/j.vetmic.2010.01.001.
- Lion C, Escande F, Burdin JC. 1996. Capnocytophaga canimorsus infections in human: review of the literature and cases report. Eur J Epidemiol 12:521–533. http://dx.doi.org/10.1007/BF00144007.
- Manfredi P, Lauber F, Renzi F, Hack K, Hess E, Cornelis GR. 2015. New iron acquisition system in *Bacteroidetes*. Infect Immun 83:300–310. http://dx.doi.org/10.1128/IAI.02042-14.

- Manfredi P, Pagni M, Cornelis GR. 2011. Complete genome sequence of the dog commensal and human pathogen *Capnocytophaga canimorsus* strain 5. J Bacteriol 193:5558–5559. http://dx.doi.org/10.1128/JB.05853 -11.
- Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. Genome Res 18:821–829. http:// dx.doi.org/10.1101/gr.074492.107.
- 11. Vallenet D, Belda E, Calteau A, Cruveiller S, Engelen S, Lajus A, Le Fèvre F, Longin C, Mornico D, Roche D, Rouy Z, Salvignol G, Scarpelli C, Thil Smith AA, Weiman M, Médigue C. 2013. MicroScope—an integrated microbial resource for the curation and comparative analysis of genomic and metabolic data. Nucleic Acids Res 41:D636–D647. http:// dx.doi.org/10.1093/nar/gks1194.
- 12 Manfredi P, Renzi F, Cornelis GR. 2015. Draft genome sequences of three *Capnocytophaga cynodegmi* strains isolated from the oral cavity of healthy dogs. Genome Announc 3(3):e00200-15. http://dx.doi.org/10.1128/ genomeA.00200-15.