Putative Novel Species of Genus Capnocytophaga, Capnocytophaga stomatis Bacteremia in a Patient with Multiple Myeloma after Direct Contact with a Cat

Koh Shinohara¹², Yasuhiro Tsuchido²¹, Michio Suzuki⁴, Kojiro Yamamoto⁵, Yasutaro Okuzawa⁶, Koichi Imaoka⁴ and Tsunehiro Shimizu¹

Abstract:
Capnocytophaga species are among the typical zoonotic pathogens causing infections following direct contact with animals. Recently, a putative novel species of zoonotic Capnocytophaga, Capnocytophaga stomatis, was reported. We herein report the first case of bacteremia caused by C. stomatis. A woman in her 80s with multiple myeloma who was receiving bortezomib and dexamethasone therapy was admitted to our hospital with a 2-day history of a fever and right calf redness. She was often licked by her cat. On a blood culture, thin, Gram-negative rods were detected, which were identified as C. stomatis by whole-genome sequencing. The patient was successfully treated with ampicillin-sulbactam treatment. Our case highlights the pathogenic potential of the putative novel Capnocytophaga, C. stomatis, in immunocompromised hosts.

Key words: Capnocytophaga stomatis, cellulitis, multiple myeloma, bacteremia, whole genome sequencing

(Intern Med 61: 2233-2237, 2022)
(DOI: 10.2169/internalmedicine.7947-21)

Introduction

Capnocytophaga species are fermentative bacteria that appear as slender, Gram-negative bacilli and are divided into two distinct groups. Capnocytophaga ochracea, C. sputigena, C. haemolytica, C. granulosa, C. leadbetteri, C. gingivalis, and C. periodontitidis are among the groups associated with the human oral cavity, while C. canimorsus, C. canis, C. cynodegmi, and C. felis are zoonotic (1-3). Recently, Zangenah et al. reported a putative novel species of the genus Capnocytophaga, Capnocytophaga stomatis, using whole-genome sequencing (4). C. stomatis has been isolated in humans who developed wound infections following animal bites and is genetically similar to C. cynodegmi. However, clinical information concerning C. stomatis is lacking, and its pathogenic potential in humans is unclear.

We herein report the first case of C. stomatis bacteremia, identified using whole-genome sequencing.

Case Report

A woman in her 80s presented to our hospital with a 2-day fever history and right calf redness. Two years earlier, she had been diagnosed with asymptomatic multiple myeloma. The patient had experienced worsening bilateral leg edema over the past six months. Furthermore, a urinalysis revealed proteinuria, consistent with nephrotic syndrome. A renal biopsy confirmed amyloid light-chain amyloidosis. Bortezomib and dexamethasone were administered four weeks before the presentation. The patient kept a cat in her house, which often licked and scratched her calves. In addition to bilateral leg edema, redness from the right ankle to the distal half of the calf was observed. Vital signs showed a low-grade fever (37.1°C) and mild tachycardia (96 beats per minute).

¹Department of Infectious Diseases, Kyoto City Hospital, Japan. ²Department of Clinical Laboratory Medicine, Kyoto University Graduate School of Medicine, Japan. ³Department of Infectious Diseases, University Hospital, Kyoto Prefectural University of Medicine, Japan. ⁴Department of Veterinary Science, National Institute of Infectious Diseases, Japan. ⁵Department of Nephrology, Kyoto City Hospital, Japan and ⁶Department of Dermatology, Kyoto City Hospital, Japan

Received: May 16, 2021; Accepted: November 17, 2021; Advance Publication by J-STAGE: March 12, 2022

Correspondence to Dr. Koh Shinohara, shinoharakoh@kuhp.kyoto-u.ac.jp
A laboratory examination on admission revealed unremarkable findings concerning the white blood cell count (6,570/μL) and serum C-reactive protein level (0.09 mg/dL). The results of laboratory tests are summarized in Table 1.

**Table 1. Results of the Laboratory Examination on Admission.**

| Variables                      | Reference range, adult | Variables                      | Reference range, adult |
|--------------------------------|------------------------|--------------------------------|------------------------|
| White blood cell count (μL)    | 6,570                  | 3,500-8,500                     |                        |
| Red blood cell count (×10^12/μL)| 339                    | 380-490                         | 391                    | 120-230                |
| Hemoglobin (g/dL)              | 11.8                   | 11.5-15.0                       | 19.7                   | 0.3-1.1                |
| Hematocrit (%)                 | 34.1                   | 34-45                           | 48.5                   | 8-21                   |
| Platelet count (×10^12/μL)     | 800                    | 1,300-3,500                     | 132                    | 135-147                |
| Total protein (g/dL)           | 4.8                    | 6.7-8.3                         | 5.3                    | 3.3-4.8                |
| Albumin (g/dL)                 | 1.8                    | 3.9-4.9                         | 104                    | 98-109                 |
| Total bilirubin (mg/dL)        | 0.6                    | 0.2-1.2                         | 8.1                    | 8.2-10.2               |
| Aspartate aminotransferase (U/L)| 31                     | 0-35                            | 0.09                   | 0-0.3                  |
| Alanine aminotransferase (U/L) | 18                     | 0-30                            | 0.09                   | 0-0.3                  |

The ANI represents the average nucleotide identity of all orthologous genes shared between any two genomes and offers robust resolution between strains of the same or closely related species. In general, an ANI value 95% is equivalent to DDH value 70%, which is regarded as the threshold for differentiation of the species. The ANI value between our isolate (HP26001) and the *C. stomatis* strain H2177 was 97.2%, whereas the values between our isolate and DSM 107251 (a type strain of *C. felis*) and between our isolate and ATCC 49044 (a type strain of *C. cynodegmi*) were 89.2% and 84.1%, respectively. A whole-genome-based phylogenetic analysis of the isolated genome was performed with the Type (Strain) Genome Server (https://tygs.dsmz.de) to confirm close phylogenetic relationships between our isolate and the H2177 strain (Fig. 2). These results suggested that our isolate and H2177 were genetically different from *C. cynodegmi*. The isolate did not exhibit hemolytic activity on sheep blood agar plates (Fig. 3). The isolates were also re-analyzed using MALDI-TOF MS 6 times, which generated a score between 1.47-1.68 for *C. cynodegmi*. An E-test strip (bioMérieux, Marcy-L’Etoile, France) and the disk diffusion test were used to determine antimicrobial susceptibility following incubation on sheep blood agar plates (5). The isolates were also resistant to ampicillin-sulbactam (3 g every 12 h) and cephalosporins (6), and the patient was discharged on admission day 13.

However, she was readmitted on day 14 because of skin erosion and a fever caused by amoxicillin-clavulanic acid, and ceftriaxone was administered instead. The 14-day therapy after the initiation of ampicillin-sulbactam was completed, and no relapse was observed thereafter. The clinical course is shown in Fig. 1.

**Discussion**

*C. stomatis* is a putative novel species of zoonotic *Capnocytophaga*, described by Zangenah et al. (4). It was found to be pathogenic in humans when isolated from an infected wound caused by a dog bite. However, no severe disease cases, including bacteremia caused by *C. stomatis*, have been reported. Our case revealed the pathogenic potential of
Figure 1. The clinical course of this case. CEZ: cefazolin, ABPC/SBT: ampicillin/sulbactam, AMPC/CVA: amoxicillin/clavuate acid, CTRX: ceftriaxone, WBC: white blood cell count, CRP: C-reactive protein, Cre: serum creatinine

Figure 2. Tree inferred with FastME 2.1.6.1 from GBDP distances calculated from whole-genome sequences of the “Capnocytophaga stomatis” strain HP26001 and related species. The tree was rooted at the midpoint using Pasteurella multocida as an outgroup sequence. The branch lengths are scaled in terms of the GBDP distance formula d5.

Capnocytophaga stomatis HP26001
Capnocytophaga stomatis H2177
Capnocytophaga felis DSM 107251^T
Capnocytophaga canimorsus ATCC 35979^T
Capnocytophaga cynodegmi ATCC 49044^T
Capnocytophaga canis LMG 29146^T
Capnocytophaga haemolytica CCUG 32990^T
Capnocytophaga ochracea DSM 7271^T
Capnocytophaga gingivalis ATCC 33624^T
Capnocytophaga spuligena NCTC 11653^T
Capnocytophaga periodontalis DSM 34126^T
Capnocytophaga granulosa NCTC 12948^T
Capnocytophaga leadbetteri DSM 22902^T
Pasteurella multocida ATCC 43137^T

C. stomatis to cause severe infection in immunocompromised hosts.

Most human infections with Capnocytophaga species are caused by C. canimorsus, and documented cases of bacteri-
nia of non-carnimorsus Capnocytophaga are limited. To our knowledge, only two cases of C. cynodegmi (9, 10) and four cases of C. canis (7, 11-14) have been reported (Table 3). Most cases of non-carnimorsus Capnocytophaga bacteremia occurred in immunocompromised patients, especially anatomical asplenic patients (7, 10, 13, 14) or those with a splenic function reduction due to liver diseases (7, 11-13). In our case, humoral immune deficiency due to multiple myeloma and nephrotic syndrome, combined with cellular immune deficiency due to steroid and bortezomib use, appeared to have caused bacteremia. In addition, all non-carnimorsus Capnocytophaga bacteremia cases, including our case, were associated with direct contact with cats or dogs, not limited to bites (7, 9-14). Physicians should carefully identify patient’s animal exposure history when managing cases of skin and soft tissue infections.

Our isolate’s MALDI-TOF MS log score was below the cut-off level of 1.7. Zangenah et al. reported the usefulness of MALDI-TOF analysis for C. canimorsus or C. cynodegmi identification (15); however, among the strains identified as C. cynodegmi, three C. stomatis and one C. canis strains were included, which generated log scores below 1.7 (4, 15). Our isolate also exhibited log scores of 1.47-1.70. Based on these results, a MALDI-TOF analysis may contribute to identifying C. stomatis and C. cynodegmi. Zangenah et al. described the colony morphologies of C. stomatis on blood agar plates. The colonies of C. stomatis were reportedly flat, larger than those of C. cynodegmi, and formed transparent/greyish colonies similar to those of C. canimorsus, which were clearly distinct from those of other zoonotic Capnocytophaga sp. In addition, the isolates of C. stomatis exhibited beta hemolytic activity on blood agar. However, our isolate showed gliding motility and no beta hemolytic activity, suggesting that the colony morphologies and hemolytic activity of C. stomatis isolates are not uniform, and attention should be paid to the identification of Capnocytophaga species by phenotypic features.

Suitable antimicrobial agents for C. stomatis have not yet been determined. Historically, zoonotic Capnocytophaga species, mainly C. canimorsus, have been broadly susceptible to most antimicrobial agents, and penicillin or penicillin/beta-lactamase inhibitor combination is recommended as first-line therapy. However, recent reports have revealed that

![Figure 3. Colony morphology of the isolate on sheep blood agar, 48 h culture, 37 °C, 5% CO₂ atmosphere.](image)

| Table 2. The Results of Antibiotic Susceptibility Test of the Isolate. |
|-----------------|-----------------|------------|
| **Antimicrobial agents** | **Disk (mm)** | **E-test** |
| Penicillin G | 27 | 0.125 |
| Amoxicillin/clavuate acid | 25 | 0.094 |
| Cefazolin | 18 | - |
| Ceftriaxone | 34 | 0.032 |
| Imipenem | 36 | 0.25 |
| Gentamycin | - | >256 |
| Minocycline | 39 | 0.047 |
| Ciprofloxacin | 23 | 0.75 |
| Azithromycin | 28 | - |
| Clindamycin | 36 | - |

| Table 3. Literature Review of the Cases with Non-carnimorsus Zoonotic Capnocytophaga Species Bacteremia. |
|-----------------------|----------|-------------|-----------------|---------------------|-----------------|-----------------|-----------------|-----------------|
| **Case No.** | **Organism** | **Age** | **Sex** | **Underlying illness** | **Animal exposure** | **Source of isolation** | **Clinical manifestation** | **Outcome** |
| 1 [9] | C. cynodegmi | 59 | Male | Diabetes mellitus | Dog bite | Blood, BALF, sputum, pus | Cellulitis, sepsis, pneumonia | Recovered |
| 2 [10] | C. cynodegmi | 72 | Female | Post-splenectomy | Dog bite | Blood, CSF | Septic shock, meningitis | Died |
| 3 [11, 14] | C. canis | 49 | Male | Chronic alcoholic consumption | Cat scratch | Blood | Sepsis | Recovered |
| 4 [7, 14] | C. canis | 67 | Female | Idiopathic portal hypertension, post-splenectomy | Cat bite | Blood | Septic shock | Recovered |
| 5 [12, 14] | C. canis | 82 | Female | Liver cancer | Contact with dog | Blood | Sepsis, multiple organ failure | Died |
| 6 [13] | C. canis | 70 | Male | Atrial fibrillation, chronic alcoholic consumption, post-splenectomy | Cat scratch | Blood | Septic shock | Recovered |
| This case | C. stomatis | 81 | Female | Multiple myeloma, nephrotic syndrome | Contact with cat | Blood | Cellulitis | Recovered |

CSF: cerebrospinal fluid, BALF: bronchoalveolar lavage fluid
some strains of *Capnocytophaga* species, mainly human *Capnocytophaga* species and zoonotic *Capnocytophaga* species, harbor beta-lactamases, such as OXA-347, and demonstrate resistance to penicillin, cephalosporins, and imipenem (16). Although our isolate appeared to be susceptible to other beta-lactams, the zone diameter of cefazolin (18 mm) is regarded as resistant when using the CLSI clinical breakpoints for Enterobacterales (8), and the clinical response to cefazolin in this case appeared partially ineffective. More clinical information and antimicrobial susceptibility data of zoonotic *Capnocytophaga* species are needed.

Our case highlights the pathogenic potential of a putative novel *Capnocytophaga*, *C. stomatis*, in immunocompromised hosts. Since 16S rRNA sequencing and species-specific PCR cannot differentiate between *C. cynodegmi* and *C. stomatis*, molecular surveillance using whole-genome sequencing will help deepen our understanding of the clinical and epidemiological features of zoonotic *Capnocytophaga* infections.

The authors state that they have no Conflict of Interest (COI).

**References**

1. Brenner DJ, Hollis DG, Fanning GR, Weaver RE. *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, 1989.
2. Suzuki M, Umeda K, Kimura M, Imaoka K, Morikawa S, Maeda K. *Capnocytophaga felis* sp. nov. isolated from the feline oral cavity. Int J Syst Evol Microbiol 70: 3335-3360, 2020.
3. Zhang Y, Qiao D, Shi W, Wu D, Cai M. *Capnocytophaga periodontitidis* sp. nov., isolated from subgingival plaque of periodontitis patient. Int J Syst Evol Microbiol 71: 2021.
4. Zangenah S, Abbasi N, Andersson AF, Bergman P. Whole genome sequencing identifies a novel species of the genus *Capnocytophaga* isolated from dog and cat bite wounds in humans. Sci Rep 6: 22919, 2016.
5. Suzuki M, Kimura M, Imaoka K, Yamada A. 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, 2010.
6. Ritcher M, Rosselló-Móra R, Glöckner FO, Peplies J. JSpeciesWS: a web server for prokaryotic species circumscription based on pairwise genome comparison. Bioinformatics 32: 929-931, 2015.
7. Taki M, Shimojima Y, Nogami A, et al. Sepsis caused by newly identified *Capnocytophaga canis* following cat bites: C. canis is the third candidate along with *C. canimorsus* and *C. cynodegmi* causing zoonotic infection. Intern Med 57: 273-277, 2018.
8. CLSI. Performance Standards for Antimicrobial Susceptibility Testing. 31st ed. Clinical and Laboratory Standards Institute, Wayne, PA, 2021.
9. Sarma PS, Mohanty S. *Capnocytophaga cynodegmi* cellulitis, bacteremia, and pneumonitis in a diabetic man. J Clin Microbiol 39: 2028-2029, 2001.
10. Khawari AA, Myers JW, Ferguson DA Jr, Moorman JP. Sepsis and meningitis due to *Capnocytophaga cynodegmi* after splenectomy. Clin Infect Dis 40: 1709-1710, 2005.
11. Yamamoto U, Kunita M, Mohri M. Shock following a cat scratch. BMJ Case Rep 2013: bcr2012007892, 2013.
12. Irie Y, Yamatogi T, Sugahara R, Suzuki M, Imaoka K. A case of septicemia caused by putative novel species of genus Capnocytophaga. Nihon Rinsho Biseibutsugaku Zasshi (J Jpn Soc Clin Microbiol) 25: S1 290, 2015 (in Japanese).
13. Donner V, Buzzi M, Lazarevic V, et al. Septic shock caused by *Capnocytophaga canis* after a cat scratch. Eur J Clin Microbiol Infect Dis 39: 1993-1995, 2020.
14. Suzuki M, Imaoka K, Haga Y, et al. Characterization of three strains of *Capnocytophaga canis* isolated from patients with sepsis. Microbiol Immunol 62: 567-573, 2018.
15. Zangenah S, Özenci V, Boräng S, Bergman P. Identification of blood and wound isolates of *C. canimorsus* and *C. cynodegmi* using VITEK2 and MALDI-TOF. Eur J Clin Microbiol 31: 2631-2637, 2012.
16. Zangenah S, Andersson AF, Özenci V, Bergman P. Genomic analysis reveals the presence of a class D beta-lactamase with broad substrate specificity in animal bite associated *Capnocytophaga* species. Eur J Clin Microbiol Dis 36: 657-662, 2017.

The Internal Medicine is an Open Access journal distributed under the Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License. To view the details of this license, please visit (https://creativecommons.org/licenses/by-nc-nd/4.0/).