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Letter to the Editor

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Fatal septic shock and Waterhouse-Friderichsen syndrome caused by serovar B *Capnocytophaga canimorsus* in an immunocompetent patient. Case report and review

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Sir,

Capnocytophaga canimorsus is a fastidious Gram-negative rod that is oral flora in 74% of dogs and 57% of cats [1]. It can be transmitted to humans by exposure to a dog or cat, usually by bites, scratches or licks. According to a recent systematic review, the case fatality rate of *C. canimorsus* infection in immunocompetent patients was 29,7% between 2002 and 2019 [2]. We report a fatal case of Waterhouse-Friederichsen Syndrome (WFS) by *C. canimorsus* in an immunocompetent patient who was microbiologically diagnosed in less than 24 hours.

A 55-year-old woman, with no relevant medical history apart from fibromyalgia, was evaluated at the emergency department for fever (40ºC) since 24h, dyspnea, greenish diarrhea and general worsening. She referred a cranioencephalic trauma against her dog 4 days before with posterior headache and left periorbital inflammation. Physical examination showed hypotension (86/45 mm Hg), generalized lividity and poor peripheral perfusion. Blood analysis revealed: acute kidney failure (creatinine 2.64 mg/dL (0.40-1.1), acute liver failure (total bilirubin 1.9 mg/dL (< 1.2), alanine aminotransferase 301 U/L (5-31)), metabolic acidosis (pH 7.08 (7.35-7.45), HCO_3- 14 mmol/L (21-26)), leukopenia (710 leukocytes/µL), thrombocytopenia (15000 platelets/µL) and disseminated intravascular coagulation (DIC) (D-Dimer 33330 ng/mL (0-500), INR 2.6 (0.9-1.2), APTT 104 s (25-40)), elevated C-reactive protein (343 mg/L (0-5)) and procalcitonin (11.53 ng/mL (< 0.5)). These data were consistent with septic shock with multiorgan failure. Wright-Giemsa stain of peripheral blood revealed extracellular rods as well as erythrocytes aggregates, forming *rouleaux* (Figure 1).

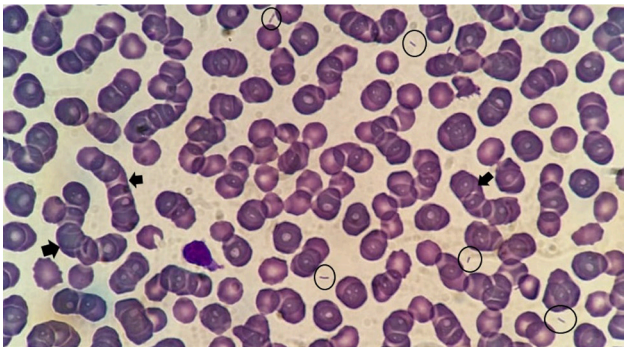

Figure 1 **Pheripheral blood smear (Giemsa stain) of the patient when she arrived at the emergencies room with rods-shaped (rounded) between the erythrocytes. In this blood smear erythrocytes aggregates were seen, forming rouleaux (arrow).**

Two sets of blood cultures were obtained before starting empiric broad-spectrum antibiotic therapy with intravenous piperacillin-tazobactam, meropenem and clindamycin (4/0,5g single dose, 2 g/8 h and 900 mg/8 h, respectively). Despite high-dose intravenous fluid replacement, noradrenaline and bicarbonate infusions, the patient didn´t improve and was transferred to intensive care unit after orotracheal intubation. A body CT scan was conducted, which described signs of hypovolemic shock with thickening of both adrenal glands with crosslinking of the peri-renal fluid, as well as liver with periportal edema and no highlight of the spleen (Figure 2A). Her condition deteriorated rapidly overnight and, in spite of cardiopulmonary resuscitation measures, patient expired 16 hours after admission. Because of multi-organ failure and septic shock of unknown origin, an autopsy was requested.

One of the anaerobic blood culture bottles (BD BACTEC™ Lytic/10 Anaerobic/F) was positive after 12 hours. Gram stain revealed fusiform gram-negative rods. It was subcultured on

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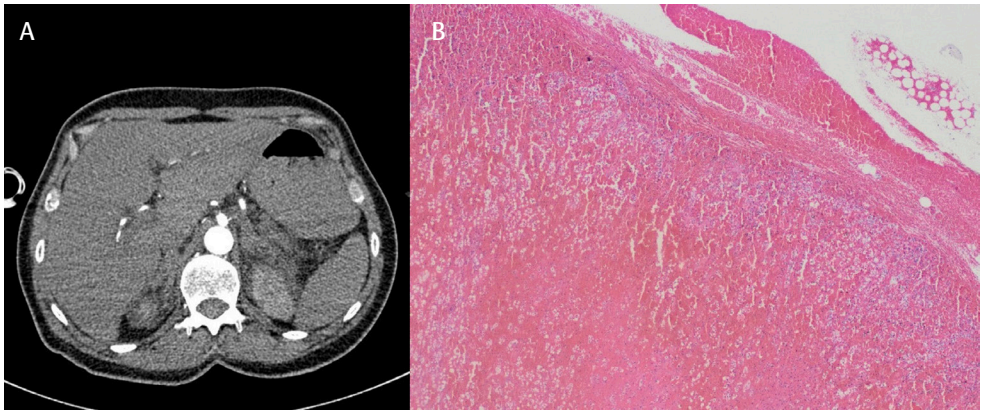

Figure 2 | **A: signs of hypovolemic shock with thickening of both adrenal glands with crosslinking of the peri-renal fluid.**
B: H-E stain suprarenal gland section with hemorrhagic replacement of suprarenal necrotic parenchyma.

chocolate agar, trypticase soy agar with 5% of sheep blood and brucella agar plates. Bottle direct bacterial identification was performed by MALDI-TOF MS (Bruker Daltonics) following this *in-house* procedure: transfer 1 mL from positive bottle, centrifuge for 2 min at 13000 rpm, discard the supernatant, apply the pellet to the polished steel target plate and, once dried, overlaid with 1uL of matrix solution (alfa-cyano-4-hydroxycinnamic acid) before MALDI-TOF MS analysis [3]). *C. canimorsus* was identified with a score of 2.15. After eight days, there was a veil-like growth with greyish colonies in TSA with 5% of sheep blood agar, being identified by MALDI-TOF as *C. canimorsus* with a score of 2.34.

Antibiotic susceptibility testing was performed by Liofilchem® MIC Test Strips on Mueller Hinton agar plates with 5% of sheep blood (Becton Dickinson, Franklin Lakes, NJ, USA) incubated in 5-10% CO_2 atmosphere. Amoxicillin, amoxicillin/clavulanic acid, ceftriaxone, meropenem, clindamycin, tetracycline, piperacillin/tazobactam and trimethoprim-sulfamethoxazole were considered susceptible according to EUCAST 2020 PK/PD guidelines, as well as the antimicrobial MIC breakpoints of HACEK microorganism group established by CLSI (exposed in the "infrequent isolated or fastidious bacteria" guideline) [4,5].

In order to perform capsular typing of our strain, the isolate was submitted to the reference centre "*Research Unit in Biology of Microorganisms*" at the University of Namur (Belgium). *C. canimorsus* identification was confirmed by 16S rRNA sequencing, following Mally M et al. protocol [6]. Capsular typing was also performed by the PCR according to Hess E et al. belonging to serovar B [7].

Autopsy findings showed extensive cutaneous purpura, bilateral adrenal hemorrhage consistent with Waterhouse-Friderichsen Syndrome (Figure 2B), multiple microthrombi in both lungs and kidneys, brain and spleen, corresponding to histological lesions of DIC.

C. canimorsus is a common host of the dog´s oral flora, but the number of human infections remains low, suggesting that other factors are necessary to produce infection. The main risk factors for increased susceptibility to *C. canimorsus* infections are male sex, aged over 50 years, splenectomy, chronic alcohol consumption, tobacco smoking, diabetes mellitus and use of immunosuppressive therapy [8]. However, more than 40% of the patients have no evident risk factor [9]. In fact, this bacteria could lead to severe complications as DIC, meningitis or fulminant sepsis, even in immunocompetent patients [9-19].

In our institution, we have reported until now three cases of *C. canimorsus* invasive infections, one of them being the presented case which was the only one with fatal outcome. All three cases occurred in 2021 in a period of 3 months, but no relationship was found between patients and pets. One of the cases was also a sepsis by *C. canimorsus* serovar B (Table 1), as in the case we describe here [17]. The third patient was an HIV patient with no history of bites who developed a septic arthritis by *C. canimorsus* serovar A [20].

According to previous reports, we review in Table 1 eleven cases of *C. canimorsus* infections in immunocompetent patients. In all cases, patients had close contact with dogs, but only seven reported a bite prior to the episode and eight patients expired despite being treated correctly. The four patients who did not show bites prior to the episode died, confirming that a previous traumatic antecedent such as a bite is not necessary for *C. canimorsus* to produce severe and even fatal infections.

According to Nakayama R et al, only four cases of progressively fatal *C. canimorsus* infection in immunocompetent patients resulting in death within the first day of hospitalization have been reported, being our case the fifth one that shares this characteristic [9,13,14,16]. The common features between our case and previously reported cases were that the patients did not respond to

Table 1 **Review of invasive *C. canimorsus* cases in immunocompetent patients.**

Case (year of publication and reference)	Gender and age (years)	Pet contact/ bite	Diagnostic method	Clinical picture and complications	Antibiotic treatment	Outcome
2015 [9]	M/53	Yes/Yes	16S rRNA sequencing	Septic shock with multiorgan failure and DIC. WFS	Vancomycin 1,750 mg and ceftriaxone 2 g	Death*
2010 [10]	M/50	Yes/Yes	16S rRNA sequencing	Meningitis	Ampicillin (2g/4h) and ceftriaxone (2g/12h)	Alive
2019 [11]	W/63	Yes (licks)/No	Blood cultures (no additional information)	Sepsis and purpura fulminans. Multiorgan failure.	Clarithromycin and piperacillin/ tazobactam (no dosage information)	Death
2019 [12]	W/62	Yes/Yes	16S rARN sequencing	Sepsis with DIC and thrombotic microangiopathy	Piperacillin/tazobactam (4,5 g/8 h)	Alive
2022 [13]	M/68	Yes/Yes	Direct MALDI-TOF MS from blood cultures	Septic shock with multiorgan failure and DIC.	Meropenem and vancomycin (no dosage information)	Death*
2000 [14]	W/47	Yes/No	Phenotypic identification from cultures	Sepsis, purpura fulminans, DIC and WFS.	No data	Death*
2018 [15]	M/54	Yes/No	MALDI-TOF MS from colony	Septic shock, DIC and purpura.	Oral ofloxacin, ceftriaxone (2 g) and metronidazole (500 mg)	Death
2018 [16]	W/80	Yes/Yes	Blood cultures (no additional information)	Purpura fulminans	Piperacillin/tazobactam (4,5 g/6 h)	Death*
2022 [17]	M/50	Yes/Yes	Direct MALDI-TOF MS from blood cultures. Serovar B.	Sepsis	Meropenem (1 g/8 h)	Alive
2008 [18]	W/36	Yes/No	Phenotypic identification from cultures	Sepsis with multiorgan failure, DIC and WFS.	Vancomycin, ceftriaxone, metronidazole and ciprofloxacin (no dosage information)	Death
1996 [19]	M/59	Yes/Yes	Subculture from blood cultures	Sepsis with multiorgan failure, DIC. WFS	Penicillin and gentamicin (no dosage information)	Death

***Death occurred within the first 24 hours of admission. DIC: disseminated intravascular coagulation; WFS: Waterhouse-Friederichsen Syndrome**

antimicrobials, fluid resuscitation, vasopressors, and had a tendency to bleed, such as purpura, which were associated with thrombocytopenia and coagulopathy. Similarly to two previous cases, our patient autopsy results revealed bilateral adrenal hemorrhage and necrosis consistent with WFS [9, 14]. Septic shock with disseminated intravascular coagulation occurs in 13-30 % of cases and mortality is estimated at 60% [15].

C. canimorsus infections may have been underestimated because of the difficulty in isolation of the bacteria, due to the slow and unique growth media requirements [8]. It has also been described an increment of *C. canimorsus* cases mainly due to improved techniques for bacterial identification, as generalization of MALDI-TOF MS [21,22]. Our diagnosis was based on blood culture bottle direct MALDI-TOF MS method as we previously described, which decreases time for etiological diagnosis allowing antibiotic treatment optimization. From the cases we have reviewed (Table 1), no one used this rapid method, except for us in another patient previously reported in our institution.

In severe septic cases, when a high-grade bacteremia is developed, multiple rods can be visible in Wright-Giemsa stain of peripheral blood smears, both inside polymorphonuclear leukocytes and extracellularly, as in our patient. This cytological element should not be overlooked in septic shock of unknown origin, because it can accelerate the suspicion of bacterial sepsis and the rapid prescription of an empirical antibiotic treatment. In our case, the blood smear was performed due to an abnormal decreased amount of platelets, justified by the DIC (Figure 1), a finding previously seen in the literature. In this blood smear, we can see only erythrocytes aggregates, forming rouleaux. Rouleaux formation occurs when the plasma protein concentration is high and it may be seen in patients with infection, multiple myeloma or diabetes mellitus, among others [9,14,23].

WFS is described as a rapid dissemination of a pathogen in blood that can lead to septic shock, DIC, cutaneous purpura and bleeding into the adrenal glands. A rapidly evolving adrenal insufficiency is developed being associated with bilateral adrenal gland necrosis, a condition typically

observed in meningococcal infection. However, this syndrome can be seen in other capsulated bacteria like *Streptococcus pneumoniae* or *Haemophilus influenza,* like in our capsulated *C. canimorsus* serovar B case [24,25]. If this syndrome is suspected, a CT scan must be performed as soon as possible. A delayed diagnosis and antibiotic treatment increases the probability of a fatal outcome. Treatment for WFS is based on glucocorticoids, volume resuscitation, appropriate antibiotic coverage, vasssopresors to ensure end-organ perfusion and other supportive care. Despite an early management, the mortality remains high, being consistent with the cases we have reviewed (Table 1, all patients with WFS died) [26,27]

Clinicians should be aware that when a dog or cat bites, scratches or licks a patient, even in immunocompetents, *C. canimorsus* must be considered as a potential cause of severe infection with fatal outcome. It must be taken in consideration that *C. canimorsus* has slower growth than other microorganisms requiring special growth conditions. MALDI-TOF MS represents an accurate, rapid and inexpensive tool for this fastidious pathogen detection directly from blood culture bottle, which allowed us the etiological diagnosis in less than 24 hours since patient admission. Incidence of *C. canimorsus* infection is expected to increase over time because of new reliable diagnostic methods, more susceptible population and rising popularity of pet ownership.

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None to declare

CONFLICT OF INTEREST

The authors declare no conflict of interest.

ETHICS APPROVAL

The Basurto Universitiy Hospital ethics committee approve the use of information in this brief case (inner code: 81.21 CEICHUB)

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