www.ijcem.com /ISSN:1940-5901/IJCEM0026570

Case Report
Spontaneous bacterial peritonitis caused by oxidase negative Campylobacter fetus subsp. testudinum isolated from the patient with decompensated liver cirrhosis: a case report and literatures review

Pengling Li1, Min Wang1, Xianping Li1, Feihu Hu2, Huan Song1, Yixin Xie1, Min Yang1, Aiguo Tang1

Departments of 1Laboratory Medicine, 2Psychiatry, The Second Xiangya Hospital, Central South University, Changsha, Hunan 410011, China
Received February 24, 2016; Accepted June 4, 2016; Epub July 15, 2016; Published July 30, 2016

Abstract: Campylobacter is a spiral microaerophilic gram-negative bacillus, which is widespread in animals and causes opportunistic infection in immunocompromised humans. Spontaneous bacterial peritonitis (SBP) is the most frequent and life-threatening infection in patients with liver cirrhosis requiring prompt recognition and treatment. The common pathogens of SBP are Escherichia coli (E. coli), Klebsiella species and streptococcus. SBP caused by Campylobacter fetus is rare in clinic, especially in those with septicemia simultaneously. Only five cases have been reported in the literature so far, yet no one was reported in China. Here, we report a case of Campylobacte fetus SBP in a 46-year-old man with decompensated liver cirrhosis and the pathogen was identified by 16S rRNA sequencing in China. It is well known that oxidase test is positive in Campylobacter. However, the organism is negative in this case, which has never been reported before.

Keywords: Campylobacter fetus, spontaneous bacterial peritonitis (SBP), oxidase

Introduction
SBP is the most frequent and life-threatening infection in patients with liver cirrhosis requiring prompt recognition and treatment [2]. The diagnosis of SBP is based on a polymorphonuclear cell count greater than or equal to 250 cells/μL in ascitic fluid without an intra-abdominal source of infection [1, 2]. Culture is the recommended diagnostic procedure. However, the negative rate of ascitic fluid classical bacterial culture is approximately up to 65% [3].

Campylobacter is a spiral microaerophilic gram-negative bacillus, which causes opportunistic infection in immunocompromised humans, such as gastroenteritis, abortion, bacteremia, endocarditis, abscess and meningitis [4]. SBP caused by Campylobacter fetus is rare in clinic, especially in those with septicemia simultaneously. Only five cases have been reported in the literature, yet no one was reported in China. In this article, we present a case of SBP caused by C. fetus subsp. testudinum.

Case report
A 46-year-old man with decompensated cirrhosis, suffering from diarrhea for two weeks and fever for one week, was admitted to our hospital in August 2015. The patient had been admitted to our hospital for SBP three months earlier.

On admission, his vital signs were as follows: blood pressure, 120/85 mmHg; heart rate, 105 bpm; respiratory rate, 22 breaths/min and body temperature, 39°C. Laboratory testing is showed in the Table 1. These laboratory data showed signs of high-grade inflammation and abnormal liver function.

On examination, the abdomen was distended with the tenderness and rebound tenderness, and shifting dullness was positive. Abdominal ultrasound revealed massive ascites and gall-bladder-wall hydrops (Figure 1). Laboratory findings of the ascites revealed a WBC count of 20500×10⁶/L (85% neutrophils). The patient
SBP caused by oxidase negative Campylobacter fetus subsp. testudinum

had been given a empirical treatment with meropenem 1.0 g every 8 hours, linezolid 600 mg every 12 hours and intraperitoneal injection with amikacin 0.4 g for three times, based on a presumptive diagnosis of SBP before culture results were available. On day 4, ascites culture was negative. On day 5, four sets of blood cultures from different parts revealed Campylobacter-like gram-negative rods by Gram staining (Figure 2). However, the kind of bacteria was not confirmed by BD Phoenix 100 automated microbial identification system. A series of traditional manual biochemical identification reactions were conducted, and except for the negative oxidase test, most of the results, such as the positive catalase test, resistance to nalidixic acid and susceptibility to cephalothin were accorded with Campylobacter. The routine typing methods were not able to identify the microorganism causing the infection, so it was further analyzed by molecular methods and whole-genome sequencing. Then the genomic DNA was extracted, and polymerase chain reaction (PCR) for 16S rRNA was performed. On day 13, the isolate was first identified as Campylobacter fetus subsp. testudinum in our hospital, as determined by 16S rRNA gene sequencing with universal primers (forward, 5'-AGAGTTTGATCCTGGCTC-3'; reverse, 5'-ACGGTTACCTTGT-TACGACTT-3'). BLAST result showed the amplified sequences is 99% identity compared with the 16S rRNA sequences of Campylobacter fetus subsp. testudinum strain logged in Genbank.

Fever of the patient subsided within 72 hours and abdominal pain disappeared with meropenem treatment. On day 5, we switched to using cefoselis instead of linezolid and meropenem according the blood culture result. On day 7, meropenem was used again for the abdominal pain and increasing ascites. On day 12, the analysis of the ascites revealed a WBC count of 405×10^6/L (41% neutrophils). On day 18, the treatment was changed to moxifloxacin 300 mg administered every 24 hours for 10 days as his clinical symptoms were improved. The sepsicaemia and spontaneous peritonitis were successfully treated with meropenem for 16 days and moxifloxacin for 10 days.

Discussion

The species C. fetus is a renowned pathogen worldwide that produces considerable economic losses, mainly in bovine and ovine productive chains for being a primary cause of ruminant infertility and abortions [5]. Campylobacter fetus can be an opportunistic pathogen of SBP among immunocompromised patients, which is rare in clinic, especially in those with sepsicaemia simultaneously. A MEDLINE search using “spontaneous peritonitis” and “Campylobacter fetus” as keywords (MeSH) only identified five cases, yet no one was reported in China. The first case of SBP caused by Campylobacter
SBP caused by oxidase negative *Campylobacter fetus subsp. testudinum*

---

**Figure 2.** Gram staining of the blood culture isolate showed spiral gram-negative rods.

*Campylobacter fetus* was reported by Targan SR [6] in 1976. In the presented case, the patient has a high risk for *C. fetus* infection as he had decompensated liver cirrhosis which led to the low immunity. Among the *Campylobacter* species, *C. jejuni* and *C. coli* are the prototypes for enteric infection, and *C. fetus* is the prototype for extraintestinal infection and only this subspecies can cause septicaemia [6]. However, in our case, the patient infected with *C. fetus* had two weeks diarrhea with watery stool.

*Campylobacter* species are the top three bacteria implicated in food-borne disease [7]. Some reports have described *C. fetus* infection cases are connected with raw meat, such as insufficiently cooked chicken, beef, sheep meat or liver [8]. Therefore, immunocompromised patients should avoid eating undercooked food.

*Campylobacter* is, to the best of our knowledge, positive for oxidase [9]. However, the organism we isolated was negative which has never been reported before. Bacteria can evolve and adapt faster to the changing environment, or some bacteria characteristics are not be found, which can mislead our judgment. In the case, although we primarily considered it to be *Campylobacter*, the oxidase test was negative. Ultimately the isolate was identified as *Campylobacter fetus subsp. testudinum* by 16S rRNA gene sequencing. So far few laboratories use molecular methods to identify strains. DNA-based methods are highly sensitive and can overcome some of the limitations of classical microbiological methods. Therefore, we encourage laboratories to use this method to identify some rare bacteria rapidly to guide the clinical medication if routine typing methods are not able to identify the causative agents of infection.

Ascitic fluid culture is important to guide antibiotic therapy and should be performed in all SBP patients. However, twice ascites cultures were negative by inoculation ascites in blood plate directly. Inoculation of ascites into blood culture bottles at the patient's bedside could increase the sensitivity of diagnosis [2]. Infection with *Campylobacter fetus* should be considered when there are spiral microaerophilic gram-negative bacillus in the ascites or blood, in cirrhosis patients with hyperpyrexia and diarrhea, and empirical antibiotic therapy must be initiated immediately since culture results are not available at this time point.

According to the available guidelines for empirical antibiotic treatment of SBP, a third-generation cephalosporin should be initiated immediately after diagnosis of SBP. However, the use of cefoselis was an unsuccessful treatment in our case. The outcome of meropenem treatment in our case was favorable as reported by Tremblay C [10]. Fluoroquinolones are recommended for the treatment of systemic campylobacteriosis [11], and in our case, moxifloxacin was effective. Comprehensive analysis of relevant cases reports, we think the effective treatment of bloodstream infection caused by *C. fetus* was combined application of fluoroquinolone and carbapenem which should be used as early as possible. Besides the therapeutic paracentesis, peritoneal lavage and abdominal antibiotic administration such as amikacin should be performed instantly as well if SBP was co-infected.

Although many cases of *C. fetus* bacteremia have been reported, the case of *C. fetus* SBP and septicemia is rare in China. Decompensated liver cirrhosis with ascitic fluid could lead to hypoproteinemia, which can cause severe infectious diseases. Except for continuous intravenous antimicrobial therapy, the administration of adequate intravenous albumin adjuvant therapy can improve the physical condition and to minimize the risk of severe infection.

To summarize, we report a case of SBP and septicemia caused by *Campylobacter fetus*...
SBP caused by oxidase negative *Campylobacter fetus subsp. testudinum*. Although we primarily consider *Campylobacter* according to the Gram staining, the oxidase test was negative. At last, the isolate was identified by 16S rRNA gene sequencing on day 13, which may delay the illness. Consequently, larger scale studies are needed to determine both diagnostic and treatment guidelines for this condition. Meanwhile, accurate identification remains a challenge in clinical microbiology laboratories and more advanced methods are needed to identify C. fetus.

**Disclosure of conflict of interest**

None.

**Address correspondence to:** Min Wang, Department of Laboratory Medicine, The Second Xiangya Hospital, Central South University, Changsha, Hunan 410011, China. Tel: +86-731-85292142; Fax: +86-731-85292142; E-mail: wangmin0000@aliyun.com

**References**


