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The *Olsenella uli* induced pneumonia: A case report

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Keywords: *Olsenella uli*, pneumonia, human infection

Abstract

Olsenella uli is a Gram-positive bacterium common in the oral cavity or gastrointestinal tract. Here we reported a first case of human pneumonia caused by the *Olsenella uli*. The identification of *Olsenella uli* was based on micromorphology, sequence analysis and mass spectrometry analysis of the bacteria recovered from sputum. Ceftazidime, one of the third generation cephalosporins was used for the anti-infection treatment of the patient. CT results showed a significant improvement of the pulmonary lesion and pleural effusion and recovery from pulmonary infection after 10 days. The mechanism underlying *Olsenella uli* induced pneumonia is unclear, our report suggests a causative role of gingival bacteria in pathogenesis of pneumonia, and the intervention by Ceftazidime may offer a therapeutic strategy for *Olsenella uli* infection.

Background

Pneumonia is the most common infectious cause of mortality and morbidity worldwide. Pneumonia infection results from a complex process where the lower respiratory tract suffers the invasion of an infective microorganism. It is estimated that lower respiratory tract infection caused 2.38 million deaths and an estimated 91.8 million disability-adjusted life-years [1]. Pneumonia can be acquired in the community or acquired in the hospital environment, and can be transmitted by the aspiration of a pathogenic microorganism or by inhalation of a pathogenic microorganism. Bacteria, viruses and fungi are the common pathogenic microorganism of pneumonia and with increasing antimicrobial resistance, pneumonia induced by known bacterium is a threat to human health [2]. In recent years, new bacteria or well-known bacteria such as *Rhodococcus defluvi*, brucellosis which were rarely infected the lung have been reported to cause pneumonia [3, 4].

The genus *Olsenella* is a group of bacteria that are microaerotolerantly (moderately obligately) anaerobic [5]. At the time of writing, the genus comprises six species, *Olsenella uli*, *Olsenella profusa*, *Olsenella umbonata*, *Olsenella scatoligenes*, *Olsenella timonensis* and *Olsenella faecalis* (www.bacterio.net/olsenella.html). *Olsenella uli*, the type species of the genus *Olsenella*, was originally designated as *Lactobacillus uli* in 1991 [6] and moved to a new genus, *Olsenella* with genomic studies in 2001 [7]. *Olsenella uli* is regularly isolated from lesion sites in the human mouth such as gingival and subgingival sites with periodontitis [5, 7]. In 2008, Shane R Durkin et al. isolated *Olsenella uli* from vitrectomy specimen of an Australian man with chronic postoperative endophthalmitis [8]. Additionally, sometimes *Olsenella uli* could be isolated from blood of humans with local oral or gastrointestinal infections [9, 10]. However, there have not been reports about *Olsenella* infection in lung with either culture or gene analysis. With clinical and laboratory work, we found that *Olsenella uli* can infect the lung and induce pneumonia for the first time and this may provide some new ideas for interventions of *Olsenella uli* infection.

Case presentation

A male, 70 years old, farmer, had previous history of hypertension with blood pressure peak at 160/110 mmHg, but had no diabetes, coronary heart disease, chronic lung disease, kidney disease and liver disease. He had a 30 year history of smoking with 15 - 20 cigarettes per day and gave up smoking for 10 years. On May 31 2020, he got a cough with bloody sputum for three times a day without obvious causes, not accompanied by fever, chest pain and distress, difficulty in breathing abdominal pain and diarrhea. After anti-infection and hemostatic treatment in the local hospital, he had no bloody sputum, but got difficulty in breathing, alleviating when lying on the right side, accompanied by fever with heat peak 38.9 °C at June 8, 2020. Without improvement, he was transferred to our hospital on June 10, 2020. The computed tomography (CT) results showed a mass shadow with uneven density, including gas and liquid density shadow in the lower lobe of the right lung, the density shadow of encapsulated liquid and gas, gas-liquid level in the right chest, multiple patchy shadows in both lungs, multiple vesicular translucent shadows in both upper lungs (Fig 1). This indicated bilateral pneumonia, pyogenic necrosis in the right lower lobe of the lung,

encapsulated pyothorax in the right chest and bilateral emphysema. Routine blood test showed white blood cells $41.74 \times 10^9/L$, neutrophil count $36.99 \times 10^9/L$, neutrophil ratio 88.60%, lymphocyte count $2.14 \times 10^9/L$, lymphocyte ratio 5.10%, platelets $533 \times 10^9/L$. Abnormal test results in liver function: Alanine aminotransferase (ALT) 61.3 U/L, Aspartate aminotransferase (AST) 46.5 U/L, Cholinesterase (CHE) 1229 U/L, Total bilirubin (TB) 36.3 $\mu\text{mol/L}$, Direct Bilirubin (DB) 20.2 $\mu\text{mol/L}$, Lactate dehydrogenase (LDH) 404 U/L; Abnormal test results in renal function: Blood urine nitrogen (BUN) 8.71 mmol/L, Creatinine (Cr) 89.0 $\mu\text{mol/L}$; Hypersensitivity C reactive protein (Hs-CRP) 175.35 mg/L. Abnormal test results in coagulation function: Prothrombin time (PT) 16.40, international normalized ratio (PT-INR) 1.40, D-dimer (DD) 1040 ng/mL; sputum anti-acid staining (–), HIV antibody (HIV-Ab) (–). The patient was diagnosed to be pneumonia with pulmonary abscess and underwent CT guided drainage and lung biopsy showed chronic inflammatory cell infiltration and focal alveolar epithelial hyperplasia in the right lung which confirmed the CT results. According to the traditional experiences, biapenem combined with piperacillin and tazobactam, was given to resist infection. At 8th day, the re-examination of chest CT plain showed that the pulmonary lesion aggravated in comparison with the previous symptoms, showing that the treatment was ineffective.

A Gram-positive bacillus was isolated from the sputum culture at 9th day. Sputum samples were inoculated on Columbia blood agar, MacConkey agar medium and chocolate agar medium containing vancomycin separately. Colonies on Columbia blood agar are small needle tip like and translucent (Fig 2) after 48 hours of incubation at 37°C with air plus 5% CO₂. The microorganism was unable to grow on McConkey agar medium and chocolate agar medium containing vancomycin. Cells were analyzed under microscopic after gram staining, and gram-positive, blue purple cells were seen singly, in pairs, in short or long chains, but the size was not uniform (Fig 3). With Brooke mass spectrometer MALDI-TOF, the bacteria were identified as *Olsenella uli* with average score 2.019 ± 0.157 (Fig 4).

For further identification, two single colony on the Columbia blood agar was selected, after genomic DNA extraction, the general primers (7F and 1540R, 27F and 1492R) for 16S rDNA were used for PCR amplification. Following gel electrophoresis, purification, recovery, cloning and sequencing of gene, the sequencing results (GenBank accession numbers MZ220349 and MZ220350) were analyzed by BLAST, and the homology to the *Olsenella uli* sequence in the gene bank was 99%, and the NCBI number referenced was CP002106.1.

As *Olsenella uli* is anaerobic or microaerobic, and sensitive to the third generation cephalosporins [8], so ornidazole combined with ceftazidime was used for anti-infection in the patient's treatment. After 10 days later, chest CT showed significant improvement in the pulmonary lesion and pleural effusion. As there were no clinical symptoms, the patient discharged in the next day. With regular follow-up visit, the patient recovered well from pulmonary infection.

Discussion

In this study, we isolated *Olsenella uli* confirmed by mass analysis and sequence analysis from sputum of a patient with pneumonia accompanied by pulmonary abscess.

Colonies of our sputum *Olsenella uli* on Columbia blood agar are small needle tip like and translucent, having some difference from *Olsenella uli* type strain DSM 7084^T [5] which is semi-translucent or opaque after cultured on FAA and PYG after 7 days. The colonies of *Olsenella uli* from sputum shared more similarity to the *Olsenella uli* strain VPI D76D-27C^T [11]. Culture process and gram staining confirmed *Olsenella uli* as gram-positive, blue purple cells. Similar to *Olsenella uli* DSM 7084^T [5] and VPI D76D-27C^T [11], *Olsenella uli* isolated from the sputum appeared singly, in pairs and in short to very long serpentine chains, and the center part of the cell may swell, but the size was not uniform. Difference in colonies and bacterium appearance of *Olsenella uli* may result from the different disease sites or the different culture condition, as we used Columbia agar and aerobic condition (air plus 5% CO₂, 48h), which may make the *Olsenella uli* appear not typical.

Although there has been significant decline, anaerobic bacteria are still the most common microorganisms of lower respiratory tract [12], which resulted in the aspiration pneumonia firstly, then purulent pneumonia and finally lung abscess or complicated empyema. Anaerobic infection of pneumonia is clinically characterized by fever, cough and stink, purulent sputum and the chest CT shows changes of pneumonia and lung abscess. *Olsenella uli* which is anaerobic or microaerobic has not been reported to be associated with lower respiratory tract infection. In this study, we reported the first pneumonia case induced by *olsenella uli* with similar clinical and laboratory characteristics to anaerobic infection of lung. This may provide new etiology in the pneumonia although further study needed.

Olsenella uli can be detected in the oral cavity or gastrointestinal tract by culture or molecular analysis [5, 7]. Interestingly, several studies have found that microorganisms colonizing the oropharynx, nasopharynx or upper gastrointestinal can be aspirated into the lung and cause aspiration pneumonia [13]. So we speculated this pneumonia case obtained the lung infection by aspirating the *Olsenella uli* of the oral cavity or upper gastrointestinal tract although a large number of further studies needed. There are multiple risk factors for aspiration pneumonia, such as age, dysphagia, reduced consciousness, neurologic disorders, gastroesophageal reflux disease, tube feeding, male sex, smoking, diabetes mellitus [13]. Our reporting pneumonia case has three risk factors, age, male sex and smoking, which confirmed our speculation in a degree with the clinical characteristics such as fever, cough, sputum and imaging diagnosis of pneumonia and pulmonary abscess. Because there's no evidence of *Olsenella uli* in the oral cavity or gastrointestinal tract of this pneumonia patient, the source and mechanism of *Olsenella uli* inducing pneumonia is not sure. However, this study still provide a new etiology for pneumonia especially with complicated pulmonary abscess, and may propose some more ideas for intervention of *Olsenella uli*.

Ethics approval and consent to participate

The study has got the approval of Medical Ethics Committee of the Affiliated Hospital of Qingdao University (QYFY WZLL 26341).

Consent for publication

The study has got the consent of the patient for publication.

Author's contributions

All authors were involved in study concept and design. Yufen Yan and Chunhua Han authored the initial draft of the manuscript, and all authors critically revised the manuscript and approved the final copy.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Financial disclosure

The authors have indicated they have no financial relationships relevant to this study to disclose.

Acknowledgement

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Figure legends

Figure 1. Computed tomography (CT) results of the *Olsenella uli*-induced pneumonia.

Take the day when the patient transferred to our hospital as the first day, A-B, CT at the second day showed bilateral pneumonia, pyogenic necrosis in the right lower lobe of the lung, encapsulated pyothorax in the right chest and bilateral emphysema; C-D, CT at 10th day showed aggravating bilateral pneumonia and emerging bilateral pleural effusion in comparison with 2nd day; E-F, CT at 20th day showed improving bilateral pneumonia and left pleural effusion in comparison with 10th day; G-H, CT at 66th day (August 13) showed improving bilateral pneumonia and no pleural effusion in comparison with 20th day.

Figure 2. Colony morphology of *Olsenella uli* on Columbia blood agar medium.
Colonies of bacteria isolated from sputum culture on Columbia blood agar were small needle tip like and translucent (Fig 2) after 48 hours of incubation at 37°C with 5% CO₂.

Figure 3. Microscopic morphology of *Olsenella uli*.
After Gram-staining, gram-positive, blue purple cells which appeared singly, in pairs, in short to long chains under microscope, and the center of some cells swelled, but the size was not uniform. Magnification ×1000

分析物名称: G11
 分析物说明:
 分析物 ID: G11
 分析物创建日期/时间: 2020-07-16T10:15:45.610
 选用的 MSP 数据库:
 选用的分类树: Taxonomy, Projects, Bruker Taxonomy

等级 (质量)	匹配模式	分值	NCBI 标识符
1 (++)	Olsenella uli ENR_0045 ENR	2.229	133926
2 (++)	Olsenella uli 68 RLT	2.096	133926
3 (++)	Olsenella uli P10fluc_i9AN USH	2.015	133926
4 (+)	Olsenella uli DSM 7084T DSM	1.904	133926
5 (+)	Olsenella uli P10fluc_i9AN USH	1.832	133926
6 (-)	Streptomyces violaceoruber B263 UFL	1.418	1935
7 (-)	Clostridium cochlearium 1080_ATCC 17794T BOG	1.346	1494
8 (-)	Clostridium cochlearium 1050_NCTC 2909 BOG	1.338	1494
9 (-)	Proteus mirabilis 13210_1 CHB	1.361	584
10 (-)	Lactobacillus pentosus DSM 20199 DSM	1.221	1589

Figure 4. Results of mass spectrometry analysis for sputum culture.

With Brooke mass spectrometer, the sputum culture of the pneumonia case was defined as *Olsenella uli* with average score 2.019 ± 0.157 .

Figures

Figure 1

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Figure 2

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Figure 3

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分析物名称:

分析物说明:

分析物 ID:

分析物创建日期/时间:

选用的 MSP 数据库:

选用的分类树:

G11

G11

2020-07-16T10:13:45.610

Taxonomy, Projects, Bruker Taxonomy

等级 (质量)	匹配模式	分值	NCBI 标识符
1 (++)	<i>Olsenella uli</i> ENR_0045 ENR	2.229	133926
2 (++)	<i>Olsenella uli</i> 68 RLT	2.096	133926
3 (++)	<i>Olsenella uli</i> P10fluc_i9AN USH	2.035	133926
4 (+)	<i>Olsenella uli</i> DSM 7084T DSM	1.904	133926
5 (+)	<i>Olsenella uli</i> P10fluc_i9AN USH	1.832	133926
6 (-)	<i>Streptomyces violaceoruber</i> B263 UFL	1.418	1935
7 (-)	<i>Clostridium cochlearium</i> 1080_ATCC 17794T BOG	1.346	1494
8 (-)	<i>Clostridium cochlearium</i> 1050_NCTC 2509 BOG	1.338	1494
9 (-)	<i>Proteus mirabilis</i> 13210_1 CHB	1.283	584
10 (-)	<i>Lactobacillus pentosus</i> DSM 20199 DSM	1.223	1589

Figure 4

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