Case study

***Pseudomonas mosselli*: A case series on its potential as a rare opportunistic pathogen in immunocompromised patients**

Abstract

Background

Pseudomonas mosselli is a gram negative, rod-shaped, aerobic, non-spore-forming and motile bacteria found mainly in soil, protecting associated plants from fungal and bacterial pathogens through the production of diverse molecules and infection caused by it is rare and presents as an opportunistic infection in immunocompromised hosts. Most common presentation of respiratory infection caused by this isolate includes fever, respiratory distress and purulent cough. Adequate antibiotic coverage and early diagnosis mostly leads to complete recovery of the patient.

Cases

Three cases have been included in our case series out of which patient from case 1 was a known case of type 2 diabetes mellitus, chronic kidney disease and hypertension who was also diagnosed of being positive for SARs- COV-2 by RT-PCR (Real time polymerase chain reaction), patient from case 2 was a known case of an immunological disorder named, Takosubo cardiomyopathy with impending heart failure and patient from case 3 was a known case of advanced renal failure due to rapidly progressive glomerulonephritis on weekly dialysis and steroid therapy. These three isolates were identified based on the culture characteristics and species identification, performed by Matrix assisted laser desorption / ionization- time of flight-mass spectrometry (MALDI-TOF-MS).

Conclusion

Pseudomonas mosselii causes opportunistic infections in immunocompromised patients. The respiratory infection being less drug resistant than *Pseudomonas aeruginosa* can be easily overcome by administration of appropriate antibiotic treatment followed by antibiotic susceptibility testing.

Keywords: Pseudomonas aeruginosa, Pseudomonas putida, Pseudomonas mosselii, Matrix assisted laser desorption / ionization- time of flight- mass spectrometry (MALDI-TOF-MS)

Introduction

The genus *Pseudomonas* is a group of gram-negative, rod shaped, aerobic, non-spore-forming and motile bacteria [1]. *Pseudomonas* strains are capable of surviving in diverse niches, ranging from terrestrial and aquatic environments to tissues of eukaryotic hosts. Many members of this genus displayed remarkable physiological and metabolic activity against different pathogens [2, 3]. *Pseudomonas mosselii* was clinically isolated in France and often found in soil and can protect associated plants from fungal and bacterial pathogens through the production of diverse molecules and is a rare opportunistic human pathogen [4, 5]. Based on 16S rRNA analysis, it has been placed in the *Pseudomonas putida* group [6]. *Pseudomonas mosselii* was formally described as a novel species in 2002 by Dabboussi et al. (2002), who examined strains of *Pseudomonas fluorescens*, *Pseudomonas putida* and *Pseudomonas species* using a polyphasic taxonomic approach including 16S rDNA phylogeny, numerical analysis, DNA–DNA hybridization, thermal stability of DNA–DNA hybrids and siderotyping methodology [4]. They reported it as a novel species due to low level of DNA–DNA relatedness to other Pseudomonas strains and that strains are phenotypically and genotypically homogeneous with characteristic phenotypic features that allow speciation [4]. Here, we present three cases of

immunocompromised patients with respiratory distress of which our case 1 was also positive for COVID-19 and suffered from severe covid pneumonia followed by secondary bacterial infection, case 2 suffered from Takosubo cardiomyopathy and impending heart failure and case 3 was a known case of advanced renal failure due to glomerulonephritis. These patients' sputum samples revealed growth of a rare *Pseudomonas species* that were confirmed as *Pseudomonas mosselii* by Matrix assisted laser desorption/ionization- Time of flight- Mass spectrometry (MALDI-TOF MS).

Case reports

Case 1

A 74 year old male, known case of type-2 diabetes mellitus, chronic kidney disease and hypertension, presented to a covid testing facility with chief complains of fever and sore throat for one week and myalgia for four days. He tested positive for SARs-COV-2 and was admitted to a covid facility at a tertiary care center, where he was administered steroids, oxygen therapy and other broad spectrum antibiotics despite which he developed moderate COVID-19 pneumonia. Being a chronic kidney disease patient, he needed regular haemodialysis sessions and was referred to the haemodialysis unit dedicated for covid 19 patients after 12 days of admission. His cough and respiratory distress had not relieved so a sputum sample was sent to the bacteriology section of the department of microbiology and a grams stained smear and culture was put up for the same on mackonkey and blood agar. On gram's stained smear, few epithelial cells, plenty pus cells and gram negative bacilli were observed and the next day culture on mackonkey agar showed non-lactose fermenting colonies with a fruity odour and blood agar showed transparent to grey colonies with haemolysis and was oxidase positive, suggestive of *Pseudomonas species*. The colonies were subjected to species identification by Matrix assisted

laser desorption / ionization- time of flight- mass spectrometry (MALDI-TOF-MS) and were identified as *Pseudomonas mosselli*. When antibiotic sensitivity testing was performed by Kirby-Bauer disc diffusion method after preparing a McFarland of 10^5 microorganisms, it showed sensitivity to Amikacin, Ceftazidime, Imipenem and Piperacillin-Tazobactam. The patient was started on Ceftazidime and his symptoms were relieved within 3 days.

Case 2

A 22 year old female, known case of Takosubo cardiomyopathy with impending heart failure, presented to the department of emergency medicine, with chief complains of on and off fever for two weeks which subsided with administration of oral antipyretics, productive cough and respiratory distress for 10 days and generalised edema of ascending type for a week. She was managed conservatively and Bi-level positive air pressure (BIPAP) was used for respiratory distress, cause of chest pain could not be ruled out, she also presented with upper or lower gastrointestinal bleed, joint pains, rashes and one unit blood was also transfused. On examination, she was observed to have pallor, cyanosis, clubbing and prominent edema. Her pulse rate was raised ranging from 120-130 per minute and respiratory rate was also raised to 32 per minute with basal crepts observed bilaterally. A COVID-19 Real Time- Polymerase Chain Reaction (RT-PCR) was performed owing to her symptoms but turned out to be negative. All other routine blood and radiological investigations were performed and a sputum sample was sent to the bacteriology section of the department of microbiology. Microscopic examination of the sputum sample showed plenty pus cells and few long and thin gram negative bacilli, next day oxidase positive, translucent colonies were observed on blood agar surrounded by a zone of haemolysis, suggestive of *Pseudomonas species*. On MALDI-TOF-MS, the colonies were

identified to be those of *Pseudomonas mosselli*. When antibiotic sensitivity testing was performed by Kirby- Bauer disc diffusion method, the microorganism showed sensitivity to Amikacin, Ceftazidime, Cefoperazone- Sulbactum, Imipenem and Piperacillin-Tazobactam. After antibiotic sensitivity testing, adequate antibiotics were administered and her fever subsided gradually over the course of 48 hours and respiratory symptoms where alleviated in 5 days of time.

Case 3

A 37 year old male, known case of advanced renal failure presented to the department of Nephrology with chief complaints of edema, hypertension and active urinary sediments for a week. A kidney biopsy was performed on admission, which diagnosed him of having rapidly progressing glomerulonephritis (RPGN) and weekly dialysis was advised. He was started on steroids as histopathology reported IgA nephropathy. His total leukocyte count was around 22,000 cells/ cubic mm, suggestive of infection. He was advised to start Oseltamivir prophylaxis as a neighboring patient tested positive for H1N1 swine flu infection during the course of hospital stay. After 10 days, he developed fever, respiratory distress and dry cough; he was tested for HINI swine flu which came negative. His sputum sample was sent for bacteriological culture and microscopy. Microscopic examination of the sputum sample showed moderate pus cells and few long and thin gram negative bacilli, next day oxidase positive, translucent colonies were observed on blood agar surrounded by a zone of haemolysis, suggestive of *Pseudomonas species*. On MALDI-TOF-MS, the colonies were identified as *Pseudomonas mosselli*. On antibiotic sensitivity testing by Kirby- Bauer disc diffusion method, it was sensitive to Amikacin, Ceftazidime, Cefoperazone- Sulbactum, Imipenem, meropenem and Piperacillin-Tazobactam.

After clinicians had started empirical antibiotics, his fever and respiratory symptoms where alleviated in 3 days of time. On discharged he was advised to take oral Cefuroxime Axetil 500 mg tables, twice a day for 5 days, steam inhalation and advised follow up in outpatient department after two weeks.

Discussion

Pseudomonads other than *Pseudomonas aeruginosa* are only occasionally isolated from sputum cultures and rarely isolates from these clinical specimens are identified to be *Pseudomonas mosselli*. This rare isolate was, in fact, only formally described as a novel species in 2002 by Dabboussi et al, who reported it as a novel species on the basis of a low level of DNA–DNA relatedness to other *Pseudomonas* strains and that *Pseudomonas mosselii* strains are phenotypically and genotypically homogeneous with characteristic phenotypic features that allow speciation [4].

Prior to classification of *Pseudomonas mosselii* as a separate species, strains would have most likely been identified as *Pseudomonas fluorescens*; however, we have come across reports of *Pseudomonas fluorescens* as a cause of respiratory tract infection. While *Pseudomonas fluorescens* has been identified in human bronchoalveolar lavage fuid (BALF), sputum specimens or throat swabs, its role in pneumonia pathogenesis is unclear. It has been previously suspected of being an etiologic agent of pneumonia in several reports [7-10]. However, clinical characteristics and drug susceptibility pattern of *Pseudomonas fluorescens* pneumonia have rarely been reported [9]. In our case reports, case 1 was positive for SARs-COV-2, receiving steroid therapy due to severe COVID pneumonia; type-2 diabetes mellitus, chronic kidney disease and hypertension, owing to severe immunosuppression may be a cause for succumbing to

an opportunistic infection by *Pseudomonas mosselli* after two weeks of hospitalization. However, case 2 was a Takosubo cardiomyopathy patient with impending heart failure having a presentation similar to that of an infectious disease in a young female who developed respiratory distress with pulmonary edema and purulent cough, sputum culture revealed growth of *Pseudomonas mosselli* after four days of hospitalization. Due to timely communication with the clinicians both our case were discharged after adequate treatment and were advised for follow up after two weeks. Case 3 was a patient of rapid progressive glomerulonephritis on weekly dialysis and steroid therapy, who developed respiratory distress during the course of hospital stay, the sputum culture revealed growth of *Pseudomonas mosselli* after two weeks of hospitalization. The clinicians had started empirical antibiotic prophylaxis, suspecting an opportunistic infection.

Conclusion

Pseudomonas mosselii causes opportunistic infections in immunocompromised patients. Although, the infection caused by this isolate is less virulent in comparison to *Pseudomonas aeruginosa*, it causes enough morbidity to already immunocompromised patients. The respiratory infection can thus be easily overcome by administration of appropriate antibiotic treatment followed by antibiotic susceptibility testing.

References

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Table 1. Cases of Pseudomonas mosselli isolated from respiratory samples of patients admitted to a tertiary care center in Northern India

Case	Age /Sex	Underlying condition	Infecting organism	Culture specimen	Procalcitonin assay	Treatment	Outcome
1	74/M	Type 2 Diabetes mellitus	*Pseudomonas mosselli*	Sputum	0.16	Moxifloxacin and Ciprofloxacin	Alive
2	22/F	Takosubo cardiomyopathy	*Pseudomonas mosselli*	Sputum	63.60	Levofloxacin	Alive
3	37/M	Advanced renal failure on dialysis, Hypertension	*Pseudomonas mosselli*	Sputum	3.93	Amoxicillin-clavulinic acid, Ceftriaxzone and Cotrimoxazole	Alive