



Case report

Are *Burkholderia* Emerging Pathogens in patients with underlying morbidity: A case series

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ABSTRACT

Burkholderia is a genus consisting of several species including the *Burkholderia pseudomallei* group, *Burkholderia cepacia* complex and other phytopathogens. *Burkholderia* species is a gram-negative bacillus with protean presentation that can be acquired from various sources, including water, soil, plant surfaces, and hospital environments. The organism on Gram staining is seen as gram-negative rod and on culture, the colonies are non-lactose fermenting. As it can mimic other diseases, it is frequently misdiagnosed and there is lack of awareness about the clinical spectrum of disease and diagnosis. This study aims to investigate varied clinical manifestations, identify potential risk factors and transmission modes and contribute to enhancing the clinical management of diseases. The increasing prevalence of *Burkholderia* infection implies its potential emergence as a significant public health concern, compounded by the growing incidence of diabetes, which has the potential to escalate the overall disease burden.

The principal finding of the case series highlighted a spectrum of clinical presentations, emphasizing the need for comprehensive diagnostic strategies and tailored therapeutic interventions. These strategies will address the diverse manifestations and challenges posed by *Burkholderia* infections.

This underscores the importance of heightened awareness among clinicians and microbiologists, given the need for extended treatment to achieve a complete cure and prevent potential relapses.

1. Introduction

The genus *Burkholderia* comprises several species, including the *Burkholderia pseudomallei* (Bps) group, the *Burkholderia cepacia* complex (Bcc) and phytopathogen species [1]. These species are phenotypically very similar and differentiation within them is very difficult. In most resource poor settings, they are identified as non-fermenting gram-negative bacilli, sometimes erroneously as *Pseudomonas* species. This may be the reason for under reporting of *Burkholderia* infections seen in India [2]. It has manifested as a concerning opportunistic pathogen, demonstrating a significant capacity to induce severe respiratory infections in individuals with

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co-morbid conditions, especially cystic fibrosis and chronic granulomatous disease [3]. *Burkholderia* infection is often seen in patients with comorbid conditions like diabetes mellitus, malignancy, chronic kidney disease, chronic lung disease and immunocompromise, with diabetes being the most important risk factor [4]. The increasing prevalence of diabetes mellitus globally has the potential to elevate the overall burden of the disease.

Clinical presentation can range from localized infection to septic arthritis, visceral abscess, sepsis and death, with pneumonia being the most common presentation [5]. The disease was earlier considered endemic in Australia and South Asian countries like Sri Lanka and Bangladesh. In India, it has been reported in various states including Karnataka, Kerala, Maharashtra, Tamil Nadu, and Puducherry with increase in prevalence during rainy weather and among rice farmers [6]. *Burkholderia* species are aerobic, can grow well on standard culture media like blood agar and MacConkey agar and exhibit positive result in oxidase and catalase tests. *Burkholderia* species can be distinguished through their growth on selective media (*B. cepacia* selective agar, Ashdown's agar), the amino acid decarboxylation test and the oxidation of sugars such as sucrose, adonitol, arabinose, among others. Additionally, serological tests like the indirect immunofluorescent antibody test, contribute to the identification of these species. Molecular-based techniques also contribute to the accurate identification of these species [7]. *Burkholderia* species are naturally resistant to commonly used antibacterial against gram-negative microorganisms like narrow spectrum cephalosporins, most penicillin, polymyxin and macrolides [8]. This raises concern about the potential severity of *Burkholderia* infection which needs early diagnosis to prevent morbidity and mortality.

The diverse array of clinical presentations and similarities to other infections often result in misdiagnosis or insufficient recognition. This collection of cases aims to heighten awareness among clinicians and microbiologists, promoting enhanced vigilance for accurate and timely diagnoses.

The increasing incidences of *Burkholderia* infection indicate its potential to evolve into a significant emerging public health issue. This underscores the necessity to augment vigilance in public health surveillance and preparedness efforts.

We present a series of cases collected over the span of a year from our tertiary care centre. Individuals were admitted for various reasons, including cervical abscess, bloodstream infection, post-surgery wound infection, and septic arthritis. The observed cases prompt concerns, highlighting the need for an investigation to monitor trends over time.

CASE/CASE SERIES PRESENTATION: MIMICKING TUBERCULOSIS.

1.1. CASE 1

A 49-year-old female diabetic patient who was an agriculturist presented with a painless enlargement of left cervical lymph nodes for a duration of three months. The patient was clinically diagnosed as Extra pulmonary tuberculosis elsewhere and started on Anti-tubercular treatment (ATT). There was no symptomatic improvement even after three months of ATT. Patient was then transferred to



Fig. 1. CT scan of neck: thick-walled hypodense lesion in left mandibular space with extensions likely represent abscess/necrotic lymph node.

our tertiary care hospital for further management. On examination, the cervical swelling measured 5×4 cm with a bosselated surface located anterior to the sternocleidomastoid muscle with erythema. Computed tomography (CT) (Fig. 1) scan of neck was done on the second day of hospital admission. The scan showed the presence of large, heterogenous enhancing hypodense nodal mass in left cervical region suggestive of pyogenic or tubercular abscess. The chest X-ray on the same day revealed no abnormalities indicative of tuberculosis. Following CT scan, Fine needle aspiration cytology (FNAC) was done which showed suppurative lymphadenitis without granulomas not suggestive of tuberculosis. The patient underwent incision and drainage of the swelling on day four of admission, and it revealed frank pus and necrotic tissue. Sample was collected and sent for GeneXpert and acid-fast staining yielded negative results for acid-fast bacilli (AFB) in the pus. Additionally, histopathological examination ruled out mycobacterial infection. Gram staining showed gram-negative bacilli with bipolar appearance. Additionally, on the second day, MacConkey agar culture plates exhibited non-lactose fermenting dry colonies with pinkish hue (Fig. 2). The organism was identified through conventional methods. Antibiotic susceptibility test showed polymyxin B resistance, raising suspicion of *Burkholderia* infection. The amino acid decarboxylation test showed positive results for Arginine dehydroxylase and Ornithine decarboxylase. Additionally, the automated VITEK2 GN ID card system confirmed the identification as *Burkholderia pseudomallei* on the third day. This was further confirmed by PCR based DNA sequencing targeting the 16s rRNA region and sequencing. VITEK 2 antimicrobial susceptibility testing showed isolated strain susceptible to co-trimoxazole, tetracycline, ceftazidime, amoxicillin-clavulanate and meropenem. The obtained sequence was subsequently submitted to GenBank and assigned Accession number: OQ748076. The patient was diagnosed with melioidosis, and ATT was stopped immediately and was treated with intravenous (IV) ceftazidime 2g thrice a day for two weeks in intensive phase of therapy and showed improvement. Following a six-month regimen of oral co-trimoxazole 480mg twice a day during the eradication phase, the patient achieved complete recovery and remained free of infection.

2. Septicaemia

2.1. CASE 2

A 51-year-old female patient with diabetes who was undergoing chemotherapy for breast cancer experienced fever on and off after chemotherapy. One week later 5 ml blood sample was collected aseptically and was inoculated in BACTEC bottle. It was transported promptly to the laboratory, ensuring appropriate temperature throughout transport. Blood culture bottles are incubated in BACTEC™ 9120 an automated continuous monitoring blood culture system. Blood culture bottle signalled for growth after 72 hours, was processed for Gram stain and culture. The Gram staining revealed gram-negative bacilli. After 48 hours, culture plates grew non-lactose fermenting colonies. They were diagnosed as *Burkholderia vietnamiensis* using MALDI – TOF MS (matrix-assisted laser desorption ionization-time of flight mass spectrometry). VITEK 2 antimicrobial susceptibility testing showed isolated strain susceptible to meropenem, levofloxacin, ceftazidime, doxycycline and resistant to co-trimoxazole. Subsequently, the chemotherapy port, a device implanted under the skin to administer chemotherapy drugs directly into the bloodstream, was removed. The tip of the port was sent for culture testing. The testing confirmed the presence of the same organism. The patient received intravenous ceftazidime at a dosage of 2g thrice daily for a duration of two weeks. This resulted in noticeable improvement. The patient was then discharged with advice to continue doxycycline 100mg twice daily for 3 months as organism showed resistance to co-trimoxazole. Unfortunately, the patient was lost to follow up after that. Further testing through colony PCR and sequencing confirmed the organism as *Burkholderia vietnamiensis*. Sequencing was submitted to GenBank with Accession no [OQ748077](https://www.ncbi.nlm.nih.gov/nucl/OQ748077).

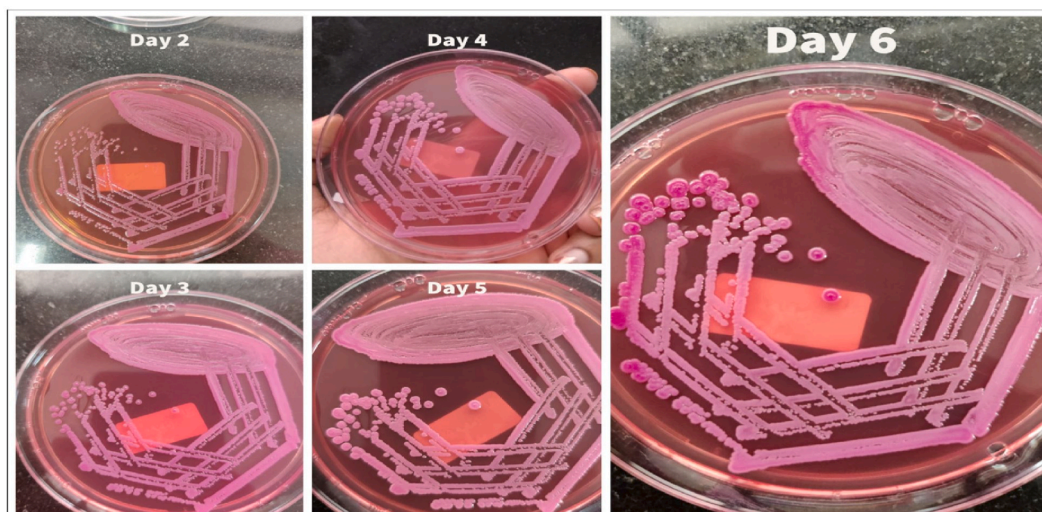


Fig. 2. MacConkey agar showing non-lactose fermenting colonies with pinkish hue showing cornflower head appearance on further incubation.

2.2. CASE 3

A 50-year-old patient with Systemic lupus erythematosus on immunosuppressants and Type 2 diabetes presented with fever of 1 month duration, dyspnoea, and cough for 20 days. Patient was treated with ceftazidime and azithromycin elsewhere and referred here for tertiary care. On presentation to our hospital, right upper limb cellulitis was observed along with fever and cough. Since the clinical presentation was suggestive of pulmonary tuberculosis, the patient underwent investigations including sputum AFB and chest X-ray. The X-ray revealed lobar consolidation (Fig. 3), while the sputum AFB was negative. The patient was subsequently diagnosed with community-acquired pneumonia. The blood culture was sent on the day of admission, which grew non-lactose fermenting colonies with pinkish hue on MacConkey agar culture plates by third day. It was further confirmed by VITEK 2 GN ID card as *Burkholderia pseudomallei*. The most probable reason for limb cellulitis might be *Burkholderia* infection which might have got seeded into blood causing septicaemia. However, no pus sample was received for cultivation. The patient was immediately started on IV ceftazidime 2g thrice daily and IV meropenem 2g twice daily combination therapy. Patient recovered clinically in two weeks and got discharged and was asked to continue oral co-trimoxazole at 480mg twice a day for six months. The history showed that the patient received IV ceftazidime 2g for 2 weeks elsewhere without any clinical improvement. The most probable reason was biofilm formation for the monotherapy which was rectified by giving combination therapy in our tertiary care centre. We lost the follow up of the patient.

3. Surgical site infection/wound infection

3.1. CASE 4

A 50-year-old diabetic male patient who had a road traffic accident (RTA) suffered from an open fracture of the right lower limb. Examination revealed a laceration measuring 6 × 3cm on the medial aspect of the leg, exposing underlying bone and deeper tissue. Active bleeding and local tenderness were also noted at the site. However, distal foot vascularity remained uncompromised, and sensation was intact. The wound was washed immediately using 3 L of saline solution and Betadine. Emergency wound debridement and bone fixation procedures were then performed on the day patient was admitted. On the fifth postoperative day, the patient was clinically diagnosed with surgical site infection (SSI) promoting the collection of pus sample for Gram stain, culture and sensitivity. Gram staining showed gram-negative bacilli. After 48 hours, culture plates grew non-lactose fermenting colonies. The VITEK 2 GN ID card confirmed it to be *Burkholderia cepacia*. The contact with soil by this individual may have resulted in direct inoculation through the skin. Additionally, inadequate wound cleaning before surgery may be regarded as a factor contributing to surgical site infection. The patient recovered after receiving treatment with IV meropenem 2g each twice daily for two weeks. The patient was discharged and asked to continue oral co-trimoxazole 480 mg twice a day for six months. Patient made full recovery.

3.2. CASE 5

A 35-year-old male with newly diagnosed diabetes who had a road traffic accident presented with multiple fractures of limbs



Fig. 3. X-ray chest AP view: Diffuse bilateral heterogenous consolidation predominantly involving left upper lobe and right lower zone.

Table 1
Review of *Burkholderia* cases in Diabetic patients.

	Our case with various reported studies	Place	Organism	Clinical picture	Profession	Antibiogram	Treatment	Outcome
Mimicking Tuberculosis	Our case Case 1	Chennai	<i>B. pseudomallei</i>	Unilateral Suppurative Cervical lymphadenitis	Farmer	Susceptible-co-trimoxazole, tetracycline, ceftazidime, amoxicillin-clavulanate, meropenem, imipenem, doxycycline	IV Ceftazidime 2g thrice a day for two weeks followed by oral cotrimoxazole 480mg twice a day for six-months	two weeks in intensive therapy showed improvement. Following a six-month of eradication phase, the patient achieved complete recovery and remained free of infection.
	Reports from India [9]	Chennai		Suppurative Vertebral osteitis	Farmer		IV ceftazidime 2g thrice daily for six weeks and oral Co-trimoxazole 480 and doxycycline 100mg twice daily	Recovered
	Reports from worldwide [10]	Bangladesh		Brain abscess	Forest officer		IV ceftazidime for 2 g three times daily for 2 weeks followed with oral co-trimoxazole 960 mg two times daily and oral doxycycline 100 mg twice daily for 3 months	Recovered
Septicaemia	Our case Case 2 Case 3	Chennai	<i>B. vietnamiensis</i> <i>B. pseudomallei</i>	High grade Fever Fever, dyspnoea and cough	Housewife Paddy field worker	Case 2 Susceptible -meropenem, levofloxacin, ceftazidime, doxycycline Resistant to co-trimoxazole. Case 3 Susceptible – to meropenem, ceftazidime, doxycycline, amoxicillin-clavulanate and co-trimoxazole.	Case 2 IV ceftazidime 2g thrice daily for a duration of two weeks, advice to continue doxycycline 100mg twice daily for 3 months Case 3 IV ceftazidime 2g thrice daily and IV meropenem 2g e twice daily combination therapy followed by oral cotrimoxazole at 480mg twice a day for six months	Both cases were Lost to follow up
	Reports from India [11]	Kerala	<i>B. pseudomallei</i>	Fever with altered sensorium	Farmer		IV meropenem 1g eight hourly	Recovered
	Reports from worldwide [12]	Colombia	<i>B. pseudomallei</i>	Fever with chills, rigor and right leg pain	Gardner		IV meropenem 2g 12 hourly and vancomycin 1g 12 hourly	Recovered
SSI/wound infection	Our case Case 4 Case 5	Chennai	<i>B. cepacia</i> <i>B. cenocepacia</i>	Right lower limb fracture treated with emergency surgery presented with SSI on 5th post-operative day Multiple open fracture treated with emergency surgery presented with SSI	Shopkeeper Field worker	Susceptible- ceftazidime, Minocycline, cotrimoxazole, Levofloxacin, Meropenem	IV Meropenem 2g twice daily two weeks and to continue oral co-trimoxazole 480mg twice daily for six months.	Patient completely recovered after follow-up remained free of infection.
	Reports from India [13]	New Delhi	<i>B. cepacia</i>	Post-operative pyogenic spondylo-discitis	Not mentioned		IV meropenem 1 g thrice a day for six weeks followed by oral clindamycin six weeks	Recovered
	Reports from worldwide [14]	Brazil	Burkholderia cepacian complex *	Injury with glass piece	Muddy field worker		IV Meropenem 2g six hourly for 2 weeks	Recovered

(continued on next page)

Table 1 (continued)

	Our case with various reported studies	Place	Organism	Clinical picture	Profession	Antibiogram	Treatment	Outcome
Septic arthritis	Our case Case 6	Chennai	<i>B. pseudomallei</i>	Bilateral knee joint effusion	Businessman	Susceptible-co-trimoxazole, tetracycline, ceftazidime, amoxicillin-clavulanate, meropenem, doxycycline	IV meropenem therapy 1 g every 6 h for two weeks and was recommended oral co-trimoxazole 480mg twice a day for six months.	Patient made full recovery returns to full time work after 6 months of therapy
	Reports from India [15]	Andaman and Nicobar Island		Knee swelling	Farmer		IV ceftazidime 8 hourly for 3 weeks followed with oral co-trimoxazole and doxycycline for 5 weeks	Recovered
	Reports from worldwide [16]	Oman		Left Knee joint septic arthritis	Farmer		IV meropenem and cefuroxime tablet	Recovered

B-*Burkholderia*; Hb-Haemoglobin; SSI-Surgical site infection; IV-Intravenous.

The case outlined in the table demonstrates a significant association with the specified condition.

* After meticulous search in literature there was no article that showed *Burkholderia cenocepacia* as causative agent of SSI. Follow-up appointments were scheduled for our reported cases at regular intervals to monitor the progress of treatment. With the exception of cases 2 and 3, all other patients adhered to regular follow-up appointments and showed improvement.

involving both tibia and radius, leading to exposure of deeper tissues. The patient received a rapid infusion of 500 mL of normal saline solution through an intravenous (IV) line alongside the administration of a tetanus toxoid injection at the time of hospital admission. Subsequently, the patient underwent emergency surgery involving wound debridement followed by suturing. The patient was clinically diagnosed with SSI on the third postoperative day. Pus and tissue cultures collected from site on third postoperative day showed *Burkholderia cenocepacia* on VITEK 2 GN ID card. The patient was treated with IV Meropenem 2g twice daily two weeks and was discharged on improvement and asked to continue oral co-trimoxazole 480mg twice daily for six months. Patient completely recovered after follow-up.

To our knowledge, this is an unusual case of SSI by *Burkholderia cenocepacia*. According to the manufacturer's instructions manual, the VITEK 2 GN ID card has broadened the VITEK 2 identification database and identifies *Burkholderia cenocepacia* without misidentification, however molecular methods were not employed for confirmation in this instance.

4. Septic arthritis

4.1. CASE 6

A 33-year-old male with diabetes presented with a two-week history of fever and bilateral knee joint swelling. Routine investigations on the day of admission unveiled iron deficiency anaemia, reflected by an Hb level of 7.1g%. Consequently, on day two of admission, packed cell transfusion was administered. The patient underwent bilateral arthrotomy on day five following anaemia correction. Intraoperatively, synovial fluid, and paired blood samples were collected and sent for culture and sensitivity. Both grew non-lactose fermenting colonies with pinkish hue on MacConkey agar culture plates after 48 hours, which on VITEK2 GN ID card confirmed the presence of *Burkholderia pseudomallei*. The patient was started on IV meropenem therapy 1g every 6 h for two weeks and was discharged on satisfactory improvement. Patient was recommended oral co-trimoxazole 480 mg twice a day for six months. Patient made full recovery returns to full time work after 6 months of therapy.

Clinical details, co-morbid conditions and follow-up of each case were explained in [Table 1](#) and the cases highlighted in the table illustrates a notable correlation with the specified condition.

5. Discussion

The *Burkholderia* genus encompasses more than 80 gram-negative species, comprising both plant and human pathogens. The latter can be categorized into either the *Burkholderia pseudomallei* complex (Bpc) or the *Burkholderia cepacia* complex (Bcc). Advancements in Bcc taxonomy unveiled nine phenotypically similar species or genomovars—*Burkholderia cepacia*, *Burkholderia multivorans*, *Burkholderia cenocepacia*, *Burkholderia stabilis*, *Burkholderia vietnamiensis*, *Burkholderia dolosa*, *Burkholderia ambifaria*, *Burkholderia anthina* and *Burkholderia pyrrocinia*, which can be differentiable through molecular and biochemical tests. Among which *B.cenocepacia* is associated with the highest mortality, followed by *B.multivorans* and *B.dolosa* [17]. It is commonly isolated in soil, water and moist environments. The mode of transmission includes contact, droplets, ingestion, or exposure to contaminated medical devices and medications. This infection can arise from various factors, such as contaminated hospital water sources, contact with polluted surfaces, person-to-person transmission within healthcare settings, and underlying health conditions. Additionally, these pathogens are frequently found in hospital fluids, such as irrigation solutions and intravenous fluids [3,18]. *Burkholderia* infections are very commonly seen in diabetes patients as these patients has got reduced expression of few cytokines and impaired T-cell function leading to impaired phagocytosis [19]. All of our patients had a history of Type 2 diabetes mellitus which may also be one of the factors to contribute to the susceptibility to infection. *Burkholderia species* secrete high affinity iron chelating compounds which capture iron from the local environment which can present as iron deficiency anemia in infected individuals which is also seen in our cases [20]. As India is endemic for tuberculosis, most of the cases of lymphadenitis and cold abscess are treated as tuberculosis and presumptive antitubercular drugs are often prescribed. These are designated as smear negative tuberculosis due to lack of awareness [21]. Patients with musculoskeletal melioidosis often need operative intervention and longer hospital stay, as *Burkholderia pseudomallei* is one of the causes for septic arthritis. In patients with similar symptoms, it is important to maintain a high index of suspicion for *Burkholderia*, and the prompt recovery of the organism from samples will aid in early management and a positive outcome for the patient. It has been shown that 10 % of relapses occur even after 20 weeks of treatment. Most cases of melioidotic septic arthritis in India have been reported from Kerala and Tamil Nadu [22].

We reported two cases of road traffic accident undergoing emergency surgery with *Burkholderia cepacia* and *Burkholderia cenocepacia* growth. This infection may result from various factors, including contamination of hospital water sources, contact with contaminated surfaces, person-to-person transmission in healthcare settings, and the presence of underlying health conditions. This underscores the critical importance of promoting appropriate wound care practices to prevent such infections and emphasizes the necessity for increased awareness among healthcare professionals. Long-term indwelling catheters used for chemotherapy in cancer patients provide a favorable environment for the propagation of *Burkholderia*. There have been several reports stating that long term indwelling catheters meant for chemotherapy provide favorable milieu for colonization of *Burkholderia* [23]. In our case series we had one patient on chemotherapy who grew *Burkholderia* in the blood as well as catheter tip. Global reports say that the sources of *Burkholderia* outbreak in hospitals are from hospital water, ultrasound gel, nasal spray, medications meant for nebulization, and lipid immersions [24].

Identification errors often stem from the phenotypic similarities between *Burkholderia* and other bacterial genera, including *Cupriavidus*, *Ralstonia*, *Achromobacter*, *Brevundimonas*, *Comamonas*, *Pandoraea*, and *Delfia*. These genera are prone to being

misidentified as *Burkholderia cepacia* complex (Bcc) species. Challenges in distinguishing between *B. pseudomallei* and closely related species, such as *B. thailandensis*, may be encountered by commercial bacterial identification kits, potentially resulting in identification failures. The incorporation of the new colorimetric-based GN card has broadened the VITEK 2 identification database, enhancing the precision in identifying *Burkholderia* species [25].

The manifestation of multidrug resistance (MDR) and prolonged antibiotic therapy, a formidable challenge for clinicians in treatment of *Burkholderia* infection. Timely isolation and an antibiotic susceptibility report are essential for guiding appropriate patient management. Given the requirement for extended treatment to achieve full recovery and prevent potential relapses, counselling the patient about the necessity of prolonged chemotherapy becomes crucial [26]. In this context, a prophylactic vaccine may be a complementary approach for targeted population [27].

Recent advances in the diagnostic arena of *Burkholderia* look promising. Several molecular targets like *hisA*, *rpsU*, *recA*, 16S rRNA and 23S rRNA and restriction fragment length polymorphism (RFLP). RFLP is beneficial for distinguishing genera, species, and complexes at the molecular level within a short timeframe [28].

The limitation of our study is that 16SrRNA and RFLP is available only in reference laboratories because of a need for specific infrastructure and technical expertise. This presents a dual challenge. It encompasses both the need for specialized facilities and the high costs associated with these techniques. Consequently, it hinders our ability to validate other isolates through molecular methods.

The *Burkholderia* genus comprises numerous species, each with distinct diagnostic and management considerations. While this case series alone may not encompass all variations, it serves as a valuable contribution. It aids in understanding and addressing the complexities associated with the diagnosis and management of different *Burkholderia* species.

To substantiate the emerging nature of the disease, a comprehensive multicentric study with a substantial sample size is required.

6. Conclusion

The early isolation of *Burkholderia* and obtaining an antibiotic susceptibility report are critical steps for clinicians facing the challenges of multidrug resistance and prolonged antibiotic therapy. These measures enable appropriate patient management. They also facilitate informed antibiotic selection. Effective counselling emphasizes the importance of prolonged chemotherapy in treating *Burkholderia* infections. The increasing incidences of *Burkholderia* infection indicate its potential to evolve into a significant emerging public health issue. This underscores the necessity to augment vigilance in public health surveillance and preparedness efforts.

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Ethical approval statement

The current study was approved by the institutional ethical committee. Written informed consent was obtained from all the patients included in the study and for the publication of all their data and/or images.

Data availability statement

GenBank with Accession number: OQ748076 and OQ748077.
OQ748076 - <https://www.ncbi.nlm.nih.gov/nuccore/OQ748076.1/>
OQ748077 - <https://www.ncbi.nlm.nih.gov/nuccore/OQ748077.1/>

CRediT authorship contribution statement

Swati Kumari: Writing – review & editing, Writing – original draft, Validation, Supervision, Resources, Project administration, Methodology, Formal analysis, Conceptualization. **Marleena Banu:** Writing – original draft, Data curation, Conceptualization. **Krishnapriya Ramanatha:** Data curation. **Ramy Barani:** Methodology, Formal analysis. **Kopula Sathyamurthy Sridharan:** Validation, Formal analysis.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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