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First molecular approach to diagnose paediatric pulmonary lophomoniasis: A case series

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Abstract

A prospective study was conducted from 2017 to 2021 at Bouali Hospital in Mazandaran province, Sari, Iran. Out of 58 patients who were enrolled in our study, lophomoniasis was diagnosed in bronchoalveolar lavage fluid of nine patients, for the first time, using an in-house polymerase chain reaction technique. All patients were treated with metronidazole at 7.5 mg/kg/day every 12 h for 14 days. After 6 months of follow-up, symptoms were fully resolved.

KEYWORDS
Lophomonas blattarum, lophomoniasis, paediatrics, PCR

INTRODUCTION

Respiratory infections are one of the most common reasons for children being admitted to paediatric wards.[1] With increasing travel and migration, rates of parasitic lung and pleural diseases are increasing in the immunocompetent population.[2] *Lophomonas* is an emerging protozoan parasite that infects upper and lower respiratory system.[3,4] Humans are infected by aerosol-containing cysts (airborne route) that are excreted by the stool of some insects, mainly cockroaches as reservoir host.[3] Low-grade fever, chronic cough, expectoration and dyspnoea were the most common symptoms among the patients.[3–5] *Lophomonas* trophozoite is mainly diagnosed by microscopic examination of respiratory secretions of patients, mainly bronchoalveolar lavage fluid (BALF),[3–7] but because of its similarity to the respiratory epithelial cells, it might be missed.[3,7]

For the first time, unlike all prior studies that used microscopic examination to diagnose lophomoniasis in children,[8–11] we presented a case series in which all paediatric pulmonary lophomoniasis patients were diagnosed using the polymerase chain reaction (PCR) approach, which was first described by Fakhar and colleagues.[12]

METHODS

Participants and data collection

This was a prospective study that was conducted from 2017 to 2021 at Bouali Hospital in Mazandaran province, Sari, northern Iran. Fifty-eight patients with respiratory symptoms who were bronchoscopy candidates were enrolled in our study after their parents signed an informed consent form. Each parent was given a questionnaire that included demographic information such as age, gender, underlying disease and symptoms such as cough, shortness of breath, sputum excretion, and so on.

TABLE 1 Characteristics of the patients diagnosed with *Lophomonas* infection

Case	Age	Gender	Living place	Underlying disease	Radiology findings	Clinical symptoms	Severity index
1	2.5 years	Male	Urban	None	Consolidation, patchy nodular infiltration	Chronic cough, dyspnoea, sputum production	Severe
2	3 months	Female	Urban	None	Normal	Wheezing, weight loss	Moderate
3	4 years	Female	Urban	Asthma, allergic rhinitis	Consolidation, ground-glass opacity	Chronic cough	Mild
4	6 years	Female	Rural	Asthma, allergic rhinitis	Patchy nodular infiltration	Chronic cough, dyspnoea	Severe
5	11 years	Female	Urban	Bronchitis	Normal	Chronic cough, dyspnoea	Mild
6	17 years	Female	Urban	Asthma, allergic rhinitis, bronchitis	Bronchiectasis	Chronic cough	Mild
7	17 years	Female	Rural	Asthma	Patchy nodular infiltration	Chest discomfort, chronic cough	Mild
8	7 years	Male	Urban	Asthma, allergic rhinitis	Normal	Chronic cough	Mild
9	11 months	Female	Urban	None	Ground-glass opacity	Wheezing	Severe

Specimen collection and testing

Fibreoptic bronchoscopy was done for all patients in a bronchoscopy room under a completely sterile situation with general anaesthesia. After a saline wash, BALF was collected and instilled into different vials, and then delivered to the Iranian National Registry Center for Lophomoniasis (INRCL). We used two different methods for diagnosing *Lophomonas* infection. First, the BALF sample was analysed by just one operator under a light microscope at 10× and 40× magnifications to detect a motile and live flagellated protozoa. Second, samples were then tested by an in house-PCR[12] to confirm the results of microscopic examination by detection of the *Lophomonas* DNA.

Definition

Lophomoniasis was considered when an ovoid-shape parasite with granular cytoplasm and tufted flagella, and a positive PCR result were detected . Based on our experience in the INRCL, an innovative severity index (SI) for *Lophomonas* infection was scored as mild to severe parasite density by counting parasites per high-power microscopic fields (HPF) (400×). Accordingly, mild density was defined as 1–10 parasites/100 HPF, moderate as 1–10 parasites/10 HPF and 1–10 parasites/HPF were considered as severe.[5]

RESULTS

During this 4-year study period, nine (two males and seven females) patients with an average age of 7.29 ± 6.4 years were diagnosed with lophomoniasis using microscopic examination and an in-house PCR test. These patients mostly live in urban areas (7:2 urban:rural ratio). Chronic cough was the most common clinical symptom (77.8%; $n = 7$ cases). Moreover, in our patients, the most prevalent (55.5%; 5/9) underlying disease was asthma, and they utilized inhaled glucocorticoids (data not shown) to treat the disease. Radiology findings revealed that patchy nodular infiltration and consolidation were the frequent presentations of our patients. According to the SI, more than half of the patients had a mild infection. The details are shown in Table 1. All patients were given metronidazole at 7.5 mg/kg/day every 12 h for 14 days. Each patient's symptoms were fully resolved after 6 months of follow-up.

DISCUSSION

Lophomoniasis is a neglected tropical infectious disease and its true global burden remains unknown, particularly among children.[3] The majority of cases have been reported from Asian countries such as Iran and China.[3–7,9,10] It has been shown that children's immunity is less effective than adults.[13] Furthermore, due to poor hygienic habits, children are more exposed to pathogens in their surroundings.[14] Considering all aspects, it seems that children are more susceptible to *Lophomonas* infection.

Due to the challenge of lophomoniasis diagnosis, its prevalence is ambiguous and controversial. As previously stated, this protozoan is easily mistaken for an airway epithelial cell under a light microscope, and this is crucial in reporting positive cases. In our study, for the first time, all lophomoniasis patients were confirmed by a PCR-based test.[4] The majority of our cases live in urban areas, and the people living there are more exposed to cockroaches due to the high population of these insects. On the other hand, cockroaches are considered the main reservoir host for the *Lophomonas* parasite.[3,15] As a result, those who live in urban areas may be more at risk.[5,15] Inhaled corticosteroids were utilized to control the underlying condition in more than half of our patients. Although immunosuppression status and/or underlying diseases were observed in a relatively high number of lophomoniasis cases,[4,6,8,16] they were not present significantly in the majority of reported cases worldwide.[3,5,10,12] In

this regard, Fakhar et al. claimed that the parasite mainly affects immunocompetent individuals rather than immunocompromised ones and *Lophomonas* infection is not an opportunistic parasitic agent.[3] As a result, based on what we know so far, corticosteroid therapy and other immunosuppressive status cannot be risk factors for this emerging parasitic infection.[3]

It is noted that the high prevalence (40.4%) of *Lophomonas* infection, which has been reported among children in a study in Iran,[9] is questionable because the microscopic examination has several diagnostic pitfalls: low sensitivity and specificity due to low parasite density in the clinical samples; similarity with other respiratory cells; and low expertise of laboratory technicians.[3–5] Thus, to avoid misdiagnosis or overdiagnosis, it needs to be checked by an expert microscopist, and then, in order to approve it, a PCR test is recommended. However, in our study, lophomoniasis was found in 15.5% (9/58) of the children, based on both microscopic examination and a PCR test. Moreover, nine (77%) of the *Lophomonas*-positive patients were female, which contrasts with several evidence that showed that males were infected in greater numbers.[3,9–11] Chronic cough, low-grade fever, dyspnoea and haemoptysis were the most commonly reported clinical symptoms in paediatrics with lophomoniasis,[9–11] which are similar to our findings with the exception of fever, which none of our patients experienced. As a whole, because some findings of this study differ from that of others, we strongly advise using the PCR approach to diagnose lophomoniasis in large paediatric populations, which may help to clarify the exact burden and unknown clinical aspects of this infection. Finally, increasing global collaborations among researchers on lophomoniasis can lead to overcome challenges regarding many aspects of the disease in the present era.[17]

CONFLICT OF INTEREST

None declared.

AUTHOR CONTRIBUTION

Farzad Masiha and Ali Sharifpour were involved in BAL sampling and patient management. Amirmasoud Taheri and Mahdi Fakhar wrote the manuscript. All authors contributed to editing of the manuscript and approved the final version of the manuscript.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

ETHICS STATEMENT

The authors declare that appropriate written informed consent was obtained for the publication of this manuscript.

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REFERENCES

1. Bourgeois F, Valim C, Wei J, McAdam A, Mandl K. Influenza and other respiratory virus–related emergency department visits among young children. Pediatrics. 2006;118(1):e1–8.
2. Kunst H, Mack D, Kon O, Banerjee A, Chiodini P, Grant A. Parasitic infections of the lung: a guide for the respiratory physician. Thorax. 2010;66(6):528–36.
3. Fakhar M, Sharifpour A, Nakhaei M, Banimostafavi ES, Ghasemi M, Abedian S, et al. Lophomonas and lophomoniasis: biology, etiology, epidemiology, pathogenesis, diagnosis and treatment. 1st ed. Gorgan: Noruzi Publisher; 2021.
4. Fakhar M, Safanavaei S, Nakhaei M, Esmaeili S, Banimostafavi ES, Spahbodi F, et al. Molecular evidence of upper and lower respiratory infection due to *Lophomonas* in a post-kidney transplantation patient. Clin Case Rep. 2022;10(2):e05492. https://doi.org/10.1002/ccr3.5492
5. Fakhar M, Nakhaei M, Sharifpour A, Safanavaei S, Abedi S, Tabaripour R, et al. Morphological and molecular identification of emerged *Lophomonas blattarum* infection in Mazandaran Province, northern Iran: first registry-based study. Acta Parasitol. 2021;66(4):1510–6.
6. Xue J, Li YL, Yu XM, Li DK, Liu MF, Qiu JF, et al. Bronchopulmonary infection of *Lophomonas blattarum*: a case and literature review. Korean J Parasitol. 2014;52(5):521.
7. Li R, Gao Z. *Lophomonas blattarum* infection or just the movement of ciliated epithelial cells? Chin Med J (Engl). 2016;129(6):739–42.
8. Saldaña N, Javier F, Larrauri F, Trujillo D, Montoya E, De La Garza E, et al. Bronchopulmonary infection by *Lophomonas blattarum* in a pediatric patient after hematopoietic progenitor cell transplantation: first report in Mexico. J Thorac Dis. 2017;9(10):E899–902.
9. Ghafarian N, Bakhtiari E, Berenji F, Nakhaei M, Nakhaei B, Jamali-Behnam F, et al. The study of *Lophomonas blattarum* infection in children with respiratory symptoms: a descriptive clinical study in north east of Iran. Int J Pediatr. 2018;6(6):7797–802.
10. Ding Q, Shen K. Pulmonary infection with *Lophomonas blattarum*. Indian J Pediatr. 2021;88(1):23–7.
11. Failoc-Rojas VE, Iglesias-Osores S, Silva-Díaz H. Lophomonas sp. in the upper and lower respiratory tract of patients from a hospital in Lambayeque, Peru: clinical case studies. Respir Med Case Rep. 2020;31:101142.
12. Fakhar M, Nakhaei M, Sharifpour A, Kalani H, Banimostafavi ES, Abedi S, et al. First molecular diagnosis of lophomoniasis: the end of a controversial story. Acta Parasitol. 2019;64(2):390–3.
13. Cummins AG, Eglinton BA, Gonzalez A, Roberton DM. Immune activation during infancy in healthy humans. J Clin Immunol. 1994;14(2):107–15.
14. Nwachuku N, Gerba CP. Health risks of enteric viral infections in children. Rev Environ Contam Toxicol. 2006;186:1–56.
15. Motevalli-Haghi SF, Shemshadian A, Nakhaei M, Faridnia R, Dehghan O, Malekzadeh Shafaroudi M, et al. First report of Lophomonas spp. in German cockroaches (*Blattella germanica*) trapped in hospitals, northern Iran. J Parasit Dis. 2021;45(4):937–43.
16. Sharifpour A, Zarrinfar H, Fakhar M, Zakariaei Z, Soleymani M, Banimostafavi ES, et al. First report of Lophomonas infection in a patient with AML-2 from Qeshm Island, Persian Gulf, southern Iran. Respirol Case Rep. 2022;10(2):e0906.
17. Keighobadi M, Nakhaei M, Sharifpour A, Khasseh AA, Safanavaei S, Tabaripour R, et al. A bibliometric analysis of global research on *Lophomonas* spp. in Scopus (1933–2019). Infect Disord Drug Targets. 2021;21(2):230–7.

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