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## Authenticity of pulmonary *Lophomonas blattarum* infection: A case report

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**Author contributions:** Meng SS and Lin XH designed the report; Meng SS, Dai ZF, Wang HC, Li YX, and Yang RL collected the patient's clinical data; Wei DD and Lin XH took pictures of light and electron microscopy; Meng SS, Dai ZF, and Lin XH wrote the paper.

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### Abstract

Pulmonary protozoal infections are rare. A 28-year-old woman was admitted to hospital with chief complains of cough, sputum, and dyspnea. The clinical laboratory tests for blood revealed an increased eosinophil percentage of 31.3% and significantly elevated total IgE. The chest computed tomography scan revealed that bilateral bronchial walls were thickening, accompanied with patchy spots scattered throughout bilateral lungs. A suspected multiflagellated protozoan was observed under a light microscope. But some different features were observed by electron microscopy, such as the orientation of flagella and nucleus. Besides, both bronchoalveolar lavage fluid and bronchoscopic brush smears underwent Gram staining and Pap staining, which revealed that numerous respiratory ciliated cells were scattered or accumulated in the sample. Finally, she was diagnosed with eosinophil pneumonia. Metronidazole, bronchodilators, and mucolytics were taken for 5 d and symptoms and pulmonary ventilation function improved. We herein report a case of chronic eosinophilic pneumonia, which was misdiagnosed as multiflagellated protozoan infection, and it is suggested that reliable diagnosis approaches are necessary, rather than clinical symptoms and morphological features.

**Key words:** Multiflagellated protozoan; Respiratory disease; Parasitic infection; Case report

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**Core tip:** *Lophomonas blattarum* is a rare cause of respiratory infection. Nonspecific clinical symptoms and signs confuse diagnosis. On the other hand, it is easily

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misdiagnosed probably because of a set of common morphological features between multiflagellated protozoan and ciliated epithelial cells. Therefore, we reviewed the difficulties encountered during the diagnosis in order to improve the understanding of this disease and reduce the incidence of incorrect and missed diagnoses.

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## INTRODUCTION

*Lophomonas blattarum* (*L. blattarum*) is a protozoan that can cause infection in a variety of tissues and organs<sup>[1,2]</sup>. Clinically, the most common tissue infected is the respiratory tract, and the concomitant symptoms (including cough, sputum, and dyspnea) are similar to those of other respiratory conditions, such as bronchial asthma, pneumonia, bronchitis, or acute exacerbation of chronic obstructive pulmonary disease (AECOPD)<sup>[1,3-5,10,11]</sup>. Clinically significant pulmonary protozoal infections are rare but have been increasingly recognized in the past decades<sup>[2]</sup>. Predisposing factors contain immunosuppression and aging process<sup>[1-2,6-9,11]</sup>. This endocommensal usually parasitizes the intestine of specific arthropods, such as termites and roaches<sup>[12-14]</sup>. Inhalation of aerosols containing *L. blattarum* cysts has been proposed to infect human beings<sup>[14-16]</sup>, but this hypothesis has not been confirmed.

More than 100 cases of *L. blattarum* infection have been reported since the first case emerged in 1993. The vast majority of studies reported *L. blattarum* infection based on morphology under a light microscope. Since 2011, several reports have indicated that a few studies misidentified respiratory ciliated cells as *L. blattarum* or multi-flagellated protozoans<sup>[17-20]</sup>. The researchers assumed that despite some similar features between *L. blattarum* and respiratory ciliated cells, which are difficult to differentiate, a set of morphological features are unique to *L. blattarum* or respiratory ciliated cells under a light microscope. The morphological features observed by light microscopy are insufficient. Here, we present a case of chronic eosinophilic pneumonia that was initially misdiagnosed as a multi-flagellated protozoan infection.

## CASE PRESENTATION

A 28-year-old female college teacher was admitted to Huaihe Hospital affiliated to Henan University on October 21, 2016, with chief complaints of cough, sputum, and dyspnea for 3 years without fever. Past medical history included allergic rhinitis for 5 years, and her father had a history of bronchial asthma. Physical examination of the patient showed that breath sounds from both lungs were rough, without rales or rhonchi, and other vital signs were as follows: blood pressure, 16.7/9.3 kPa, pulse rate, 84 bpm, respiratory rate, 21 breaths/min, and body temperature, 36.6 °C. The clinical laboratory blood tests revealed that the white blood cell count was  $8.73 \times 10^9/L$ , with 31.3% of eosinophils. The C-reactive protein was 0.1 mg/L (0-8.2mg/L), the erythrocyte sedimentation rate was 13 mm/hr (0-20mm/hr), and total IgE significantly increased. A chest computed tomography scan revealed that the bilateral bronchial walls showed thickening, accompanied with patchy spots scattered throughout the bilateral lungs (Figure 1), but enlarged mediastinal lymph nodes were not observed. Pulmonary function suggested a slightly weakened pulmonary ventilation function. The laboratory studies for tuberculosis and fungi were negative; however, the serum procalcitonin level was slightly increased. Meanwhile, the bronchoalveolar lavage fluid (BALF) was collected with a bronchofiberscope, and bronchoscopic brush smears were generated for cytologic and etiological diagnosis.

Bronchofiberscope analysis indicated that the bronchial mucosa showed hyperemia, edema, and congestion, especially in the right superior lobe (Figure 2). Under a light microscope, the cilia on the top of the cell oscillated rapidly to drive cell migration, and thus, observation of possible multi-flagellated protozoans in the BALF was reported (Figure 3). Subsequently, the patient received treatment with a bronchodilator agent and metronidazole. To investigate the ultrastructures of the



**Figure 1** High resolution axial computed tomography image showing bilateral bronchial wall thickening accompanied with patchy spots scattered throughout bilateral lungs.

potential multi-flagellated protozoans, we performed scanning electron microscopy (Figure 4), and columnar cells with a cluster of cilia at the top were found. A spherical nucleus was located in the basal region, with cilia at the apical end of the cell. Both BALF and bronchoscopic brush smears underwent Gram staining and Pap staining (Figure 5). Instead of multi-flagellated protozoan, numerous respiratory ciliated cells were scattered or accumulated in the sample.

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## FINAL DIAGNOSIS

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Eventually, she was diagnosed with chronic eosinophilic pneumonia.

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## TREATMENT

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Intravenous metronidazole was administered for 5 d. Given the cytologic diagnosis, metronidazole was contraindicated, and bronchodilator, mucolytic, and nutritional therapies were used.

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## OUTCOME AND FOLLOW-UP

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Intravenous metronidazole was administered for 5 d. The patient reported that the symptoms were improved. Metronidazole was contraindicated, and bronchodilator, mucolytic, and nutritional therapies were used. Three days later, pulmonary ventilation function improved substantially, and tests for tuberculosis, fungi, and *Aspergillus fumigatus* were negative three times. However, the eosinophil number and percentage were highly increased to  $2.57 \times 10^9/L$  and 29.3%, respectively.

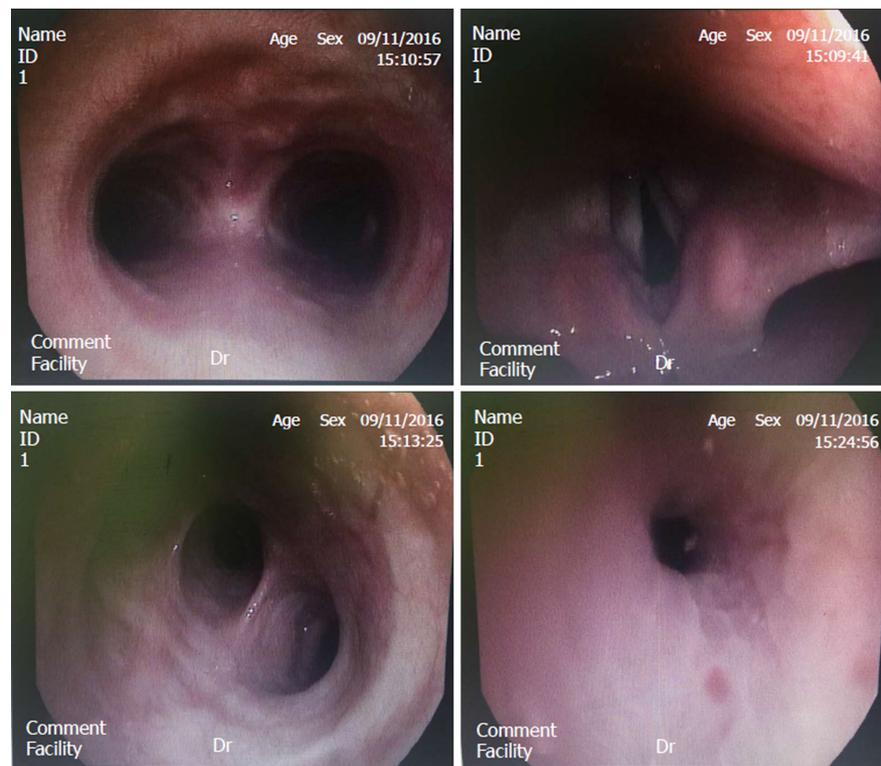
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## DISCUSSION

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In recent decades, more than 100 cases of *L. blattarum* infection, most of which were observed in Chinese adults, have been reported. The possible links between *L. blattarum* infection and insects in the domestic environment have been explored<sup>[13-16,21,22]</sup>. *L. blattarum* is widely believed to parasitize the colon of cockroaches<sup>[23]</sup>, and the parasite could be discharged through the secretions and excrement of the host, which are spread by contaminated clothing and food. Inhalation of *L. blattarum* cysts was suggested to be a route of transmission of this protozoan from cockroaches to human beings<sup>[1]</sup>.

In this case, the patient had been abroad in Belarus from 2010 to 2012. Some cockroaches were found in garderobe, and insecticides were used. Due to the patient's history of allergic rhinitis, long-term usage of corticosteroids could cause immunosuppression, which has been demonstrated to increase the risk of bacterial, viral, and fungal infections<sup>[2,6-9,24]</sup>. Based on these factors, we initially misidentified the respiratory ciliated cells as multi-flagellated protozoans. It is difficult to distinguish *L. blattarum* and ciliated epithelial cells based on morphology under a light microscope. There are several common morphological features. First, both are round and oval in



**Figure 2** Bronchofiberscopic images showing that the bronchial mucosa had hyperemia, oedema and congestion, especially in the right superior lobe.

shape, but the cells are sometimes columnar<sup>[1]</sup>. Second, both cell sizes are similar, with *L. blattarum* ranging from approximately 15  $\mu\text{m}$  to 40  $\mu\text{m}$ , comparable to that of the ciliated epithelial cells<sup>[25,26]</sup>. Third, movements of flagella and cilia, which are found in *L. blattarum* and ciliated epithelial cells, respectively, are observed by light microscopy in fresh samples. Fourth, due to the degeneration of ciliated cells in the bronchial secretion, the nuclei become misty and may even disappear, and the nuclei of *L. blattarum* are not always present. Due to these morphological similarities, ciliated bronchial cells may have been erroneously identified as multi-flagellated protozoans. In addition, nonspecific symptoms, such as cough, sputum, and dyspnea, can not help with the identification, although specific pathogens, such as bacteria and fungi, were not detected. All of these symptoms could be attributed to AECOPD or interstitial pneumonia. Specific diagnosis cannot be made based on laboratory studies and imaging analyses (only subsidiary diagnoses).

Judgements based on light microscopic structures and clinical symptoms without the confirmation of electron microscopy or molecular evidence are inadvisable. By electron microscopy, the morphological features of flagellated protozoa and ciliated epithelial cells are clear<sup>[27]</sup>. The nuclei of ciliated epithelial cells are located at the basal end, with cilia at the apical end of the cell, while flagellated protozoa show granular cytoplasm, phagocytic particles, and a tuft of flagella. In addition to these characteristics, the cilia are regularly oriented, whereas the flagella have an irregular arrangement. Significantly, electron microscopy showed some ultrastructures of flagellated protozoa that are not found in ciliated epithelial cells, such as the calyx, axial filament, and parabasal body<sup>[27,28]</sup>. These peculiar structures could promote temporary motility, while the ciliated epithelial cells could show similar mobility under a light microscope, even if detached from the airway epithelium for a long period<sup>[29]</sup>. Thus, movement detected by light microscopy is not a distinguishing feature.

Most reported cases in recent decades showed a positive response after metronidazole treatment, as in this case. The improvement was not only attributed to the broad spectrum property of this antibiotic, especially against anaerobic bacteria, but also to the administration of bronchodilator agents and physiotherapeutics, such as noninvasive ventilation. Although eosinophilia and increased total IgE indicated parasite or protozoan infection, direct evidence, such as isolation and culture, was inadequate, and images of light microscopy were not sufficient for a conclusive identification. Approximately 30 cases had eosinophilia<sup>[29]</sup>, accounting for one-third of the total cases. Protozoan infection in these cases, which would explain the eosinophil



**Figure 3** Living cells with cilia in bronchoalveolar lavage fluid. Left arrow: The cilia on the top of the cell oscillate rapidly to drive cell migration; Right arrow: The body of the cell.

counts, could be re-examined. Another possible issue is that the source of samples, sputum and throat swabs, could be contaminated with oral flora and food materials. Finally, except for 10 cases from abroad<sup>[1]</sup>, the majority of cases were reported in China; however, there is no evidence that flagellated protozoan infection is an endemic disease.

In this case, we used approaches including light microscopy, scanning electron microscopy, BALF and bronchoscopic brush smears staining for diagnosis. Although multi-flagellated protozoan morphological features were distinguished from bronchial ciliated epithelia cells by scanning electron microscopy, transmission electron microscopy is essential for exploring internal structures.

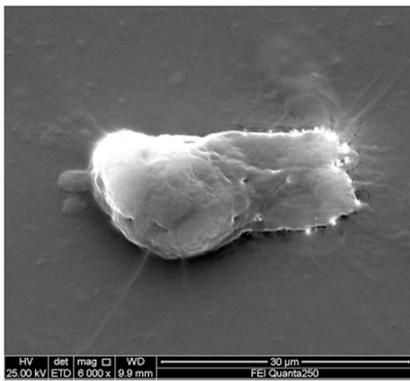
In conclusion, this case illustrated the misdiagnosis of multi-flagellated protozoan infection, although predisposing factors were compatible with those of published cases, including eosinophilia, immunocompromised respiratory symptoms, and suspected protozoa under a light microscope. It is difficult to distinguish these protozoa from normal bronchial ciliated epithelia cells only *via* light microscopy, electron microscopy, or molecular evidence, but isolation and culture may be worth pursuing. In contrast to these characteristics, clinical responses may confuse the diagnosis. Metronidazole is used to treat a wide range of anaerobic bacteria, which are poorly detected by routine testing. However, a combination with multiple agents improved clinical manifestations. Furthermore, the route of transmission, pathogenesis, and molecular techniques should be further investigated.

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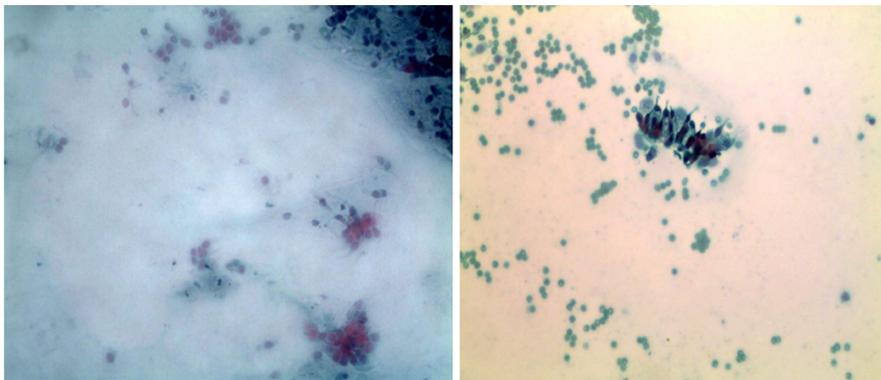
## CONCLUSION

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*L. blattarum* is a rare cause of respiratory infection. Nonspecific clinical symptoms and signs, even laboratory routine test and positive treatment, could not help for identification. Microscopic examination of sputum smear is fast for initial and presumptive diagnosis, but it is not credible due to morphological similarities of ciliated epithelial cells. Combination of smear staining, electron microscopy, and even molecular techniques should be applied for diagnosis.



**Figure 4 Morphological characteristics under a scanning electron microscope.** The nucleus of ciliary active cells is far away from the ciliate tip of the cell, and is located at the bottom of the cell.



**Figure 5 Brush smears stained with Pap stain.** Numerous scattered or gathered respiratory ciliated cells are visible.

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