

## Paracoccidioidomycosis due to *Paracoccidioides lutzii* complicated with adrenal injury and pulmonary arterial hypertension

Gilberto Gambero Gaspar<sup>1</sup>, Tiago Alexandre Cocio<sup>1</sup>, Fernanda Guioti-Puga<sup>1</sup>, Erika Nascimento<sup>1</sup>, Alexandre Todorovic Fabro<sup>2</sup>, Marcia Regina von Zeska Kress<sup>3</sup>, Eduardo Bagagli<sup>4</sup>, Roberto Martinez<sup>1</sup>

### ABSTRACT

Paracoccidioidomycosis caused by *Paracoccidioides lutzii* is endemic in the Midwest of Brazil and its clinical spectrum is still little known due to the recent identification of this fungal species. A patient resident in Southeast Brazil, but who had lived for many years in the Midwest region, presented with skin injuries, chronic cough and bilateral adrenal involvement. *Paracoccidioides* spp. was isolated in culture from a skin lesion biopsy. This isolate was later identified as *P. lutzii* using gene sequencing. A favorable initial response to treatment with itraconazole was observed, but a few weeks later, the patient developed respiratory failure and worsening of lung lesions. Evaluation by computed tomography and echocardiography were suggestive of pulmonary arterial hypertension, and a bronchoscopic biopsy showed peribronchial remodeling. The patient completed the antifungal treatment but maintained the respiratory dysfunction. The reported case shows that *P. lutzii* can be isolated from patients in a geographic area far from the place of infection acquisition and that, as *P. brasiliensis*, it can cause adrenal injury and cardio-respiratory complications as a consequence of excessive necrosis and fibrosis.

**KEYWORDS:** Paracoccidioidomycosis. *Paracoccidioides lutzii*. Pulmonary arterial hypertension.

### INTRODUCTION

Since the report of the first case in 1908, paracoccidioidomycosis infection and disease have been attributed to a single species of the dimorphic fungus *Paracoccidioides brasiliensis*. At the beginning of the 21<sup>st</sup> century, some isolates of *Paracoccidioides* spp. had enough genetic polymorphism to be characterized as a new species, named *P. lutzii*<sup>1</sup>. This species has been isolated predominantly from patients in the Midwest region and around the Amazon region of Brazil<sup>2</sup>. The endemic area of *P. lutzii* is still little known and may have expanded, as there is molecular evidence of its presence in rural environments in other regions of Brazil<sup>3</sup>.

Regarding clinical manifestations, paracoccidioidomycosis by *P. lutzii* has been hidden for a long time within the disease caused by *P. brasiliensis*, either due to the lack of knowledge on new species or the similarity of the clinical manifestations related to *Paracoccidioides* species<sup>4</sup>. The number of reported cases of paracoccidioidomycosis caused by *P. lutzii* is still relatively small to fully establish the clinical spectrum of this disease. However, an atypical serological response, with absence or low antibody titer against *P. brasiliensis* antigens, has already

<sup>1</sup>Universidade de São Paulo, Faculdade de Medicina de Ribeirão Preto, Departamento de Clínica Médica, Ribeirão Preto, São Paulo, Brazil

<sup>2</sup>Universidade de São Paulo, Faculdade de Medicina de Ribeirão Preto, Departamento de Patologia e Medicina Legal, Ribeirão Preto, São Paulo, Brazil

<sup>3</sup>Universidade de São Paulo, Faculdade de Ciências Farmacêuticas de Ribeirão Preto, Departamento de Análises Clínicas, Toxicológicas e Bromatológicas, Ribeirão Preto, São Paulo, Brazil

<sup>4</sup>Universidade Estadual Paulista 'Júlio de Mesquita Filho', Instituto de Biociências, Departamento de Ciências Químicas e Biológicas, Botucatu, São Paulo, Brazil

**Correspondence to:** Gilberto Gambero Gaspar  
Universidade de São Paulo, Faculdade de Medicina de Ribeirão Preto, Departamento de Clínica Médica, Av. Bandeirantes, 3900, Monte Alegre, CEP 14049-900, Ribeirão Preto, SP, Brazil  
Tel: +55 16 3602-2468

**E-mail:** [ggaspar@hcrp.usp.br](mailto:ggaspar@hcrp.usp.br)

**Received:** 5 August 2020

**Accepted:** 22 October 2020

been observed in cases of paracoccidioidomycosis in the Midwest region of Brazil<sup>5</sup>. Current evidence suggests that these patients were infected by *P. lutzii*, whose antigens are partially different from those of *P. brasiliensis* used in conventional serological tests<sup>6</sup>.

This report of paracoccidioidomycosis by *P. lutzii* shows that this fungal species can be isolated from patients living far from the known endemic area and shows clinical aspects that have not yet been reported in the disease caused by *P. lutzii*.

## CASE REPORT

A 55-year-old man was admitted to the hospital presenting with widespread skin lesions, fever, chills, asthenia, and a loss of 16 kg in his body weight. The lesions looked like erythematous papules that progressively became painful ulcers. In the past few days, he had been experiencing diffuse pain throughout his body. He has also

reported having dry cough for many years, but without dyspnea or other respiratory complaints.

The examination of the patient revealed numerous skin lesions, including erythematous papules and ulcerations on the head, face, trunk and limbs, which had a slightly elevated and erythematous border, with a depressed center covered with a crust. The lesions were generally circular and had a diameter between 0.5 and 1.5 cm, but the frontal region of the face presented with an ulcer of approximately 3.0 × 1.5 cm (Figure 1). He had a blood pressure of 100 × 70 mmHg, heart rate of 82/min and respiratory rate of 20/min. No lymphadenomegaly, hepatomegaly or splenomegaly were found, and pulmonary auscultation did not reveal any changes.

The patient was born in Realeza, Parana State, Brazil, and his family moved to Corpus Christi, Paraguay, when he was 9 years old, where he stayed for about 10 years. Then, he returned to Brazil, residing in Sete Quedas, Mato Grosso do Sul State, Marcelandia, Mato Grosso State, and



**Figure 1** – Skin lesions caused by the spread of *P. lutzii*: a) Larger ulcer covered by crusts on the forehead and minor lesions on the root of the nose, zygomatic region and extremities of the labial commissure; b) Ulcers on the nape, back and left shoulder; c) Lesions at different stages on the shoulder and right arm; d) Forearm showing nodule and ulcers presenting with high and erythematous edges; e) Lesion with ulcerative crust on the distal phalanx of the middle finger of the left hand; f) Lower limbs showing several different lesions ranging from papular to small nodules with central ulceration.

Ribeirao Preto, Sao Paulo State, several years in each of these municipalities. He has worked for a long time as a truck and tractor driver, transporting manioc and wood, and has lived in a rural area, close to a deforestation area. He had contact with sawmills and prolonged exposure to wood dust. The patient claimed to be a heavy smoker since his youth and that he is used to drinking from 50 mL to 1 L of distilled alcoholic beverages per day. He has also reported a cutaneous leishmaniasis 16 years earlier.

Blood count and blood biochemical tests were normal, except for a high C-reactive protein (12.3 mg/dL). Serological tests for HIV infection, leishmaniasis, Chagas disease, and hepatitis C virus were negative, but serum anti-HBc Ag IgG antibodies were detected. Serological screening for systemic mycoses by the counterimmunoelectrophoresis (CIE) method was negative for histoplasmosis and aspergillosis, but positive for paracoccidioidomycosis with a 1:8 titer against *P. brasiliensis* antigen.

A chest X-ray showed diffuse infiltrates in both lungs, especially in the perihilar region, with a reticular and micronodular appearance and areas of alveolar opacities. The investigation through computed tomography (CT) showed that both adrenals were enlarged and had hypodense areas. Plasma cortisol (7.8 µg/dL) was just above the lowest level of the normal range (5.0 µg/dL). The histopathological examination of a skin lesion collected by biopsy showed granulomatous inflammatory infiltrates with giant cells and yeasts presenting with exosporulation. The culture of the biopsied skin lesion led to the isolation of *Paracoccidioides* spp.

The patient was diagnosed with the chronic form of paracoccidioidomycosis, presenting with lung, skin and adrenal lesions. The antifungal treatment was carried out with itraconazole (400 mg/day), leading to a progressive improvement, except for the appearance of dyspnea during intense physical activities. About 40 days after the beginning of the treatment, the patient had accentuated dyspnea, dry cough and worsening of his physical condition. The skin lesions were healed, but there were crepitations in both lungs, a respiratory rate of 36/min, and arterial O<sub>2</sub> saturation between 87% and 92%. No clinical signs of heart failure were detected and echocardiography was not performed at this time. The radiological pattern of the lungs had worsened (Figure 2). Trimethoprim-sulfamethoxazole, at a dose of 160 mg of trimethoprim was introduced intravenously every 8 h and prednisone was associated at a dose of 20 mg/day, orally. The dyspnea decreased and treatment with itraconazole was reintroduced after 5 days in association with the corticosteroid therapy, maintained for 50 days.

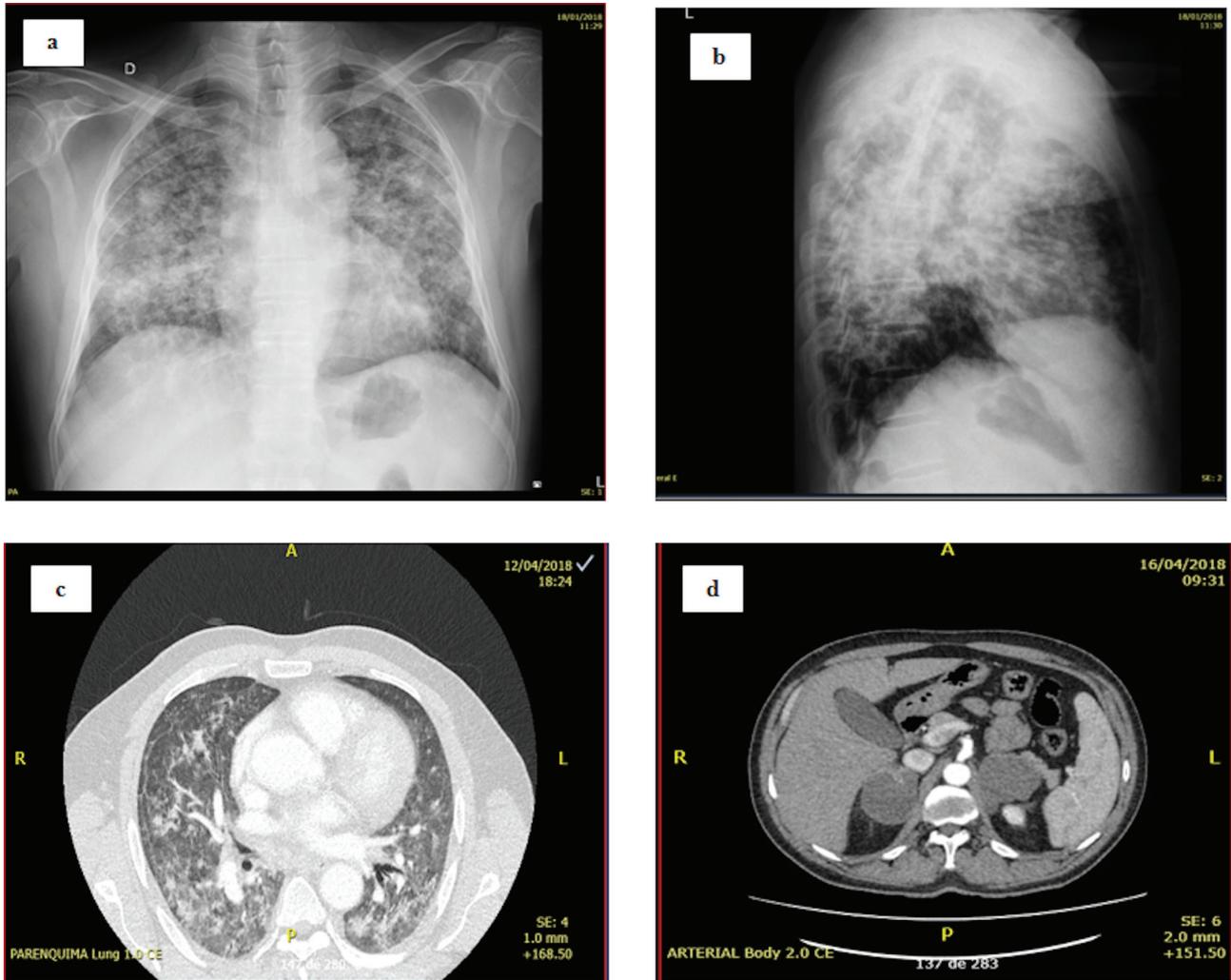
After four months of antifungal therapy, pulmonary vital capacity and airway flow were normal in the spirometry evaluation. A chest CT scan showed thickened bronchi walls, some with dilation, lungs with a diffuse pattern that looked like micronodules, ground-glass opacities and cardiomegaly (Figure 2). Histological examination of tissues obtained by bronchoscopy revealed lymphoplasmacytic infiltrates associated with bronchial and vascular remodeling, aside from distortion of the tissue architecture (Figure 3). Doppler echocardiography showed slight dilation of the left atrium, ascending aorta and pulmonary artery, in addition to pulmonary arterial hypertension (pulmonary artery systolic pressure estimated at 42 mmHg, normal limit < 25 mmHg). The performed tests led to the conclusion that the patient had sequelae of paracoccidioidomycosis, functional changes in the lung and pulmonary circulation. The dose of itraconazole was reduced to 200 mg/day, completing the 17 months of antifungal use.

In the clinical follow up, the patient maintained dyspnea of varied intensities. A new chest CT scan performed eight months after the end of treatment showed a decrease in pulmonary opacities, the presence of bronchiectasis, a slight increase in the right ventricle and dilation of the pulmonary artery trunk (32 mm, normal range: < 29 mm).

Total genomic DNA of *Paracoccidioides* spp. isolated from the patient's lesions was extracted and ITS1-5.8S-ITS2 rDNA, as well as *gp43* exon 2 gene regions were amplified by PCR, purified and sequenced. Gene sequencing was carried out on the platform of the Human Genome and Stem-Cell Research Center, Institute of Biosciences of the University of Sao Paulo. The comparison of the Pb 01 (*P. lutzii*) nucleotide sequences revealed similarities of 99.5% with the ITS1-5.8S-ITS2 rDNA sequence (accession N° EU870297.1) and 100% with the *gp43* exon 2 loci sequence (accession N° EU870196.1). The fungus isolated from the patient was therefore identified as *P. lutzii* and the obtained gene sequences (ITS1-5.8S-ITS2 rDNA and *gp43* exon 2) were deposited in GenBank (accession N° MK909806 and MK886790, respectively)<sup>7</sup>.

## DISCUSSION

The reported case is relevant due to *P. lutzii* successful isolation and unprecedented clinical characteristics regarding this fungal species. The patient lived in recent years in the region of Ribeirao Preto, SP, a municipality located in the Southeast of Brazil, and this fact initially suggested the acquisition of the infection in a place far from the endemic area of *P. lutzii*. This species was also isolated from a patient with the chronic PCM form who had a current and past residence in Londrina, Parana State,



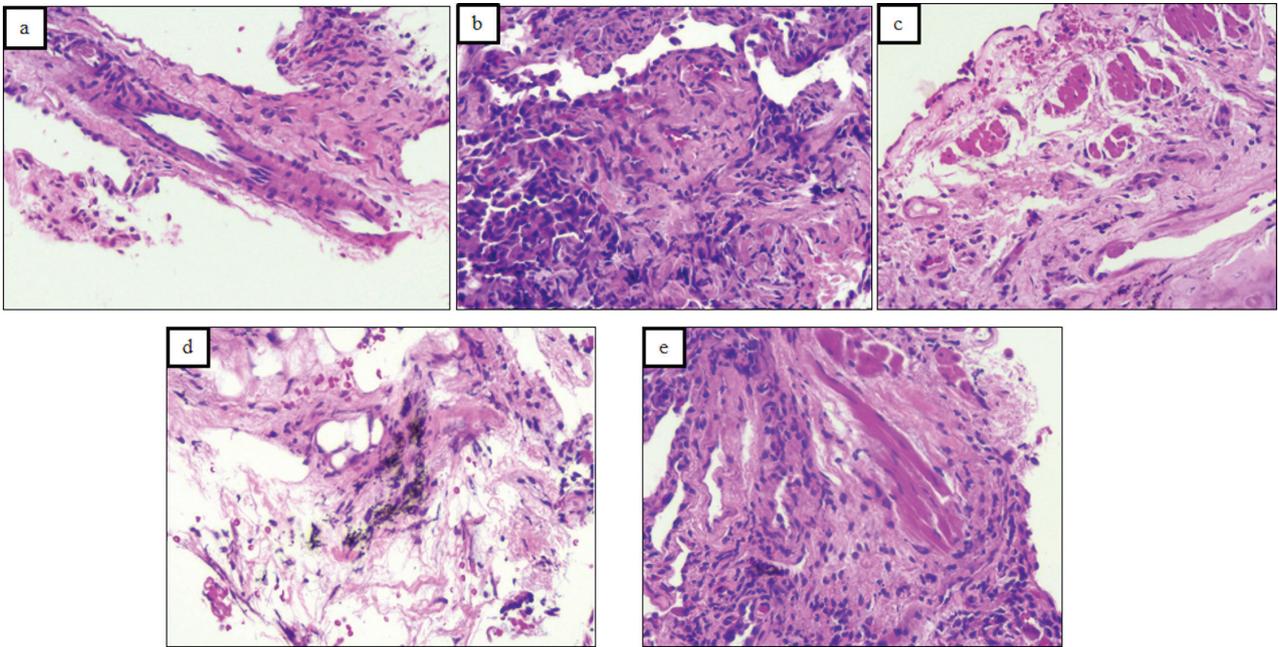
**Figure 2** - Posteroanterior chest radiography (a) lateral position chest radiography (b) after 40 days of antifungal treatment: extensive bilateral interstitial and alveolar opacities, predominating in the middle thirds and bases of the lungs; c) chest computed tomography: ground-glass and reticular diffuse opacities through the lungs, micronodules, bronchiectasis and cardiomegaly; d) computed tomography of the abdomen: very enlarged adrenals with hypodense areas.

and Botucatu, Sao Paulo State, respectively, suggesting that *P. lutzii* infection was acquired in Southeast of Brazil<sup>8</sup>. In the case reported here, it is more likely that the patient acquired *P. lutzii* in the Midwest of Brazil, where he lived for many years and performed a professional activity with prolonged exposure to aspiration of *Paracoccidioides* spp. propagules. Paracoccidioidomycosis caused by *P. brasiliensis* can have a long incubation period, which is apparently valid for *P. lutzii*, allowing this species to be isolated much later outside its endemic region.

According to the classification adopted for *P. brasiliensis*, the patient had the chronic form of paracoccidioidomycosis. He had widespread skin lesion, lung lesion and, as an incidental finding, injury in both adrenals. In a series of cases of *P. lutzii* disease, all patients were classified with the chronic form of paracoccidioidomycosis<sup>9</sup>. The adrenals were supposed

to be compromised by *P. lutzii* due to the extensive fungal dissemination in the patient and also by analogy with the disease caused by *P. brasiliensis*, whose chronic form is more associated with injury and even adrenal insufficiency<sup>10</sup>. The patient did not have Addison's disease, but a frequent subclinical reduction of adrenals function has been observed in paracoccidioidomycosis<sup>11</sup>.

Another interesting aspect of the reported case is the evolution of lung lesions and respiratory symptoms, culminating in pulmonary arterial hypertension. The patient had severe dyspnea, increased pulmonary radiological changes and relative hypoxia a few weeks after the beginning of the antifungal therapy, that improved with the use of corticosteroids. At that time, paracoccidioidomycosis had already been controlled by using itraconazole, considering the regression of skin lesions. Heart failure is a rare adverse affect of itraconazole<sup>12</sup>, but the patient's clinical



**Figure 3** – Histopathological examination of a transbronchial biopsy (HE staining): a) Vascular remodeling: dense adventitious and low cell layer; b) Vascular remodeling: exuberant thickening of the intima layer associated with endothelial hyperplasia; c) Bronchial remodeling: intense deposition of the extracellular matrix in the bronchial structures, low cell mucosa, with distortion of the architecture of the bronchial smooth muscle layer associated with thickening of the basement membrane, slight histiocytic anthracotic pigmentation (lower right quadrant) and secondary lymphatic dilation; d) Histiocytic black anthracotic pigmentation with a lymphatic distribution in the bronchovascular axis; e) Mild lymphoplasmacytic bronchitis associated with an architectural distortion of the bronchial remodeling and anthracotic pigmentation in histiocytes.

conditions excluded this hypothesis. An accentuation of the inflammatory process may take place at the initial stage of the paracoccidioidomycosis treatment, probably due to the pharmacological blocking of the infectious agent, necrosis and immunological recovery. The use of corticosteroids is recommended in patients with clinical complications due to excessive inflammation in *Paracoccidioides* spp. lesions<sup>2</sup>. The evolution towards the cure of the patient was accompanied by histological alterations evidenced in a bronchoscopic biopsy, culminating in fibrosis that caused structural alterations in the lungs, pulmonary arterial hypertension and functional pulmonary impairment. Pulmonary sequelae and respiratory dysfunction in paracoccidioidomycosis by *P. brasiliensis* are not uncommon<sup>13</sup>, as well as pulmonary arterial hypertension<sup>14</sup>. The outcome in the described case shows that it can also occur in the disease caused by *P. lutzii*. Cases progressing to severe lung injury and respiratory dysfunction in paracoccidioidomycosis by *P. lutzii* have already been reported<sup>15,16</sup>.

The reported case shows new aspects of the disease caused by *P. lutzii*, expanding the clinical spectrum related to this species of *Paracoccidioides*. *P. lutzii* cases can occur far from the endemic area of this species, requiring a cautious analysis of data on the geographic distribution of *Paracoccidioides* species.

## ACKNOWLEDGMENTS

Cocio TA and Nascimento E have a doctoral and a post-doctoral scholarship awarded by the Coordination for the Improvement of Higher Education Personnel (CAPES Brazil – Finance Code 001), grants N° 88882.180015/2018-1 and 88882.317609/2019-1, respectively. *Fundação de Apoio ao Ensino, Pesquisa e Assistência* of the *Hospital das Clínicas da Faculdade de Medicina de Ribeirão Preto – USP* (FAEPA) have also provided financial support.

## CONFLICT OF INTERESTS

The authors declare no conflict of interests.

## CONSENT FOR PUBLICATION

The patient signed an informed consent for the publication of the case.

## REFERENCES

1. Teixeira MM, Theodoro RC, de Carvalho MJ, Fernandes L, Paes HC, Hahn RC, et al. Phylogenetic analysis reveals a high level of speciation in the *Paracoccidioides* genus. *Mol Phylogenet Evol.* 2009;52:273-83.

2. Shikanai-Yasuda MA, Mendes RP, Colombo AL, Queiroz-Telles F, Kono AS, Paniago AM, et al. Brazilian guidelines for the clinical management of paracoccidioidomycosis. *Rev Soc Bras Med Trop.* 2017;50:715-40.
3. Arantes TD, Theodoro RC, Teixeira MM, Bosco SM, Bagagli E. Environmental mapping of *Paracoccidioides* spp. in Brazil reveals new clues into genetic diversity, biogeography and wild host association. *Plos Negl Trop Dis.* 2016;10:e0004606.
4. Arantes TD, Bagagli E, Niño-Veja G, San Blas G, Theodoro RC. *Paracoccidioides brasiliensis* and *P. lutzii*, a secret love affair. *Rev Inst Med Trop Sao Paulo.* 2015;57 Suppl 19:25-30.
5. Batista Jr J, Camargo ZP, Fernandes GF, Vicentini AP, Fontes CJ, Hahn RC. Is the geographical origin of a *Paracoccidioides brasiliensis* isolate important for antigen production for regional diagnosis of paracoccidioidomycosis? *Mycoses.* 2010;53:176-80.
6. Gegembauer G, Araujo LM, Pereira EF, Rodrigues AM, Paniago AM, Hahn RC, et al. Serology of paracoccidioidomycosis due to *Paracoccidioidomycosis lutzii*. *Plos Negl Trop Dis.* 2014;8:e2986.
7. Cocio TA, Nascimento E, von Zeska Kress MR, Bagagli E, Martinez R. Phylogenetic species of *Paracoccidioides* spp. isolated from clinical and environmental samples in a hyperendemic area of paracoccidioidomycosis in Southeastern Brazil. *J Fungi (Basel).* 2020;6:E132.
8. Takayama A, Itano EN, Sano A, Ono MA, Kamei K. An atypical *Paracoccidioides brasiliensis* clinical isolate based on multiple gene analysis. *Med Mycol.* 2010;48:64-72.
9. Hahn RC, Rodrigues AM, Della Terra PP, Nery AF, Hoffman-Santos HD, Góis HM, et al. Clinical and epidemiological features of paracoccidioidomycosis due to *Paracoccidioides lutzii*. *Plos Negl Trop Dis.* 2019;13:e0007437.
10. Bellissimo-Rodrigues F, Bollela VR, Fonseca BA, Martinez R. Endemic paracoccidioidomycosis: relationship between clinical presentation and patients' demographic features. *Med Mycol.* 2013;51:313-8.
11. Tóbon AM, Agudelo CA, Restelpo CA, Villa CA, Quiceno W, Estrada S, et al. Adrenal function status in patients with paracoccidioidomycosis after prolonged post-therapy follow-up. *Am J Trop Med Hyg.* 2010;83:111-4.
12. Ahmad SR, Singer SJ, Leissa BG. Congestive heart failure associated with itraconazole. *Lancet.* 2001;357:1766-7.
13. Queiroz-Telles FV, Peçanha Pietrobom PM, Rosa Júnior M, Baptista RM, Peçanha PM. New insights on pulmonary paracoccidioidomycosis. *Semin Respir Crit Care Med.* 2020;41:53-68.
14. Tuder RM, el Ibrahim R, Godoy CE, De Brito T. Pathology of the human pulmonary paracoccidioidomycosis. *Mycopathologia.* 1985;92:179-88.
15. Marques-da-Silva SH, Rodrigues AM, de Hoog GS, Silveira-Gomes F, Camargo ZP. Occurrence of *Paracoccidioides lutzii* in the Amazon region: description of two cases. *Am J Trop Med Hyg.* 2012;87:710-4.
16. Hahn RC, Rodrigues AM, Fontes CJ, Nery AF, Tadano T, Queiroz LP, et al. Fatal fungemia due to *Paracoccidioides lutzii*. *Am J Trop Med Hyg.* 2014;91:394-8.