

Value of exfoliative cytology in diagnosis of oral paracoccidioidomycosis - case report

Wartość cytologii eksfoliatywnej w rozpoznawaniu parakokcydioidomikozy jamy ustnej - opis przypadku

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ABSTRACT:

Paracoccidioidomycosis is a systemic mycosis caused by inhalation of the fungus *P. brasiliensis*. The primary infection involves the lungs and can spread to other organs and systems resulting in secondary lesions in the mucosa, lymph nodes, skin and adrenal glands. Oral lesions are insidious in progression and can be multifocal. The aim of this study was to report a case of paracoccidioidomycosis in a patient who was misdiagnosed with pulmonary tuberculosis. A 40-year-old male, HIV-positive, was admitted with complaints of weight loss, asthenia, diarrhea, coughing and dyspnea. Physical examination revealed the presence of caries, coated tongue, residual roots, oral candidiasis and ulcerated, granulomatous lesions in the hard palate and alveolar ridge. Laboratory tests revealed anemia, neutrophilia and an increased erythrocyte sedimentation rate. Cytology and incisional biopsy were performed. The cytological examination revealed the presence of fungus and the pathological examination confirmed the diagnosis of paracoccidioidomycosis. The doctors who previously made the diagnosis of tuberculosis were informed of the new diagnosis, as the patient was being treated for tuberculosis. The treatment for paracoccidioidomycosis was initiated and the patient is under clinical monitoring.

KEYWORDS:

mouth mucosa; Paracoccidioidomycosis; Cytodiagnosis; Dermatomyces

STRESZCZENIE:

Parakokcydioidomikoza jest grzybicą układową, wywołaną przez inhalację grzyba z gatunku *P. brasiliensis*. Zakażenie pierwotne obejmuje płuca i może rozprzestrzeniać się do innych narządów i układów w wyniku wtórnych zmian obejmujących błony śluzowe, węzły chłonne, skórę i nadnercza. Zmiany w obrębie jamy ustnej są podstępne w progresji, wieloogniskowe i mogą dotyczyć różnych obszarów anatomicznych. Celem niniejszego badania było opisanie przypadku parakokcydioidomikozy u pacjenta, u którego błędnie rozpoznano gruźlicę płuc. Czterdziestoletni mężczyzna, murarz, zakażony wirusem HIV, został przyjęty do szpitala Oswaldo Cruz (Curitiba) z powodu zgłaszanej utraty masy ciała, osłabienia, biegunki, kaszlu i duszności. Badanie przedmiotowe wykazało obecność próchnicy, nalotu na języku, pozostałości korzeni zębów, kandydozy jamy ustnej oraz zmian owrzodzeniowych o charakterze ziarniniakowym w obrębie podniebienia twardego i wyrostka zębodołowego. Badania laboratoryjne wykazały niedokrwistość, neutrofilie i podwyższony odczyn OB (szybkość opadania krwinek czerwonych). Wykonano cytologię oraz biopsję wycinkową. Badanie cytologiczne wykazało obecność grzyba, a badanie patologiczne potwierdziło rozpoznanie parakokcydioidomikozy. Lekarze zostali poinformowani o diagnozie u pacjenta, który był leczony z powodu gruźlicy. Ustalono sposób leczenia parakokcydioidomikozy, a chory pozostaje pod kontrolą kliniczną.

SŁOWA KLUCZOWE:

błona śluzowa jamy ustnej, parakokcydioidomikoza, cytodiagnostyka, grzybice skóry

INTRODUCTION

Paracoccidioidomycosis is a chronic, fungal, granulomatous disease with an insidious presentation. *Paracoccidioides brasiliensis* is the etiological agent, and it is present in plants and soil. This infection occurs more frequently in countries in South America where contact with the ground is more frequent. The most common mode of entry for the fungus is through an inhalation into the respiratory tract of the host. The lungs and upper airways are the initially affected sites. However, paracoccidioidomycosis may spread to many organ systems and tissues of the human body via blood or lymph [1].

Oral lesions of paracoccidioidomycosis manifest as exulcerations and ulcers with irregular borders. Their surfaces are granulomatous with a yellow background interspersed with bleeding points. They cause a lot of pain during brushing of teeth and eating, which leads to anorexia and weight loss [2].

The histological aspect is characterized by a granulomatous, inflammatory reaction, suppuration, presence of fungi of multiple sporulation, which produces the pathognomonic signs of a “steering wheel”, “wheel” and “Mickey Mouse ears”. Paracoccidioidomycosis is endemic and one of the ten most common causes of morbidity and mortality in Brazil. It infects children and teenagers in its acute form and adults over thirty years of age in a subacute or chronic form. Brazil has the largest number of cases, followed by Colombia and Guatemala. Regions with tropical and subtropical forests, with temperatures between 14 to 20° C, relatively high humidity and higher rainfall favor the development of the fungus [3].

Paracoccidioidomycosis has a male-to-female ratio of 5.4. However, it can affect both sexes equally after 50 years of age. People with suppressed immunity, especially with HIV/AIDS, as well as land workers in their various fields of activity are most susceptible. The mortality rate of paracoccidioidomycosis ranges from 2 to 30% in HIV/AIDS patients [3].

The diagnosis of this disease is based on signs and symptoms and it is confirmed by laboratory tests. In some cases, differential diagnosis of paracoccidioidomycosis becomes difficult, because the lymphatic involvement can simulate Hodgkin's disease and other malignant diseases. Additionally, lesions may resemble leishmaniosis, histoplasmosis or tuberculosis. The treatment of paracoccidioidomycosis is still widely performed with sulfamethoxazole, trimethoprim combined with itraconazole. Amphotericin B is used for the treatment of more severe cases. The aim of this study was to report a case of paracoccidioidomycosis in a patient with HIV/AIDS [4,5].



Fig. 1. Flat, ulcerated lesions on the hard palate

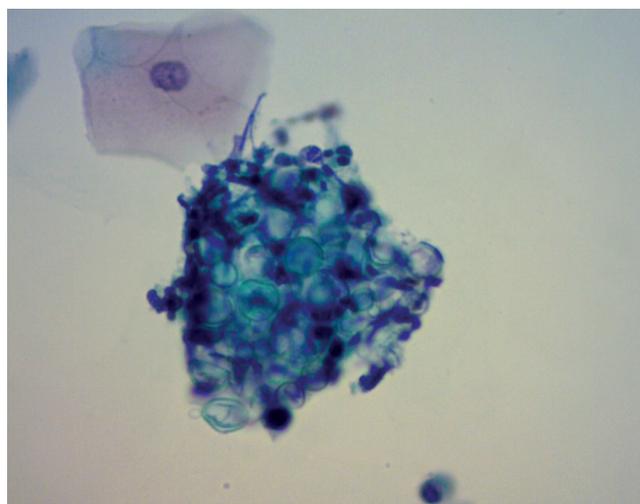


Fig. 2. Oral smear showing cluster-like structures representing fungi (Papanicolaou x400).

REPORT OF CASE

A 40-year-old, black male, a mason, was admitted with complaints of weight loss, asthenia, diarrhea, coughing and dyspnea. According to the patient, he lost almost 40 kilograms (kg) over two months. The patient had HIV/AIDS for over 10 years and used antiretroviral therapy irregularly. Medical doctors requested additional tests to investigate pneumocystosis and tuberculosis. The dental examination revealed that the patient had a precarious oral condition with the presence of caries, residual roots, tongue coating and pseudomembranous oral candidiasis in the soft palate, bilateral buccal mucosa and tongue dorsum. In addition, two large ulcerated lesions of a granulomatous character were identified in the hard palate and

alveolar ridge (Figure 1). These lesions were flat, painful and had irregular edges. Laboratory tests revealed anemia (erythrocytes 2.61 M/ μ L, and hemoglobin = 9.30 g/dL), neutrophilia (leukocytes = 880 k/ μ L, neutrophils = 90%) and an elevated erythrocyte sedimentation rate (ESR = 120 mm).

Exfoliative cytology and incisional biopsy were carried out on the oral lesions. The cytological examination was performed with a cytological kit (Liqui-PREP™ - Cellular base solution). The collected material was centrifuged, which was followed by slide preparation and Papanicolaou staining. The evaluation of the smear was performed under a light microscope (Eclipse E200, Nikon, Japan) with a 10x and 40x objective lens. Oral cytology stated that the patient was negative for squamous dysplasia, but accentuated inflammatory changes were present. Yeast budding structures suggestive of *Paracoccidioides* spp. were observed (Figure 2) along with leukocytes and histiocytes with moderate presence of inclusion bodies.

A chest x-ray revealed the presence of pronounced, diffuse reticulonodular opacities in the middle lobes with a consolidation with butterfly wing features, suggestive of paracoccidioidomycosis. Cytology revealed the presence of cell structures compatible with fungi, and the pathological examination confirmed the diagnosis of paracoccidioidomycosis (Figures 3, and 4). The doctors who previously made the diagnosis of tuberculosis were informed of the new diagnosis, as the patient was being treated for tuberculosis. The treatment of paracoccidioidomycosis was instituted (80mg trimethoprim + 400mg sulfamethoxazole) and the patient is under clinical monitoring.

DISCUSSION

Ulcerative lesions in the mouth can be a manifestation of a spectrum of clinical conditions, such as: a) an epithelial damage resulting from trauma, b) an immune attack as in the case of Lichen planus, pemphigoid or pemphigus, c) autoimmune deficiency, including AIDS and leukemia, d) infection, including tuberculosis, syphilis and herpes simplex, or e) nutritional changes, including vitamin deficiencies and some gastrointestinal diseases [6].

The patient in this case report had multiple lesions in the mouth and systemic symptoms (weight loss, asthenia, diarrhea, coughing and dyspnea). In addition, he was infected with HIV and did not take antiretroviral medications on a regular basis. Given these findings, the clinician may initially try to associate clinical signs with oral lesions found in the course of HIV/AIDS that are part of the EC-Clearinghouse classification. In this classification, oral lesions are divided into three categories ac-

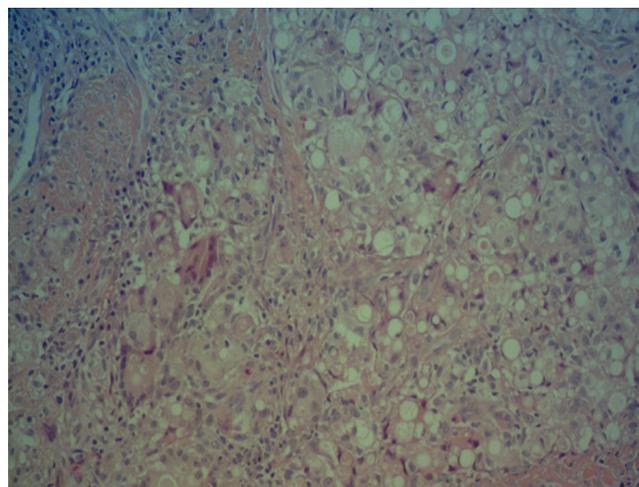


Fig. 3. Granulomatous inflammatory reaction with giant cells and rounded structures similar to fungi (HE x100)

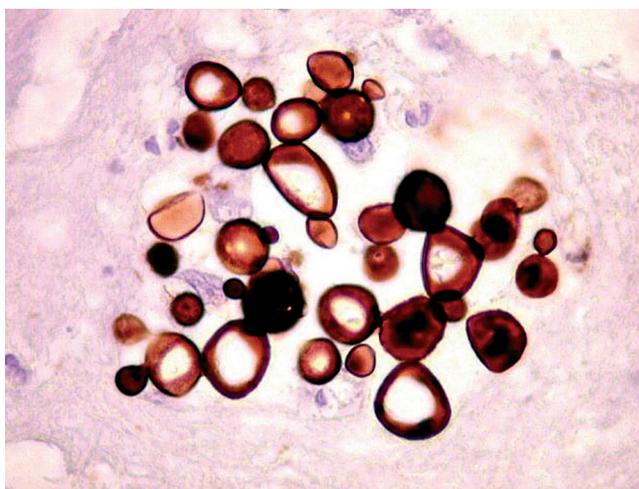


Fig. 4. Fungal structures in sporulation with the sign of "steering wheel" (Gomori-Grocott x400).

ording to their degree of association with HIV infection. The first category includes lesions that are strongly associated with HIV, such as candidiasis, hairy leukoplakia, Kaposi's sarcoma, non-Hodgkin's lymphoma and periodontal disease [7]. However, none of these diseases causes ulcerated lesions similar to those found in our patient. The second category includes some bacterial and viral lesions [7]. As the patient had granulomatous ulcers, this excluded a viral nature of the lesions. Therefore, only oral lesions induced by tuberculosis would be the remaining hypothesis. Oral lesions are uncommon in tuberculosis, and when they occur, they present as nodular, granular or ulcerated areas or as firm leukoplakia [8].

The lesions seen in HIV-infected patients represent the third category of the EC-Clearinghouse classification that includes

some bacterial, fungal and neurological disorders [7]. According to this classification, lesions associated with histoplasmosis and cryptococcosis should be the first differential diagnoses, as they both initially affect the lungs and then spread to other tissues. However, cryptococcosis rarely affects the oral cavity [8]. On the other hand, the classification of EC-Clearinghouse does not include oral lesions induced by paracoccidioidomycosis.

Paracoccidioidomycosis is a tropical, fungal disease caused by *Paracoccidioides brasiliensis*. It has a high prevalence in Latin America, especially in Brazil, where it represents a public health problem due to its highly incapacitating potential [9]. With the advent of HIV infection in the world, a large number of paracoccidioidomycosis/HIV coinfection cases was to be expected. Fewer cases of paracoccidioidomycosis/HIV coinfection have been described in comparison to other mycoses such as candidiasis, histoplasmosis and cryptococcosis. This is perhaps because paracoccidioidomycosis is not a disease that defines HIV/AIDS. Moreover, the use of trimethoprim combined with sulfamethoxazole as a prophylaxis for pneumocystosis is also very effective against *Paracoccidioides brasiliensis* [10]. Another fact to consider is that this disease affects predominantly individuals from rural areas and agricultural workers who are in direct contact with the soil, such as farmers, gardeners and construction workers.

Oral paracoccidioidomycosis has been diagnosed mostly by histopathological examinations of biopsied lesions. Serological tests have also been used as a diagnostic tool in patients

without oral or cutaneous disease, whose specimens are difficult to obtain, visualize or culture, especially in the case of deep-seated lesions [11]. Cytology is a simple, inexpensive and fast examination that can be used in the diagnosis of various oral diseases, such as squamous cell carcinoma, herpes simplex virus, candidiasis, pemphigus vulgaris and various forms of anemia. [12 -16]. Furthermore, it also can be used to evaluate the effect of external agents on the oral mucosa [17,18]. Several authors have shown that swabbing of oral lesions is a useful aid in the diagnosis of paracoccidioidomycosis [18-20]. Cabral et al. (2010) reported a case of a male patient with HIV who presented with mulberry-like lesions on the palate. The diagnosis was made by exfoliative cytology and Papanicolaou staining. According to these authors, swabs from paracoccidioidomycosis lesions exhibit fungal structures with birefringent walls and exosporulation conferring an airplane, radial, motor appearance or even bowel-like or goblet-like forms compatible with *Paracoccidioides brasiliensis* [21]. All these microscopical features were observed in smears collected from this patient. In addition, some authors argue that due to the formation of microabscesses in the epithelium, it is possible to observe various forms of *P. brasiliensis* in cytological preparations [8,22].

Another advantage of using exfoliative cytology is that it can be used in immunocompromised patients who are unable to undergo a more invasive procedure (biopsy). Furthermore, the smears were stained with Gomori-Grocott staining, allowing for an easy and quick visualization of the fungus and enabling monitoring during and after treatment [17].

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