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CLINICAL AND MYCOBIOLOGICAL PROFILE OF OTOMYCOSES IN THE AMAZON, BRAZIL

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Abstract: Objective: To comprehend the mycobiological profile of patients presenting with otologic complaints of pruritus, otalgia, auricular fullness, and hypoacusis at the Ambulatório Araújo Lima in Manaus. **Methods**: Initially, the sample was selected through appointments at the otolaryngology outpatient clinic of the service in Manaus, obtaining informed consent, filling out a questionnaire, evaluating inclusion and exclusion criteria, followed by the collection of otologic secretion samples, treatment prescription, and after one month, treatment outcomes were assessed, and new samples were collected.

Results: 50 patients were included in the study, with 26 (52%) being female and 24 (48%) male, with pruritus being the most prevalent symptom present in 74%. Aspergillus was the most prevalent fungus, found in 46% of the sample**. Conclusion**: This study demonstrated that the most affected population were individuals manipulating the external auditory canal or using headphones, with females being more prevalent. Aspergillus was the most commonly found fungus.

Keywords: Otomycosis, otitis, external otitis.

INTRODUÇÃO

The state of Amazonas has, due to its geographical location and hot, humid climate, characteristics that favor the emergence of mycoses in general. Therefore, the study of otomycoses in the city of Manaus is of paramount importance because these features predispose to the occurrence of the pathology in question, often not receiving effective treatment due to being underdiagnosed.

Otomycosis is a term used to describe infections of the epithelium of the external auditory canal (EAC) caused by yeasts and filamentous fungi[1]. The main symptoms include intense otalgia, purulent otorrhea, and varying degrees of hypoacusis. A recent increase in the incidence of fungal otitis may be associated with the widespread use of broad-spectrum systemic antibiotics and the growing use of topical fluoroquinolone-based antibiotics.[2]

Although several fungal species are found in otomycosis, Aspergillus sp and Candida sp are the most common species isolated from the external auditory canal of patients. Otomycosis has a global distribution with prevalence ranging from 9% to 30% among patients presenting signs and symptoms of external otitis and ear discharge in otorhinolaryngology clinics.[3]

It should be noted that even in the ear considered normal, there may be growth of saprophytic fungi with low pathogenicity. However, pathological proliferation is found mainly when there is chronic bacterial infection and obstructive entities such as a foreign body or tumor of the acoustic meatus[4]. Other predisposing factors include the use of hearing aids or individual sound amplification devices, self-induced trauma (use of cotton swabs for ear cleaning), swimming, broad-spectrum antibiotic therapy, use of corticosteroids and cytostatics, neoplasms, immune alterations, or debilitating illnesses[5].

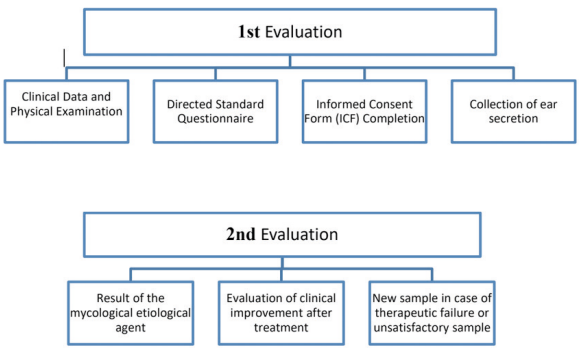
METHODS

In the present study, 50 patients aged between 14 and 78 years old with reports of otologic symptoms such as pruritus, hypoacusis, auricular fullness, otorrhea, and tinnitus, along with an otoscopy compatible with otomycosis visualized through mycelia in the external auditory canal, were included.

Patients with otologic symptoms not compatible with otomycosis were excluded. Patient recruitment was based on spontaneous demand, through appointments at the otorhinolaryngology outpatient clinic at the Ambulatório Araújo Lima (AAL) in the city of Manaus. Subsequently, questionnaires were

filled out regarding otologic symptoms and habits that could influence the condition, such as the use of cotton swabs, hearing aid use, swimming habits, and/or diving, as well as manipulation of the canal with objects. During the in-person appointments for the objective and subjective analysis of the patients' dysfunction in the study, safety protocols and social distancing measures were appropriately observed. The in-person assessment consisted of three different moments. After the initial contact, material collection and proposed treatment, a follow-up assessment was scheduled for the following month.

During the initial assessment, demographic data such as gender, age, city, profession, occupational occupation, clinical data related to otologic complaints, and the recording of comorbidities were collected. Comorbidities included hypertension, diabetes mellitus, immunosuppression, chronic corticosteroid use, presence of mycosis in other areas of the body, previous or current treatment with antibiotics, corticosteroids, or antifungals, drug allergies, and continuous use medications.



Flowchart detailing the study's stages:

SAMPLE SELECTION

Following the approval by the Research Ethics Committee of HUGV, patients were recruited based on spontaneous demand for care at the Otorhinolaryngology Service of AAL, UFAM Faculty of Medicine, in the city of Manaus. Upon meeting the inclusion criteria for sample selection, a clinical analysis and physical examination were conducted following a standard data collection questionnaire, provided in the Informed Consent Form (ICF). Subsequently, questionnaires regarding otological and general symptoms, prior treatments, medication usage, and habits were completed. The questionnaires were validated based on the inclusion and exclusion criteria. Following this, the research team collected ear material from the patient, placing it in a sterile container for subsequent laboratory analysis.

The affected auricle and external auditory canal were cleaned with alcohol (70% isopropanol) prior to sample collection, which was performed using properly sterilized curettes and Hartmann forceps. Approximately 2 to 3 samples were taken and placed in sterile containers, then immediately transported to the mycology department of Getúlio Vargas University Hospital (HUGV) for direct examination and culture.

The samples were mounted on slides using a 20% potassium hydroxide (KOH) solution. After an incubation period of 2 to 24 hours, the slide is examined under a microscope using 10X and 40X objectives to identify fungal mycelia, budding yeast, and associated structures.

Various fungal forms become detectable in direct smears, depending on the type of organisms. Septate hyphae, yellow or black-brown conidia, occasionally with fruiting heads, are fungal elements associated with Aspergillus species, while broad, non-septate, ribbon-like hyphae are common for zygomycetes fungi. On the other hand, septate hyaline hyphae with reproductive structure indicate hyalohyphomycetes such as Penicillium sp., when associated with other causative agents of otomycosis. In contrast, yeasts, particularly those of Candida species, are characterized by the presence of clusters of

blastoconidia and pseudohyphae (SANGARÉ *et al.*, 2021).

Another portion of the sample was inoculated onto the surface of two tubes containing Sabouraud dextrose agar supplemented with 0.05 mg/mL of chloramphenicol and mycosel agar. The tubes were incubated at 37°C and 27°C for 2-3 weeks with daily examination for the presence of growth from days 3-4.

All patients diagnosed with otomycosis underwent cleaning of the external auditory canal by aspiration and removal of fungal debris. Following this procedure, patients received prescriptions for topical miconazole cream once daily or drops, using 4-5 drops, 3 times a day for a minimum of 7 days and a maximum of 14 days. In cases of associated external otitis or acute exacerbation of chronic otitis, weekly cleaning of the external auditory canal was performed along with the use of topical or oral antibiotic therapy.

A new sample was collected when previous results were unsatisfactory or in case of therapeutic failure.

The subjective and objective data were associated for the analysis of otological changes. Statistical tests will be performed using the Statistica program (Version 7.0; StatSoft Inc., Tulsa, OK, USA), adopting a significance level of 5%. Continuous variables will be compared using t-Student or Mann-Whitney tests. Categorical variables will be assessed by the Chi-square test or Fisher's test, and the questionnaire's test-retest will be evaluated by the Kappa index. Results will be considered statistically significant at a 5% level, with a p-value < 0.05 and a 95% confidence interval. This study was submitted to the Research Ethics Committee of the Federal University of Amazonas via Plataforma Brasil (CAAE: 68910423.3.0000.9167; Opinion Number: 6.182.872, approved on 07/14/2023).

RESULTS

A total of 50 patients were included in the study, with 26 (52%) being female and 24 (48%) male. On average, the patients had a mean age of 43.5 years, ranging from 14 to 78 years old.

Within the sample, 3% reported using cotton swabs, 42% reported manipulating ears with objects, 8% reported having fungi in other parts of the body, 36% reported having used or currently using topical or oral antibiotics, 14% reported using oral or topical corticosteroids, 2% reported using a hearing aid, 20% reported having associated comorbidities, 26% reported using headphones, 6% reported swimming, 56% presented with associated chronic otitis media, 2% presented with diffuse external otitis, 0% presented with necrotizing external otitis, and 0% presented with acute otitis media.

In relation to associated signs, it was observed that 22% of the sample's patients presented external auditory canal edema, 36% exhibited canal hyperemia, 14% had tympanic perforation, 16% showed canal desquamation, 40% displayed secretion in the external auditory canal, and 36% presented mycelia in the canal. **Table 1**.

In relation to symptoms, 74% presented pruritus, 44% otalgia, 24% auricular fullness, 36% otorrhea, 34% hypoacusis, and 6% tinnitus. **Table 2.**

Regarding the most prevalent fungus, Aspergillus sp was found in 46% of patients. Followed by Candida in 28%, with 78.57% Candida sp; 7.14% Candida tropicalis; 7.14% Candida krusei; and 7.14% Candida albicans. Followed by the association of Candida sp and Aspergillus sp in 6% of the sample. Association of Penicillium sp and Aspergillus sp in 6% of patients. Observed dichotomous hyphae and budding blastospores in 6%. Bacteria in 2% and negative for fungus in 6%. **Table 3. Figure 1.**

As an additional finding, two types of treatments were administered. The first involved 3% boric acid and 10-volume hydrogen peroxide for 21 days, and the second involved topical miconazole for 21 days. The first treatment proved to be more effective, showing a good response in patients who used the medication. However, no control group was performed to confirm its efficacy.

Signs	Quantity	Percentage
EXTERNAL AUDITORY CANAL EDEMA	11	22%
TYMPANIC PERFORATION	7	14%
SECRETION	20	40%
MYCELIA	18	36%
EXTERNAL AUDITORY CANAL HYPEREMIA	18	36%
EXTERNAL AUDITORY CANAL DESQUAMATION	8	16%

Table 1. Signs Presented by Study Patients

Symptom	Quantity	Percentage
PRURITUS	37	74%
OTALGIA	22	44%
AURICULAR FULLNESS	12	24%
OTORRHEA	18	36%
HYPOACUSIS	17	34%
TINNITUS	3	6%
ASYMPTOMATIC	1	2%

Table 2. Symptoms Presented by Study Patients

PATHOGEN	N	%
CANDIDA	14	28
ASPERGILLUS	23	46
CANDIDA+ ASPERGILLUS	3	6
ASPERGILLUS + PENICILLIUM	3	6
BACTERIA	1	2
OTHERS	3	6
NEGATIVE	3	6
TOTAL	50	100

Table 3. Most Prevalent Pathogens

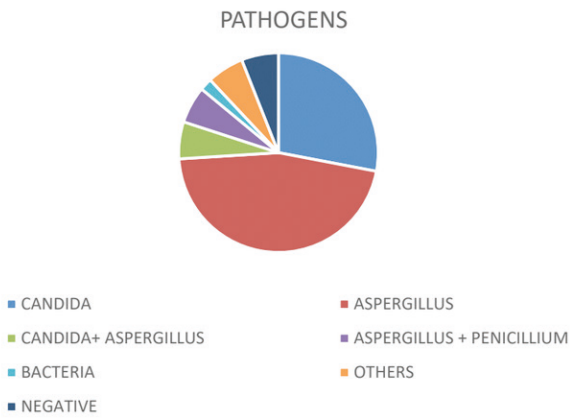


Gráfico 1. Patógenos mais prevalentes

DISCUSSION

This study was designed to gather more information about the most prevalent pathogen in otomycosis, gain a better understanding of the clinical profile of otomycosis patients in the state of Amazonas, as well as the influencing factors contributing to the onset of this pathology, its persistence after the proposed treatment, associated complaints, and treatment outcomes. The motivation behind this study stemmed from the high prevalence of this pathology in the region due to the warm and humid climate, as well as the observation that its incidence increased during the winter.

A higher incidence of cases was observed during the Amazonian winter, accounting for 90% of the sample. The relative air humidity is considered high throughout the year, averaging 90% during the months of January to June, which correspond to the Amazonian winter (SEGOVIA, 2020, p. 35).

MOUSSALLE, S. K. et al. (2023) defined otomycosis as a fungal infection affecting the external auditory meatus and the tympanic membrane, first documented in 1841. The incidence of otomycosis tends to be more common in the age group of 40-50 years (NAJAFLOO R, et al., 2021), coinciding with the average age found in the present study, which was 43.5 years.

Studies indicate that diabetic patients are more prone to otomycosis (MOUSSALLE. K.S, et al., 2019). However, in our study, we found that only 6% (3 patients) were diabetic, which reinforces the significant climatic influence on this condition. Additionally, women show a greater predisposition to develop the disorder compared to men. The justification may be associated with different innate immunities, expression of steroid hormones, and factors related to sex chromosomes (NAJAFLOO R, et al., 2021).

A relevant risk factor was the presence of chronic otitis media, accounting for 56% of the sample.

Another factor contributing to otomycosis is the manipulation of the external auditory canal with an object, observed in 36% of the sample.

The intense and unnecessary use of antibacterial medications in the treatment of middle and external ear infections can cause fungal overgrowth in this region. Therefore, one of the adverse effects of the abusive use of broad-spectrum antibiotics might be the secondary overgrowth of fungi and the increased prevalence of otomycosis" (JACKMAN. A, et al., 2005). This reinforces the finding of the study, demonstrating that the previous use of topical or oral antibiotics was observed in 36% of patients affected by otomycosis.

No significant association was observed between fungal diseases in other parts of the body and fungal external otitis.

CONCLUSION

This study demonstrated that females were the most affected, presenting pruritus and otalgia as the main symptoms, with the most affected age group being 25 years old (Graph 3). The group that manipulates the external auditory canal or uses earphones was more affected. The most effective treatment resulting in resolution without recurrences was the use of boric acid and hydrogen peroxide treatment in relation to the objective otoscopy test, demonstrating the absence of mycelium after 21 days of treatment.

Thus, we can conclude that the most prevalent fungus in the region was Aspergillus, which responded well to treatment with boric acid and hydrogen peroxide, showing resolution within 21 days of medication application and without recurrences. However, further case-control studies would be necessary to confirm the treatment's efficacy.

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