

Original Article

Fungal identification in the air and water of a hemodialysis unit in Brazil

Identificação fúngica no ar e na água de uma unidade de hemodiálise no Brasil

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Abstract

The presence of fungi in healthcare settings, including hemodialysis units, represents a significant risk for immunocompromised patients. This study aimed to investigate the occurrence of fungi in the air and water of a hemodialysis unit located in a tertiary public hospital in Maceió, Alagoas, Brazil. Over a period of three consecutive months, monthly air samples were collected and analyzed using the spontaneous sedimentation technique on Petri dishes containing Sabouraud Dextrose Agar (SDA). Simultaneously, water samples (100 mL) were collected from four specific water distribution points and subjected plating on SDA. Fungi were phenotypically identified based on their macroscopic and microscopic characteristics. In total, 498 colony-forming units (CFUs) of fungi were isolated, with 86 CFUs originating from the air and 412 CFUs from the water. Regarding the water samples, a higher concentration of fungal CFUs was observed in the potable water from the supply network (229 CFUs). Unexpectedly, 23 CFUs were identified in the reverse osmosis samples and 11 CFUs in the storage tank, which are post-treatment points where the presence of microorganisms is not desired. The fungus *Cladosporium* spp. was the most prevalent in both air and water samples, followed by *Penicillium* spp. in the air and *Rhodotorula* spp. in the water. These findings underscore the need to implement effective control and monitoring measures for fungi in the hemodialysis unit to ensure patient safety.

Keywords: hemodialysis, fungi, water, air, environmental health.

Resumo

A presença de fungos em ambientes de saúde, incluindo unidades de hemodiálise, representa um risco significativo para pacientes imunocomprometidos. Este estudo teve como objetivo investigar a ocorrência de fungos presentes no ar e na água de uma unidade de hemodiálise localizada em um hospital público terciário na cidade de Maceió, Alagoas, Brasil. Ao longo de três meses consecutivos, amostras mensais de ar foram coletadas e analisadas por meio da técnica de sedimentação espontânea em placas de Petri contendo Ágar Sabouraud Dextrose (ASD). Paralelamente, amostras de água (100 mL) foram coletadas em quatro pontos específicos de distribuição de água e submetidas ao plaqueamento por esgotamento em meio ASD. Os fungos foram identificados fenotipicamente por meio de associação de suas características macroscópicas e microscópicas. No total, foram isoladas 498 unidades formadoras de colônias (UFC) de fungos, sendo 86 UFC provenientes do ar e 412 UFC da água. Em relação à água, observou-se uma maior concentração de UFC de fungos na água potável proveniente da rede de abastecimento (229 UFC). Inesperadamente, foram identificados 23 UFC nas amostras da osmose reversa e 11 UFC no reservatório de armazenamento, pontos de pós-tratamento nos quais a presença de microrganismos não é desejada. O fungo *Cladosporium* spp. foi o mais prevalente tanto no ar quanto na água, seguido por *Penicillium* spp. no ar e *Rhodotorula* spp. na água. Esses achados enfatizam a necessidade de implementar medidas eficazes de controle e monitoramento de fungos na unidade de hemodiálise, visando garantir a segurança dos pacientes.

Palavras-chave: hemodiálise, fungos, água, ar, saúde ambiental.

1. Introduction

Fungi can be dispersed through various pathways, including air, water, food, insects, humans, and animals, with those dispersed through the air referred to as anemophilous, along with their fungal components such as spores or propagules. These elements serve as

aeroallergens, which, when inhaled, can cause respiratory allergic manifestations such as asthma and rhinitis, with severity directly related to the intensity of exposure. Additionally, it is crucial to recognize that fungi play multifaceted roles in the ecosystem, being naturally

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ubiquitous as saprophytes, contributing significantly to organic matter decomposition and nutrient cycling. In recent decades, some fungal species have evolved into opportunistic pathogens, posing a growing concern for public health (Mezzari et al., 2003; Venceslau et al., 2012; Silva et al., 2021).

The microbiological analysis of environmental fungi, particularly of hospital airborne microbiota, is of great importance in the prevention of allergic diseases and healthcare-associated infections (HAI) in hospitalized patients, especially those with immunosuppression, whether acquired or induced by different therapies, and patients undergoing invasive procedures such as hemodialysis. In this regard, the lack of air quality control through environmental monitoring can lead to prolonged hospital stays and compromise patient recovery (Schiavano et al., 2014; Durán Graeff et al., 2017; Rostami et al., 2017; Sajjadi et al., 2018; Calumby et al., 2019; Zhao et al., 2022).

In addition to the significance of airborne fungal pathogens, ensuring water quality is a critical factor in the hemodialysis procedure. Hemodialysis is the established treatment for patients with Chronic Kidney Disease (CKD), typically administered three times a week, with sessions lasting 3 to 4 hours, depending on the patient's clinical condition. In this therapy, water is utilized in dialysate production and dialyzer reprocessing, exposing patients through the semipermeable membrane of the dialyzer to a considerable volume of dialysis fluid, approximately 400 liters per week or 20,000 liters per year. Given this substantial exposure, the quality of water used in the process plays a pivotal role in mitigating any additional health risks for the patients (Nazemi et al., 2016; Totaro et al., 2017; Anversa et al., 2021).

Water quality guidelines for hemodialysis procedures have been established in various regions worldwide (Anversa et al., 2021). However, it is important to highlight that in Brazil, current guidelines do not include the enumeration of filamentous fungi and yeasts as part of the recommended microbiological parameters (Brasil, 2014). This absence of specific guidelines for the assessment of fungi in the water used during hemodialysis may create a gap in ensuring patient safety.

Most fungi are naturally found in soil, air, and water, and rarely act as pathogens in a healthy host. Nonetheless, patients undergoing hemodialysis have a compromised immune system, which increases their susceptibility to various pathogens (Pires-Gonçalves et al., 2008). It is relevant to note that infectious diseases represent the second leading cause of mortality in patients with chronic kidney disease, and the incidence of fungal infections has shown a progressive increase in individuals undergoing hemodialysis or peritoneal dialysis (Crowe et al., 2021; Heerspink et al., 2021; Gowher et al., 2022; Kanjanabuch et al., 2022). However, the knowledge regarding the epidemiology of these infections, their associated factors, and the impact they entail is still limited, underscoring the urgent need for microbiological studies to assess water quality in these facilities (Anversa et al., 2021).

Given the significance of these fungi in the development of infections that can lead to severe complications in dialysis

patients, this study aimed to investigate the occurrence of fungi in the air and water of a hemodialysis unit in a tertiary public hospital in the city of Maceió, state of Alagoas, Brazil.

2. Material and Methods

2.1. Study site

The study was conducted in a hemodialysis unit of a public tertiary-level hospital in the city of Maceió, Alagoas, Brazil.

2.2. Sample collection

Over the course of three consecutive months, spanning from November 2019 to January 2020, monthly samples of ambient air (six samples each month) and water (eight samples each month) were collected, resulting in a total of eighteen air samples and twenty-four water samples.

2.2.1. Air

For the collection of airborne fungi, the exposure method of Petri dishes containing Sabouraud Dextrose Agar (SDA) supplemented with Chloramphenicol at a concentration of 50 mg/L was used. This method is suitable for assessing fungal bioaerosols that settle on the culture medium, simulating the exposure of surfaces to these contaminants (Calumby et al., 2019). The samples were collected during the hemodialysis procedures of the patients, with the plates exposed for 30 minutes at strategic locations with constant access, such as tables, sinks, shelves, and counters. The placement of the plates took into account the dimensions of the environment, maintaining an equidistance of 3 meters between the plates. After the exposure period, the samples were identified, refrigerated, and transported to the Mycology Laboratory of Centro Universitário Cesmac, where they were maintained at 28°C in a microbiological incubator for a duration of up to seven days. They were observed every 24 hours until the appearance of isolated colonies.

2.2.2. Water

The water samples used in hemodialysis were collected following the guidelines of the APHA (2012) at four specific points, including the reservoir of potable water from the local water supply company, the pre-treatment reservoir, the reverse osmosis system, and the treated water reservoir where all filtered water is stored. Prior to collection, the external water outlet point was cleaned with 70% alcohol and then left open for 2 minutes to allow water to flow, as indicated by Varo et al. (2007). A volume of 100 mL of water was collected from each point in sterile bottles, to which 0.1 mL of 10% sodium thiosulfate solution was added to neutralize residual chlorine in the water and prevent its antimicrobial action, as described by Silva et al. (2017). The samples were placed in insulated boxes and transported to the Mycology Laboratory of the Centro Universitário Cesmac, where they were processed within two hours after collection.

In the controlled environment of the laminar flow hood, 1 mL of each sample was inoculated using the surface seeding technique with a Drigalski loop in duplicate onto Petri dishes containing Sabouraud Dextrose Agar (SDA) supplemented with Chloramphenicol (50 mg/L). The plates were incubated at 28°C in a microbiological incubator for up to seven days and examined every 24 hours during this period.

2.3. Isolation and identification of fungi

After observing fungal growth on the plates, a quantitative analysis was performed by counting the number of Colony Forming Units (CFUs), distinguishing between yeast-like and filamentous forms. For the water samples, the results were expressed as CFUs per milliliter (CFU/mL).

The identification of filamentous fungi was based on the combination of macroscopic and microscopic characteristics of the culture. Macroscopic features such as texture, shape, and colony color on the front and back were evaluated, along with microscopic characteristics including reproductive structures, chlamydospores, presence or absence of septa in hyphae, and color of hyphae and spores (hyaline or dematiaceous). These characteristics were compared to the criteria adopted by Hoog et al. (2000), Lacaz et al. (2002), Sidrim and Rocha (2010), and Zaitz et al. (2010). For better identification and visualization of filamentous fungal structures, microcultures were performed on slides using Lactrimel agar to stimulate sporulation (Calumby et al., 2022).

For the identification of yeast-like fungi, macroscopic analysis of the colonies and microscopic analysis using the microculture technique on cornmeal agar were performed. The results were associated with the germ tube test, chlamydospore formation on Tween 80 agar (Difco), and the pattern of assimilation and fermentation of various carbon and nitrogen sources (Zaitz et al., 2010; Silva et al., 2021).

2.4. Statistical analysis

The data were entered into a spreadsheet using the statistical software GraphPad Prism 8.0®, and subsequently presented in the form of tables.

3. Results

During the analyzed period, a total of 498 CFUs were detected in both samples under analysis, with

356 (71.5%) representing filamentous fungi and 142 (28.5%) representing yeast fungi. Out of the 18 air samples collected, 86 CFUs were identified, of which 85 were filamentous fungi and one was yeast fungi. As for the 24 water samples, 412 CFUs were isolated, comprising 271 filamentous fungi and 141 yeast fungi, as shown in Table 1.

Regarding the fungi identified in the ambient air where hemodialysis procedures were performed, a higher prevalence of *Cladosporium* spp. was observed with 32 CFUs (37.2%), followed by *Penicillium* spp. with 18 CFUs (20.9%), and *Aspergillus* spp. with 9 CFUs (10.5%). Additionally, 24.4% (21 CFUs) of the fungi were unidentified due to the absence of reproductive structures, as observed in Table 2. The risk classification assigned to the identified airborne fungi was consistent, with all of them being categorized as Class 2. This categorization implies a moderate individual risk and a low likelihood of dissemination to the surrounding community. It is crucial to emphasize that, despite this moderate risk, effective prophylactic and treatment measures are available to counteract the diseases that these fungi can cause in humans. Regarding pathogenicity, all the identified fungi were classified as either emerging or opportunistic. Furthermore, it is important to mention that the fungi *Cladosporium*, *Penicillium*, and *Aspergillus* have allergenic potential, with *Aspergillus* and *Penicillium* also being recognized for producing toxic substances known as mycotoxins.

In the analysis of samples collected from the water supply network and three different points of the dialyzer, a higher occurrence of *Cladosporium* spp. with 175 CFU/mL was observed, followed by *Rhodotorula* spp. with 141 CFU/mL and *Trichoderma* spp. with 87 CFU/mL. The highest number of isolated fungal CFUs was found in the potable water from the supply network (55.6%), followed by the pre-treatment water (36.1%), which removes impurities, chlorine, and water hardness. However, in post-treatment points such as reverse osmosis and the storage reservoir, where the presence of microorganisms should not be detected, 23 CFU/mL and 11 CFU/mL were isolated, respectively. *Rhodotorula* spp. was more prevalent in reverse osmosis water, with 12 CFU/mL, and in the treated water tank, with 6 CFU/mL, followed by *Trichoderma* spp., with 7 CFU/mL and 3 CFU/mL, respectively (Table 3).

In Figure 1, the macroscopic and microscopic aspects of the filamentous fungi identified in the water and ambient air of the hemodialysis unit are presented, which are

Table 1. Prevalence of CFUs of filamentous fungi and yeasts-like in the air and water of a hemodialysis unit in a tertiary public hospital from Maceió, state of Alagoas, Brazil.

Fungi	Samples				Total	%
	Air		Water			
	AF (CFU)	RF (%)	AF (CFU)	RF (%)		
Filamentous	85	17.1	271	54.4	356	71.5
Yeasts	1	0.2	141	28.3	142	28.5
Total	86	17.3	412	82.7	498	100.0

CFU: Colony Forming Unit; AF: Absolute Frequency; RF: Relative Frequency.

Table 2. Absolute and relative frequency of fungi identified in the ambient air of the hemodialysis room in a tertiary public hospital from Maceió, state of Alagoas, Brazil.

Fungi	Ambient Air		Risk class	Pathogenicity
	AF (UFC)	RF (%)		
<i>Acremonium</i> spp.	1	1.2	2	E
<i>Aspergillus</i> spp.	9	10.5	2	A, E, T
<i>Cladosporium</i> spp.	32	37.2	2	A, E
<i>Curvularia</i> spp.	2	2.3	2	E
<i>Penicillium</i> spp.	18	20.9	2	A, E, T
<i>Rhodotorula</i> spp.	3	3.5	2	E
Unidentified	21	24.4	-	-
Total	86	100.0	-	-

Risk Class 1: low individual risk for the worker and the community, with low probability of causing disease in humans. Risk Class 2: moderate individual risk and low probability of dissemination to the community. They can cause diseases in humans for which there are effective means of prophylaxis or treatment. Pathogenicity: (A) Allergenic; (E) Emerging or Opportunistic; (T) Toxicogenic. CFU: Colony Forming Unit; AF: Absolute Frequency; RF: Relative Frequency.

Table 3. Frequency and distribution of fungi identified in water samples collected at different treatment stages in a hemodialysis unit of a tertiary public hospital from Maceió, State of Alagoas, Brazil.

Fungi	Water samples								Total (CFUs)	%
	Water supply network		Pre-treatment reservoir		Reverse Osmosis		Treated water tank			
	AF (CFU/mL)	RF (%)	AF (CFU/mL)	RF (%)	AF (CFU/mL)	RF (%)	AF (CFU/mL)	RF (%)		
<i>Cladosporium</i> spp.	98	23.8	71	17.2	4	1.0	2	0.5	175	42.5
<i>Penicillium</i> spp.	2	0.5	1	0.2	-	-	-	-	3	0.7
<i>Rhodotorula</i> spp.	76	18.4	47	11.4	12	2.9	6	1.5	141	34.2
<i>Trichoderma</i> spp.	49	11.9	28	6.8	7	1.7	3	0.7	87	21.1
Unidentified	4	1.0	2	0.5	-	-	-	-	6	1.5
Total	229	55.6	149	36.1	23	5.6	11	2.7	412	100.0

CFU/mL: Colony forming unit per milliliter; AF: Absolute frequency; RF: Relative frequency. Conventional symbol used: - Numeric value equal to zero, not resulting from rounding.

essential for the accurate identification of these fungi through detailed morphological analysis.

4. Discussion

The hospital atmosphere is susceptible to various types of airborne microorganisms, including viruses, bacteria, fungi, and their metabolites. Microbial contamination can originate from both the external environment and patients and visitors, who disseminate them through coughing, sneezing, and breathing (Zhao et al., 2022). Fungi, in particular, have a high capacity to disperse in the air, which can pose a significant risk to susceptible patients, especially those who are immunocompromised, such as hemodialysis patients. Exposure to pathogenic fungi present in the air can result in infections with adverse consequences for patient health (Venceslau et al., 2012; Sajjadi et al., 2018; Calumby et al., 2019; Souza et al.,

2019). Therefore, minimizing the dispersion of aerosols containing pathogenic fungi in the hospital environment is essential to ensure the health protection of both patients and medical staff.

Several studies have been conducted to assess the air quality in hospital environments; however, most of them have focused on intensive care units (ICUs), where critically ill patients are highly susceptible to opportunistic infections. On the other hand, few studies specifically concentrate on the hemodialysis department, making this study particularly relevant. Furthermore, methodological and sampling differences among studies limit the comparison of results. Gonçalves et al. (2018) and Pereira et al. (2014) conducted studies aiming to detect the presence of airborne fungi in Brazilian hospital environments. In the first study, 40 Petri dishes were exposed in the ICU of a university hospital in Pelotas-RS, with the identification of 79 CFUs of fungi. In the second study, 19 plates were exposed in a hospital in the city of

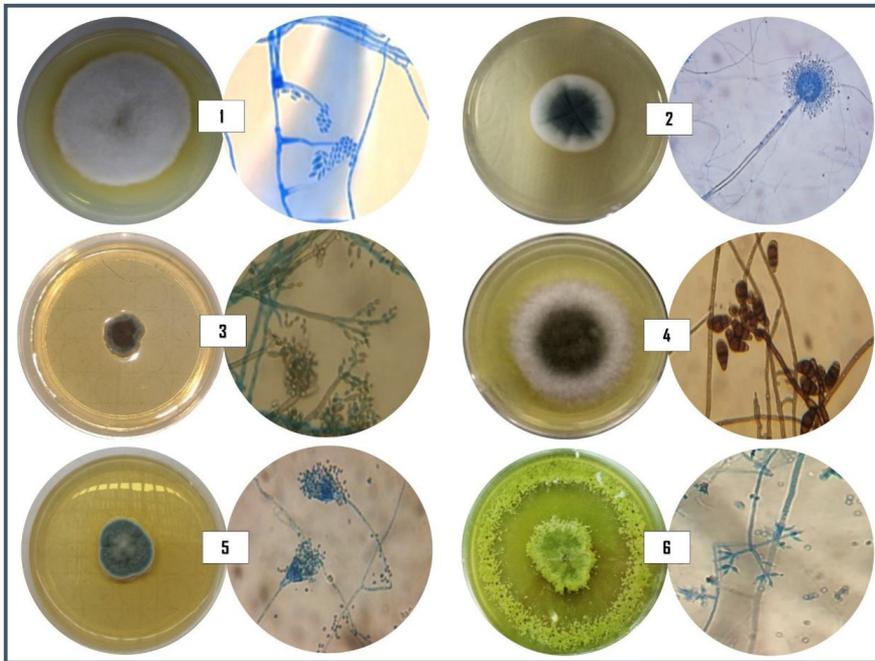


Figure 1. Macroscopic and microscopic characteristics of filamentous fungi identified in the water and ambient air of the hemodialysis unit, cultivated on Petri dishes containing Sabouraud Dextrose agar at 28°C; Slides stained with lactophenol cotton blue. Identified fungi: 1) *Acremonium* spp.; 2) *Aspergillus* spp.; 3) *Cladosporium* spp.; 4) *Curvularia* spp.; 5) *Penicillium* spp.; 6) *Trichoderma* spp.

Ariquemes-RO, with the detection of 50 CFUs. Both results were lower than those found in the present study, where 89 CFUs of fungi were isolated in 16 exposures. A more recent study conducted by Zhao et al. (2022) evaluated the presence of microorganisms in the air of different departments of a hospital in Kunming, China, before and after the SARS-CoV-2 pandemic, observing higher fungal contamination in the hemodialysis department, with an average of 200 CFU/m³ in that unit. However, air sampling was performed with an air impactor, preventing the use of this study for comparison. The findings of these studies confirm that atmospheric air is an effective and successful medium for the dispersion of fungi in hospital environments.

The predominance of fungal genera *Cladosporium* spp., *Penicillium* spp., and *Aspergillus* spp. in the air samples of the hospital environment is consistent with previous studies that highlighted the adaptation of these microorganisms to diverse environments (Menezes et al., 2006; Lobato et al., 2009; Shams-Ghahfarokhi et al., 2014; Calumby et al., 2019; Pedrosa et al., 2022). Furthermore, research conducted in Brazil and other countries, such as those by Mezzari et al. (2003), Martins-Diniz et al. (2005), Flores and Onofre (2010), Souza et al. (2019), and Gao et al. (2022), confirmed the high occurrence of *Cladosporium* species in microbiological surveys of indoor airborne fungi. In 2019, we conducted a study in the ICU of the same hospital to identify the airborne fungal microbiota present in the healthcare unit (Calumby et al., 2019). The results of this study revealed a high prevalence of *Cladosporium* spp., suggesting a possible environmental contamination by this microorganism in the mentioned hospital.

Among the fungi present in the air, all identified genera have been classified as class 2 and are considered opportunistic, with particular emphasis on the presence of *Aspergillus* spp. and *Penicillium* spp. This may represent a potential health risk to patients since several species within these genera are epidemiologically important due to their ability to cause infections in humans (Sidrim and Rocha, 2010). *Aspergillus* spp., in particular, has been reported as the main filamentous fungus responsible for infections in immunocompromised patients. The predominant route of entry is through the respiratory system, with the nasal sinuses and the bronchopulmonary tree being the preferred sites for the development of aspergillosis. Additionally, this fungus can contaminate injectable solutions such as saline and peritoneal dialysis fluid, presenting an additional risk (Sidrim and Rocha, 2010). The main clinical conditions observed include cutaneous aspergillosis, otomycosis, onychomycosis, aspergilloma, and allergic conditions such as allergic bronchopulmonary aspergillosis and allergic sinusitis. Furthermore, mycotoxicosis can occur due to exposure or ingestion of its mycotoxins. In more severe cases, invasive pulmonary aspergillosis can occur, especially in immunocompromised patients (Cadena et al., 2021; Moldoveanu et al., 2021).

Regarding *Penicillium* spp., an increase in the incidence of fungal infections caused by this fungus has been observed in recent years, covering several clinical manifestations, such as keratitis, otitis, sinusitis, urinary infections, pulmonary infections, allergic conditions, mycotoxicosis and hyalohyphomycosis. Immunocompromised patients, especially those with the Human Immunodeficiency Virus (HIV), are the main victims of severe systemic

infections related to *Penicillium* (Sidrim and Rocha, 2010; Hakeem et al., 2023).

With respect to the water samples, a high percentage of CFUs was observed in samples from the water supply network and the pre-treatment reservoir, indicating the presence of contamination sources throughout the water supply system that reaches the hospital unit. Additionally, although in smaller proportions, fungi were isolated at post-treatment points such as the reverse osmosis unit (23 CFUs/mL) and the treated water tank (11 CFUs/mL). The detection of microorganisms in these locations points to possible deficiencies in the treatment and filtration systems used in the hemodialysis unit.

In addition to the issues already mentioned, other factors can also influence water contamination and treatment efficacy. Temperature is a relevant factor, as its increase can stimulate the growth and proliferation of microorganisms, including pathogenic fungi, and promote the formation of biofilms in equipment and reservoir piping systems (Yin et al., 2019). Another aspect to be considered is the concentration of electrolytes and glucose in the water. Inadequate concentrations can create a favorable environment for fungal growth, compromising water quality and treatment effectiveness. Furthermore, the existence of stagnation zones within the structure of the water treatment and distribution system in the hemodialysis unit is a critical issue that needs to be addressed. These low-circulation areas may favor the formation of biofilms, where fungi can adhere and multiply, becoming more resistant to cleaning and disinfection procedures, resulting in greater difficulty in eradicating these pathogenic agents (Bugno et al., 2007). Given these factors, it is crucial for dialysis services to carry out comprehensive microbiological control, regularly monitoring water quality, and implementing effective strategies to minimize the risk of contamination. This includes adopting rigorous protocols for cleaning, disinfection, and continuous monitoring of the water used (Sehulster et al., 2003; Totaro et al., 2017; Anversa et al., 2021).

In the present study, 412 CFUs were detected in 24 water samples, with the presence of fungi observed in 100% of the samples. These results are consistent with previous studies conducted by Varo et al. (2007) and Pires-Gonçalves et al. (2008) in Brazil, and Arvanitidou et al. (2000) in Greece, which reported isolation rates of fungi of 100%, 89%, and 95.3% in dialysis water samples. However, other studies conducted in Brazil and other countries have reported lower results. For example, Figel et al. (2013) observed that 66% of water samples from six hemodialysis units in Curitiba, Paraná, Brazil, showed the presence of fungi, with black fungi detected in 46% of all samples. Additionally, Anversa et al. (2021) detected fungi in 24.1% of dialysis water and dialysate samples from three hemodialysis units in Brazil, with an average count of 16.3 CFU/mL. Similar results were observed by Bambauer et al. (1994) in Germany, where 10% of treated water samples and 20% of dialysate water samples from 30 hemodialysis centers showed the presence of filamentous fungi. In a study conducted in Iran, Mahmoudabadi et al. (2011) reported the presence of fungi in 27.4% of the analyzed samples, while Schiavano et al. (2014) isolated these microorganisms

in 12.1% of water samples from eight hemodialysis units in Italy. It is important to note that due to the diversity of methodologies used, including different isolation techniques, applied volumes, culture media, and incubation periods, direct comparisons between studies are not always possible.

Filamentous fungi, particularly *Cladosporium* spp. and *Trichoderma* spp., were the main fungal contaminants found in the water samples, especially those from the water supply network and the pre-treatment reservoir. These microorganisms have also been reported in previous studies investigating the presence of fungi in dialysis water, such as the studies by Varo et al. (2007), Mahmoudabadi et al. (2011), Figel et al. (2013), Schiavano et al. (2014) and Anversa et al. (2021), with varying frequencies. Additionally, *Rhodotorula* spp. was the only yeast identified, showing a significant prevalence in post-treatment water samples, as observed in reverse osmosis (12 CFU/mL) and the treated water tank (6 CFU/mL). The presence of this yeast has also been documented in previous studies investigating the fungal microbiota in water used for hemodialysis procedures (Arvanitidou et al., 2000; Montanari et al., 2018).

Opportunistic nosocomial fungal infections have become more frequent, and both air and water distribution systems can serve as pathways for the dissemination of these fungal pathogens (Pires-Gonçalves et al., 2008). *Cladosporium* spp. has been identified as the most prevalent fungus in air and water samples. This microorganism is widely distributed in the environment and is often associated with allergic rhinitis and localized cutaneous and subcutaneous lesions. However, it can also cause deeper and disseminated infections (Sandoval-Denis et al., 2015; Anversa et al., 2021).

Trichoderma spp. has been mentioned as an emerging pathogen in immunocompromised patients, especially in association with risk factors such as peritoneal dialysis, organ transplantation, chronic kidney disease, chronic pulmonary diseases, and hematological disorders (Oliveira et al., 2016; Calumby et al., 2022). At least nine species of this genus are associated with infections, ranging from allergic sinusitis, keratitis, and otitis to superficial and subcutaneous infections, peritonitis, endocarditis, cerebral abscess, and deep lung infection (Sandoval-Denis et al., 2014).

In relation to *Rhodotorula* spp., this yeast is associated with cutaneous diseases, invasive fungal infections, and mucosal infections, especially in immunocompromised patients (Wirth and Goldani, 2012; Cespedes et al., 2022). Additionally, this fungus exhibits a special metabolic activity in highly eutrophicated waters and heavily contaminated municipal wastewater. This characteristic indicates its association with human activities, and its presence indicates significant pollution of aquatic systems by industrial and municipal wastewater (Caicedo-Bejarano et al., 2023).

5. Conclusions

Based on the obtained results, it was observed that the analyzed hemodialysis unit exhibited a high prevalence of fungi in both the air and water, representing a potential risk to the health of patients and professionals in the sector.

The presence of *Cladosporium* spp., *Aspergillus* spp., and *Penicillium* spp. in the air samples necessitates a careful assessment of their potential implications, considering their possible association with the onset of respiratory allergic manifestations or, in more challenging scenarios, the development of invasive infections, depending on the individual's immune status.

The identification of *Rhodotorula* spp. and *Trichoderma* spp., especially in the water from the reverse osmosis points and the treated water reservoir, translates into potential issues within the treatment and filtration systems utilized in the hemodialysis unit. It is crucial to underscore that the presence of these fungi in the water signifies unsatisfactory practices concerning equipment and pipeline hygiene and cleanliness, thereby compromising the quality of care provided during the hemodialysis procedure. Although the research parameters for filamentous fungi and yeast in dialysis water are not currently stipulated in Brazilian legislation, it is of utmost importance that the monitoring of these microorganisms be included to ensure the integrity and safety of patients.

In light of these observations, the establishment of cleaning and disinfection protocols for equipment becomes imperative, such as the implementation of HEPA filters in air systems, along with environmental surveillance programs to routinely monitor air and water quality. Additionally, healthcare professionals must undergo training in hygiene practices and the adoption of preventive measures. While these actions may entail additional costs, they are indispensable in ensuring the safety and well-being of patients during the hemodialysis procedure.

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