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Research Article

Adaptations of filamentous fungi to extreme conditions: a study in mud from the El Totumo volcano in Colombia

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Background: Microorganisms are present throughout the environment and play a very important role in different ecosystems, including soil functions and composition. The presence of fungi in sludge raises concerns about potential skin infections or allergies if they come into contact with the skin. This is a mud-filled volcanic cone located in the rural area of the municipality of Santa Catalina in the Caribbean Region of Colombia, at coordinates (10°44′40′′ N and 75°14′29′′ W). The objective of this study was to identify filamentous fungi present in liquid and dry sludge from El Totumo volcano.

Methods: Plates were prepared with SDA, RBA, and Mycosel culture media. For sample analysis, one gram of each sample was weighed in triplicate and spread onto each plate. The plates were incubated for 7 to 14 days at 25°C. The species were then transferred to tubes containing the same culture medium and incubated for 7 days at the same temperature. Species were identified by their microscopic and macroscopic characteristics. Colonies (CFU) were counted, and the average was obtained for each medium. For the microscopic sample, they were transferred to a slide containing lactophenol and observed under a microscope. Taxonomic keys were used.

Results: The average number of colonies in the sludge was 25 CFU in the dry sludge and 44 CFU in the liquid sludge. Six genus were identified in the taxonomic analysis: *Aspergillus, Penicillium, Trichoderma, Fusarium, Culvularia,* and *Rhizopus*. Six fungal species were isolated from the liquid sludge: *Rhizopus, Aspergillus fumigatus, Aspergillus glaucus, Aspergillus flavus, Penicillium sp.,* and *Aspergillus terreus*. Seven fungal species were isolated from the dry sludge: *Aspergillus niger, Culvularia, Fusarium, Aspergillus flavus, Trichoderma, Penicillium sp.* and *Aspergillus terreus*. The fungal genus with the most species identified was *Aspergillus*.

Conclusion: More colony and species growth were found in the liquid mud than in the dry mud. Most of the isolated fungi represent skin and respiratory diseases that can affect humans. Understanding these findings is critical to preventing potential illnesses in visitors to this site.

Keywords: *mud, filamentous fungi, volcano, microorganisms*

Introduction

The Totumo Volcano, located in the municipality of Santa Catalina, in the department of Bolívar, Colombia, is a prominent tourist site known for its healing thermal mud. This volcano, unlike others, does not expel lava or gases, but rather emits thermal mud, whose therapeutic properties have been valued by both locals and tourists. The volcano is located in an area rich in natural resources and biodiversity, making it an interesting setting for microbiological studies, especially those related to fungal flora. Totumo's mud presents a unique environment, with two predominant forms: dry mud and wet mud. The former, found at the top of the volcano, are dense and have a muddy texture. The wet mud,

found in the crater and used for therapeutic baths, is rich in minerals such as sulfur, which gives it antibacterial and antifungal properties (EFE: Verde, 2015; Dill & Kaufhold, 2018). However, their interaction with the local microbiota, including fungi, remains an underexplored topic in the scientific literature. The study of fungi in geothermal and thermal environments, such as the muds of the Totumo volcano, has gained interest in recent years due to the extreme adaptations of organisms to conditions of high temperature, low water availability, and a high concentration of mineral compounds. Several studies have identified a wide variety of fungal species in these environments, some of which have significant biotechnological applications due to their unique properties, such as resistance to extreme temperatures and the production of specialized enzymes (González & Guzmán, 2019). In studies carried out in geothermal environments, the presence of fungal genus such as *Aspergillus*, *Penicillium*, *Cladosporium*, and *Alternaria* has been documented, which have shown a remarkable ability to adapt to environmental stress conditions, such as high mineral concentrations and temperature fluctuations (Gostinčar & Turk, 2012). These fungi have a symbiotic relationship with the extreme conditions of the mud, allowing them to survive and thrive in these harsh environments. Although Totumo Volcano has not been the subject of specific research regarding the identification of fungi in its mud, it could be inferred that the fungal community in the region could present species similar to those found in other geothermal environments (Robledo-Mahón et al., 2020). These studies suggest that thermal muds may be a suitable niche for thermophilic and halophilic fungi. The identification of fungal species in Totumo muds is of both scientific and applied importance. First, these fungi can offer unique insight into how microorganisms adapt to extreme environments, which may have implications for biotechnology, such as the development of heat-resistant industrial enzymes. Furthermore, research on microbial biodiversity in these muds could provide relevant information about their interaction with humans during thermal baths and their potential therapeutic or adverse effects. In summary, the Totumo Volcano and its thermal muds represent an ecosystem of interest for the identification and study of fungal species in extreme environments. The study of these fungi could open new perspectives for understanding microbial biodiversity in geothermal ecosystems and its applications in medicine, biotechnology, and conservation (Hmad & Gargouri, 2024).

Materials and methods

Liquid and dry sludge samples were taken in January 2024. As shown in Figure 1, the top of the volcano is covered with liquid sludge. The liquid sample was collected from individuals on-site. The dry sample was collected from a package they sell, which they package themselves for sale and use as a skin mask. Plates containing the following culture media were used: Sabouraud dextrose agar (SDA), Rose Bengal Agar (RBA), and Mycosel. Three media were used to recover more fungal isolates. A negative control (uninoculated medium) and a positive control were prepared from each culture medium. The positive control was inoculated with *Fusarium* species. One gram of each sample was weighed in triplicate and spread onto each plate containing the three-culture media (Brandão et al., 2002). The plates were incubated for 7 to 14 days at 25°C. After this time, the colonies on each medium were counted, and the average was tabulated. Species are isolated in tubes of each culture medium and incubated for 7 days at 25°C. For microscopic identification, slides are prepared using lactophenol reagent, the fungus is placed in the tube, and observed under a microscope. Taxonomic keys are used to identify the genus and species of each isolate (Echevarría, 2019, 2022, 2025).

Figure 1. Totumo mud volcano in Colombia

Results

Mud volcanoes offer a humid, warm environment with a high concentration of decomposing organic matter, ideal conditions for the growth of various pathogenic fungi, especially those of the genus *Aspergillus* and *Fusarium* (Warnock et al., 2001). These fungi, which can be opportunistic pathogens, can cause respiratory, skin, or systemic infections in immunocompromised individuals. For example, *Aspergillus fumigatus* and *Aspergillus flavus* are well documented as causative agents of aspergillosis, an infection affecting the lungs, sinuses, and other parts of the body, which can be severe if not treated properly (Latgé & Chamilos, 2019; Arastehfar et al., 2021). Exposure to volcanic mud could increase the risk of inhalation of airborne fungal spores, which could lead to respiratory infections. People immersed in the mud could come into direct contact with these pathogenic fungi through wounds in the skin or mucous membranes. Positive controls grew on all media, while the negative control showed no growth, indicating that the media was free of organisms (negative) and that the organism could grow (positive). Figure 2 shows the filamentous fungal species found in both the dry and liquid sludge. Six genus of fungi were identified in both the dry and liquid samples: *Aspergillus*, *Penicillium*, *Trichoderma*, *Fusarium*, *Culvularia*, and *Rhizopus*. Six species of fungi were identified in the liquid sludge: *Rhizopus*, *Aspergillus fumigatus*, *Aspergillus glaucus*, *Aspergillus flavus*, *Penicillium* sp., and *Aspergillus terreus*. Seven fungal species were found in the dried sludge: *Aspergillus niger*, *Culvularia*, *Fusarium*, *Aspergillus flavus*, *Trichoderma, Penicillium* sp., and *Aspergillus terreus*. The *Rhizopus* species *A. fumigatus* and *A. glacus* were identified in the liquid sludge samples, but not in the dried sludge. The species *A. niger*, *Curvularia*, *Fusarium*, and *Trichoderma* were isolated only from the liquid sludge. The species common to both samples (dry and liquid sludge) were *A. flavus*, *Penicillium* sp., and *A. terrus*.

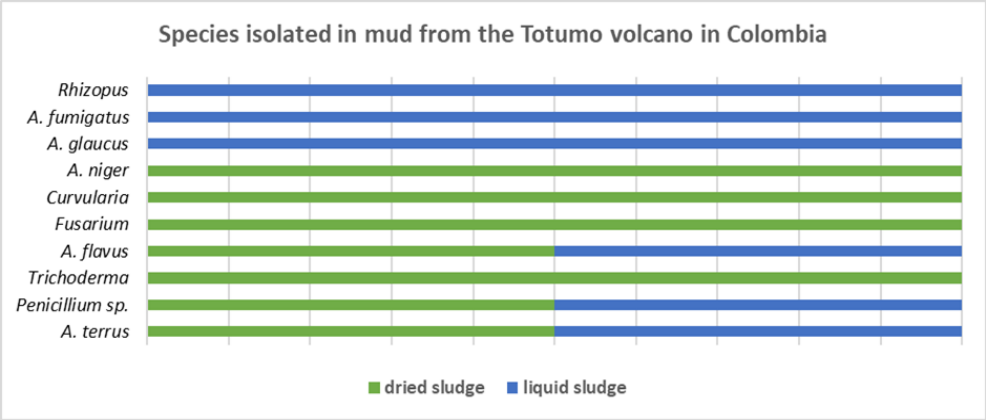

Figure 2. Species isolated in mud from the Totumo volcano in Colombia

Prolonged exposure to volcanic mud can result in inhalation of fungal spores present in the environment. These can cause allergic reactions or infections, especially in people with compromised immune systems. Symptoms may include nasal congestion, eye irritation, or skin irritation. Figure 3 shows the average number of colonies (CFU) for each culture medium in triplicate samples. The greatest amount of colony growth was observed in the liquid mud samples. The average number of colonies in the dry mud samples averaged 25 CFU. The liquid mud samples averaged 44 CFU. If we compare the results using the table of values: maximum allowable value (MAV) and maximum recommended value (MRV) for beach sand quality indicator, we can indicate that the mud would be at an average quality MRV level (Brandão et al., 2007). A rich diversity of filamentous fungi adapted to metal-rich soils, volcanic ash, or hot mud has been recorded. For example, in volcanic soils, saprophytic and phytopathogenic species such as *Chaetomium* sp., *Fusarium* sp., *Coprinus* sp., *Cladosporium* sp., and *Curvularia* sp., associated with soil decomposition and regeneration processes, have been reported (González & Guzmán, 2019).

Similarly, in sewage sludge and humid urban environments, recent studies have isolated fungal strains with the ability to tolerate high concentrations of heavy metals, suggesting their potential in bioremediation (de Mesquita et al., 2024). Fungi such as *Aspergillus niger*, *Penicillium chrysogenum*, and *Trichoderma harzianum* are common in enriched organic matrices, where they contribute to the degradation of organic matter, enzyme production, and resistance to toxic conditions (Wang et al., 2021b).

Although Totumo does not expel volcanic ash like magmatic volcanoes, the muds emitted by deep geological pressure share geochemical characteristics with secondary volcanic soils, which could allow colonization by halotolerant, metallotolerant or thermotolerant filamentous fungi (Dishliyska et al., 2025).

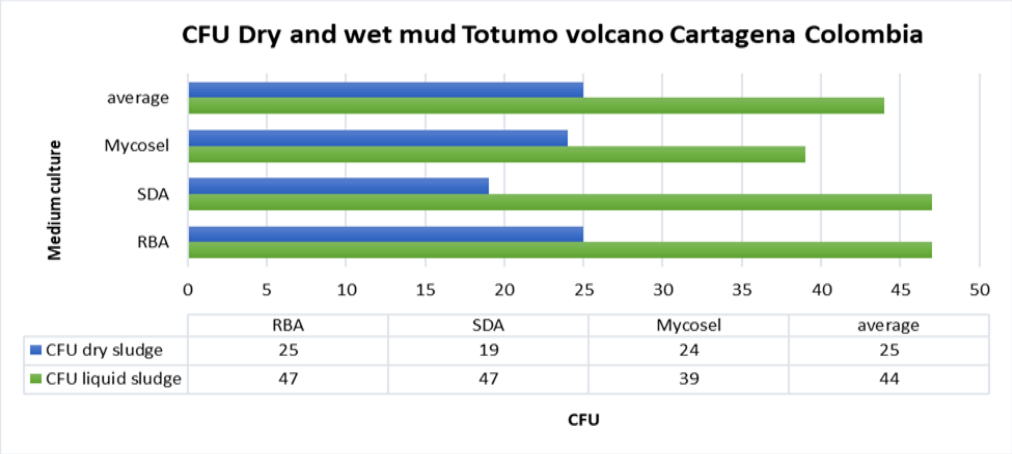

Figure 3. CFU Dry and wet mud Totumo volcano, Cartagena, Colombia

It's important to note, given these results, that although there isn't much information available on diseases directly caused by these fungal species in volcanic mud, the presence of various microorganisms in these environments suggests the need for preventive measures. Implementing safety practices and paying attention to personal hygiene are essential to minimizing the risks associated with exposure to these materials (CDC; Ameen, 2010; Berger, 2015).

Discussion

The results demonstrate that the El Totumo mud volcano hosts a diverse community of filamentous fungi adapted to an alkaline environment rich in mineral salts. The predominant presence of genus such as *Aspergillus* and *Penicillium* is consistent with studies in other volcanic soils and alkaline muds (Isola et al., 2025), indicating their ability to colonize extreme ecosystems. Filamentous fungi are key organisms in volcanic ecosystems due to their ability to decompose organic matter and recycle nutrients. In the case of volcanic soils, their presence is fundamental to soil ecology, as they act as decomposers of organic matter, which favors soil regeneration and nutrient cycling. The fungi found in these environments, such as *Aspergillus* sp., *Penicillium* sp., *Rhizopus* sp., *Fusarium* sp., among others, are adaptive to the extreme conditions of these environments and play an important ecological role by maintaining biological balance. One of the most common groups in volcanic soils is the genus *Aspergillus*, with species such as *A. fumigatus*, *A. flavus*, *A. terreus*, and *A. niger*. These fungi are known for their ability to thrive in warm and humid environments, such as volcanic soils. In addition to their ecological relevance, several *Aspergillus* species, such as *A. fumigatus* and *A. flavus*, have important clinical implications, as they are opportunistic pathogens responsible for respiratory infections, mainly in immunocompromised individuals (Arastehfar et al., 2021). The ability of these fungi to produce spores that are easily dispersed through the air increases their pathogenic potential, as spores can come into contact with humans and animals living in these areas, increasing the risk of infection, especially in volcanic areas with high geothermal activity. The fungus Rhizopus sp. is another important member of volcanic ecosystems, particularly in soils with high organic matter. Although it is also known for its ability to cause mucormycosis in humans, in its natural habitat it plays a crucial role in the decomposition of organic matter and nutrient retention. This fungus is found in soils rich in decomposing organic matter, making it common in volcanic areas containing large amounts of organic debris (Flores-Amaro et al., 2024). Furthermore, Penicillium sp. have a significant relationship with these ecosystems, as they participate in the decomposition of organic matter, promoting nutrient recyclability. It is relevant to note that Fusarium sp. and *Curvularia* sp., also present in volcanic soils, have a significant impact on both the environment and human health. These fungi are known for their pathogenic capabilities, causing human infections such as fusariosis, which include respiratory and skin infections (Stępień 2020, 2023). The presence of these species in volcanic soils suggests that extreme temperature and humidity conditions may be a factor in their adaptation and proliferation, while also increasing the risk of disease in exposed individuals. On the other hand, fungi of the genus *Trichoderma*, although not major pathogens for humans, play a vital role as biocontrol agents in the environment. *Trichoderma* is widely used for the biological control of other plant pathogenic fungi (Harman, 2006). In their volcanic habitat, these fungi can help control other fungal species, promoting a healthy ecological balance. In general, the fungi found in volcanic soils are a mixture of species that perform both critical ecological functions and pathogenic roles in human health. Adaptations to the extreme conditions of the volcanic environment, such as high temperatures, humidity, and high mineral concentrations, allow these fungi to thrive in conditions where other organisms cannot survive (Wang & Pecoraro, 2021a). However, their ability to generate highly dispersible spores also makes them potential agents of infection in exposed people, especially in volcanic areas with high geothermal activity. Identifying and understanding fungal

species in volcanic ecosystems is of great importance for public health, especially in tourist areas and in communities near these ecosystems. The dispersal of fungal spores, such as those of *Aspergillus*, *Fusarium*, and *Rhizopus*, can increase the risk of respiratory infections, especially in people with compromised immune systems. These infections can be difficult to treat due to the resistance of some of these fungi to conventional treatments. As tourism related to volcanic mud grows in popularity, studies on the fungal microbiota in these environments may be essential for developing prevention and control strategies to protect human health. Additional research on the biology and ecology of these fungi is needed to better understand their interactions with humans and their impact on public health. (Isola, 2025).

Conclusion

Taxonomy analysis revealed six genus: *Aspergillus*, *Penicillium*, *Trichoderma*, *Fusarium*, *Culvularia*, and *Rhizopus*. The fungal genus with the most species identified was *Aspergillus*. Data were collected from six fungal species in the liquid sludge: *Rhizopus*, *Aspergillus fumigatus*, *Aspergillus glaucus*, *Aspergillus flavus*, *Penicillium* sp., and *Aspergillus terreus*. Seven fungal species were found in the dry sludge: *Aspergillus niger*, *Culvularia*, *Fusarium*, *Aspergillus flavus*, *Trichoderma*, *Penicillium* sp., and *Aspergillus terreus*. The average colony count in the sludge was 25 CFU in the dry sludge and 44 CFU in the liquid sludge. More colony and species growth were found in the liquid sludge than in the dry sludge. Most of the isolated fungi cause skin and respiratory diseases that can affect humans. Although mud volcanoes may have temporary therapeutic benefits, as believed in some cultural contexts, they also pose potential health risks, particularly regarding fungal and bacterial infections, dermatological effects from minerals, and exposure to gases and heavy metals (CNN, 2024: Goel & Bhardawaj, 2014). It is essential that tourists and visitors to these areas take appropriate precautions, such as avoiding prolonged exposure and contact with hot mud on open wounds. Monitoring the microbiological and chemical quality of volcanic mud is crucial to ensure visitor safety.

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Author contributions

Designed. performed the experiments. wrote and revised the paper.

Conflict of interests

The author declares this article content has no conflict of interest.

Ethics approval

Not applicable.

AI tool usage declaration

No AI tool was used in manuscript preparation.

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