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Full Length Research Paper

The Physiological Characteristics of Halophilic and Halotolerant Fungus, as well as Possible Uses for Them

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This Organisms that can survive in harsh environments are known as extremophiles. The halophilic and halotolerant bacteria are two of the many varieties of extremophiles that need various environments and growth conditions to flourish. According to reports, these bacteria thrive in high-salinity environments such as the sea, sediments, lakes, mines, plants, and soil. To attain the most livable conditions for their survival, they require a high concentration of salt and carbon. Osmotic and ionic stress, which are controlled by the genetic expression of enzymes, proteins, cell wall components, and transporters, are the mechanisms underlying these microorganisms' high salinity survival and tolerance. Because of their resilience, halophiles and halotolerant fungi have demonstrated great promise in a variety of fields, including genetics, bioremediation, nanoparticle creation, enzyme production, antibacterial and anticancer activity, and more. The current study set out to investigate the halophilic and halotolerant fungi, which have received the least attention in terms of their development requirements, habitats, and mechanisms for tolerance and resistance to salt. As a result of the emerging multi-drug resistant pathogenic microorganisms, their biotechnological applications centered on the biomedical business will come next.

Key words: Extremophiles, Fungi, Halophiles, Halotolerant, Physiology.

INTRODUCTION

Because of their physiological and metabolic processes, extremophilic microorganisms may thrive in a variety of harsh environments, including hot, cold, salty, sandy, extremely acidic, and alkaline ones. Extremophilic microorganisms are more popular as sources of new bioactive chemicals and for learning about the origins of life's evolution because to their resilience (Chung et al., 2019). According to Chamekh et al. (2019), halophilic bacteria have been investigated for their biotechnological uses and stress adaptability mechanisms. In the past, halophilic fungi have been isolated from a variety of environments, such as terrestrial, aquatic, decomposing debris, and dried foods (González-Abradelo et al., 2019; Tafer et al., 2019; Pérez-Llano et al., 2020). All kinds of microorganisms, including bacteria, algae, fungi, and protozoa, have been researched from various environmental and geographical samples, and groupings that can withstand high salt levels are more varied (Chamekh et al., 2019). Compared to halophilic fungus, halophilic bacteria are the most extensively researched halophiles. According to the literature and reports that are now available, halophilic fungi have more potential for the discovery of new species with unique bioactivities. Utilizing halophilic and halotolerant fungi, with an emphasis on their physiological and biotechnological uses, was the aim of the current study.

2. Habitats

According to an earlier study by Ali et al. (2019), the classification of halophiles into three groups based on salt content is as follows: minor halophiles (2–5%), moderate halophiles (5–20%), and extreme halophiles (20–30%).

Fungal variety is significant, because geographical location affects both physiological behavior and metabolic secretions. For their investigations of biodiversity, researchers have investigated the solar salterns, dead sea, arid desert, sebkha, soil, and terrestrial ecosystems mud (Moubasher et al., 2018; Chamekh et al., 2019). From the enormous Sebkha of Oran, Algeria, the biodiversity research investigated the fungi Aspergillus sp. strain A4, Chaetomium sp. strain H1, Penicillium vinaceum, Gymnoascus halophilus, Wallemia sp., and Ustilago cynodontis (Chamekh et al., 2019). According to Qiu et al. (2020), Aspergillus glaucus was recently identified as "China Changchun halophilic Aspergillus (CCHA)" after being isolated from the surface of plants growing close to a salt mine in Jilin, China. Aspergillus chevalieri, Pleosporaceae spp., Alternaria tenuissima, and Alternaria alternata were identified as the fungi isolated from Pakistan's Miani-Hor Mangrove Forest Soil (Khan et al., 2020). From the Lake dirt in Algeria, G. halophilus and Wallemia spp. were isolated (Chamekh et al., 2019). According to a prior study by González-Martínez et al. (2017), halophilic fungi can be found in sediment samples. From the Gulf sediment in North America, Scopulariopsis spp., Aspergillus spp., Peniophora spp., and Cladosporium spp. have been isolated (González-Martínez et al., 2017). Similarly, P. rubens and A. protuberus were isolated from Bonna sediment in New England, according to Corral et al. (2018). Since halophiles are known to withstand salt, research into salt mines and salterns has led to the discovery of a variety of species, such as Yarrowia lipolytica recovered from solar saltern saline (Alamillo et al., 2017), Wallemia ichthyophaga and Paranerita triangularis from solar saltern (Primožič et al., 2019), and A. salisburgensis isolated from a salt mine (Tafer et al., 2019). Plants like A. montevidensis ZYD4 from Medicago sativa L. Plant (Liu et al., 2017a) and A. glaucus isolated from the leaf surface (Qiu et al., 2020) are examples of other habitats. Halotolerant fungi like A. sydowii have also been found to live in sugarcane bagasse (González-Abradelo et al., 2019). The findings in Table (1) show the halophilic and halotolerant fungus habitats, with Aspergillus species being more prevalent and found in each regional environment.

3. Identification of the halophilic fungus and their nutritional needs

It is difficult to separate halophilic fungi from microbial communities with high salt levels; this process mostly relies on enrichment and cultivation with varying nutritional conditions (Anteneh et al., 2019). The physiological needs and environments of halophilic and halotolerant fungi determine their differences. According to Ruginescu et al. (2020), these bacteria employ a wide range of strategies to deal with the osmotic pressure imposed by the high salt content of their environment. As evidenced by their variability across the various communities, the halophilic microbes' capacity to adapt to a broad range of settings is ascribed to physicochemical circumstances, such as temperature, salinity, and nutritional state (Menasria et al., 2019). Halophiles have been found to thrive in a variety of salty environments, including lakes, rivers, salterns, soils, salted foods, some plant leaves, and wall paintings (Ruginescu et al., 2020).

The carbon supply, medium pH, temperature, and salt content are among the halophilic fungi's nutritional and cultivation parameters. Potato dextrose agar (Chamekh et al., 2019; Qiu et al., 2020), yeast extract peptone dextrose agar (González-Martínez et al., 2017), and malt extract agar (Pérez-Llano et al., 2020) are the most often used media for isolating halophilic fungi; the incubation period has been reported to be seven days (González-Martínez et al., 2019). The carbon sources in the medium ingredients have a high sugar content, which promotes fungal development. According to a 2019 study by Anteneh et al., the lower acid pH created during fermentation aids in bacterial inhibition; occasionally, antibiotics are added to the medium to further aid in bacterial inhibition. Based on their nutritional and cultivational characteristics, Table (2) illustrates the isolation of the various halophilic fungi at various sites. According to González-Martínez et al. (2017), most halophilic fungi grow in 7–15 days. However, two contradictory reports from the Gulf Sediments, Mexico, indicate that Cladosporium spp., Aspergillus spp., Peniophora spp., and Scopulariopsis sp. require a longer growth period of 2 months, while Cladosporium spp., Aspergillus sp., and Talaromyces spp. require a shorter growth period of 48 hours. Magnuscella marinae, which was isolated from Australian marine sponge samples, needed two months to be cultivated in various media supplemented with increased salt concentrations, according to another study by Anteneh et al. (2019). The primary roles for each fungal physiology are thus held by the geographical areas, which also have distinct culture and nutritional needs.

4. Mechanisms by which halophilic fungal species tolerate salt

Halo tolerant fungi are fungal species that can survive high salinity. They can resist ionic stress (an increase in the amount of Na^+) and high osmotic pressure (water loss from the fungal cells and solute accumulation in the cytosol). According to Gunde-Cimerman et al. (2018), fungal adaptation requires that they be able to withstand increasing salt concentrations and variable salinities. According to Plemenitas et al. (2014), the most characteristic method for salt adaptation in Hortaea werneckii and Wallemia ichthyophaga is the employment of suitable solutes. In order to maintain intracellular Na^+ levels below hazardous levels, fungi cultivated in saline-containing conditions accumulate suitable solutes in the cytosol, according to a prior work by Chung et al. (2019). A. sydowii's osmoprotective mechanisms under both ideal and excessive saline environments have been the subject of much research. Changes were seen in the lamellar structure and cell wall thickness, as well as a decrease in chitin content and an increase in α and β-glucan content. Additionally, it was noted that excessive salinity altered the expression of the hydrophobin gene (Pérez-Llano et al., 2020). In comparison to the proteome of the non-halophilic species, the proteome of A. sclerotialis exhibited a higher proportion of alanine, glycine, and proline (Tafer et al., 2019). Higher salt concentrations and decreased water activity in hypersaline environments lead to the production of vital industrial enzymes by halophilic bacteria. Fewer investigations concentrated on the enzymes recovered from the obligate halophilic fungi, although there have been numerous reports on the halophilic hydrolases produced by the halophilic fungi, including cellulases, lipases, and proteases, as well as amylases (Chamekh et al., 2019; Ruginescu et al., 2020). The bacteria with the greatest enzymatic indices were Aspergillus sp. strain A4, Chaetomium sp. strain H1, P. vinaceum, Gracilibacillus halophilus, Wallemia sp., and Ustilago cynodontis (Chamekh et al., 2019). To create an ion gradient, halophilic fungi were shown to use a variety of transporters, such as K+ efflux, K+ uptake, P-type ATPase, and Na+ efflux (Gunde-Cimerman et al., 2018). This explains why halophilic fungi may survive in extremely salinized environments. The cellular, genetic, enzyme, and/or metabolic pathways are the mechanisms by which halophilic fungus survive extreme salinity, as shown schematically in Fig. (1). Additionally, Table (3) provides a summary of the many halophilic fungi that were isolated from various extreme habitats along with their salt tolerance concentrations.

5. Future characteristics and applications of halophilic and halotolerant fungus

Figure (2) illustrates the various uses and potential future developments of halophilic and halotolerant fungi in several fields, including health care, antibacterial and anticancer activities, nanoparticle creation, enzyme production, genetics, bioremediation, and other areas.

5.1. Healthcare

Modern humans are in the process of developing several breakthroughs in every industry that improve people's lives and/or health. People are battling a variety of illnesses (Stansberry et al., 2019). The historical medical systems of Ayurveda, Siddha, Unani, and Chinese medicine were traditionally used to treat human ailments; however, their contemporary drug systems include homeopathy, naturopathy, and allopathy (Dhingra, 2020). However, modern lifestyles, eating patterns, and ecological shifts have wreaked havoc on human lives and made it possible for harmful microorganisms to infiltrate. According to a recent study by Flandroy et al. (2018), multidrug-resistant organisms are now frequently heard of and common to witness, putting human lives at higher risk and making them more susceptible to illness. This has made it challenging for researchers to conduct in-depth genetic analyses of dangerous microbes. As a result, developing novel medications that can alter the course of drug resistance has become vital. It should be mentioned that the majority of medications that have received FDA approval are derived from microorganisms, such as bacteria and fungus (Andrei et al., 2019). This offers information for further research into halophilic fungi, which have broader uses in the medical realm.

5.1.1. Anti-microbial activity

For the creation of novel compounds with possible uses in biomedicine, the utilization of extremophiles is especially crucial (Giordano, 2020). The primary goal of efforts is to address pressing health requirements, especially those related to cancer and resistant bacteria, two of the biggest worldwide threats (Aslam et al., 2018). The spread of antibiotic resistance is endangering public health worldwide (Ben et al., 2019). Fungi offered a broader basis for the identification of antimicrobials throughout the ongoing study of natural goods. According to Ruginescu et al. (2020), halophilic and halotolerant fungal species that thrive in naturally occurring hypersaline settings don't need salt since they can develop and adapt to a variety of salinities, including freshwater and injected NaCl solutions. Table (4) lists the halophilic fungus as sources of bioactive substances with antibacterial properties.

5.1.2. Anticancer potential

According to a recent study by Abdel-Razek et al. (2020), natural products, such as anticancer medications made by microbes, are referred to as bioactive molecules. The majority of well-known anticancer natural products have been derived from plant cells, but microorganisms are also great substitutes due to the following reasons: 1) the diversity of the microbial world, 2) their ease of manipulation, and 3) the ease of physiological screening to find new natural products with antitumor properties (Pham et al., 2019). Even though bacterial cells interact with tumor cells in different ways than metabolites do in a lab setting, bacterial metabolites are thought to be the most effective means of stopping cancer cells from surviving (Sedighi et al., 2019). As fresh sources of unique biomolecules, extremophiles have received more attention recently (Corral et al., 2019). It is believed that the halophilic and halotolerant bacteria that thrive in hypersaline conditions are trustworthy sources of metabolites that fight tumors. The functions of halophilic bacteria' metabolites in the treatment of cancer have been documented in a number of research. Ali et al., 2019; Ruginescu et al., 2020; Corral et al., 2018; Rani and Kalaiselvam, 2013).

5.1.3. Role in nanoparticle synthesis

Nanotechnology is now widely used in practically every application area. The creation of nanoparticles (NPs), the tiniest particles with sizes ranging from 1 to 100 nm, is one of the innovations (Rajput, 2017). Depending on their intended use, these NP are created using three different processes, as shown in Fig. (3): physical, chemical, and green synthesis.

The process by which microorganisms produce NPs is known as "green synthesis" (Salem and Fouda, 2021). According to Jeevanandam et al. (2018), nanoparticles are extensively utilized in a variety of industries, including textile, healthcare, food, agriculture, electronics, the environment, renewable energy, and numerous manufacturing processes. NPs are being used in a lot of healthcare applications, mostly drug delivery systems, because they don't damage any organs or tissues while delivering the medication to the body (Chauhan et al., 2020). In addition to having strong antibacterial properties, the NPs made from halophilic fungus also function as inhibitory agents against a variety of microorganisms (Wang et al., 2017). Fig. (4) illustrates the procedure for the green synthesis of the NPs.

5.2. Biotechnological applications (Enzymes production)

The varied groupings of microorganisms known as extremophilic fungus possess a wide range of adaption qualities, including metabolic processes and genetic diversity, which enable them to thrive in environments that contain high levels of salt. These fungi have a wide range of uses in biotechnology, including genetics, medicine, and agriculture. There are a variety of uses for halophilic and/or halotolerant fungus, mostly because of their capacity to generate a large number of enzymes (Satyanarayana et al., 2005). The study conducted by Chamekh et al. (2019) examined the enzymatic activities of halophilic fungi that were isolated from Sebkha in Oran, Algeria. It was noted that in the presence of a medium rich in NaCl, they were able to produce a variety of enzymes, including lipases, amylases, proteases, and cellulases. The halophilic fungus A. flavus produces a halotolerant protease that can be employed in a variety of industrial processes. Normally, the activity of the regular proteases is inhibited by salty solutions (Razzaq et al., 2019). The halophilic fungus Engyodontium album produces α-amylase, which has several uses in the food, pharmaceutical, and detergent industries and whose enzyme activity increases with increasing NaCl content (Elyasi Far et al., 2020). The halophilic cellulases produced by A. flavus KUB2 have a wide range of commercial uses (Namnuch et al., 2021). Table (5) provides a summary of the various halophilic fungal species linked to the synthesis of particular enzymes and their respective uses.

5.3. Agriculture (Genetic)

By examining their biogeochemical cycles, which include sulfur, nitrogen, carbon, and phosphorus, the halophilic fungus help humans understand how survival is feasible in harsh environments (Martínez-Espinosa, 2020). Understanding gene expression and repression in live organisms is greatly aided by transcriptomics research. Transport-related genes differed between the halophilic fungus A. salisburgensis and the halotolerant fungus A. sclerotialis (Tafer et al., 2019). Additionally, research on A. sydowii under various salinity conditions revealed expression profiles based on genes linked to stress, including a rise in the expression of the gene encoding the solute transporter (Pérez-Llano et al., 2020). The genes that code for superoxide dismutase, catalase, and peroxiredoxin—also referred to as the oxidative stress response genes—were linked to increased salinity tolerance in comparative studies between Wallemia ichthyophaga, Hortaea werneckii, and Aureobasidium pullulans. These genes are in charge of both antioxidant activity and salt tolerance (Gostinčar and Gunde-Cimerman, 2018).

Halotolerant and halophilic fungi have emerged as new sources of target genes that can be utilized to genetically enhance plants' resistance to salt due to the growing worldwide issue of agricultural salinization (Egamberdieva et al., 2019). Plants having salt tolerance mechanisms are advantageous because they can prevent plant death due to rising soil contamination (Kamran et al., 2019). According to a recent study by Gupta et al. (2021), it is also possible to modify the model microorganisms so that they exhibit a symbiotic relationship with the plants and so aid in their resistance to elevated salt concentrations.

5.4. Environment (Bioremediation)

The technique of using living creatures, mostly bacteria, to remove contaminants from the environment is known as bioremediation (Abatenh et al., 2017). The bioremediation capacity of halophilic microorganisms is yet unknown, despite their study as promising biological agents for the degradation of contaminants at high salt concentrations (González-Abradelo et al., 2019). Pollutants are found in practically all ecosystems, including the air, water, and land, and their presence is becoming detrimental to the local populations. The environment needs to be cleared of dangerous pollutants, which can be done by bioremediation (Liu et al., 2017b). Extremophilic, halophilic, and halotolerant fungi are hardy and play significant roles in bioremediation applications, even in severe environments.

Heavy metals are a component of marine pollution and are challenging to eliminate, according to Briffa et al. (2020). In general, fungi are adapted to the contaminated environment and have the ability to clean up heavy metal-polluted soil. According to Jin et al. (2021), these fungi may have developed defense mechanisms against the harm caused by heavy metals. Halophilic fungi with the capacity to consume copper (Cu) metal include A. flavus, A. restrictus, and Sterigmatomyces halophilus (Bhattacharjee and Goswami, 2018). In the meantime, A. flavus and S. halophilus can break down zinc and cadmium, which are likewise broken down by A. gracilis and S. halophilus (Kalpana et al., 2018).

The overuse of pesticides, herbicides, and other dangerous chemicals has contaminated agricultural regions. Because of the chemicals' buildup inland, these contaminants have totally diminished the soil's capacity to supply essential elements for plant growth (Meena et al., 2020). According to Fowzia and Fakhruddin (2018), massive amounts of petroleum pollutants infiltrate the environment and seriously harm the ecosystems of the land and water. Although microbial biodegradation of petroleum hydrocarbon pollutants has long been regarded as an environmentally acceptable, economical, and effective biological treatment, its capabilities are diminished in harsh settings. Therefore, enzymes and/or chemicals that can function in harsh environments are highly needed (Li et al., 2019). The various halophilic fungi generate enzymes such lipase, amylase, protease, cellulase, β-glucosidase, and chitinase, which have more uses in the bioremediation process (Beltagy et al., 2018; Chamekh et al., 2019; Primožič et al., 2019; Namnuch et al., 2021). Table (5) lists the halophilic fungus along with their sources and the enzymes they produce.

5.5. Otherapplications (Cosmetology/ Food/ Textile)

In the biotechnology industries of food, textiles, medicine, and cosmetics, colors are much sought for (Sajid and Akbar, 2018). As secondary metabolites, fungi are microorganisms that can produce potent pigments that can be utilized as food coloring or colors. Pigments that have been isolated from halophilic and halotolerant fungus enable this application. According to Heer and Sharma (2017), these pigments are substances that absorb light with a certain wavelength in the visible spectrum.

Melanin, anthraquinones, hydroxyanthraquinones, azaphilones, carotenoids, oxopolyene, quinones, and naphthoquinone are among the many pigments that fungi are known to make (Kalra et al., 2020). In addition to its application in cosmetics (Sajid and Akbar, 2018), melanin pigment derived from the halophilic fungus Hortaea wernecki is also utilized in the food and textile industries (Heer and Sharma, 2017). According to Chadni et al. (2017), Talaromyces verruculosus produces red pigment, while Trypethelium eluteriae produces 5-hydroxytrypethelone, (-)-trypethelone, (+)-trypethelone, and (+)-8-hydroxy-7-methoxytrypethelone as dark violet-red pigments. Additionally, a red pigment is produced by the marine fungus Talaromyces albobiverticillius 30548 (Venkatachalam et al., 2019). One of the three compounds produced by the bioactive molecule Funiculosone's isolation from the endolichenic fungus Talaromyces funiculosus is ravenelin, a yellow, uniform powder with strong antimicrobial properties that finds use in the food and pharmaceutical industries (Padhi et al., 2019). An orange pigment, a derivative of the carotenoids, is produced by Penicillium sp. (GBPI_P155), which was isolated from the Himalayan region (Pandey et al., 2018). Similarly, the orange, red, or yellow pigment known as Azaphilone can be produced by the fungus Monascus ruber M7 (Chen et al., 2017). Fungi are thought of as the cell factories for the manufacture of pigments, where researchers can test out their functionalities, when taking into account the total advantages of fungal diversity (Kalra et al., 2020).

Conclusion

Beginning with a basic understanding of fungal habitats, this study examined how halophilic and halotolerant fungi survive in extreme environmental conditions. They are categorized as extremophiles based on these fungal nutritional needs. Because of their halophilic nature, they need NaCl to grow and survive. They use several strategies to endure harsh environments. Additionally, these fungi's isolated enzymes exhibit increased activity at elevated salt concentrations and have a broad range of industrial uses. Halophilic fungi are a boon to the medical field in the fight against multidrug-resistant infections because of their ability to synthesize bioactive chemicals and their inhibitory effects. The largest innovation is the synthesis of nanoparticles, which are useful in medication delivery systems and have a wide range of applications. Furthermore, halophilic fungi play a crucial role in the cosmetics sector. To sum up, a lot of fungi are found in various settings but have not yet been well studied. As a result, more study is required to identify and isolate new halophilic and halotolerant fungi, which have greater potential to benefit humanity through their varied products, mostly in the area of human health.

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Conflict of interest

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