Biological features of fungi of the genus Mucor

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Abstract. Mucor belongs to the most prominent order Mucorales, a phylogenetically ancient group of fungi referred to as "early divergent fungi". The genus Mucor contains several species. The most common are Mucor amphibiorum, M. circinelloides, M. hiemalis, M. indicus, M. racemosus and M. ramosissimus. Mucor has fast growing colonies and is characterized by tall, simple, unbranched sporangiophores without basal rhizoids, non-apophyseal sporangia, and pigmented zygosporangial walls. The walls are covered with granules, and the swollen apex contains spores that are white or yellow when immature, becoming brownish gray or dark gray when mature. .Colonies usually have a fluffy appearance, the height of the colony is 3-4 cm, similar to cotton candy.

1 Introduction

The genus Mucor belongs to the zygomycotic order Mucorales Taxonomic classification of mucor fungi: Kingdom: Mushrooms; Phylum: Zygomycota; Order: Mucorales; Family: Mucoraceae; Genus: Mucor.

Mucor has a filamentous form, it is an inhabitant of the soil, plants, decaying plant residues, it is widely distributed in nature. Mucor spp. can cause infections in humans, amphibians, cattle and pigs [2,3].

Mukor fungus belongs to the genus called mold. Food, soil, organics of plant origin - the place where it can occur in case of improper and careless storage of products.

The initial stage is characterized by the appearance of a whitish fluff, which is why it is called white mold. Further reproduction is determined by the formation of sporangia with the maturation of the bunch. The sporangia are grayish or beige in color and are black at maturity (Fig. 1).

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Fig. 1. Mucor sporangia

Division - Zygomycota. Some experts are of the opinion that such a variety existed during the Proterozoic era. The moldy species tends to feed on unnecessary waste products of humans, plants and animals. The genus Mucor belongs to the most conspicuous order Mucorales, a phylogenetically ancient group of fungi referred to as “early divergent fungi” [1]. From the first microscopic observation of a Mucor specimen in 1665 to the present, several hundred potential Mucor species have been described [2]. Mucor species are widespread and are predominantly saprotrophs [3]. Representatives of the genus Mucor have an ambiguous effect on human activity. With regard to their negative effects, several Mucor species, in particular the thermotolerant species M. indicus, M. ramosissimus, and seven members of the M. circinelloides complex [4], have been recognized as human and animal pathogens that cause mucormycosis [2].

2 Research Methodology

To conduct studies to identify the effect of aloe juice on bacterial cells of pathogens of pneumonia, 15 strains obtained from sputum from patients with pneumonia were used. For the cultivation of pneumococci, both conventional culture media and media supplemented with blood or serum are suitable. Growth is much faster on the latter media, as they prefer media supplemented with 0.1% glucose or 5% blood.

The juice of the Aloe vera plant, considered one of the most medicinal species, was the object of our research. Since the use of aloe aged from 3-5 years is desirable, it was these plants that were used. At this age, the concentration of nutrients and medicinal substances is considered the highest in aloe. Fresh leaves were used and had not been watered for seven days prior to pruning for study. For medicinal purposes, the lower or middle leaves, the largest and most fleshy, are usually used to obtain more juice. These are the leaves we used. Thoroughly washing the leaves, the peel was removed, carefully separating the gel. The aloe juice thus obtained in the amount of 5 grams was used for further research.

We used a universal nutrient medium, that is, meat-peptone agar, inoculated the obtained strains into Petri dishes, strictly observing all the rules. For incubation, they were placed in a thermostat at 37°C for a day. 1 ml of gel was added to the test sample, and the

same medium, but without gel, was used as a control. The sensitivity of bacteria to aloe gel was determined as follows. Working dilutions of aloe juice were prepared, which were applied dropwise into Petri dishes, pre-distributed into sectors.

The conducted studies gave the following results. Tender, translucent colonies were obtained on dense nutrient media. The structure of the colonies is very clear, as if outlined, they do not merge with each other, the diameter of the colonies is about 1 mm.

Streptococcus pneumonia often forms short chains of ovoid diplococci, and α-type colonies on blood agar. Chains usually consist of 2-8 pairs of cocci. Pneumococci are surrounded by a polysaccharide capsule. They are rarely spherical, more often they resemble short artillery shells in shape, touching their bases. The causative agent of diplococcal infection has the form of paired cocci, the latter being somewhat elongated in length, their outer ends are lanceolate-pointed (they are also compared with the contours of a candle flame). In the pathological material (smears from affected tissues, sputum, blood), pneumococci have a clearly visible capsule covering both cocci. In cultures of diplococcus, the capsule is usually absent and often the microbe forms short chains, while acquiring similarities with streptococcus. The size of individual cocci ranges from 0.5 to 1.5 microns.

Pneumococci stain well with conventional aniline dyes, are gram-positive, immobile, do not form spores. The capsule with the usual color of pneumococcus remains colorless (they do not perceive paint); it is well stained according to Romanovsky - Giemsa.

3 Results and Discussions

Mucormycosis ranks third among the fatal fungal infections, and, moreover, the negative significance of mucormycosis is due to the fact that they lead to spoilage of animal feed and food. On the other hand, fungi of this genus, having the activity of enzymes that break down proteins and fats, are good biotechnological objects; mucosal fungi are also used in the fermentation of certain foods, for example, ragi, tempeh, mureha, and in the maturation of French cheeses [5,8,9].

The genus Mucor contains several species. The most common are M.amphibiorum, M. circinelloides, M. hiemalis, M. indicus, M. racemosus and M. ramosissimus.

Mucor colonies grow rapidly at 25-30°C and rapidly cover the surface of the agar. With their fluffy appearance at a height of several centimeters, they resemble cotton candy (Fig. 2). On the front side, the color is initially white, with time it becomes grayish-brown. The reverse side is white. M. indicus is an aromatic species and can grow at temperatures up to 40°C. M. racemosus and M. ramosissimus, in contrast, grow poorly or not at all at 37°C.

Unsepted or rarely septated, wide (6-15 microns) hyphae, sporangiophores, sporangia and spores are visualized. Intercalary or terminal arthrospores (oidia) located through or at the end of the hyphae and a few chlamydospores can also be produced by some species. Apophysis, rhizoid and stolon are absent. Sporangiophores are short, erect, tapering towards the apex and may form short sympodial branches. The columellae are hyaline or demaciaceous and hardly visible unless the sporangium is ruptured. Smaller sporangia may lack columellae. Sporangia are round, 50-300 microns in diameter, gray or black, filled with sporangiospores. After rupture of the sporangium, the sporangiospores are freely distributed. Sometimes a collarette may remain at the base of the sporangium after its rupture. Sporangiospores are round (4-8 microns in diameter) or slightly elongated. Zygospores, if present, arise from the mycelium [13].

Fig. 2. Mucor colonies

Branching of sporangiophores (branched or unbranched), shape of sporangiospores (round or elongated), maximum growth temperature, presence of chlamydospores, ethanol assimilation, and molecular analysis help differentiate Mucor spp. from each other [13].

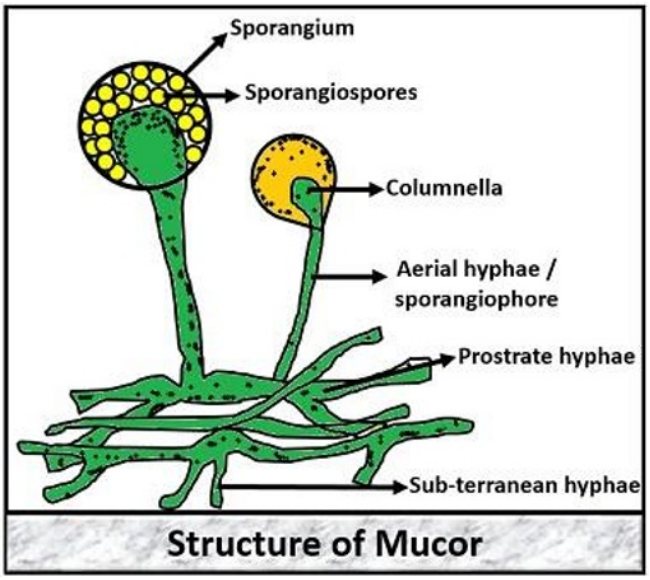

Fig. 3. Typical morphological characteristics of fungi belonging to the genus Mucor.

Mucor has fast growing colonies and is characterized by tall, simple, unbranched sporangiophores without basal rhizoids, non-apophyseal sporangia, and pigmented zygosporangia walls (Fig. 3) [11,12]. The walls are covered with granules, and the swollen top contains spores that are white or yellow when immature, and become brownish-gray or dark gray when mature [8,13]. Colonies usually have a fluffy appearance, the height of the colony is 3-4 cm, similar to cotton candy. Temperature has a significant effect on the growth and appearance of M. mucedo, for example, at 30 °C fungi do not grow, temperatures from 5 to 25 °C notice that fungi grow and sporulation is observed, temperatures of 15 °C and below give small with curved edges sporangiophores, as well as narrow cylindrical columella and large sporangiospores [13].

Representatives of the genus Mucor reproduce both sexually and asexually. Large tall sporangiophores grow in daylight and 20 °C, or sporangiophores do not grow at all. The best media for the growth of mycelium of mucosal fungi are pumpkin and sweet potatoes, fungi also grow well on potato dextrose agar, this medium includes potato starch and dextrose. Dextroses are the main sources of carbon. On a phospholipid medium, mucor gives apical growth and hyphae branch [10,12].

These fungi are characterized by eucarpic, mostly koinocytic thalli containing haploid nuclei. Asexual reproduction is characterized by the production of one to many sporangiospores in the mitosporangium. Sexual reproduction occurs by the conjugation of two identical gametangia, resulting in the formation of a zygospore. In general, the families and other taxa of Mucorales can be distinguished from each other by the morphology of the asexual reproductive structures, in particular by the characteristic features of sporangiophores, sporangia, columellae, and sporangiospores. The family Mucoraceae, which includes Mucor, is characterized by columellar, multispored sporangia. In addition, rhizoids and stolons in representatives of this family are either greatly reduced or completely absent. Arthrospores are formed under unfavorable nutritional or environmental conditions by septation of usually koinocytic hyphae followed by fragmentation of the hyphae. Typical morphological characteristics of the genus Mucor are shown in the figure.

Many types of mucor have polymorphism; under certain conditions, they grow as spherical multipolar budding yeasts. Yeast growth always requires the presence of fermentable hexose, and although this is not a requirement for all species, anaerobiosis is preferred. In addition, hexose concentration, CO2 partial pressure, and nitrogen source may also be important dimorphism factors in some species. Mucor, like many other mucor fungi, are usually the first saprophytic colonizers on dead or decaying plant material. They are able to quickly use a limited number of simple carbohydrate molecules before other complex carbohydrate-using fungi such as cellulose and lignin dominate the decomposition process. Mucor species are also capable of aerobically using a wide range of carbon sources, fermenting carbohydrates, and using ammonia or organic nitrogen. In addition, species of the genus Mucor are able to grow at temperatures ranging from 40 °C, in the case of M. recurvus, to 0 °C for strains of M. flavus, M. piriformis, M. plasmaticus, and M. racemosus; occur at pH values from 4 to 8; and have a water activity limit of 0.92 to 0.93. This genus is considered ubiquitous in nature and has therefore been isolated from numerous sources, including various processed and unprocessed foods.

Asexual reproduction occurs by the formation of non-nuclear, haploid sporangiospores in sporangia, at the terminal ends of aerial sporangiophores. In sporangia, the accumulation of nutrients, cytoplasm and nuclei occurs. An extension of the sporangiophore called the columella protrudes into the sporangium, and once the sporangiospores have matured, the rupture of the sporangium allows the spores to disperse, with wind being the main mode of dispersal. Under unfavorable environmental conditions, preference may be given to asexual reproduction, since unfavorable conditions do not allow conjugation of sexual individuals, in the “female” strain (-) the sexual ability is more likely to disappear faster than in the “male” strain (+) [11,13].

When strains (+) and strains (-) come into contact, elongated hyphae are formed, called progametangia, at the ends of which most of the nuclei and cytoplasm accumulate. Septa are formed near the point of contact, and the terminal component, the gametangium, is visible, to which elongated cells called suspensoria are attached. As the wall of the gametangium grows, and the gametangium itself, after repeated division by mitosis, disappear, then the germ cells are released and united, forming a zygote. The zygote is either black or grey. The spores mature, the walls of the zygosporangium rupture and the spores spill out, this occurs when favorable conditions occur [11,13].

Mucor is susceptible to the fungicide captafol (terazol), which inhibits hyphal apical growth and, at lower concentrations, thickens the fungal cell wall. Terrazol, having a fungistatic effect, causes the release of phospholipases in mitochondria and other membranes, which leads to complete lysis of mitochondria. The only known antidote against the action of terrasol is impure sucrose, which contains phospholipase inhibitors [13].

With a decrease in phosphorylation in mitochondria, the cell wall of mucor fungi thickens. Pentachloronitrobenzene (PCNB) leads to the death of the mitochondrial matrix. In the species M. mucedo, pentachloronitrobenzene leads to an increase in the perinuclear space, the number of vacuoles in the cell also increases, and the cell wall thickens significantly. Cell wall thickening is influenced by fungicides, molecular nitrogen and high glucose content, while the mycelial form becomes yeast.

Mucor mucedo is a widespread fungus. M. mucedo can be isolated from horse manure in early spring, permanent habitat - soil, manure, water, nasal cavity of cattle, grain in granaries, most plants and their fruits [10].

Fungi of the genus Mucor are able to break down soil pollutants - polycyclic aromatic hydrocarbons and this plays a very important role in the fight against organic pollutants. Mucor fungi synthesize exopolymer substances that decompose PAHs [10].

4 Conclusions

M.mucedo produces oxalate, or oxalic acid, a simple dicarboxylic acid that is one of the metabolic end products of many fungi and plants. It is known to be toxic to higher animals, including humans, due to its local corrosive effect and affinity for calcium ions, with which oxalate reacts to form water-insoluble calcium crystals. M. mucedo also produces aflatoxins known to cause liver cancer and other complications of the digestive, urinary, endocrine, hematopoietic, reproductive, and circulatory systems, although this requires further confirmatory studies as aflatoxins are mostly characteristic of Aspergillus species The ability of mycotoxins to diffuse from mycelium into the environment depends on their solubility in water. Foods with a high water content, in particular cheese and dough, allow significant diffusion of mycotoxins. It has been observed that aflatoxins diffuse into food products without extensive mycelial growth in them [13].

References

1. N. G. Almyroudis, D. A. Sutton, A. W. Fothergill, M. G. Rinaldi, S. Kusne. In vitro susceptibilities of 217 clinical isolates of Zygomycetes to conventional and new antifungal agents, Antimicrob, **51**, 2587-2590 (2007).
2. E. Alvarez, Molecular phylogenetic diversity of the emerging mucoralean fungus Apophysomyces: proposal of three new species, **27**, 80-89 (2010).
3. E. Alvarez, Spectrum of Zygomycetes species identified in clinically significant specimens in the United States, **47**, 1650-1656 (2009).
4. E. Alvarez, Molecular phylogeny and proposal of two new species of the emerging pathogenic fungus Saksenaea, **48**, 4410-4416 (2010).
5. M. Ameen, R. Arenas, E. Martinez-Luna, M. Reyes, R. Zacarias, The emergence of mucormycosis as an important opportunistic fungal infection: five cases presenting to a tertiary referral center for mycology, **46**, 380-384 (2007).
6. E. Anaissie, New spectrum of fungal infections in patients with cancer, **11**, 369-378 (1989).

7. M. M. Bagy, A. Y. Abdel-Mallek, Thermophilic and thermotolerant fungi of animals' hair. Acta Microbiol. Hung, **38**, 117-120 (1991).
8. S. A. Balajee, Sequence-based identification of Aspergillus, Mucorales species in the clinical mycology laboratory: where are we and where should we go from here? J. Clin. Microbiol, **47**, 877-884 (2009).
9. V. P. Baradkar, S. Kumar, Cutaneous zygomycosis due to Saksenaea vasiformis in an immunocompetent host, Indian J. Dermatol, **54**, 382-384 (2009).
10. V. P. Baradkar, M. Mathur, S. Taklikar, M. Rathi, S. Kumar, Fatal rhino-orbito-cerebral infection caused by Saksenaea vasiformis in an immunocompetent individual: first case report from India, **26**, 385-387 (2008).
11. E. A. Bearer, P. R. Nelson, M. Y. Chowers, C. E. Davis, 8 zygomycosis caused by Saksenaea vasiformis in a diabetic patient, J. Clin, **32**, 1823-1824 (1994).
12. C. R. Benjamin, C. W. Hesseltine, The genus Actinomucor, Mycologia, **49**, 240-249 (1957).
13. M. Bjorkholm, Liposomal amphotericin B and surgery in the successful treatment of invasive pulmonary mucormycosis in a patient with acute T-lymphoblastic leukemia, **33**, 316-319 (2001).