EPRA International Journal of Socio-Economic and Environmental Outlook (SEEO) ISSN: 2348-4101
Volume: 11 | Issue: 1 | January 2024 | SJIF Impact Factor (2023): 8.153 | Journal DOI: 10.36713/epra0314 | Peer-Reviewed Journal

STUDY OF THE BIOLOGICAL CHARACTERISTICS OF SOME SPECIES OF YEAST FUNGI OF THE CANDIDA GENUS

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ANNOTATION

The cultural and proteolytic properties of yeast fungi of the Candida genus were studied. It was found that the natural variability of the morphology of C. albicans colonies depends on the long-term storage of the strains and the amount of replanting. In addition to the typical S and atypical K forms, the R form can also be detected in collector and hospital strains. Negative results for proteolytic activity in the third reseeding for collection strains and in the first reseeding for hospital strains.

INTRODUCTIONS

Yeast fungi of the genus Candida are divided into several species, the clinical and epidemiological significance of which is not clear (Rebrova R.N., 1989; Dekhkankhodzhaeva N.A., 1996; Elinov N.P., 2002; Osipova S.O., 2002; Fotos PG, Lilly JP, 1998; Gautret P. et al., 2000). The need for high-quality diagnostics, and accordingly, the volume of research on microorganisms of the genus Candida is increasing at present. Yeast-like fungi of the genus Candida are a separate genus in the classification of microorganisms, which includes more than 80 species [Koch H., 1973], but only some species can cause various diseases in humans. These include Candida albicans, C.tropicalis, C.pseudotropicalis, C.krusei, C.guillermondi, C.pelliculosa, C.parapsilosis. Fungi of the genus Candida belong to opportunistic plant fungi; they are quite often isolated from the surface of the skin and mucous membranes of humans [Iskhakova Kh.I. et al., 1986; Bazhenov L.G. et al., 2002, 2005]. In clinically healthy individuals, candidate carriage reaches 5%, and in individuals with inflammatory processes of the mucous membranes, 53.2% of cases [Rebrova R.N., 1979].

Cultivation and quantitative recording of yeast-like fungi of the genus Candida is important in diagnosing candidiasis infections and determining the degree of dysbiosis of various biotopes of the body. Changes in the living conditions of fungi of the genus Candida lead to changes in their various biological properties, including cultural, proteolytic properties, adhesiveness and a number of others. This will lead to a decrease in their detection in qualitative and quantitative terms.

In this regard, the purpose of this study was to study the cultural and proteolytic properties of yeast-like fungi of the genus Candida, which are most susceptible to changes under the influence of various factors.

MATERIALS AND METHODS

We studied the morphological, cultural, proteolytic properties of the main representative of yeast-like fungi of the genus Candida - Candida albicans. Identification and differentiation of isolated microorganisms was carried out according to Bergi [1997]. To compare the results, 3 passages of collection and hospital strains of Candida albicans were carried out.

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All studies were carried out at the Urgench branch of the Tashkent Medical Academy and the Research Institute of EMIZ of the Ministry of Health of the Republic of Uzbekistan using generally accepted bacteriological methods. Statistical processing of the material was carried out on a personal computer using the Excel program.

RESEARCH RESULTS

Taking into account the growth properties of Candida albicans colonies grown on nutrient media was assessed using the scale proposed by Nuraliev N.A. and co-authors [2004]:

Level I - good growth (colonies are typical, juicy, full-fledged, lush. Abundant growth during incubation at 37 C for 18-24 hours, meets nomenclature requirements, morphological, tinctorial, enzymatic and other biological properties of microorganism cultures do not change);

Level II - moderate growth (colonies are small, dry, lagging behind in development to typical forms by 4-6 hours when incubated at 37 C for 18-24 hours, while maintaining the morphological, tinctorial, enzymatic and other biological properties of microorganism cultures);

Level III - weak growth (colonies are very small, difficult to determine visually and do not meet nomenclature standards when cultivated under conditions recommended by generally accepted methods);

Level IV - no visible growth.

The growth properties of different collection strains of Candida albicans were studied using rice bran aqueous extract (RBAE), which was prepared in two versions: I - in nutrient broth; II - on an isotonic 0.5% NaCl solution. The results obtained show that the sown cultures grow equally well on this medium at concentrations of 104 and 102 after 48 hours of cultivation in a thermostat - 370C (Table 1).

Table 1
Growth properties of museum cultures of yeast-like fungi of the genus Candida when cultivated in rice bran aqueous extract (RBAE).

Culture, registration number	ROVE on nutrient broth				ROE at 0.5% NaCl			
	After 24 hours		After 48 hours		After 24 hours		After 48 hours	
	Concentration							
	104	102	104	102	104	102	104	102
Candida albicans 7 003838	II	IV	I	II	II	IV	I	I
Candida albicans 10 003848	II	IV	I	II	II	IV	I	I
Candida albicans 5 003818	I	II	I	I	I	II	I	I
Candida albicans 723 003592	I	II	I	I	I	II	I	I

Note: Growth properties according to degrees I, II, III, IV according to Nuraliev N.A. (2014).

In addition, we studied the natural variability in the morphology of grown colonies of collection and hospital strains of Candida albicans (Table 2). It has been established that during primary sowing (I - passage), the Candida albicans population consists of the following types of colonies: a colony with a typical morphology (S form); the colony is atypical in morphology - dwarf form (K form).

Typical colonies are smooth, convex, shiny with smooth edges, white; on the third day of growth at 37 C on Sabouraud's medium with 4% glucose, the dimensions are 3-7 mm in diameter. In the atypical form, all the signs are similar, only on the third day of growth the diameter of the colonies is up to 2 mm.

The ratio of these colonies has certain quantitative expressions for different strains. This ratio of colonies in the population of the studied strains is natural and depends on the duration of storage and the number of passages. In addition, colonies of a different morphological type (R form) may appear in the third passage. These colonies are radially folded, raised from the surface of the medium, white in color, with a diameter of up to 4 mm. The frequency of occurrence is insignificant - 0.4-0.8%.

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Table 2
Natural variation in colony morphology in a population Candida albicans (in %).

Passage	Frequency of morphological forms of colonies					
	Collection strains			Hospital strains		
	S	K	R	S	K	R
I	76±2.8	24±1	0	95.2±1.4	4.8±1.2	0
II	82±2.4	18±1.3	0	96.7±1.5	3.3±1.4	0
III	82±1.8	17.2±1.8	0.8±0.6	88.4±2	11.2±1.6	0.4±0.4

It should be emphasized that with increasing generation in the population of "dwarf" forms, the quantitative ratio of these forms significantly decreases and the number of S forms increases. This indicates that the "dwarf" forms of colonies are not stable and are phenotypically variable.

The next stage of the work was to determine the variability of the proteolytic activity of the same collection and hospital strains of Candida albicans in a comparative aspect. All results were obtained after each passage (Table 3 and 4).

Table 3
Variability of proteolytic activity of various forms of collection strains of Candida albicans (in %).

Passage	Degree of proteolytic activity					
	High		Average		Short	
	S	K	S	K	S	K
I	10±3.2	19.3±3	18±4	42.2±6.3	72±5.2	38.5±4
II	0	6.5±2.8	30.4±4	39.4±5	69.6±5.3	54.1±3.8
III	0	0	0	50±8.3	96±8	50±8

It was found that in the first passage the percentage of high proteolytic activity was higher in the K form compared to the S form of collection strains - 19.3±3% and 10±3.2%, respectively. In the second passage of the same cultures, this trend continued. It is interesting to note that negative results for proteolytic activity were established in the third passage for the S form of the collection strains (4±3.3%), this peculiar change was not observed in the K forms.

Table 4
Variability of the proteolytic activity of various forms of hospital strains of Candida albicans (in %).

Passage	Degree of proteolytic activity					
	High		Average		Short	
	S	K	S	K	S	K
I	39.2±7.2	21.1±6	22.1±6	18±5.3	28.6±6.7	32±6.6
II	0	0	40±7.3	30±6.7	60±7.7	70±5.8
III	0	7.3±3.2	74.2±6	60.4±8.2	25.8±4.8	32.3±6.2

During further studies, we observed a different picture in hospital strains. High proteolytic activity was observed in the S form, 1.86 times more than in the K form of colonies (39.2±7.2% versus 21.1±6%). Negative results for proteolytic activity were detected for hospital strains already in the first passage - for the S form in 10.1±4.5% and for the K form in 28.9±4.7% of cases. But with an increase in the number of passages, the proteolytic activity of the S form gradually decreases and in the third passage, practically no high activity was detected.

CONCLUSION

1. Assessment of growth properties according to the proposed scale indicates good and moderate growth of yeast-like fungi of the genus Candida at concentrations of 104 and 102 degrees after 48 hours in rice bran aqueous extract.
2. Natural variability in the morphology of Candida albicans colonies depends on the duration of storage of the strains and the number of passages. In addition to the typical S forms and atypical K forms, in the third passage, R forms can be detected in both collection and hospital strains. Their frequency of occurrence is 0.4-0.8%.
3. Negative results for proteolytic activity were established for collection strains in the third passage for the S form in 4±3.2% of cases, for hospital strains in the first passage for the S form in 10.1±4.5% of cases, for the K form in 28.9±4.7% of cases.

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