Methods of Laboratory Diagnostics of Candidiasis Stomatitis

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Abstract: Relevance. Due to the significant increase in the frequency of mycoses in recent years, the treatment of these diseases is of particular importance and remains a difficult task. Antifungal agents used in the treatment are in most cases of synthetic origin, some of which cause a number of side effects. Individual intolerance to specific drugs creates significant difficulties in the treatment of this nosology. High resistance and rapid adaptation of pathogens to therapeutic drugs necessitate the constant search for new antifungal agents.

Thus, in modern dentistry, the development and introduction into practice of new methods and means for the treatment of candidiasis stomatitis is relevant.

According to the literature data, celandine grass has a fungistatic and fungicidal effect. Calendula has antimicrobial, anti-inflammatory, regenerating effect. Leucea safflower, Phytoecdysteroids cause an increase in protein-synthesizing processes, stimulation of immune processes in the body, and an increase in the activity of enzyme systems. According to a number of authors, ecdysteroids exhibit a membrane-stabilizing effect, which underlies their anti-inflammatory effect, as well as regulate mineral, carbohydrate, lipid and protein metabolism, and exhibit antioxidant properties. It has been studied that phytoecdysteroids stimulate the proliferation of epithelial cells, the proliferation and activation of fibroblasts, collagen synthesis by fibroblasts, and neoplasm of blood vessels.

Currently, laboratory diagnostics of candidiasis stomatitis includes microscopic, mycological, serological, histological, allergological methods [1.22].

Microscopic examination of the material is one of the most frequently used methods of examination of patients with candidiasis stomatitis. The material for the study of candidiasis stomatitis is scrapings and flushes from the mucous membrane of the mouth, the red border of the lips, dentures. A positive result of microscopy is considered when a set (more than 10) of blastoconidia, pseudogyphs or true hyphae is detected in one or more fields of vision. In chronic forms of candidiasis stomatitis caused by C. albicans, old forms of mycelium predominate. It should be noted that according to a number of literature sources, in 96% of cases, in smears, except for fungi of the genus Candida, concomitant microflora is detected, the presence of which contributes to a more pronounced manifestation of the disease. It was revealed that a positive result indicates candida colonization and confirms the diagnosis only in the presence of clinical manifestations.

According to the literature, candidiasis stomatitis is mainly a superficial process and in most cases is
more often localized in the epithelial layer of the oral mucosa. In this regard, the condition of epithelial cells is being investigated, their ability to provide resistance to the aggression of various microorganisms, including fungi of the genus Candida. Analysis of microscopic data reveals a violation of the differentiation process of surface and subsurface epithelial cells in patients with candidiasis stomatitis, there is a decrease in the number of surface epithelial cells. Mature surface epithelial cells are characterized by yellow cytoplasm, dark, oblong nuclei, and a sharp decrease in their number indicates a violation of the plastic function of the oral mucosa and, as a consequence, a decrease in its barrier properties [17.18.20.21.22.23].

According to the literature data, mycological examination is used to confirm the diagnosis - sowing on Saburo medium. Colony the white color has rounded outlines and clear borders, a convex shape, a shiny and smooth surface, but the isolation of the fungus is considered insufficient for laboratory diagnosis, it is necessary to quantify the number of cells in the medium. The specific definition of isolated crops is an important diagnostic criterion, has a complex character. So, a sign of candidiasis stomatitis is the sowing of up to 1,000 colonies in 1.0 of the material, 1-100 colonies in 1.0 of the material - this is the norm and a signal for observation. The culture study makes it possible to quantify the microorganisms contained in the obtained material, and identification determines the type of fungus, which is necessary in clinically difficult cases. Studies on media also allow us to determine the sensitivity of fungi to certain antifungals, which contributes to obtaining more effective results in the treatment of mycoses [19.21.22.23].

According to modern research, an express technique for the primary isolation and identification of Candida fungi has been proposed, which includes microscopic and cultural studies using Hi Crome Candida Agar chromogenic medium (Himedia, India). Quantitative determination of fungi C.albicans, C.glabrata, C.tropicalis makes it possible to monitor the results of antimicrobial therapy and, if necessary, adjust it [20.23].

One of the methods of identifying yeast fungi is a seedling test, or a test for the formation of seedling tubes. A colony of yeast fungus after incubation is placed on a slide and examined, if there is a formation of germ tubes without narrowing at the base of the tube, then the pathogen is identified as C.albicans, but 10-15% of strains are not able to form germ tubes.

According to Khmelnitsky O.K. (2005), in candidiasis stomatitis, the detection of germ tubes, and especially pseudomycelia, even on the surface of tissues usually indicates the invasive nature of the growth of the fungus. The detection of only blastospores on the surface of the epithelium in the complete absence of filamentation indicates in favor of fungal saprophytism [14.15.16.17].

According to modern literature, the biochemical identification of yeast fungi is based on the ability of Candida fungi to assimilate and ferment carbohydrates. Traditional biochemical methods are reliable, but time-consuming, so currently automated pathogen identification systems and special commercial kits are included in practice, for example, the ARCOS system testing the assimilation of carbohydrates. The kit uses a flatbed with 25 substrates, into which a liquid medium containing the studied microorganisms is introduced, the assimilation of the substrate is judged by the turbidity of the medium. The results are recorded photometrically using a computer, the strain is determined according to the numbering-code principle [1.3.5.7.9.11.13].

Ready-made tests on chromogenic media (representatives of "Candi Select 4", "Albicans ID2") identify C. albicans. The material is placed immediately on the medium, the basis of the test is that specific enzymes of certain fungi hydrolyze a special substrate, forming colored colonies. C. albicans forms colonies of bluish color, and Candida non-albicans — unpainted, the result is evaluated after 24-48 hours. Chromogenic media in terms of specificity are between the seedling sample and assimilation tests, referring to screening, since they only allow to confirm or refute the belonging of fungi to the
species C. Albicans.

The metabolism of mannose and arabinitol is determined in serum by gas-liquid chromatography. This test is highly specific up to 93-95%. Determination of the serum content of arabinitol is based on the ability of some species of C. albicans, C. Tropicalis to synthesize this substance [2,4,6,8,10,12,14,16].

According to the literature data, the method of quantitative research is the determination of candida antigens in the separated by a latex agglutination test. The Bichro-Latex Albicans kit includes a reagent containing latex particles coated with monoclonal antibodies to the components of the cell wall of C. albicans. Agglutination of red particles on a green background is considered a positive result [17,18,19,20].

In deep mycoses, along with the microbiological method of investigation, serological blood tests are carried out when antibody titers increase in the blood. However, with superficial mycoses, which include candidiasis stomatitis, such an increase does not occur. The search for antigens in the blood is a rather expensive and time-consuming method, and therefore has not been widely used in practical medicine [21,22,23].

Currently, polymerase chain reaction (PCR) is used to diagnose candidiasis stomatitis. With the help of amplification of a DNA fragment located inside the gene, the fungus is detected in PCR within 6 hours from the time of receipt of the material. This method is very sensitive, which limits its use in the diagnosis of candidiasis stomatitis, the result is positive in the presence of minimal amounts of fungal DNA with a carrier that is widespread.

A positive result of histological examination of the material with Gram, Romanovsky, hematoxylin or PAS staining of the biopsy confirms the diagnosis of candidal stomatitis.

According to modern literature sources, intradermal tests are carried out with polysaccharide antigen of various types of fungi. The sample is put according to the type of Mantoux reaction. The reaction is taken into account 24-48 hours after the test. Intradermal tests are considered more specific in the chronic course of candidiasis.

**USED LITERATURE**

19. Mirsalikhova Ф. Л. Особенности биофизических свойств и минерализующей функции слюны у детей в период прорезывания постоянных зубов //Клиническая стоматология. – 2016. – №. 4. – С. 4-6.