

випадках – дисплазія високого ступеня (0,22%). Аденокарциному стравоходу встановлено у 4 пацієнтів (0,05%, ДІ 0,01-0,12%). Продемонстровано низьку ймовірність виникнення аденокарциноми при всіх типах метapлазії епітелію стравоходу. Лише наявність дисплазії метapлазованого епітелію повинна викликати підвищену настороженість щодо виникнення аденокарциноми. Стравохід Барретта без дисплазії не повинен розцінюватися як передраковий стан. Перебільшення його значення призводить до додаткової травматизації слизової, підвищення ризику кровотеч та стенозів.

Ключові слова: стравохід Барретта, метapлазія, дисплазія, аденокарцинома стравоходу.

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случаях - дисплазія високої ступені (0,22%). Аденокарциному пищевода діагностовано у 4 пацієнтів (0,05%, ДІ 0,01-0,12%). Была продемонстрована низька вероятность виникнення аденокарциноми при всех типах метapлазии. Только наличие дисплазии метapлазированного епітелію должно вызывать повышенную настороженность в отношении виникнення аденокарциноми. Пищевод Барретта без дисплазии не должен оцениваться как предраковое состояние. Преувеличение его значения приводит к дополнительной травматизации слизистой, повышению риска кровотечений и стенозов.

Ключевые слова: пищевод Барретта, метapлазія, дисплазія, аденокарцинома пищевода.

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EFFECT OF NATURAL MINERAL BISCHOFITE CONTAINING PREPARATION ON THE ORAL MICROFLORA

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The oral cavity can be considered as an ecological system complex in which external factors (biological, individual, social) interact with internal ones (periodontium, bacterial community, local immune system, oral epithelium). If the favorable conditions arise, one or another disease of the oral mucosa may be developed and must be adequately treated. The arsenal of drugs is large and requires the doctor's knowledge, skills in using of the most optimal means and providing the appropriate recommendations to the patient. We studied the efficacy of the oral care product Antiqua Mare MAX, containing natural mineral Poltava's Bischofite, on the representatives of the oral microflora. To achieve this goal, museum strains of *E. coli* ATCC 25922, *S. aureus* ATCC 25923, *S. epidermidis* ATCC 14990, *E. faecalis* ATCC 29212, *M. lysodeicticus* ATCC 4698 and *C. albicans* ATCC1023 were used. As a control, a well-known antimicrobial agent 0.02% aqueous solution of chlorhexidine bigluconate was used. The ability of the Antiqua Mare MAX drug to inhibit the growth of museum cultures of the yeast-like fungi, colon bacilli, enterococci, micrococci, epidermal and golden staphylococci in the liquid medium was shown to coincide completely in three repeated determinations during the study of the effect of Bischofite containing oral care product. The fungistatic activity of the studied preparation Antiqua Mare MAX exceeded the effect of 0.02% solution of chlorhexidine bigluconate by 4 times ($p < 0,001$), but the fungicidal effect did not differ.

Keywords: OCMM microflora, chlorhexidine bigluconate, Antiqua Mare MAX.

The work is a fragment of the research project "Dental health restoration in patients with underlying diseases and their rehabilitation", state registration No. 0116U004191.

Oral mucosa diseases are common lesions of the human body among dental diseases. They reflect changes in organs and tissues of the body. Notwithstanding the diversity of causes, mechanisms of development and clinical course of the disease, the majority of these diseases are characterized by some common features that can be combined into separate related groups.

The oral microflora is represented by numerous types of aerobic and anaerobic bacteria, among which anaerobes dominate (in dental plaque the anaerobic/aerobic ratio is 1000/1) [7]. The permanent oral microflora is composed of representatives of several groups of microorganisms: bacteria, fungi, spirochetes, protozoa, viruses. The role of microorganisms in the development of periodontitis, candidiasis, ulcerative necrotic gingivitis, etc., is undoubtful [5]. The development of oral inflammatory diseases alters composition of the microflora of different biotopes that are part of the oral cavity [10].

The oral cavity can be considered as a complex ecological system in which the external factors (biological, individual, social) interact with internal ones (periodontal, bacterial community, local immune system, oral epithelium) [9]. Similar to the outer environment, all components of the system are in dynamic equilibrium.

In case of favorable conditions, any of the oral diseases, which required treatment, may develop [9]. The drug arsenal is large and requires the dental professional to be expert in their usage and giving recommendations.

Jardin Cosmetics LLC (Ukraine) has developed the oral care product Antiqua Mare MAX that is composed from natural products only: Poltava's Bischofite mineral complex, propolis, decoctions of stevia leaves, liquorice, oak bark (Sanitary-Hygienic Official Letter No. 602-123-20-1/781 as of 21.01.2019).

The spray contains eco-mineral Poltava's Bischofite, which is a bromine chloride-magnesium brine solution with a high content of salts and microelements of magnesium, potassium, calcium, sodium, iodine, copper, iron, etc., which has anti-inflammatory, absorbent and analgesic properties, based on the penetration of micro- and macroelements through the mucous membranes. Propolis has antimicrobial, antioxidant, anti-inflammatory, immunomodulatory and cardioprotective effects. Decoction of stevia leaves is a safe substitute for sugar. Stevia is 10-15 times sweeter than sugar and is low calorie; its intake has no negative effects. It is used as a natural sweetener to reduce blood pressure, increase immunity. Liquorice contains active substances glycyrrhizic acid with its anti-inflammatory, hypo-sensitizing effect; flavonoids, which reduce inflammation, normalize the level of vascular permeability; natural surfactants (saponins) with their anti-inflammatory, disinfectant properties.

The purpose of the work was to study the positive effect of Antiqua Mare MAX oral care product, containing natural mineral Poltava's Bischofite, on the oral bacterial species.

Materials and methods. To gain the objective of the study the museum strains of *E. coli* ATCC 25922, *S. aureus* ATCC 25923, *S. epidermidis* ATCC 14990, *E. faecalis* ATCC 29212, *M. lysodeicticus* ATCC 4698 and *C. albicans* ATCC 10231, obtained from the L.V. Gromashevskiy Institute of Epidemiology and Infectious Diseases of the NAMS of Ukraine (Kyiv) have been used. The above strains were used to prepare daily cultures on an MPA slanting nutrient agar or Sabouraud's slope agar ("Pharmaktyv" LLC, Ukraine), which inocula were brought to 0.5 McFarland. The sensitivity of standard strains of microorganisms to the preparations was studied by quantitative method in accordance with the Order of the Ministry of Health of Ukraine No. 167 as of 05.04.2007 "On approval of guidelines: Determination of sensitivity of microorganisms to antibacterial drugs" [4].

The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of Antiqua Mare MAX (*Jardin Cosmetic* LLC, Ukraine) was determined for each test-culture of standard strains of microorganisms [6]. Conventional antimicrobial agent, 0.02% chlorhexidine bigluconate (*Chervona Zirka*, Pharmaceutical Plant, Ukraine) was used as a control. Thereafter, all subsequent dilutions were made from the main solution of the preparation in nutrient broth (LLC "Pharmaktyv", Ukraine), adding 10^6 microbial bodies/1 ml of standard strains of microorganisms to each dilution. 1 ml of broth and 0.1 ml of bacterial suspension (CC - culture control) was added into the last test tube. Bacterial inoculations were incubated at + 37 ° C or + 30 ° C for *C. albicans* during 24 h, after which the results were determined by the degree of turbidity of the nutrient medium. The last test tube with a transparent medium indicated a delay in the growth of microorganisms under the influence of the MIC of the study preparation. To determine the MBC, the above determination of MIC was supplemented with a second step during which inoculation was made onto agar sectors or Sabouraud's medium in Petri dishes from the test tubes with no evidence of bacterial mass accumulation, and after 24 h of incubation, the presence of microbial growth was determined, taking the lowest concentration of the preparation for MBC, which showed bactericidal effect [6]. The above studies were repeated three times. The combined effect of Antiqua Mare MAX and chlorhexidine was studied quintuply by diffusion method [7]. The substance of the study preparation was applied onto the clean paper disks to the amount of 25 µg /disk. The antiseptic chlorhexidine bigluconate in the form of 0.02% aqueous solution was applied onto the clean disks to the amount of 25 µg /disk. The disks were dried at a room temperature and used to determine the sensitivity of museum strains. The areas of inhibition of growth of microorganisms around the disks were measured after 24 h. The results obtained by the disk-diffusion method were statistically processed by standard computer programs. The probability of difference between the groups was determined using the Student's t-test.

Results and Discussion. The findings of the study of the effect of Antiqua Mare MAX on museum strains of *E. coli*, enterococci, micrococci, *S. epidermidis* and *S. aureus* and yeastlike fungi have shown the ability of the preparation to inhibit the growth of these microorganisms in a liquid medium that completely coincided with the results of three repeated tests (table 1).

Visual accumulation of bacterial mass of *E. coli* ATCC 25922 test-culture was absent in 1:1 dilution. The repeated inoculation onto solid medium showed a complete absence of growth of microorganisms. This meant that MBC of Antiqua Mare MAX for *E. coli* ATCC 25922 coincided with the MIC. The findings of the study of the effect of the preparation on the *S. aureus* museum strain after incubation have shown no growth of the test-culture in dilutions of 1:1–1:2 (table 1). Under the influence of the preparation, a complete sterility of the medium was achieved in 1:1 dilution. The sensitivity of the museum strain of *S. epidermidis* to the study preparation was similar to that of *S. aureus*.

Table 1

The effect of Antiqua Mare MAX on the museum strains of the microorganisms in liquid medium

Museum strain	Dilution, concentration of the preparation, μL								C/C
	1:1/500	1:2/250	1:4/125	1:8/62.5	1:16/31.3	1:32/15.7	1:64/7.8	1:128/3.9	
E. coli ATCC 25922	–ster	+	+	+	+	+	+	+	+
S. aureus ATCC 25923	–ster	–	+	+	+	+	+	+	+
S. epidermidis ATCC 14990	–ster	–	+	+	+	+	+	+	+
E. faecalis ATCC 29212	–ster	–	–	–	+	+	+	+	+
M. lysodeicticus ATCC 4698	–ster	–	–	+	+	+	+	+	+
C. albicans ATCC10231	–ster	–ster	–	–	–	+	+	+	+

Notes: 1. «–» – no growth of microorganisms; 2. «+» – regular growth of microorganisms; 3. ster. – sterility in repeated inoculation onto solid nutrient medium (bactericidal effect).

E. faecalis ATCC 29212 museum strain was sensitive to Antiqua Mare MAX at a maximum dilution of 1:32; however, the MBC was 1:1. The preparation delayed the growth of the test-culture of M. lysodeicticus ATCC 4698 in 1:4 titre and showed a bactericidal effect in 1:1 dilution. Antiqua Mare MAX showed inhibition of C. albicans growth in dilutions of 1:1–1:64 (Table 1). The repeated inoculation of the above dilutions onto solid nutrient medium showed no growth of fungi at a maximum dilution of 1:2. Apparently, in our study, slightly higher values of bacteriostatic concentrations can be explained by a visual evaluation of the results, whereas in the works of A.G. Miroschnichenko, et al. hardware densitometric determination of the optical density of bacterial suspensions was used [2]. These authors consider the effect of the preparation to be bacteriostatic and suggest that its mechanism is to inhibit protein synthesis [3]; however, bactericidal property of certain concentrations of the preparation and its effect on both bacteria and fungi is controversial. In our opinion, most likely, in a microbial cell, the preparation acts on the same target as in the cells of a macroorganism, namely, energy processes, structure and functions of membranes.

Thus, Antiqua Mare MAX demonstrated a suppressive effect on museum strains of gram-positive cocci, Escherichia coli and yeastlike fungi, indicating antimicrobial properties and wide range. The most sensitive to the preparation were strains of C. albicans ATCC10231 and E. faecalis ATCC 29212. The MIC of chlorhexidine bigluconate in relation to E. coli ATCC 25922 was $12.5 \mu\text{g} / \text{ml}$, the MBC was $25 \mu\text{g} / \text{ml}$ (Table 2). Growth of Staphylococcus after incubation of test-cultures was absent in dilutions of 1:1–1:4, which corresponded to $25 \mu\text{g} / \text{ml}$ MIC (table 2). Under the influence of the preparation, complete sterility of the medium was achieved at a dilution of 1:1, and MBC of chlorhexidine bigluconate was $100 \mu\text{g} / \text{ml}$.

Museum strain of E. faecalis ATCC 29212 was sensitive to the antiseptic at a maximum dilution of 1:8, though the MBC was $50 \mu\text{g} / \text{ml}$. Chlorhexidine delayed the growth of the M. lysodeicticus ATCC 4698 test-culture at 1:4 titer and showed a bactericidal effect in 1:1 dilution, similar to Antiqua Mare MAX.

Table 2

The effect of 0.02% chlorhexidine bigluconate on the museum strains of the microorganisms in liquid medium

Museum strain	Dilution / concentration of the preparation, μL								C/C
	1:1/100	1:2/50	1:4/25	1:8/12.5	1:16/6.25	1:32/3.12	1:64/1.56	1:128/0.78	
E. coli ATCC 25922	–ster	–ster	–ster	–	+	+	+	+	+
S. aureus ATCC 25923	–ster	–	–	+	+	+	+	+	+
S. epidermidis ATCC 14990	–ster	–	–	+	+	+	+	+	+
E. faecalis ATCC 29212	–ster	–ster	–	–	+	+	+	+	+
M. lysodeicticus ATCC 4698	–ster	–	–	+	+	+	+	+	+
C. albicans ATCC10231	–ster	–ster	–	+	+	+	+	+	+

Notes: 1. «–» – no growth of microorganisms; 2. «+» – regular growth of microorganisms; 3. ster. – sterility in repeated inoculation onto solid nutrient medium (bactericidal effect).

Under the influence of antiseptic, inhibition of growth of C. albicans was observed in dilutions of 1:1–1:4, which is typical for MIC of $25 \mu\text{g} / \text{ml}$ (Table 2). The repeated inoculation onto solid nutrient

medium showed no growth of fungi at a maximum dilution of 1: 2, corresponding to MBC of 50 µg / ml. After 24 hours of incubation, the zones of absence of growth of all bacteria around the disks with Antiqua Mare MAX had no significant differences compared to the disks with chlorhexidine, except for two cases (Table 3). First, *E. coli* ATCC 25922 strain was by 3.2 times ($p < 0.001$) more sensitive to chlorhexidine, and, secondly, the antifungal effect of Antiqua Mare MAX exceeded the activity of the conventional antiseptic by 1.4 times ($p < 0.01$).

Table 3

Combined effect of Antiqua Mare MAX and chlorhexidine, (M±m), n=5

Museum strain	The diameter of the zone of growth inhibition, mm		
	Antiqua Mare MAX	Chlorhexidine	Antiqua Mare MAX + Chlorhexidine
<i>E. coli</i> ATCC 25922	7.3±0.8	23.3±2.9*	22.6±3.3
<i>S. aureus</i> ATCC 25923	12.1±2.1	14.2±2.5	25.8±2.8**
<i>S. epidermidis</i> ATCC 14990	12.3±2.1	14.6±2.1	15.2±0.8
<i>E. faecalis</i> ATCC 29212	17.0±2.1	20.6±3.3	20.0±3.3
<i>M. lysodeicticus</i> ATCC 4698	11.2±2.5	12.5±0.8	12.2±3.0
<i>C. albicans</i> ATCC 10231	23.4±2.1*	17.2±2.5	23.9±3.4

Notes: 1. * – reliable compared to chlorhexidine, ($p < 0,01$); 2. ** – reliable compared to combination of preparations, ($p < 0,01$)

The effect of chlorhexidine, supplemented with study preparation, showed that the diameter of the zone of inhibition of *Staphylococcus aureus* growth increased by 2.1 times ($p < 0.01$) compared to that without Antiqua Mare MAX. The combined effect of the study preparation and chlorhexidine showed no increase in the zones of growth inhibition of other museum strains compared to this antiseptic, not supplemented with Antiqua Mare MAX (table 3). Apparently, the oral care product Antiqua Mare MAX increased the sensitivity of the museum strain of *S. aureus* ATCC 25923 to 0.02% chlorhexidine bigluconate. Obviously, given the antimicrobial effect of Antiqua Mare MAX, such synergy can be regarded as a summation [8]. Thus, the antimicrobial effect of Antiqua Mare MAX revealed synergy with another antimicrobial agent, chlorhexidine, which is a natural component of the pharmacodynamics of the novel preparation of Antiqua Mare MAX, which should be considered in the clinical use of the preparation, especially in combination therapy of oral infections, which is consistent with other researchers [1].

Conclusion

Fungistatic activity of the study preparation Antiqua Mare MAX exceeded the effect of 0.02% chlorhexidine bigluconate by 4 times ($p < 0.001$), though with similar fungicidal effect. For the *M. lysodeicticus* museum strain, the antibacterial effect of both preparations was the same, whereas for museum strains of *E. coli*, *S. aureus* and *S. epidermidis* antibacterial effect of Antiqua Mare MAX was weaker compared to 0.02% chlorhexidine bigluconate. The antifungal activity of Antiqua Mare MAX exceeded the activity of the conventional antiseptic to *C. albicans* ATCC 10231 museum strain by 1.4 times ($p < 0.01$), which was demonstrated by the method of paper disks.

The combination of the study preparation with chlorhexidine contributed to the tendency of increase the effect against the museum strain of *S. aureus* ATCC 25923.

In conclusion, the findings of the study have shown that Antiqua Mare MAX preparation can be recommended for the treatment of fungal stomatitis caused by *C. albicans*.

Prospects of further research will encompass the use of the Antiqua Mare MAX preparation in the comprehensive treatment of oral mucosa diseases and development of algorithm for its usage.

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Реферати

ВПЛИВ ПРЕПАРАТУ, ЩО ВМІЩУЄ ПРИРОДНИЙ МІНЕРАЛ БІШОФІТ, НА МІКРОФЛОРУ ПОРОЖНИНИ РОТА

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Порожнину рота можна розглядати як комплексну екологічну систему, в якій зовнішні фактори (біологічні, індивідуальні, соціальні) взаємодіють із внутрішніми (пародонт, бактеріальне співтовариство, локальна імунна система, епітелій порожнини рота). При виникненні сприятливих умов може розвинути те чи інше захворювання яке необхідно лікувати. Ми вивчали ефективність впливу засобу по догляду за ротовою порожниною Antiqua Mare MAX, що вміщує природний мінерал Бішофіт Полтавський, на представників мікрофлори порожнини рота. Для вирішення поставленої мети використовували музейні штами E. Coli ATCC 25922, S. Aureus ATCC 25923, S. Epidermidis ATCC 14990, E. faecalis ATCC 29212, M. Lysodeicticus ATCC 4698 та S. Albicans ATCC1023. У якості контролю використовували відомий антимікробний засіб - 0,02% водний розчин хлоргексидину біглюконат. Фунгіостатична активність дослідного препарату перевищувала дію 0,02% розчину хлоргексидину біглюконату у 4 рази ($p < 0,001$), але фунгіцидна дія не відрізнялась.

Ключові слова: мікрофлора СОПР, хлоргексидин біглюконат, Antiqua Mare MAX.

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ВЛИЯНИЕ ПРЕПАРАТА СОДЕРЖАЩЕГО ПРИРОДНЫЙ МИНЕРАЛ БИШОФИТ НА МИКРОФЛОРУ ПОЛОСТИ РТА

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Полость рта можно рассматривать как комплексную экологическую систему, в которой внешние факторы (биологические, индивидуальные, социальные) взаимодействуют с внутренними (пародонт, бактериальное сообщество, локальная иммунная система, эпителий полости рта). При возникновении благоприятных условий может развиваться то или иное заболевание слизистой оболочки полости рта которое необходимо адекватно лечить. Мы изучали эффективность воздействия средства по уходу за полостью рта Antiqua Mare MAX, вмещающий природный минерал бишофит Полтавский, на представителей микрофлоры полости рта. Для решения поставленной цели использовали музейные штаммы E. coli ATCC 25922, S. aureus ATCC 25923, S. epidermidis ATCC 14990, E. faecalis ATCC 29212, M. lysodeicticus ATCC 4698 и S. albicans ATCC1023. В качестве контроля применяли известное антимикробное средство - 0,02% водный раствор хлоргексидина биглюконат. Фунгиостатическая активность исследуемого препарата Antiqua Mare MAX превышала действие 0,02% раствора хлоргексидина биглюконата в 4 раза ($p < 0,001$), однако, фунгицидное действие не отличалась.

Ключевые слова: микрофлора СОПР, хлоргексидин биглюконат, Antiqua Mare MAX.

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EMG-ACTIVITY OF MUSCLES OF THE CRANIO-MANDIBULAR SYSTEM DURING FUNCTIONS OF THE DENTO-FACIAL REGION

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Electromyographic (EMG) activity of the temporal, masseter, orbicularis oris, mentalis, and sternocleidomastoid muscles was studied in 30 patients aged 20-28 years without somatic pathology, morphological, functional and aesthetic disorders in dento-facial region. The results of the study proved the involvement of the mimic and neck muscles in the teeth clenching, movements of the mandible and swallowing. The obtained data indicated the functional unity of the neuromuscular component of the stomatognathic system and the need to study the bioelectric activity of these muscles in subjects with functional disorders, especially associated with movements of the mandible and tongue at all stages of orthodontic treatment.

Key words: dento-facial region, electromyography, cranio-mandibular muscles, functions.

The work is a fragment of the research project "An interdisciplinary approach to the diagnosis, prevention and treatment of patients with dental malocclusion and jaw deformations", state registration No. 0018U004343.

Within the framework of the modern human functional development during the evolution of functions, such as speech, mental activity, self-awareness and social behavior, we can conclude: Masticatory organ is a highly organized multifunctional cybernetic system that works independently and interacts with various internal and external components, adapts permanently to the changing environmental factors, maintains a constant state of instable homeostasis (R. Slavicek, 2016) [2]. The dento-facial region is an integral part of the whole stomatognathic system of the person. The structural components of this cybernetic system are the cranio-mandibular (TMJ), neuro-muscular systems of the dento-jaw area and occlusion. The cranio-mandibular system, as a component of the neuro-muscular system of the human body, consists of the muscular apparatus (temporalis, masseter, lateralis and medialis pterygoideus muscles) and ligaments (spheno-mandibularis, stylo-mandibularis, stylo-hyoideus) [2, 3, 4]. These