SOME ASPECTS OF LABORATORY DIAGNOSTICS OF OPHTHALMODEMODECOSIS

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ABSTRACT

Demodicosis occupies a leading position among all dermatoses. According to ICD-10, it is not distinguished as a separate disease; it belongs to the class of parasitic diseases. The relevance of studying the problem of ophthalmodemodecosis is caused by its high prevalence and contagiousness, chronic course, an open issue of the role of the Demodex mite in the occurrence of inflammatory eye diseases, as well as the lack of effective methods for treating and preventing this pathology. In addition, Demodex causes discomfort and contributes to the occurrence of cosmetic defects, which in turn worsens the patient's quality of life.

The aim. *To present a review of the literature data and our own results of laboratory diagnostics of ophthalmodemodecosis.*

Materials and methods. *The article presents clinical cases of demodectic eyelid lesions with different disease outcomes in case of similar treatment. Laboratory diagnostics included drawing up an acarogram. Epilated eyelashes were used as a material for detecting mites on eyelids. Counting of individuals was carried out using light microscopy; all forms of mite development were taken into account. The work presents the statistics on the frequency of examination of patients with suspected demodicosis at different times of the year, confirming the seasonality of this disease. The literature review included data on the history of studying the Demodex mite, existing hypotheses and theories about its pathogenesis, as well as the information on domestic and foreign methods of treating ophthalmodecosis, including modern hardware techniques.*

Results. *Demodex mites play a significant role in the development of blepharitis and blepharoconjunctivitis. It is important to consider that demodicosis can occur against the background of inflammatory eye diseases of another etiology. Therapy for ophthalmodemodecosis currently remains complex, lengthy and ineffective. When assessing the results of an acarogram, any detected stages of a mite are clinically significant, and there isn't a direct relationship between the number of detected mites and the severity of clinical manifestations in all cases.*

Key words: *Demodex, blepharoconjunctivitis, laboratory diagnostics, ophthalmodemodecosis, IPL therapy*

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НЕКОТОРЫЕ АСПЕКТЫ ЛАБОРАТОРНОЙ ДИАГНОСТИКИ ОФТАЛЬМОДЕМОДЕКОЗА

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РЕЗЮМЕ

Демодекоз занимает лидирующее положение среди всех дерматозов. Согласно МКБ-10, он не выделяется как отдельное заболевание, относится к классу паразитарных болезней. Актуальность изучения проблемы офтальмодемодекоза обусловлена его высокой распространённостью и контагиозностью, хроническим течением, открытым вопросом о роли клеща Demodex в возникновении воспалительных заболеваний глаз, а также отсутствием эффективных методов лечения и профилактики данной патологии. Помимо этого, Demodex вызывает дискомфорт и способствует возникновению косметических дефектов, что в свою очередь ухудшает качество жизни пациента.

Цель работы. *Представить обзор литературных данных и собственных результатов лабораторной диагностики офтальмодемодекоза.*

Материалы и методы. *В статье представлены клинические случаи демодекозного поражения век с различными исходами заболевания при аналогичном лечении. Лабораторная диагностика включала в себя составление акарограммы. Материалом для обнаружения клещей на веках служили эпилированные ресницы. Подсчёт особей осуществлялся методом световой микроскопии, учитывались все формы развития клеща. Приведена статистика частоты обследования пациентов с подозрением на демодекоз в различное время года с подтверждением сезонности данного заболевания. Литературный обзор включал в себя данные об истории изучения клеща рода Demodex, существующие гипотезы и теории о его патогенезе, а также информацию об отечественных и зарубежных методах лечения офтальмодемодекоза, включая современные аппаратные методики.*

Результаты. *Клещ рода Demodex играет значительную роль в развитии блефаритов и блефароконъюнктивитов. Важно учитывать то, что демодекоз может возникать на фоне воспалительных заболеваний глаз другой этиологии. Терапия офтальмодемодекоза в настоящее время остаётся сложной, длительной и малоэффективной. При оценке результатов акарограммы клинически значимыми являются любые обнаруженные стадии клеща, а также не во всех случаях наблюдается прямая зависимость между количеством обнаруженных клещей и тяжестью клинических проявлений.*

Ключевые слова: *Demodex, блефароконъюнктивит, лабораторная диагностика, офтальмодемодекоз, IPL-терапия*

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Inflammatory eye diseases of demodectic aetiology have a high prevalence worldwide *Demodex* mites are known to play a role in the development of various ophthalmic diseases, as well as complicate their course [1]. These diseases include blepharitis, blepharoconjunctivitis, chalazion, episcleritis, and marginal keratitis. According to domestic studies, asymptomatic carrier state occurs in a significant number of patients. The incidence of demodecosis is similar in men and women. In persons over 70 years of age, the probability of detecting a mite is almost 100 % [2].

Demodecosis is an infectious disease caused by the parasitisation of the opportunistic mite of the genus *Demodex*. The mite reproduces in hair follicles, sebaceous glands and mainly affects the face and external auricles, and rarely the skin of the chest and back [3].

The *Demodex* mite was first described as a worm by Jacob Henle in 1841. It was later correctly classified as the human mite *Acarus folliculorum* by dermatologist Carl Gustav Theodore Simon. In 1843 Richard Owen calls the mite *Demodex*. In the course of studies in 1970, L.H. Akbulatova discovered and described two forms of the mite, *Demodex folliculorum brevis* and *Demodex folliculorum longus*, – parasitising humans [4].

Demodex folliculorum longus has a long body and its predominant site of localization is hair follicles. The second form of *Demodex folliculorum brevis* has a shorter body and parasitizes the meibomian and Zeiss glands. The entire body of the mite is covered with a chitinous shell, legs are short, with the presence of fingernails at the ends (Fig. 1).

After fertilization, males die and females lay eggs in the cavity of hair follicles. The development cycle of the mite consists of five phases and lasts about 15–25 days. *Demodex* feeds on the secretion of sebaceous glands and the cytoplasm of epithelial cells. The viability of mites is not affected by low temperatures or low humidity, but at temperatures below 14 °C they fall into a state of torpor. They are viable in water for up to 25 days (at a temperature of 12–15 °C), in pus and dead skin layers – 20 days, in dry air – 36 hours. *Demodex* mites are most active at temperatures of 30–40 °C, and at high temperatures (53–55 °C) they die after 18 days. In cosmetic cream, vegetable oil, petroleum jelly mites retain their vital activity for a long time [5].

There are several main hypotheses regarding the pathogenicity of the *Demodex* mite. Some authors believe that the mite manifests its pathogenicity by transporting microorganisms into the sebaceous glands and hair follicles. As an example, *Bacillus oleronius*, found on the tick surface, can activate both the ticks themselves and other microorganisms, and its proteins cause an increase in pro-inflammatory cytokines (interleukin (IL) 6 and IL-1b).

The following hypothesis is based on the fact that mites become active under the influence of endogenous and exogenous factors that contribute to a decrease in immunity (diabetes mellitus, gastrointestinal diseases, thyroid disorders, cardiovascular diseases, bad habits and stress) [6].

FIG. 1.
Demodex folliculorum longus mite. Photo was taken using Lomo MC-8.3 digital camera (Lomo JSC, Russia); magnification × 400

Foreign authors cite data reflecting the link between the rise in incidence in the spring and summer period, which is explained by increased synthesis of cathelicidin molecules supporting the inflammatory process (LL-37-natural antimicrobial barrier) as a result of the body's production of vitamin D under the influence of ultraviolet radiation [7].

According to the data of studies carried out in the Irkutsk branch of the Federal State Autonomous Institution «S.N. Fedorov Eye Microsurgery Centre» of the Ministry of Health of Russian Federation, it can also be observed that clinical manifestations of demodecosis have their seasonality (Fig. 2).

Demodex mites and their waste products cause chemical and mechanical irritation. With their jaws (chelicerae) they destroy skin cells. This leads to inflammation in the form of infiltrates and deposition of keratin protein and lipids in the stratum corneum. According to Yu.N. Koshevenko (2008), as a result of mite vital activity, the accumulation of metabolic products creates conditions for the attachment of various infections [8].

A.Ya. Varapetov (1972) asserts that «one of the results of mite activity is stagnation of sebum, which leads to prolonged irritation of the nerve-receptor apparatus of the sebaceous-hair follicle» [9].

Following another theory, mites of the genus *Demodex* are capable of inducing various immune responses. In the scientific literature, there is evidence of violations of the cellular link of immunity, manifested in a decrease in phagocytosis and expression of $CD3^+$, $CD4^+$, $CD8^+$, increased production of $CD22^+$, IgM, tumor necrosis factor α, IL-4, IL-6, increased phagocytosis and expression of $CD25^+$, $CD95^+$, HLA-DR+ [10].

There are studies upon which the theory of genetic predisposition to demodecosis disease is based [11].

To date, there is no consensus on the etiology and pathogenesis of the mite genus *Demodex*.

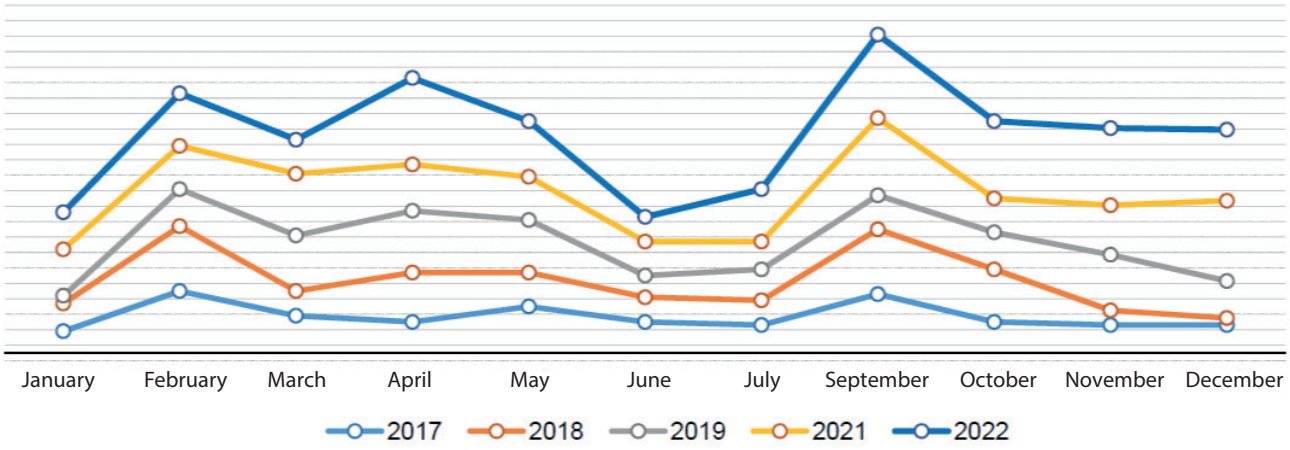

FIG. 2.
Distribution of the number of demodecosis laboratory tests (acarograms) during the year

All existing hypotheses and theories are contradictory and diverse.

In 1979, N.D. Zatsepina proposed the following classification of demodecosis of the visual organ: «asymptomatic carrier, obliterated forms, demodecosis blepharoconjunctivitis (uncomplicated, complicated), demodecosis episcleritis, demodecosis keratitis, demodecosis iridocyclitis» [12]. It is known that ophthalmodemodecosis can either act as an independent disease or be combined with demodecosis of other parts of the body.

At an ophthalmologist's appointment, patients have the following complaints: a feeling of a «foreign body» in the eyes, periodic discomfort, rubbing, itching of the eyelids, sometimes eyebrows, thick discharge in the corners of the eyes [13]. When examining the patient, the following is observed: eyelid margins are thickened, hyperaemic, meibomian gland ducts are filled with thick secretion, superficial plaque in the form of «muffs» at the root of the eyelashes is observed [14].

Only after laboratory diagnosis can a diagnosis of demodecosis be confirmed. For 1 day before the research the patient is recommended not to use decorative and medicinal cosmetics, as well as if possible not to use eye drops. The material for the study is epilated eyelashes, 4 pieces from each eyelid. This material is placed on a slide, in a drop of glycerol, covered with a coverslip and examined in a light microscope at a magnification of ×100.

The identification of the parasite is based on an acarogram by counting the different developmental stages (larvae, eggs and adults) (Fig. 3).

Acarogram is an objective diagnostic criterion. The presence of more than 4 specimens in the preparation is an indication of mite activity. As clinical practice demonstrates, large numbers of mites may be found on the eyelid margins in the obliterated clinic of blepharitis, and, conversely, in the vivid course of the disease that requires treatment, mites may be detected in small numbers.

The course of treatment for ophthalmodemodececosis may last 30–40 days, followed by retreatment in 2–3 months. The three-layer cuticle, which densely covers the surface of the *Demodex* mite, creates an obstacle to the action of the preparations.

Today the main method of blepharitis treatment in Russia is a complex application of anti-inflammatory, antibacterial, antiparasitic therapy with therapeutic eyelid hygiene and the use of tear substitutes.

Anti-inflammatory therapy is used to eliminate swelling and hyperaemia. Corticosteroid ointments, by reducing local immunity, increase the number of mites, so their use is not recommended [15].

Therapeutic eyelid hygiene is important in the treatment of ophthalmodemodecosis. This method of treatment was proposed by Professor G.S. Polunin in 2007 [16]. Three-stage eyelid hygiene is carried out: first, warm compresses improve metabolism; eyelid massage allows the evacuation of viscous secretions; then the eyelid margins are treated with disinfectants and acaricides.

Nowadays, there is a large number of modern devices for eyelid heating, moisturising and massage on the world market: in Russian ophthalmological practice, the most widely used are the Blephasteam mask [17] and the LipiFlow system, with the help of which the simultaneous effect on the inner surface of the upper and lower eyelid by heat and pulsating mechanical pressure is carried out.

In Europe, Optima IPL is used to treat dry eye disease and restore meibomian gland function. In 2002, V.G. Prieto et al. found that IPL (intense pulsed light)

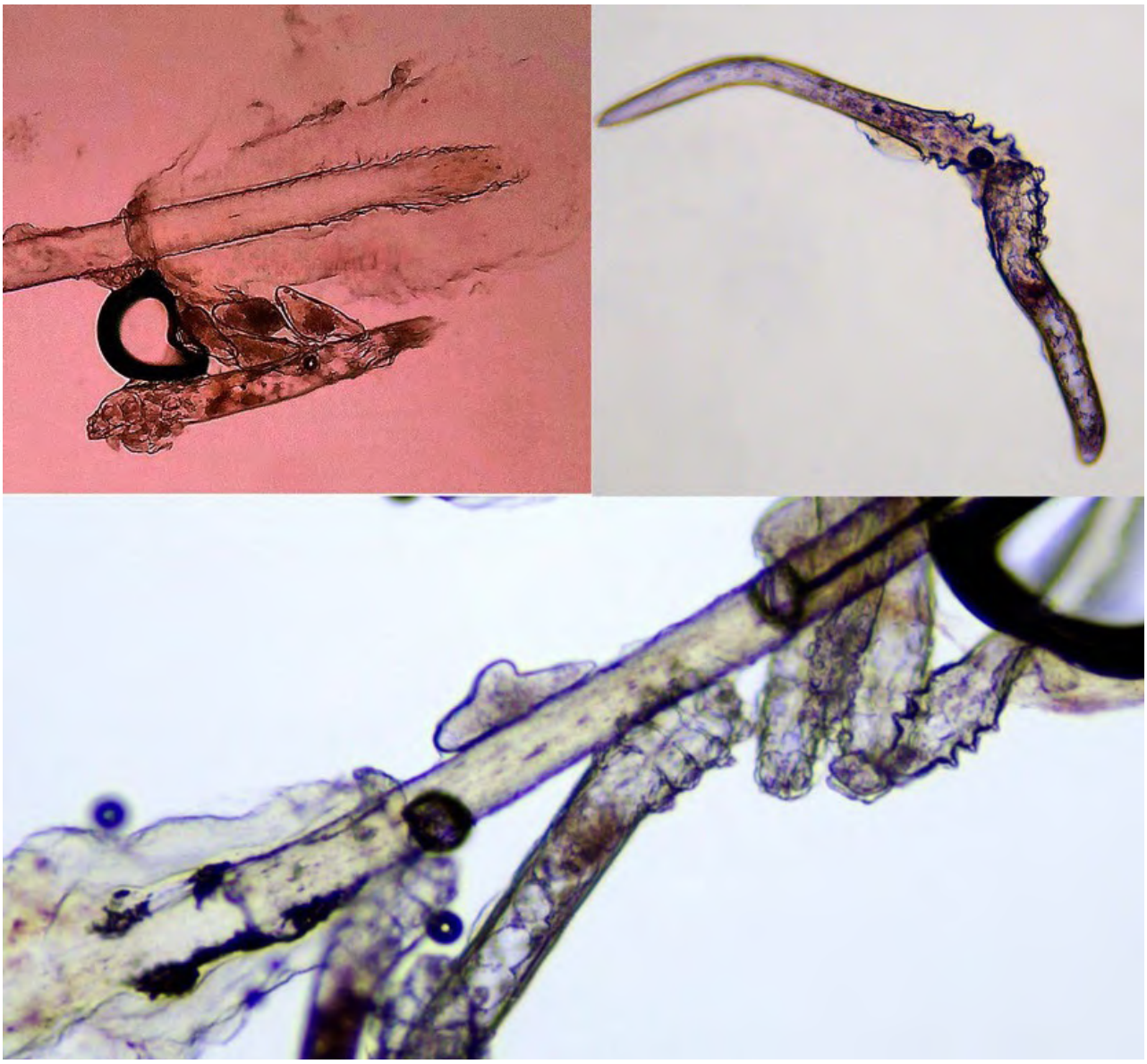

FIG. 3.
Developmental stages of the mite Demodex folliculorum longus: а – adult and 4 eggs; б – adult and larva; в – adult, larva and egg (magnification ×100)

therapy resulted in coagulation necrosis of *Demodex* mites and reduction of perifollicular lymphoid infiltrate [18]. Dr. Rolando Toyos has perfected a specific treatment protocol that maximises the long-term elimination of the *Demodex* mite [19].

After a month from the beginning of treatment a control acarogram is carried out, and in the absence of dynamics or an increase in the number of specimens, antiparasitic (acaricidal) drugs are used. Chloroform, carbolic acid, tar, cresol, ether and dichlophos are known to kill *Demodex* instantly. Preparations based on these substances are used in veterinary medicine and dermatology, but are not used in ophthalmology due to their toxicity. Metronidazole has proven to be highly effective over the years. The treatment of demodecosis with metronidazole, however, has not been successful in recent years [20]. Ornidazole may also be the drug of choice.

There are many data about the successful use of fluoroquinolones in the treatment of blepharitis [21]. Additionally, for the purpose of antibacterial therapy, benzildimethyl-myristoylamino-propylammonium-based preparations are prescribed as instillations into the conjunctiva [22].

It has also been observed that the consequence of impaired outflow of meibomian gland secretion is a decrease in the lipid layer of the tear film, which accelerates its evaporation and causes the development of «dry eye» syndrome [23]. The use of multicomponent lacrimal substitute preparations (LSPs), which include carmellose and hydroxypropylguar, has an advantage over the use of single-component LSPs, which has been statistically proven [24]. Alcoholic solutions of calendula, wormwood, tea tree oil, which have antimicrobial and antiseptic effect, are of great importance in the treatment of blepharitis of demodectic etiology.

It has been observed that cholinomimetics used in the treatment of glaucoma paralyses the musculature of mites due to muscarinic and nicotine-like action. Such drugs include 0.02 % phosphacol, physostigmine, 0.5 % tosmilene, 0.01 % armin [2].

The *Demodex* mite does not tolerate an alkaline environment and therefore alkaline eye drops and zinc sulphate drops in boric acid are used as symptomatic treatment.

Sanitation of foci of infection, treatment of concomitant diseases, limitation of sun exposure and dietary intake of polyunsaturated fatty acids such as Omega-3 are important in the treatment of ophthalmodemodecosis [25].

The above presented modern approaches to the diagnosis and treatment of ophthalmodemodecosis can be demonstrated by clinical cases.

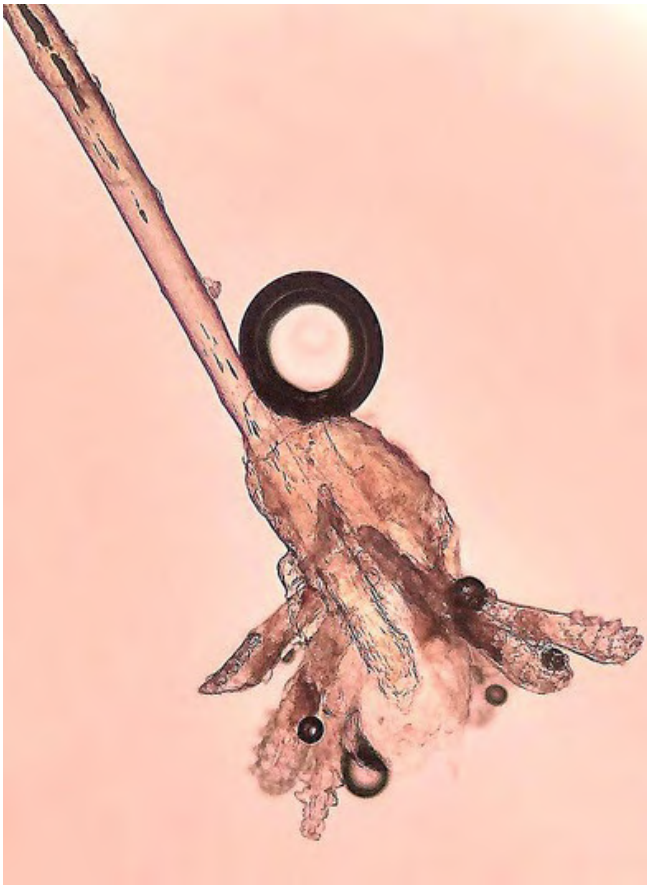

FIG. 4.
Demodex folliculorum longus mites. Magnification ×100; lens 10/0.25; eyepieces WF 10×/22

Clinical case No. 1

Patient L., 66 years old, complained of decreased near vision, dryness, discomfort, and itching of the eyelids of both eyes.

Biomicroscopy revealed the following: intermarginal eyelid margins thickened; meibomian glands filled with their own content. There are small gray muffs on the eyelashes.

The patient was examined for demodecosis; an acarogram revealed 8 adult specimens, 1 larva and 1 egg (Fig. 4).

Diagnosed with chronic demodectic blepharitis in both eyes.

Treatment was prescribed: eyelid massage No. 1–2; washing with tar soap; taking Trichopol; treatment of eyelid margins with Blepharogel-2, calendula alcohol solution. Oxyal drops (or Chilo-comod, Thealoz) 1 drop 3 times a day in both eyes for 1 month, then – as needed. Taking vitamin preparations with lutein, zeaxanthin, Omega-3, 6, 2 courses per year. Wearing sunglasses.

The patient came for a follow-up examination 2 months later. Acarogram results revealed 4 adults, 2 larvae and 1 egg.

Continuation of treatment was recommended; consultation with a dermatologist in the place of residence.

The patient came for a follow-up examination 2 months later.

Subjectively: after the treatment she observed improvement – disappearance of itching of eyelids of both eyes.

Objectively: eyelid skin without peculiarities; no *Demodex* mites were found according to the results of acarogram.

Clinical case No. 2

Patient V., 49 years old, complained of low vision of both eyes in the distance, discomfort, itching of the eyelids of both eyes. Visited a dermatologist at the place of residence for treatment of acne.

Concomitant diseases: euthyroidism.

When examined, the intermarginal margins of the eyelids are thickened.

The patient was tested for demodecosis; an acarogram revealed 18 adult specimens, 8 larvae and 7 eggs (Fig. 5).

Diagnosed with demodectic blepharitis in both eyes.

Treatment was prescribed: Stillavit drops (or Systein Ultra, Vizmed) 1 drop 3 times a day in both eyes for 1 month, further – as the circumstances require. Trichopol reception, eyelid treatment with Blepharolosione, Blepharogel-2. Re-examination after the course of treatment.

FIG. 5.
Demodex folliculorum longus mites. Photo taken using Lomo MC-8.3 digital camera (Lomo JSC, Russia); magnification ×100; objective 10/0.25; eyepieces WF 10×/22

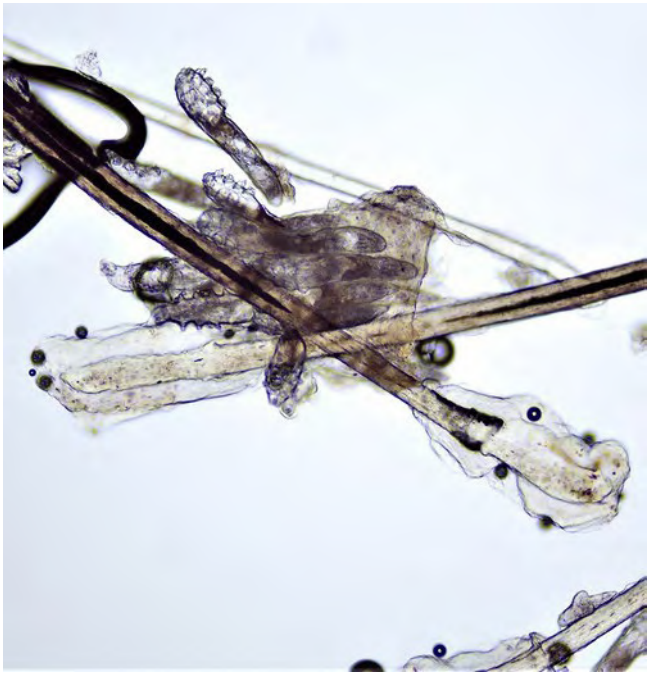

FIG. 6.
Demodex folliculorum longus mites (adults, larva and egg). Photo taken using Lomo MC-8.3 digital camera (Lomo JSC, Russia); magnification ×100; objective 10/0.25; eyepieces WF 10×/22

The patient came for a follow-up examination 1 month later.

Subjectively: observes a decrease in complaints.

Objective: intermarginal margins of eyelids thickened.

A repeat acarogram revealed 21 adults, 1 larva and 1 egg.

It is recommended to continue local treatment of demodectic blepharitis.

The patient came for a follow-up examination 3 months later.

Subjective: observes disappearance of complaints.

Objectively: eye adnexa without peculiarities.

Acarogram result: 5 adults and 1 larva were revealed.

Clinical case No. 3

Patient N., 49 years old, complained of insufficient vision in both eyes, periodic redness and peeling of the eyelid skin.

Examination: eyelid skin moderately hyperemic, single scales on eyelashes.

Acarogram result: 5 specimens were revealed.

Treatment of demodecosis was prescribed at the place of residence: heat compresses on eyelid skin 2 times a day for 2 months; Blefarolotion – treatment of eyelid skin edges 2 times a day for 2 months; Blefarogel-2 – treatment of eyelid skin edges 2 times a day for 2 months.

The patient came for a follow-up examination 2 months later.

Acarogram result: 14 specimens were revealed (Fig. 6).

Trichopol 0.25 mg regimen for 1 month was added to the treatment.

The patient came in after treatment for demodecosis for a follow-up examination 2 months later.

Examination: eyelid skin without peculiarities.

Acarogram result: 16 specimens were revealed.

Continued treatment was recommended.

The patient came for a checkup 4 years later.

Examination: eyelid skin pink, single «muffs» on eyelashes.

Acarogram result: 19 specimens were revealed.

Therefore, despite the ongoing therapy, no positive dynamics was obtained.

CONCLUSIONS

The data presented in this review suggest that despite the wide variety of drugs and cosmetics available for the treatment of ophthalmodemodecodosis, therapy remains difficult, time-consuming, and ineffective.

In treatment, in an attempt to treat the problem in a holistic manner, it is worth avoiding the abundance of prescribed medications, as this reduces patient adherence to treatment.

In assessing acarogram results, any mite stages that were found are clinically significant.

The use of ointments, tear-replacement drugs can aggravate the course of the disease.

The importance of *Demodex* mites in the development of blepharitis and blepharoconjunctivitis should not be underestimated, and it should be considered that demodecosis may occur against the background of inflammatory eye diseases of other etiologies.

Conflict of interest

The authors of this article declare no conflicts of interest.

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