Efficiency of 0.01% Dexamethasone Solution in Comprehensive Therapy of Dry Eye Disease

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Abstract: *Introduction:* At present anti-inflammatory therapy of patients with dry eye disease is based mainly on glucocorticoids' instillations. In spite of the fact that dexamethasone in officinal dosage (0.1%) has a marked local anti-inflammatory effect, its wide use is limited by the presence of a destructive process in the cornea. Taking this into account, the authors developed a drug containing 0.01% dexamethasone phosphate in combinations with 6% polyvinylpyrolidone and 1.5-5.5% dextrose solution.

Objective: To study the impact of the developed medication on the inflammatory process dynamics in the tissues of the eye surface in patients with dry eye disease of various etiology.

Materials and Methods: The material of this study was based on the results of the examination and treatment of 25 volunteers (50 eyes) with corneal-conjunctival xerosis developed on the background of 7 cases of meibomian blepharitis (14 eyes), 8 cases of perimenopause (16 eyes), and 10 cases of Sjögren's syndrome (20 eyes).

All patients used the developed medication in the form of eye drops 3-4 times daily on the background of tear replacement therapy. Prior to the therapy and on day 28 of the study the following parameters were assessed, lower tear meniscus index, precorneal tear film production, stability and osmolarity, staining of eye surface epithelium with sodium fluorescein solution, as well as bengal rose and lissamine green. The quantity of cytokines was determined in the tear fluid and blood plasma with ELISA method: interleukins-1β, 2, 4, 6, 8, 10, 17A, interleukin 1 receptor antagonist, TNF α, interferons α and γ. Besides, all the subjects were asked to fill in a questionnaire to evaluate subjective signs of the ocular surface epithelium xerosis or Ocular Surface Disease Index (OSDI).

Results: By day 28 of the study statistically valid increase of the tear meniscus index, precorneal tear film stability, main and total tear production and decrease of tear film osmolarity were observed. Besides, the staining degree of the ocular surface epithelium with bengal rose and lissamine green (van Bijsterveld scale) and with sodium fluorescein (Oxford scale) decreased. Also, the positive dynamics of the objective parameters of the ocular surface epithelium is confirmed by the subjects' patients evaluation of their quality of life.

Conclusion: The results of the study performed prove the high clinical efficiency of the developed medication that has a marked local anti-inflammatory effect in the therapy of dry eye disease of various etiology.

Keywords: Anti-inflammatory therapy, dry eye disease, the quantity of cytokines in the tear fluid.

1. INTRODUCTION

For many years, the dry eye disease (DED) occupies an important place in the structure of opthalmic pathologies. On the one hand, this is connected with wide prevalence of the disease under consideration, on the other hand, with the severity of the clinical course and outcomes of some of its clinical forms [1].

At that, DED clinical manifestations, viz. the development of the so called corneal-conjunctival xerosis, are often followed by irreversible morphological changes of the conjunctiva, mainly the cornea. And, as demonstrated by the practice, those may be seen in a wide range: from minimal dystrophic changes of the epithelium to deep destructive process, progressive corneal ulcer or even keratomalacia [1, 2].

As known, the central link of DED pathogenesis is the precorneal tear film stability violation followed by the increase of its vaporization ability and osmolarity, and as the result, development of inflammatory processes in ocular surface tissues [1]. Hence, the important vector of pathogenetically oriented DED therapy is the use of anti-inflammatory medications.

Traditionally, anti-inflammatory therapy is based on the use of glucocorticoids, in particular, dexamethasone phosphate. This is connected with the fact that the latter blocks such transcription factors as nuclear factor kβ (NF-kβ) and protein activator 1 (AP–1), thus repressing DNA binding and inhibiting further transcription of IL-2 key cytokine that regulates

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cellular immune response. Besides, under the impact of dexamethasone phosphate the number of T-cells decreases, as well as their influence on B-cells, the production of immunoglobulins slows down, while the formation of the components of the complement system goes down and their degradation goes up.

However, long-term use of dexamethasone phosphate in officinal dosage (0.1%) is limited by a wide range of its side effects: IOP increase, development of steroid cataract and glaucoma, thinning of xerotically changed cornea, that lead to the progression of the ulcerative process and the development of corresponding complications. Taking this into account, the authors developed a drug containing 0.01% dexamethasone phosphate in combinations with 6% polyvinylpyrolidone and 1.5-5.5% dextrose solution [4].

As is commonly known, polyvinylpyrolidone is a polimer base of many modern "artificial tear" preparations, since it stimulates endogenous interferon generation, increases the wettability of the hydrophobic corneal epithelium and conjunctiva and improves tolerability of pharmacologically active drugs when used as drops into conjunctiva cavity. The addition of dextrose solution is substantiated by its property to stabilise cellular membranes of the ocular surface epithelium.

This study deals with the efficiency of the use of our worked out medication, in comprehensive therapy of patients with typical corneal-conjunctival xerosis pathologies.

2. OBJECTIVE

Study the impact of the developed medication on the inflammatory process dynamics in the tissues of the eye surface in patients with dry eye disease of various etiology.

3. MATERIALS AND METHODS

The material of the study consisted of the results of examination and therapy of 25 volunteers (50 eyes): 6 males (24%) and 19 females (76%) at the age from 40 to 80 years (on average, 60.44±11.1 yrs) with dry eye disease of various etiology: 7(14 eyes)–due to meibomian blepharitis, 8(16) - perimenopause, and 10(20) - Sjögren's syndrome.

It is known that most important in DED pathogenesis in case of meibomian blepharitis is precorneal tear film stability violation, in case of perimenopause - tear film lipids and mucins production drop down, and in case of Sjögren's syndrome - the simultaneous decrease of secretion of all components of precorneal tear film. Thus, the subjects included patients with all the main pathogenetic DED types.

Control group amounted to 25 volunteers (50 eyes): 5 males (20%) and 20 females (80%) at the age from 37 to 78 years (on average, 62.5±10.9 yrs) with DED of the same etiology: 6 subjects (12 eyes) with DED due to meibomian blepharitis, 9 (18) - due to perimenopause, and 10(20) - due to Sjögren's syndrome.

All the subjects from both groups received instillations into conjunctival cavity with preservative-free tear replacement agent on the basis of polyvinylpyrrolidone and polyvinyl alcohol–Ophtholique BK® (Sentiss, Pvt. Ltd., India), on average, 3-4 times daily, and the subjects from the main group received also the medication developed by the authors, 2-3 times daily.

Comprehensive examination was carried out prior to the study and on day 28. In particular, subjective signs of epithelium xerosis were assessed with the help of the Ocular surface disease index (OSDI) [5]. Functional examination included evaluation of the lower tear meniscus index, precorneal tear film stability according to M.S. Norn (1969), pronouncement of the bulbar conjunctiva fold at the eyelid free margin according to H. Hoh (2006) technique, measuring the main and total tear production values according to L. Jones (1966) and O. Schirmer (1903). In the course of ocular surface biomicroscopy the degree of its staining with bengal rose and lissamine green solutions using the 4-point van Bijsterveld score, and with sodium fluorescein using the Oxford score was studied [6-11]. In order to increase the accuracy and comparability of the study results, the authors limited themselves to quantitative assessment of staining degree of cornea only.

Besides that, osmolarity of precorneal tear film was measured in all the subjects with TearLab Osmolarity System device (TearLabCorp., USA). Interleukins 1β, 2, 4, 6, 8, 10, 17A, IL-1 receptor antagonist, tumour necrosis factor α, interferons α and γ (IL-1β, IL-2, IL-4, IL-6, IL-8, IL-10, IL-17A, IL-1Ra, TNF-α, INF-α, INF-γ, respectively). To carry out lab tests, 0.5ml of tear fluid was needed. Provided that usually one cannot get sufficient amount of tear fluid from DED patients, the authors used the following technique.

After local epibulbar anaesthesia with 0.4% oxybuprocaine solution (Inocaine®, Sentiss, Pvt. Ltd., India) 1.0ml of 0.9% sodium chloride was instilled drop by drop into conjunctival cavity with the help of insulin syringe (without a needle) and immediately collected lavage fluid with disposable pipette into sterile and labelled eppendorf. In the result of these manipulations, 0.5ml of tear fluid was collected in eppendorf ("diluted" in standard amount of isotonic sodium chloride) [12]. The tear fluid from the conjunctival cavity of the second eye was collected in a similar way. Prior to lab tests stage, the material thus collected was stored in the freezer at -60.0°C.

In recent years, thanks to intensive studies of lymphoid tissue associated with mucous membranes (MALT), it became evident that it is involved in all the processes that take place in mucous membranes, physiological, as well as pathological, including inflammatory ones. lymphoid tissue associated with gastrointestinal and urogenital tracts, bronchi, as well as the ocular surface. Eye-associated lymphoid tissue (EALT) is represented by lymphoid tissue associated with lacrimal gland, conjunctiva, and lacrimal passages. It belongs to peripheral parts of the immune system organs and is closely connected with it by way of blood-ocular barrier [13-15]. Due to this very reason, the authors defined the levels of cytokines listed above in blood plasma as well using ELISA test.

4. RESULTS OBTAINED AND DISCUSSION.

The dynamics of the clinical and physiological signs of the corneal-conjunctival xerosis in patients with DED of various pathogenetic types in both groups under comparison is given in Tables **1-3**.

In DED patients on the background of meibomian blepharitis (see Table **1**) on day 28 of therapy with medication containing 0.01% dexamethasone phosphate in combination with 6% polyvinylpyrolidone and 1.5-5.5% dextrose pronouncement of ocular surface xerosis subjective signs decreased with statistical significance ($p<0.001$). At the same time, statistically valid increase of the tear meniscus index and total tear production were found ($p<0.01$), as well

Table 1: The Dynamics of Clinical and Functional DED Parameters in Patients with Meibomian Blepharitis on the Background of the Performed Therapy

Evaluated Parameter	Observational Stages (days)							
	Main Group(n=14)				Control Group(n=12)			
	Initial Data	Day 28 of Therapy	t	p	Initial Data	Day 28 of Therapy	t	P
Ocular surface disease index	84.52±4.96	30.05±5.08	18.8	<0.001	73.61±4.91	37.15±4.72	12.0	<0.001
Osmolarity, mOsm/l	320.86±11.39	293.86±13.25	5.6	< 0.05	323.83±8.73	302.58±10.02	5.3	>0.05
Tear meniscus index	1.07±0.25	2.28±0.45	8.7	< 0.01	0.91±0.25	1.41±0.49	2.4	>0.05
Pronouncement of the bulbar conjunctiva fold at the eyelid free margin	1.78±0.67	0.57±0.62	4.9	>0.05	1.83±0.55	0.91±0.49	4.1	>0.05
Precorneal tear film stability, s	5.43±1.65	9.28±0.72	7.7	<0.05	5.09±0.49	8.71±0.61	15.7	<0.05
Main tear production, mm/5 min	2.03±0.81	3.54±0.70	5.2	>0.05	2.64±0.36	4.47±0.69	8.0	<0.05
Total tear production, mm/5 min	4.43±1.14	9.60±1.71	9.1	<0.01	7.74±0.56	8.76±0.21	5.7	>0.05
Epithelium assessment by vanBijsterveld score using bengal rose solution, points	4.64±0.61	1.14±0.63	14.6	<0.001	4.58±0.49	2.58±0.40	9.5	<0.01
Epithelium assessment by vanBijsterveld score using lissamine green solution, points	4.50±0.98	1.42±0.49	10.2	<0.001	4.75±0.72	2.41±0.41	9.0	<0.01
Epithelium assessment by Oxford score using sodium fluorescein solution, points	3.07±0.70	0.64±0.48	10.6	<0.05	2.33±0.47	1.75±0.43	3.1	>0.05

* Ratio between the tear meniscus height and base - tear meniscus index value: 1(decrease of moisture amount in the conjunctiva cavity); 2 (normal moisture amount); 3 (increase of moisture amount) (L.P. Prozornaya, V.V. Brzheskiy, 2006);

** 0 – no staining; 1 – weak; 2 – moderate; 3 – diffuse staining;

*** 0 – no staining; 1 – minimal; 2 – weak; 3 – moderate; 4 – diffuse; 5 – total.

as the decrease of the degree of staining of conjunctival and corneal epithelium with bengal rose, lissamine green and sodium fluorescein solutions ($p<0.05$-0.001). Besides, subjects from this group while receiving the medication under study demonstrated significant decrease of precorneal tear film osmolarity ($p<0.05$). It should be mentioned, that the pronouncement of bulbar conjunctiva fold at the eyelid free margin and total tear production were statistically insignificant on the background of the performed therapy ($p>0.05$).

In the control group, on day 28 the authors also found statistically significant decrease of the intensity of ocular surface epithelium xerosis subjective signs ($p<0.001$), significant increase of precorneal tear film stability, total tear production and decrease of the staining degree of ocular surface epithelium with vital staining agents ($p<0.01$-0.05). But the above mentioned clinical and functional parameters were less pronounced compared to those of the patients of the main group that regularly took the developed medication.

Besides, the tear fluid osmolarity, tear meniscus index, pronouncement of the bulbar conjunctive fold at the eyelid free margin, total tear production and staining degree of corneal epithelium were statistically insignificant ($p>0.05$) on the background of the performed therapy with preservative-free tear replacement agent containing polyvinylpyrrolidone and polyvinyl alcohol.

Thus, after the instillations of the medication offered, patients with DED due to meibomian blepharitis demonstrated significant decrease of the intensity of xerosis subjective signs, which is confirmed by objective research methods (See Figure **1** and **2**).

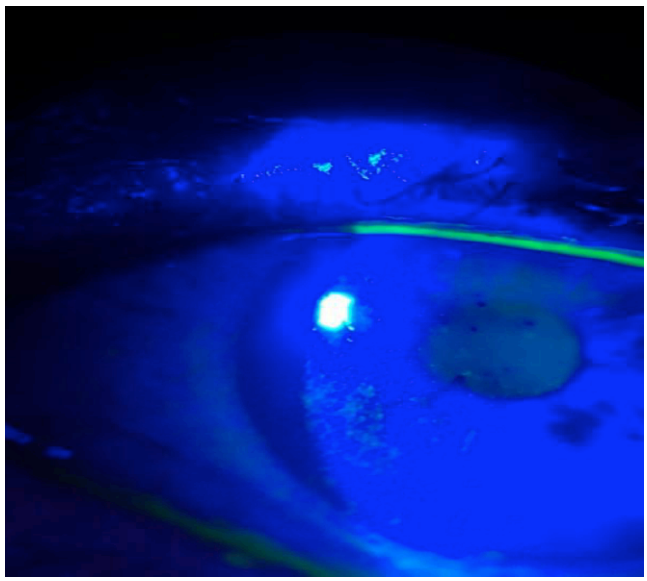

Figure 1: Degree of staining of corneal epithelium with sodium fluoroscein prior to therapy.

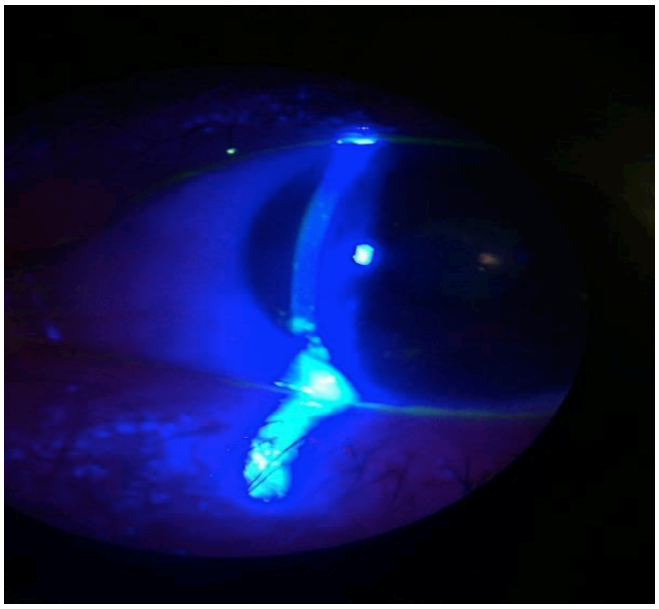

Figure 2: Degree of staining of the corneal epithelium with sodium fluoroscein on Day 28 of therapy.

Similar dynamics of the evaluated parameters was also found when treating females with DED on the background of perimenopause (see Table **2**). Thus, by day 28, on the background of the therapy performed there was a significant increase of the tear meniscus index, precorneal tear film stability, as well as main and total tear production ($p<0.05$-0.001). Besides, the degree of staining of the corneal and conjunctival epithelium with vital staining agents (See Figure **3**, **4**), as well as the intensity of the ocular surface epithelium xerosis subjective signs were significantly lower compared to the initial data ($p<0.01$-0.001). At the same time, precorneal tear film osmolarity and pronouncement of bulbar conjunctiva fold at the eyelid free margin, not statistically significant compared to the initial values ($p>0.05$), though they had a trend to decrease.

In the control group, the subjects showed statistically valid increase of precorneal tear film stability, tear production values and decrease of the staining degree of ocular surface epithelium with bengal rose and lissamine green ($p<0.01$-0.001). The objective data were followed by the reduction of corneal-conjunctival xerosis subjective signs ($p<0.01$). In spite of this, the pronouncement of mentioned clinical and functional parameters prevailed in the main group of patients. At the same time, precorneal tear film osmolarity, as well as tear meniscus index and pronouncement of bulbar

Table 2: The Dynamics of Clinical and Functional DED Parameters in Perimenopausal Females on the Background of the Performed Therapy

Evaluated Parameter	Observational Stages (days)							
	Main Group(n=16)				Control Group(n=18)			
	Initial Data	Day 28 of Therapy	t	p	Initial Data	Day 28 of therapy	t	p
Ocular surface disease index	75.51±5.29	25.51±4.51	19.1	<0.001	77.77±3.40	42.59±4.51	17.7	<0.01
Osmolarity, mOsm/l	315.06±13.41	301.81±11.27	2.9	>0.05	328.44±12.44	309.11±6.72	5.7	>0.05
Tear meniscus index	0.81±0.39	2.06±0.65	6.6	< 0.05	0.94±0.53	1.66±0.47	4.3	>0.05
Pronouncement of the bulbar conjunctiva fold at the eyelid free margin	1.87±0.78	0.50±0.50	6.0	>0.05	2.00±0.66	1.05±0.52	4.7	>0.05
Precorneal tear film stability, s	5.79±1.20	10.34±0.96	11.7	< 0.01	4.72±0.83	8.73±1.12	12.2	<0.01
Main tear production, mm/5 min	1.99±0.67	4.50±0.48	12.0	<0.001	2.27±0.75	4.83±0.53	11.6	<0.01
Total tear production, mm/5 min	5.20±1.05	9.50±1.13	11.1	<0.01	7.71±0.33	9.12±0.39	11.8	<0.01
Epithelium assessment by vanBijsterveld score using bengal rose solution, points	4.43±0.99	1.37±0.69	9.9	<0.001	5.16±0.50	2.44±0.49	16.1	<0.001
Epithelium assessment by vanBijsterveld score using lissamine green solution, points	4.56±1.41	1.75±0.83	6.7	<0.001	5.05±0.52	2.94±0.52	12.5	<0.001
Epithelium assessment by Oxford score using sodium fluorescein solution, points	3.06±0.74	0.56±0.49	10.9	<0.01	2.66±0.57	1.61±0.48	5.9	>0.05

*Ratio between the tear meniscus height and base - tear meniscus index value: 1(decrease of moisture amount in the conjunctiva cavity); 2 (normal moisture amount); 3 (increase of moisture amount) (L.P. Prozornaya, V.V. Brzheskiy, 2006);

**0 – no staining; 1 – weak; 2 – moderate; 3 – diffuse staining;

***0 – no staining; 1 – minimal; 2 – weak; 3 – moderate; 4 – diffuse; 5 – total.

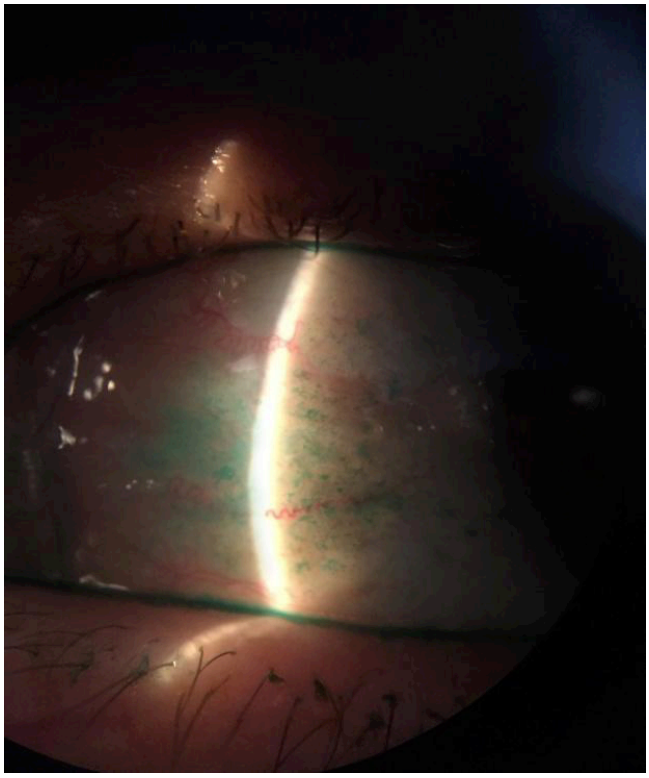

Figure 3: Degree of staining of corneal epithelium with lissamine green solution prior to therapy.

conjunctiva fold at the eyelid free margin were statisctically insignificant compared to initial values (p>0.05).

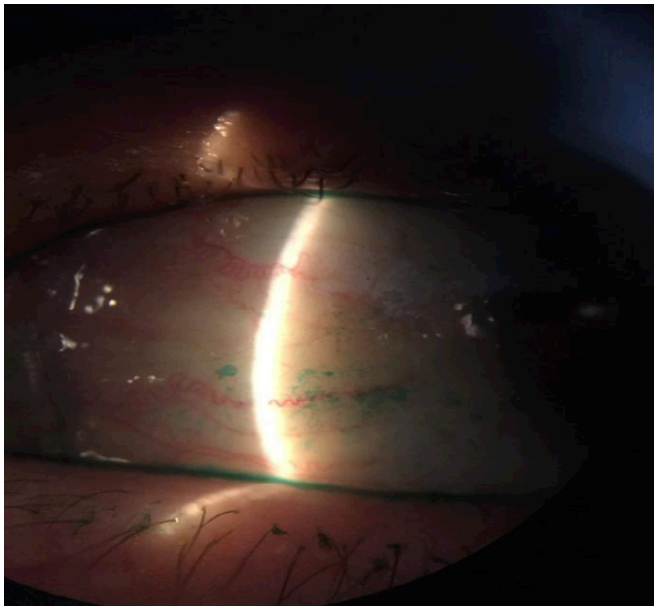

Figure 4: Degree of staining of corneal epithelium with lissamine green solution on Day 28 of therapy.

Table 3: The Dynamics of Clinical and Functional DED Parameters in Patients with Sjögren's Syndrome on the Background of the Performed Therapy

Evaluated Parameter	Observational Stages (days)							
	Main Group(n=20)				Control Group(n=20)			
	Initial Data	Day 28 of Therapy	t	p	Initial Data	Day 28 of therapy	t	p
Ocular surface disease index	74.17±6.79	23.75±5.60	17.2	<0.001	82.08±4.85	44.58±3.86	18.2	<0.01
Osmolarity, mOsm/l	342.40±12.11	316.95±6.82	8.0	< 0.05	340.60±11.76	321.85±6.10	6.2	>0.05
Tear meniscus index	0.20±0.40	0.85±0.57	4.1	>0.05	0.45±0.49	0.80±0.50	2.2	>0.05
Pronouncement of the bulbar conjunctiva fold at the eyelid free margin	1.90±0.63	0.40±0.58	7.9	< 0.05	2.15±0.48	0.85±0.36	5.9	>0.05
Precorneal tear film stability, s	5.86±1.34	9.57±1.20	9.0	< 0.05	4.96±0.69	8.07±1.22	9.7	<0.05
Main tear production, mm/5 min	2.01±0.64	4.11±0.67	10.0	<0.01	1.69±0.43	3.82±1.01	8.6	<0.01
Total tear production, mm/5 min	5.31±1.43	9.93±2.10	8.0	<0.05	5.83±0.50	8.60±0.44	11.8	>0.05
Epithelium assessment by vanBijsterveld score using bengal rose solution, points	4.50±0.50	1.70±0.46	18.7	<0.001	5.30±0.45	3.40±0.66	10.9	<0.05
Epithelium assessment by vanBijsterveld score using lissamine green solution, points	4.65±1.35	2.10±0.70	7.5	<0.01	5.20±0.40	3.40±0.48	12.9	<0.05
Epithelium assessment by Oxford score using sodium fluorescein solution, points	3.05±0.67	0.55±0.58	12.5	<0.01	2.80±0.50	1.85±0.49	7.8	>0.05

* Ratio between the tear meniscus height and base - tear meniscus index value: 1(decrease of moisture amount in the conjunctiva cavity); 2 (normal moisture amount); 3 (increase of moisture amount) (L.P. Prozornaya, V.V. Brzheskiy, 2006);

**0 – no staining; 1 – weak; 2 – moderate; 3 – diffuse staining;

***0 – no staining; 1 – minimal; 2 – weak; 3 – moderate; 4 – diffuse; 5 – total.

Observation results with patients having DED due to Sjögren's syndrome and receiving the study drug demonstrated a trend similar to the one described above (see Table **3**). Thus, on day 28 of therapy there was a statistically valid decrease of DED objective and subjective signs intensity ($p<0.05$-0.001). At the same time, though the values of tear meniscus index increased with the instillations of study drug, but these changes were not statistically significant compared to the initial values ($p>0.05$).

In the control group, patients with DED due to Sjögren's syndrome demonstrated in day 28 of therapy statistically significant increase of precorneal tear film stability, decrease of staining degree of the epithelium with bengal rose (See Figure **5** and **6**) and lissamine green solutions, followed by the decrease of intensity of corneal-conjunctival xerosis subjective signs ($p<0.01$-0.05). However, tear fluid osmolarity, tear meniscus index, pronouncement of the bulbar conjunctiva fold at the eyelid free margin, as well as total tear production and staining degree of corneal epithelium with sodium fluorescein not differ significantly from initial values ($p>0.05$).

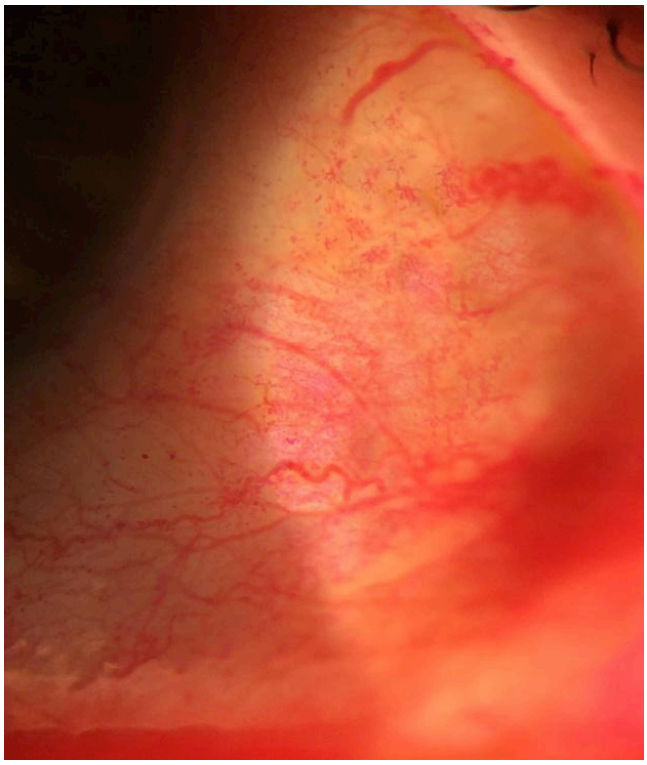

Figure 5: Degree of staining of the corneal epithelium with bengal rose solution prior to therapy.

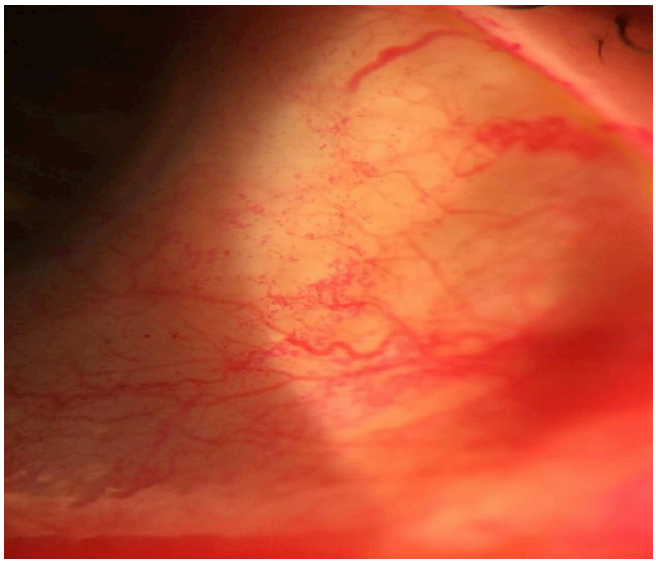

Figure 6: Degree of staining of the corneal epithelium with bengal rose solution on Day 28 of therapy.

In order to study anti-inflammatory activity of the medication offered, immunological study of the cytokunes' level in tear fluid and blood plasma was performed in patients with main pathogenetic types of corneal-conjunctival xerosis.

The values of tear fluid immunological parameters of the patients from the main and control groups are given in Table **4**.

It was found, that on the background of the therapy performed in the group of patients with DED due to meibomian blepharitis using the medication containing 0.01% dexamethasone phosphate in combination with 6% polyvinylpyrolidone and 1.5-5.5% dextrose the level of pro-inflammatory cytokines decreased. In particular, the level of IL-1β, IL-2, IL-6, IL-8, IL-1Ra and TNF-α was 1.4-2.4 times lower compared to the initial data ($p<0.05$-0.001). In spite of the decrease of pro-inflammatory cytokines IL-4 and IL-17A in the tear fluid, the differences of these levels and initial ones were not statistically significant ($p>0.05$). However, the level of IL-10, that has a marked immunosuppressive effect, outstandingly increased 2 times ($p<0.05$). Production of INF-α following the performed therapy decreased 3.2 times ($p<0.001$), which indirectly confirms the activation of innate immunity. Thus, the developed medication has a pronounced anti-inflammatory effect against the chronic inflammatory process in the eye surface epithelium that is manifested by the decrease of main pro-inflammatory cytokines production and the increase of anti-inflammatory cytokines level.

In the control group of patients with DED of the same etiology, on Day 28 of therapy statistically significant decrease of IL-6, IL-8, IL-17A, IL-1Ra, TNF-α levels is observed, however, only 1.1-1.5 times ($p<0.05$). The content of IL-1β, IL-2, IL-4, IL-10 and INF-γ on the background of therapy performed was not statistically significant against the initial levels ($p>0.05$).

Similar results were obtained in lab testing of the patients with DED of a different etiology.

Thus, in patients with DED on the background of premenopause the content of IL-1β, IL-2, IL-6, IL-8, IL-17A, IL-1Ra, TNF-α, INF-α in the tear fluid was decreased 1.1-2.7 times ($p<0.05$-0.001), the level of anti-inflammatory cytokine IL-10 was increased 1.7 times ($p<0.01$), while the content of IL-4 was not significantly changed ($p>0.05$).

In the control group of patients with DED of the same etiology, statistically valid 1.2-1.5 times decrease of IL-6, IL-8, IL-1Ra, TNF-α, INF-α levels was observed ($p<0.05$). However, the levels of IL-1β, IL-2, IL-4, IL-17A, as well as IL-10 on the background of the therapy performed did not change significantly ($p>0.05$).

In the group of patients with DED due to Sjögren's syndrome on the background of therapy with the medication under study the 1.3-2.9 times decrease of the IL-1β, IL-2, IL-6, IL-8, IL-17A, IL-1Ra, TNF-α, INF-α levels ($p<0.05$-0.001), statistically insignificant decrease of IL-4 level ($p>0.05$) and 2 times' increase of IL-10 level ($p<0.05$) were observed.

In the control group of patients, on the background of the therapy only 1.2-1.5 times' decrease of IL-1β, IL-2, IL-6, IL-8, IL-1Ra, TNF-α levels was observed ($p<0.05$-0.01). As in the previous control groups, the decrease of IL-4, IL-17A, INF-α, as well as the increase of pro-inflammatory IL-10 was not significant ($p>0.05$).

The level of INF-γ in the tear fluid on the background of the therapy performed in the main and control groups did not change significantly compared to the initial levels ($p>0.05$).

Thus, all the patients with DED of various etiology following the therapy with the drug being studied containing 0.01% dexamethasone phosphate combined with 6% polyvinylpyrolidone and 1.5-5.5% dextrose demon-strated the change of local immunity in the form of pro-inflammatory cytokines' levels' decrease and anti-inflammatory cytokines' levels increase.

Table 4: Dynamics of Lab Test Values of Tear Fluid from Patients with DED of Various Pathogenetic Types on the Background of Performed Therapy

Observational Stages			Evaluated Parameter, pg/ml										
			IL-1β	IL-2	IL-4	IL-6	IL-8	IL-10	IL-17A	IL-1Ra	TNF-α	INF- α	INF- γ
DED Due to Meibomian Blepharitis	Main Group (n=14)	Initial data	29.36± 4.15	275.78± 24.53	29.44± 3.14	39.01± 8.06	69.75± 4.11	12.00± 3.04	177.84± 9.85	338.43± 15.38	96.05± 6.75	17.37± 2.82	41.12± 12.76
		Day 28 of therapy	17.53± 3.89	131.81± 22.39	19.41± 4.61	24.13± 6.06	29.30± 5.52	24.96± 4.63	159.42± 20.81	229.74± 41.01	66.39± 9.15	5.47± 2.50	38.36± 10.16
		t	7,5; p<0.05	15,6; p<0.001	6,5; p>0.05	4,6; p<0.05	21,3; p<0.001	8,5; p<0.05	2,9; p>0.05	9,0; p<0.001	9,4; p<0.01	11,4; p<0.001	0.6; p>0.05
	Control group (n=12)	Initial data	24.25± 4.88	239.83± 9.32	26.66± 2.43	36.66± 3.17	63.75± 8.54	9.66± 2.11	177.91± 6.82	315.92± 4.29	98.42± 5.56	18.42± 2.89	22.16± 7.26
		Day 28 of therapy	19.92± 2.36	187.58± 12.24	24.50± 4.21	26.91± 2.78	42.83± 5.75	14.33± 3.88	153.33± 9.45	247.16± 10.05	70.42± 7.56	12.75± 10.74	30.25± 4.04
		t	2,7; p>0.05	11,3; p>0.05	1,5; p>0.05	7,7; p<0.05	6,7; p<0.05	3,6; p>0.05	7,0; p<0.05	20.9; p<0.001	9,9; p<0.05	5,6; p<0.05	3,2; p>0.05
DED Due to Perimenopause	Main group (n=16)	Initial data	21.93± 3.68	232.82± 30.28	28.41± 5.38	35.98± 6.90	70.63± 4.93	9.95±3.1 9	177.58± 11.95	308.43± 24.59	85.07± 5.49	18.43± 2.98	38.37± 11.31
		Day 28 of therapy	11.09± 3.77	122.08± 13.47	18.73± 4.18	25.63± 4.93	30.55± 5.64	17.55± 3.59	155.02± 6.48	228.39± 29.66	59.26± 6.43	6.74± 1.34	25.51± 5.67
		t	8,0; p<0.05	13; p<0.001	5,5; p>0.05	4,7; p<0.01	20.8; p<0.001	6,1; p<0.01	6,4; p<0.05	8,1; p<0.05	11,8; p<0.01	13,9; p<0.01	4,3; p>0.05
	Control group (n=18)	Initial data	25.44± 5.42	206.05± 20.35	28.55± 3.89	38.88± 4.43	61.33± 8.01	9.50± 1.89	166.72± 10.36	314.77± 5.67	95.44± 6.37	17.55± 2.79	24.55± 8.54
		Day 28 of therapy	17.72± 2.99	149.27± 13.59	25.55± 3.79	27.88± 3.99	37.94± 3.49	14.61± 4.08	140.38± 13.95	248.83± 16.57	68.27± 6.43	11.72± 2.44	25.94± 7.92
		t	5,1; p>0.05	9,6; p>0.05	2,3; p>0.05	7,6; p<0.05	11,1; p<0.01	4,5; p>0.05	6,3; p>0.05	15,6; p<0.05	12,4; p<0.05	6,5; p<0.01	0.5; p>0.05
DED Due to Sjögren's Syndrome	Main group (n=20)	Initial data	32.08± 6.57	303.23± 33.54	27.37± 5.78	65.14± 8.57	79.73± 6.99	7.65± 1.97	225,25± 15.27	316.48± 30.55	101.54 ±18.17	14.50± 3.40	33.03± 14.85
		Day 28 of therapy	12.37± 1.79	124.70± 25.7	21.66± 4.75	36.89± 4.77	27.23± 4.38	16.02± 3.76	171.04± 8.27	189.89± 46.34	65.05± 7.10	4.72± 2.05	26.87± 5.53
		t	12,6; p<0.01	18,4; p<0.001	3,3; p>0.05	12,6; p<0.01	27,7; p<0.01	9,8; p<0.05	7,3; p<0.001	10.0; p<0.05	8,2; p<0.05	10.7; p< 0. 01	2,3; p>0.05
	Control group (n=20)	Initial data	33.85± 5.54	305.60± 28.87	29.10± 4.84	70.10± 5.74	76.95± 4.65	10.35± 2.33	229,25± 9.74	325.20± 8.53	99.85± 7.47	17.25± 2.80	23.65± 8.55
		Day 28 of therapy	21.65± 2.95	198.15±1 8.12	26.60± 4.94	51.85± 4.26	39.95± 3.44	14.40± 3.57	194.40± 6.06	260.30± 11.23	76.75± 6.02	11.4± 2.52	24.55± 8.63
		t	8,7; p<0.05	13,7; p<0.05	1,6; p>0.05	11,1; p<0.05	28,0; p<0.05	4,2; p>0.05	13,3; p>0.05	20.1; p<0.05	10.5; p<0.01	6,8; p>0.05	0.3; p>0.05

The dynamics of the corresponding parameters of systemic immunity of the same groups of patients is given in Table **5**.

As seen from the data in the Table, after the therapy with the drug under study a noticeable change of controlled parameters in peripheral blood was observed. In particular, in patients with DED on the background of meibomian blepharitis a 1.2-1.7 times decrease of IL-1β, IL-2, IL-4, IL-6, IL-8, IL-17A, IL-1Ra, TNF-α levels was observed in blood plasma compared to the initial levels (p<0.05-0.001) and 1.6 times increase of anti-inflammatory cytokine IL-10 level (p<0.01). In the control group, a 1.1-1.3 times decrease of IL-1β, IL-2, IL-6, IL-17A, IL-1Ra, INF-α levels (p<0.05) and 1.2 times increase of anti-inflammatory cytokine IL-10 level (p<0.01) in blood plasma was observed. However, the decrease of IL-4, IL-8 and TNF-α was not statistically significant (p>0.05).

In patients with DED on the background of perimenopause the levels of IL-1β, IL-2, IL-6, IL-8, IL-17A, IL-1Ra, TNF-α, INF-α decreased 1.3-2.6 times (p<0.05-0.001), while IL-4 decrease was statistically insignificant (p>0.05). At that, the level of IL-10 increased 2.1 times (p<0.01). In the control group of patients, the levels of IL-2, IL-4, IL-6, IL-8, IL-1Ra decreased in blood plasma 1.2-1.4 times compared to the initial levels (p<0.05-0.01). The decrease of IL-1β, IL-17A, TNF-α and INF-α levels, as well as the increase of anti-inflammatory cytokine IL-10 level were not statistically significant (p>0.05).

In patients with DED on the background of Sjögren's syndrome the levels of IL-1β, IL-2, IL-6, IL-8, IL-17A, IL-1Ra, TNF-α, INF-α decreased 1.3-3 times (p<0.05-0.001), while IL-10 increased 2.3 times (p<0.05). As in the previous groups, the decrease of IL-4 levels was not statistically significant (p>0.05). In the

Table 5: Some Immunity Parameters in the Blood Plasma of Patients with DED of Various Etiology Following the Therapy Performed

Observational Stages			Evaluated Parameter, pg/ml										
			IL-1β	IL-2	IL-4	IL-6	IL-8	IL-10	IL-17A	IL-1Ra	TNF-α	INF- α	INF- γ
DED Due to Meibomian Blepharitis	Main group (n=7)	Initial data	36.14± 3.27	242.78± 13.27	18.60± 2.30	16.06± 1.44	79.46± 4.24	12.86± 1.93	200.80± 7.16	319.73± 17.20	46.80± 5.33	30.33± 3.63	37.4± 6.76
		Day 28 of therapy	24.64± 3.06	168.21± 42.28	10.53± 1.58	6.06± 1.29	31.93± 2.74	21.13± 1.70	154.73± 4.40	252.87± 8.79	25.40± 2.73	10.53± 2.36	29.93± 5.02
		t	9,3; p<0.01	6,1; p<0.05	10.9; p<0.01	19,6; p<0.001	35,5; p<0.001	12,1; p<0.01	20.6; p<0.001	13,0; p<0.01	13,4; p<0.01	17,2; p<0.05	3,3; p>0.05
	Control group (n=6)	Initial data	35.33± 1.97	237.50± 6.32	19.33± 1.49	16.16± 1.35	79.00± 2.58	15.33± 1.97	205.00± 10.50	334.67± 16.07	47.50± 5.37	26.33±1. 49	31.83±6. 42
		Day 28 of therapy	28.33± 2.11	207.33± 6.30	15.16± 1.43	11.16± 2.03	52.50± 3.90	24.16± 2.67	161.33± 11.05	276.83± 17.47	37.33± 4.85	15.66±2. 36	31.00±3. 02
		t	5,0; p<0.05	7,6; p<0.01	4,5; p>0.05	4,6; p<0.05	12,7; p>0.05	6,0; p<0.05	6,4; p<0.05	5,5; p<0.05	3,1; p>0.05	8,6; p<0.05	0.2; p>0.05
DED Due to Perimenopause	Main group (n=8)	Initial data	30.44± 3.92	244.81± 8.35	23.69± 1.61	20.19± 3.07	82.87± 4.06	13.56± 3.84	212.94± 22.56	326.06± 7.89	41.62± 3.69	18.43± 2.98	31.44± 11.20
		Day 28 of therapy	16.94± 1.64	187.12± 5.90	9.56± 1.58	10.25± 2.47	32.44± 2.74	28.81± 2.32	165.12± 5.95	258.68± 4.64	21.69± 2.66	6.74± 1.34	21.93± 5.14
		t	12,4; p<0.01	21,9; p<0.001	24,4; p>0.05	9,6; p<0.01	40.0; p<0.001	13,3; p<0.01	7,9; p<0.05	28,8; p<0.05	17,0; p<0.001	12.94 p<0.01	5,3; p>0.05
	Control group (n=9)	Initial data	36.11± 1.52	241.55± 7.97	29.11± 4.17	19.44± 2.26	91.11± 9.75	14.66± 1.74	204.11± 6.05	330.11± 7.15	43.77± 3.11	29.66± 5.49	35.44± 9.86
		Day 28 of therapy	29.66± 4.39	206.55± 8.19	17.22± 3.64	11.77± 1.81	54.44± 10.99	21.22± 3.94	182.88± 9.54	283.33± 12.87	30.88± 2.76	20.33± 1.15	22.55± 4.83
		t	3,9; p>0.05	8,7; p<0.01	6,1; p<0.05	7,5; p<0.05	7,1; p<0.05	4,3; p>0.05	5,3; p>0.05	9,0; p<0.05	8,8; p>0.05	4,7; p>0.05	3,2; p>0.05
DED Due to Sjögren's Syndrome	Main group (n=10)	Initial data	36.75± 3.25	309.55± 8.66	29.80± 3.04	26.15± 2.63	75.65± 15.35	9.55± 1.96	224.75* 6*03	330.65± 8.30	48.05± 2.48	21.30± 2.93	42.30± 8.70
		Day 28 of therapy	15.40± 2.33	183.15± 11.4	13.80± 2.36	14.05± 1.56	25.05± 4.05	21.75± 2.55	168.45± 4.74	266.35± 7.80	27.30± 2.59	11.35± 1.56	22.70± 6.74
		t	23,5; p<0.01	38,5; p<0.05	18,2; p>0.05	17,3; p<0.01	13,9; p<0.01	17,9; p<0.05	32,2; p<0.001	24,6; p<0.05	25,3; p< 0. 05	13,1; p< 0. 01	7,8; p>0.05
	Control group (n=10)	Initial data	35.80± 3.54	311.10± 13.26	28.80± 2.85	24.80± 1.72	75.60± 4.10	8.60± 1.57	227.40* 6*51	341.630 ±10.67	49.00± 2.53	19.33± 2.36	40.80± 11.85
		Day 28 of therapy	28.10± 2.25	252.80± 11.49	24.00± 1.95	15.70± 2.00	40.50± 5.54	16.30± 1.67	163.40± 10.99	290.10± 13.11	36.40± 3.77	10.60± 1.354	30.50± 8.17
		t	5,5; p>0.05	10.0; p<0.05	4,2; p>0.05	10.3; p<0.05	15,3; p<0.05	10.1; p<0.05	15,1; p>0.05	9,1; p<0.05	8,3; p>0.05	9,7; p< 0. 05	2,2; p>0.05

control group of patients with DED of etiology under consideration, the levels of IL-2, IL-6, IL-8, IL-1Ra, INF-α in blood plasma decreased 1.1-1.5 times ($p<0.05$), however, the decrease of IL-1β, IL-4, IL-17A, TNF-α levels and increase of IL-10 level were not statistically valid ($p>0.05$).

The level of INF-γ in blood plasma and tear fluid on the background of the therapy performed in the main and control groups was not significantly different from the initial levels ($p>0.05$).

Thus, after the perfomed therapy, all main groups of patients with DED of various etiology demonstrated the change of total immunity in the form of the decrease of pro-inflammatory cytokines' level and increase of anti-inflammatory ones.

The levels of TNF-α, IL-6, IL-2 in tear fluid are more than 2 times higher than their levels in blood plasma, thus confirming the important role of local cytokines' production in the development of chronic inflammation of the ocular surface tissues.

The results of the efficiency studies of 0.01% dexamethasone phosphate solution in treating patients with main pathogenetic types of the dry eye disease show high efficiency of the developed medication. This is confirmed by the decrease of subjective signs of the eye surface epithelium xerosis and the positive dynamics of the clinical and functional parameters. At the same time, the anti-inflammatory effect of the medication is also confirmed by the decrease of the levels of a broad range of pro-inflammatory and the increase of anti-inflammatory cytokines both in the tear fluid and blood plasma of patients with DED of various etiology.

The results obtained by the authors are comparable with the results of other studies dealing with the efficiency research of various glucocorticosteroids in treating corneal-conjunctival xerosis.

Thus, Lee J.H. *et al*. (2014) demonstrated the efficiency of instillations of 1% methylprednisolone solution in patients with DED of various etiology that had previously used the artificial tear medications. Thus, on the background of clinical and functional parameters improvement (increase of tear film bursting time, main tear production parameters, as well as decrease of the degree of eye surface epithelium staining with vital staining solutions and osmolarity of precorneal tear film), the authors observed statistically significant decrease of IL-1β, TNF-α and IL-8 levels. But the decrease of IL-6, IL-17A and IFN-γ levels in the tear fluid was statistically significant [16]. Besides, Marsh P. *et al*. (1999) и De Paiva C.S. *et al*. (2006) also observed significant decrease of pro-inflammatory cytokines IL-1β and TNF-α levels in the tear fluid in patients with DED under study who received instillations of 1% methylprednisolone solution, together with the improvement of clinical and functional parameters [17, 18].

Pflugfelder S.C. *et al*. (2004) showed that on the background of instillations with 0.5% loteprednol etabonate solution there was a decrease of IL-1, IL-6 and TNF-α levels in the tear film of DED patients, as well as improvement of clinical and functional parameters [19]. These data were confirmed by the studies of Sheppard J.D. *et al*. (2011) [20].

Jung H.H. *et al*. (2015) compared the use of 0.5% loteprednol etabonate and 0.1% fluorometholone to treat patients with DED of various etiology and showed a high efficiency of the mentioned glucocorticosteroids in stopping inflammatory process in the eye surface epithelium. 0.5% loteprednol etabonate solution demonstrated more pronounced anti-inflammatory effect [21].

In the studies of Djalilian A.R. *et al*. (2006) the efficiency of 1% dexamethasone phosphate solution against chronic inflammatory process in the eye surface epithelium was shown. The authors demonstrated the decrease of IL-1, IL-6, IL-8, TNF-α and MMP-9 levels in the tear fluid of patients with DED of various etiology on the background of the mentioned medication therapy [22].

Aragona P. *et al*. (2013) proved the efficiency of 0.1% clobetasone butyrate for patients with DED due to Sjögren's syndrome within the framework of a prospective placebo-controlled trial. They observed the improvement of clinical and functional parameters as soon as Day 15 of the therapy [23]. There are also data from Avunduk A.M. *et al*. (2003) who demonstrated high clinical efficiency of local glucocorticosteroids against DED due to Sjögren's syndrome or the increased evapouration of tear fluid [24].

The authors of all studies mentioned above did not observe any side effects of glucocorticosteroids, e.g. the increase of intraocular pressure or steroid cataract development, though they conclude that the use of artificial drugs of this group for more than 8-10 weeks is not safe.

CONCLUSION

The results of the comprehensive clinical and functional study, as well as laboratory research prove the high clinical efficiency of the developed medication that has a marked local anti-inflammatory effect in the therapy of corneal-conjunctival xerosis of various etiology.

All this gives hope for the broad perspectives of the clinical use of the medication developed by the authors containing 0.01% dexamethasone phosphate in combination with 6% polyvinylpyrrolidone and 1.5-5.5% dextrose solution to treat patients with DED of various etiology.

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