MODERN CYTOLOGICAL APPROACHES FOR THE DIFFERENTIAL DIAGNOSIS BETWEEN THE INFLAMMATORY ANTERIOR EYE SEGMENT DISEASES OF PARASITIC AND NON-PARASITIC ETIOLOGY

V.V. Ermilov, M.V. Bukatin, N.A. Kolobrodova, O.Yu. Kuznetsova, O.D. Chulkov

Federal State Budgetary Educational Institution of Higher Education

«Volgograd State Medical University» of the Ministry of Healthcare of the Russian Federation

Russia, Volgograd, pl. Pavshikh Bortsov, 1, 400131

Introduction

Differential diagnosis of inflammatory anterior eye segment diseases of various etiology is nowadays an actual problem in practical health care.

Besides allergic, bacterial and viral pathologies, parasites have also been assigned to the prevailing etiological factors of this nosologic group, including parasitizing of *Demodex folliculorum* and *Dirofilaria repens* in the eye tissues, among others [1, 6].

We have not encountered any descriptions of the clinical manifestation or quantitative cytological methods for the differential diagnosis between conjunctivitis caused by *D. repens*, mixed infectious (*D. repens* and *D. folliculorum*), and conjunctivitis of non-parasitic etiology, neither in Russian nor in international references.

The summary of the general data on demodecosis has revealed that this pathology is a widespread parasitic disease in human and animals and is characterized by a significant value in the structure of diseases of the anterior eye segment. In particular, it has been found that blepharoconjunctivites are of demodecosis etiology in 80-90% of cases [3, 7].

At the same time, conjunctivitis caused by demodecosis may be associated with the inflammatory diseases of conjunctive of other etiology, resulting in certain difficulties in differential diagnosis [2, 3, 4, 5, 6].

Material and methods

Aiming at the differential diagnosis of conjunctivitis caused by *D. repens*, *D. folliculorum*, mixed infectious (*D. repens* and *D. folliculorum*) and conjunctivitis of allergic etiology, we have carried out quantitative cytological analysis of cellular reaction and assessment of dystrophic epitheliocytes, mucilaginous filaments, and goblet cells in smears from conjunctiva of the patients with these forms of conjunctivitis.

The study included examination of the smears from conjunctiva of 16 patients with conjunctivitis caused by *D. repens*, 16 patients with conjunctivitis caused by *D. folliculorum*, 8 patients with mixed infectious conjunctivitis, and 16 patients with allergic conjunctivitis.

The total quantity of ticks (acarogramma) was estimated for the patients with conjunctivitis

caused by *D. folliculorum* and with the mixed infectious conjunctivitis, which varied from 4 to 8 on 6 eyelashes. Such acarogramma was assessed as pathological (while one tick per two eyelashes is classified as acceptable) [6].

The analysis of the data was performed using the statistical software Statistica 6.0. The differences with $p<0,05$ were rated as statistically significant.

Results and discussion

The most illustrative cellular reaction of conjunctiva could be detected by observation of various cell populations and their quantitative and qualitative changes.

The morphometric analysis of the counts of neutrophiles, eosinophiles, basophiles, lymphocytes, plasmocytes, and histiocytes in smears from conjunctivae of the patients with the investigated conjunctivites allowed to reveal the following cytological profiles (table 1):

Table 1

Quantitative analysis of the cellular reaction of conjunctivae in patients with conjunctivitis of various etiologies (p<0,05)

Conjunctiva	Neutrophiles	Eosinophiles	Basophiles	Lymphocytes	Plasmocytes	Histiocytes
Patients with dirofilariasis	1,7±0,2	0,10±0,06	0,3±0,1	1,8±0,6	2,0±0,4	7,2±0,14
Patients with demodecosis	6,0±0,5	1,2±0,08	2,6±0,72	4,2±1,2	7,6±0,12	3,6±0,22
Patients with D.repens and D.folliculorum	9,8±2,83	2,3±0,6	4,1±1,2	5,8±0,24	7,2±1,03	7,9±1,6
Patients with allergic conjunctivitis	0,7±0,4	4,4±1,3	7,0±0,5	2,4±0,5	1,9±0,1	1,5±0,7

Our morphometric analysis revealed the quantitative dissimilarity of the studied cell populations (table 2):

Table 2

Quantitative analysis of dystrophic epitheliocytes, mucilaginous filaments, and goblet cells in smears from conjunctivae of the patients with conjunctivitis of various etiologies (p<0,05)

Conjunctiva	Dystrophic epitheliocytes (karyopiknosis, karyorrhexis, karyolysis, fragmentation of nucleus, vacuolization, keratinization)	Mucilaginous filaments	Goblet cells
Patients with dirofilariasis	4,2 ± 0,4	1,55 ± 0,8	0,15 ± 0,05

(*D. repens*)			
Patients with demodecosis (*D. folliculorum*)	2,4 ± 1,2	5,2 ± 1,3	1,5 ± 0,3
Patients with *D. repens* and *D.folliculorum*	2,9 ± 0,03	6,2 ± 0,12	1,8 ± 0,13
Patients with allergic conjunctivitis	1,50 ± 0,7	9,7 ± 0,7	4,2 ± 1,3

By means of the quantitative analysis carried out for neutrophiles, eosinophiles, basophiles, lymphocytes, plasmocytes, histiocytes, dystrophic epitheliocytes, mucilaginous filaments, and goblet cells in smears from conjunctivae of the patients with conjunctivitis caused by *D. repens*, *D. folliculorum*, mixted infectious (*D. repens* and *D. folliculorum*) and conjunctivitis of allergic etiology, both quantitative and qualitative cytological characteristics of the inflammatory cellular reaction were revealed, and clinical and cytological parallels were drawn between the cases of inflammatory pathologies of human conjunctiva.

Conclusions

The algorithm developed by us for cytological investigation of smears from conjunctiva of the patients with conjunctivitis caused by *D. repens* revealed the following common trends. In case of conjunctivitis caused by *D. repens*, considerable prevalence of dystrophic epitheliocytes (karyopiknosis, karyorrhexis, karyolysis, fragmentation of nucleus) may be observed in serous exudates, with the distinct signs of keratinization and decreased ratio of nucleus to cytoplasm, as compared to conjunctivitis of other etiologies. Conjunctivitis caused by *D. repens* is characterized by the lowest density of goblet cells and the lowest proportion of eosinophiles and basophiles in cell populations of conjunctival cavity. This allows distinguishing the development of inflammation in conjunctival dirofilariasis from the hypersensibilization that manifests as allergic reactions. It may be explained by the absence of microfilaria in blood of the patients with cutaneous dirofilariasis.

Based on the presented data, it seems possible to use such evidences as increased proportion of dystrophic epitheliocytes with distinct signs of keratinization, as well as the decreased ratio of nucleus to cytoplasm, the lowest density of goblet cells, and the lowest proportion of eosinophiles and basophiles in cell populations of conjunctival cavity, as the cytological diagnostic criteria for conjunctival dirofilariasis.

Abstract

We developed an algorithm for the quantitative cytological methods for differential diagnosis between conjunctivitis caused by *D. repens*, mixed infections (*D. repens* and *D. folliculorum*), and conjunctivitis of non-parasitic etiology. The possibilities for cytological

diagnosis of ocular dirofilariasis were discovered, and the criteria for conjunctivitis caused by Dirofilaria were elaborated.

Key words

Ocular dirofilariasis, conjunctivitis caused by Dirofilaria, cytological diagnosis.

References

1. Ermilov V.V, Vorobiev A.A, Chernikov M.V, Smirnov A.V., Dolzhikov A.A, Safronova E.Ju. Dirofilariasis in the Volgograd region. – Volgograd, Izd. VolGMU, 2010. – 108s.
2. Forton E., Seys B., Marshal J.L., Song A.M. Demodex folliculorum and topical treatment: acaricidal action evaluated by standardized skin surface biopsy // Br. J. Dermatol.-1998.-Vol 138.-№3.-P.461-6.DOI:10.1046/j.1365-2133.1998.02125.x
3. Rozko T.E. Clinical features, diagnostic and treatment of blepharoconjunctivitis of a demodecosis etiology – Avtoref. Dis. kand.med.nauk. Krasnoyarsk. – 2003. – p. 24
4. Zav'yalova N.A., Vasil'eva A.M., Kochetkova L.Yu. About diseases of eyelids and eye anterior chamber in connection with a tick-borne invasion (demodecosis) – Oftal'mologicheskii zhurnal. – 1998. – № 5. – p. 275-278.
5. Zatsepina N.D., Maichuk Yu.F., Semenova G.Ya. Damage of eyes at a demodecosis: Methodical Recommendations– M. 1983. – p.17
6. Clifford C.W, Fulk G.W. Association of Diabetes, Lash Loss, and Staphylococcus aureus with Infestation of Eyelids by Demodex folliculorum (Acari: Demodicidae). Journal of Medical Entomology [Internet]. Oxford University Press (OUP); 1990 Jul 1;27(4):467–70. Available from:http://dx.doi.org/ 10.1093/jmedent/27.4.467
7. Gumerova E.I. Laboratory and diagnostic criteries of a demodecosis of eyes//Ophthalmology Questions. Anniversary educational and practical conference: Materials. Omsk, 2002. – Rel. 4. – p. 44-45.

The work is submitted to the International Scientific Conference « *Basic and applied research in medicine*», France (Paris), October 19-26, 2016, came to the editorial office on 14.10.2016