ORIGINAL ARTICLE

Contrast Sensitivity and Optical Coherence Tomography Examinations in Adolescent Patients with Diabetes Type I Preretinopathy (A Pilot Study)

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SUMMARY

Aim: To evaluate the development of retinal changes in adolescent patients with diabetes type I (T1DM) with disease's duration more than 10 years, which started before 5 years of age.
Methods: The development of the findings on the posterior pole was followed up. The retinal functions were established by means of contrast sensitivity in four space frequencies: 3 cycles/degree (c/deg) (perimacular area), 6 c/deg and 12 c/deg (macular area), and, finally, 18 c/deg (foveola). The central retinal thickness, average retinal thickness of the specified quadrant of macular area, the foveolar depth of its own, and the volume of the perimacular area (perimacular cube volume) were measured by means of optical coherent tomography (OCT).
Material: Altogether 20 patients with diabetes type I meeting the set criteria were examined, and their findings were compared with control group of healthy adolescent people. The values from the control group were used as our normative database.
Results: On the retina, there were found, during the disease's course lasting in average 13.3 years, changes of the macular area, especially tortuosity of macular final capillaries and pigmentation with disappearing of foveolar reflex, which, in 20 %, were followed by sporadic hard exsudates of the retina. Difference of the decreased values in adolescent patients, comparing to the control group, was recorded in contrast sensitivity in space frequencies of 3 c/deg (p 0.047) and 12 c/deg (p 0,0497), but statistically significant was the difference in space frequencies of 6 c/deg (p 0.0001) and 18 c/deg (p 0.0001). Using the OCT, no statistically significant difference was found in the central retinal thickness, but the values of foveolar depth in patients with diabetes type I were variable (p 0.0153); in four eyes it was much deeper, and in other four of them it was much shallower. Furthermore, there was higher the average thickness of the retina (p 0.0008) and the volume of the perimacular area (perimacular cube) (p 0,0001).
Conclusion: The findings in eight eyes out of five patients with T1DM were evaluated as diabetic preretinopathy – pre-stage of beginning stage of diabetic retinopathy in central area of the retina from the functional and structural point of view of current pathological changes of contrast sensitivity and OCT. The findings of other three patients were rated as diabetic preretinopathy according to sporadic hard exsudates of the retina and OCT changes, but. until now, without contrast sensitivity changes. The one-year profile of glycated hemoglobin (HbA1c) was higher in patients with diabetic preretinopathy than without the eye involvement, but it was not statistically significant (p 0,0314).

Key words: Contrast sensitivity (CS), Spectral Domain Optic Coherence Tomography (SD-OCT), diabetes mellitus type I (T1DM), diabetic preretinopathy (DpR), glycated hemoglobin (HbA1c)

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INTRODUCTION

The Liverpool declaration of EASDec (European Association for the Study of Diabetic Eye Complication) in 2005 set as its target a reduction of the risk of deterioration of vision as a consequence of diabetic retinopathy within 6 years, through the implementation of effective, systematic programmes of eye screening in patients with diabetes mellitus (31). Throughout Europe a total of 29 311 new patients with DM up to the age of 15 years were registered in the period of 1989-2003 in 17 countries, including the Czech Republic. The prevalence at this age presupposes an increase from 94 000 in 2005 to 160 000 in 2020 (9). The twenty year register of children with DM 1 in the Czech Republic over a period of twenty years (1989-2009) demonstrated a halting of the acceleration of the incidence of this metabolic disorder. Following a 15% increase in the period 1996-2001 there followed a stagnation in the number of newly diagnosed diabetic children in the period 2002-2009. A total of 5

155 new paediatric patients with DM were diagnosed over the course of the twenty year observation out of a child population of 1.76 million (3). In 2009 a total of 783 321 diabetic patients of both types of the disorder were registered in the Czech Republic, which represents approx. 8% of the population (26). From this there ensues a society-wide significance of the necessity of introducing screening programmes for prevention, as well as the availability of modern technologies in the treatment of diabetic retinopathy also within our region. The aim of our pilot study was to assess the functional and structural changes of the retina and to compare them with the finding on the ocular fundus, when visual acuity was within the norm also following applicable additional correction of the refractive error, at a time when no incipient changes of diabetic retinopathy were detected in the central region of the retina in the group of adolescent patients with DM 1.

METHOD OF FUNCTIONAL--STRUCTURAL EXAMINATION

Contrast sensitivity is a functional examination which is better at detecting the condition of the visual analyser than examination of visual acuity. It provides information about the quality of processing of the given optical stimulus within different spatial frequencies sent from the photoreceptor cells of the retina to the visual centre in the cerebral cortex. A total of 4 spatial frequencies were used: 3 cycles on one angular degree (c/deg) evaluating the perimacular region, also the macular region using 6 c/deg, 12 c/deg and finally a frequency of 18 c/deg, which corresponds to the foveola.

Optical coherence tomography is an analogy of a B-scan ultrasound examination, with the difference that instead of acoustic reflectivity, optical reflectivity is used. The principle is low-coherence interferometry, which measures the distance of various structures inside the tissue with high sensitivity to the light signal reflected from the structures of the eye. SD--OCT (spectral-domain optical coherence tomography) represents a method of three-dimensional evaluation of the structures of the retina and the choroid. The parameters measured by SD-OCT included central retinal thickness, average retinal thickness within a specified quadrant of the macular region with an edge of 6 mm, its difference as depth of the actual foveola and the cubic volume of the perimacular cube within a specified quadrant of 36 mm2.

Our own cohort

Within the framework of outpatient observation of patients at the 1st internal clinic and the Paediatric and Youth Clinic at the Královské Vinohrady University Hospital, as of 1 January 2013 we included a total of 172 patients with DM 1, with the duration of the disorder persisting for more than 10 years. Of this number there were 23 adolescents at the Paediatric and Youth Clinic aged up to 18 years who we had observed since the beginning of their metabolic disorder with regard to endocrinology and ophthalmology. Two patients were excluded from this number due to a primary ocular disease: unilateral microphthalmos and bilateral congenital nuclear cataract, together with a patient with a monogenic form of diabetes. Within the framework of the complex diagnostic approach, we first of all determined visual acuity with applicable optimal correction for an examination of contrast sensitivity using CSV 1000 (Vector Vision). In cycloplegia we evaluated the anterior segment of the eye on a slit lamp from the perspective of changes in the lenses. We conducted a stereoscopic examination of the ocular fundus by means of indirect ophthalmoscopy, using 90 and 66 D lenses, and supplemented this with an assessment of the macular region by means of a classic ophthalmoscope. For photo documentation of the central region we used an FF450 plus IR (Carl Zeiss) digital camera with sufficient resolution, e.g. of hard scattered deposits. Refraction was determined and spectral optical coherence tomography was conducted by means of Cirrus OCT (Carl Zeiss).

Our own examination of the group of 20 patients, which comprised 17 girls and 3 boys with DM 1, took place within the framework of regular checks over the period from November 2012 to July 2013, always in both eyes. The age of the patients at this complex examination ranged from 12.8 years to 18.4 years (median 16.4 years). DM 1 was diagnosed between the ages of 1.4 years to 4.7 years (median 3.1 years), thus before reaching the age of 5 years. The period of duration of DM 1 at the time of the ocular examination ranged from 10.1 years to 14.8 years (median 13.3 years). Theprecise time data about the individual patients, i.e. detection of the incidence of DM 1 and its duration, is presented in table 1. Of the metabolic parameters we observed the current value of HbA1c at the time of the complex ocular examination and its profile during the course of the last years of treatment of DM 1 before the ocular examination (table 1).

The control group comprised adolescents of the same sex representation and similar age composition from 13.2 to 18.1 years (median 16.8 years), who we examined as outpatients for non-ophthalmological diagnosis and whose general condition did not demonstrate any affliction in terms of metabolism, endocrinology or neurology (table 2). This concerned conditions of neurologically unexplained cephalea in 50% of cases and collapse or fainting in 40%. There was a secondary finding of asthenopic complaints (considered to constitute allergy due to lacrimation) and in some cases of headaches there was as yet uncorrected myopia in 20%. We examined visual acuity, the anterior segment of the eye and CS. In cycloplegia the examination was analogously supplemented with SD-OCT, determination of refraction and examination of the fundus with photo documentation. A condition for inclusion in the control group was visual acuity of 1.2 naturally or 1.0 with correction and physiological intraocular finding in the anterior and posterior segment of the eye.

RESULTS

From the beginning up to five years of observation of the ocular fundus, we observed isolated increased dilation and tortuosity of the end capillaries. Within the range from the 6th to the 10th year (within average thirteen year observation), capillary changes practically reached absolute values in 95% in almost half of the patients. Further ophthalmoscopic symptomatology included a change of the sketching of the actual foveolar and macular region after five years of duration of DM 1 in the form of increased irregular pigmentations. We detected a subsequent change of the sketching o the macula in the form of irregular foveolar reflex up to its disappearance, with relative thickening of the retina without manifest macular edema in 10% of patients. Another finding was hard scattered deposits of exudates in 20%, specified in detail in table 1.

Table 1 Relationship of CS and SD-OCT to incidence and duration of DM 1 (ocular finding: T – tortuosity of capillaries, M – Macula: increased pigmentation of macula, F – Foveola: loss of reflex, HE (Hard Exudates) – hard scattered deposits, values of HbA1c (mmol/mol according to IFCC).

Order	CS (c/deg) Number of targets			SD-OCT (µm)				Ophthalmological changes			Solution of refraction (vision)	Note incidence of DM1 duration	HBA1c -current -annual
	3	6	12	18	Central thickness	Average thickness	Depth of foveola	Volume (mm3)	Lenses	fundus	refraction			
1	6	7	7	7	238	298	60	10.7	Clear	M	0-75=0.25	Sine	From 3.8 years	66
F	6	7	7	6	237	297	60	10.7			0.25		i.e. 11.2 years	60.7
2	6	6	7	6	235	305	70	11	1st degree	M, T	1=1.25	With corr. 1.2	From 1.4 years	64
M	6	6	7	7	224	293	69	10.6			0.25	Sine	i.e. 11.4 years	64.3
3	7	7	7	7	240	287	47	10.1	1st degree	M, T	-0.75	Sine	From 2.7 years	85
F	7	7	7	7	238	283	45	10.2			-0.25		i.e. 13.9 years	76.5
4	6	6	5	6	234	272	38	9.8	Y-suture	M, T	.-4=0.75	With corr. bil.	From 3.3 years	60
F	6	5	7	6	236	276	40	10			-4	1.2	i.e. 13.8 years	73
5	6	7	7	7	242	283	39	10.2	Y-suture	M, T	-0.75	With corr. bil.	From 3.2 years	55
M	6	6	7	7	244	279	35	10			-0.75	1.2	i.e. 14 years	55.5
6	6	7	7	6	277	298	22	10.7	1st degree	M, T Isolated HE bilat.	.-1/170	With corr. bil.	From 2.4 years	67
F	6	7	7	6	282	306	24	11			.-0.5/175	1.2	i.e. 11.2 years	66.4
7	6	6	7	7	262	298	36	10.7	1st degree	T, F+M	0.5	Sine	From 3.5 years	88
M	6	6	7	7	256	296	40	10.7			0.5		i.e. 14.8 years	82
8	6	7	6	7	253	295	42	10.6	Y-suture	M, T	-0.75	Sine	From 4 years	83
F	6	6	7	6	257	294	37	10.6	1st degree		-0.75		i.e. 10.7 years	95.4
9	6	7	7	6	247	282	35	10.2	1st degree	M, T	-0.75	Sine	From 4.1 years	88
F	6	7	7	7	247	284	37	10.2			-0.5		i.e. 14.3 years	79.2
10	6	7	7	7	249	289	40	10.4	(1st degree)	M, T, drusis dx.	-0.75	Sine	From 2.5 years	82
F	6	7	6	7	250	288	38	10.4			-0.25		i.e. 12.1 years	74.2
11	6	7	7	7	264	280	24	9.7	Clear	M, T	0.5=0.5	Sine	From 4.7 years	92
F	6	7	7	7	263	281	24	9.7			0.75=0.25		i.e. 10.9 years	88.8
12	6	6	7	7	255	289	34	10.4	Y-suture	T, F+M	0	Sine	From 2.5 years	125
F	6	7	6	6	258	295	37	10.6			0.25		i.e. 13.4 years	114.7
13	6	7	7	7	265	309	44	11.1	(2nd degree)	M, T, HE bil.	-0.25	Sine	From 3.2 years	86
F	6	7	8	7	261	303	42	10.9			0.25		i.e. 10.4 years	76.3
14	5	7	6	5	265	297	33	10.7	(2nd degree)	M, T	0.5	With corr. bil.	From 1.9 years	67
F	6	6	7	6	261	299	38	10.8			0.25=0.5	1.0	i.e. 13 years	66.5
15	6	7	6	6	273	299	26	10.8	1st degree	M, T	.-3.0=0.5	With corr. bil.	From 4.4 years	69
M	7	7	7	7	273	295	22	10.6			-2	1.2	i.e. 10.1 years	70.4
16	6	7	7	6	233	280	47	10.1	1st degree	M, T	0.5	Sine	From 1.8 years	66
F	6	7	7	7	239	280	41	10.1			0.25		i.e. 13.5 years	72.8
17	6	6	7	6	268	305	37	11	1st degree	M, T HE bil.	-1	With corr. bil.	From 2.9 years	100
F	6	7	7	7	270	307	37	11.1			-1	1.2	i.e. 14.2 years	98.4
18	7	7	7	7	226	269	43	9.7	1st degree	M, T	.-0.5=-0.5	With corr. bil.	From 2.8 years	86
F	6	7	7	7	228	267	39	9.6			-1	1.2	i.e. 13.7 years	75
19	6	7	7	7	230	263	33	9.5	1st degree	M, (T)	-0.5	Sine	From 3 years	78
F	6	7	7	7	229	263	34	9.5			-0.5		i.e. 13.7 years	73.8
20	6	6	7	7	233	295	32	10.6	1st degree	M, T HE bil.	0.25	Sine		66
F	6	6	7	6	233	295	32	10.6			0			68.8

Table 2 Norm of CS and SD-OCT values.

Order	Age	CS (c/deg) Number of targets				SD-OCT (µm)				Refraction	Ophthalmological changes (correction)	Note incidence of DM1 duration
		3	6	12	18	Central thickness	Average thickness	Depth of foveola	Volume (mm3)			
1	17.8	6	7	8	7	260	271	11	9.8	0.5	Sine	Cephalea
F		7	7	7	8	262	272	10	9.8	0.5=0.75		
2	16.7	7	8	7	8	226	269	43	9.7	-0.25	Sine	Orthostatic fainting
F		6	7	7	7	226	269	43	9.7	-0.25		
3	17	7	7	8	7	249	284	35	10.1	0.75/90	Conjunctivas - no pathology	v.s. ocular allergy
F		6	7	7	8	251	289	38	10.3	0.5=0.25		
4	16.5	6	7	7	7	238	283	45	10.1	.-0.25=0.5	Sine	"School" cephalea
F		6	7	7	7	236	291	57	10.4	.-0.25=0.75		
5	16.7	7	7	7	7	276	298	22	10.6	0.25	Sine	Cephalea
F		6	7	7	7	273	291	18	10.4	-0.5		
6	15.2	6	7	6	7	249	266	27	9.5	1.25=0.5	Sine	Orthostatic fainting
M		6	7	7	7	249	263	24	9.4	1.0=0.25		
7	12.2	6	7	7	6	257	268	11	9.6	-3.5	Sine (rp. glasses)	Cephalea myopia
F		6	7	6	7	263	274	11	9.7	-3.5		
8	17.8	6	7	7	7	240	283	43	10.1	.-1.0=0.75	Sine (rp. glasses)	Cephalea myopia
F		6	7	7	8	244	280	36	10	.-1.0=1.00		
9	15.5	6	7	6	8	263	300	37	10.7	0.25/80	Sine	"School" cephalea
M		7	7	8	8	252	298	46	10.6	0.50/115		
10	13.4	6	7	7	7	267	277	10	9.9	-2	Sine (rp. glasses)	v.s. ocular allergy
F		6	7	7	7	264	275	11	9.9	.-1.75=0.5		
11	18	6	7	7	7	237	267	30	9.5	0.5	Sine	Orthostatic fainting
F		6	7	7	7	240	274	34	9.8	0.25=0.5		
12	17.8	6	7	7	6	273	262	9	9.3	-2.5	Sine	Collapse state
F		6	7	7	6	273	264	11	9.4	-0.25		
13	16.7	6	7	7	6	253	279	26	9.9	.-2.5=0.25	Sine (rp. glasses)	Collapse state
F		6	7	7	7	258	284	26	10.1	.-2.0=0.25		
14	15.4	6	7	7	8	234	267	33	9.5	.-1.75=-0.25	Sine (rp. glasses)	Orthostatic fainting
F		6	7	6	7	229	264	35	9.4	.-0.75=-0.75		
15	14.5	7	7	7	7	215	261	49	9.3	-2.25	Sine (rp. glasses)	Orthostatic fainting
F		7	7	7	7	212	263	51	9.4	.-2.0=0.25		
16	17.8	7	8	8	8	255	293	38	10.4	.0.25=-0.25	Sine	Cephalea
F		7	8	7	8	255	290	35	10.4	Emmetropia		
17	18.1	6	7	8	7	232	279	47	10	-1.25	Sine (rp. glasses)	Cephalea myopia
F		7	7	7	7	237	279	42	10	.-0.75=-0.5		
18	16.7	7	7	8	8	267	300	33	10.7	0.25=0.5	Sine	Cephalea
F		6	8	8	8	266	300	34	10.7	0.75=0.25		
19	15.6	7	7	7	7	261	292	31	10.4	.-0.25=-0.75	Sine	Collapse state
F		6	7	7	7	265	292	27	10.4	.-0.5=-0.25		
20	13.2	7	7	7	7	238	293	55	10.5	1.25	Sine	Cephalea
M		7	7	7	7	237	288	51	10.3	1.75=0.5		

We did not record an occurrence of ophthalmoscopic evident haemorrhages or microaneurysms in any of the patients. Table 1 also presents the findings in the lenses (presence of accentuated posterior "Y" suture, posterior subcapsular dissociation evaluated as 1st degree of changes, in the case of simultaneously present anterior and posterior dissociation as 2nd degree of changes), and the relationship of refraction to vision of the patients. A fundamental observation was the records of the results of the individual patients within the area of CS and SD-OCT examination (table 1). Table 2 presents the results of the CS and SD-OCT measurements on the control group. Finally table 2 presents the summary average values of the individual CS and SD-OCT measurements of both group with standard deviations and a mutual relationship of the results of both groups with the level of significance, which was calculated using a two-sample T-test.
In the CS examination we determined a borderline significant difference between the control group and the group of patients with DM 1 for 3 c/deg (p 0.047) and 12 c/deg (p 0.0497). In the other two measured quantities, the values differed by a decline of detected sensitivity on a fundamental level of significance (p 0.0001), in both 6 c/deg and 18 c/deg.
In the SD-OCT examination we did not determine a significant difference between the groups in terms of central retinal thickness (p 0.9614), the other values differed with varying statistical significance, least in depth of foveola (p 0.0153), more fundamentally in average retinal thickness (p 0.0008). We demonstrated a pronounced statistical difference in the volume of the central part of the retina on a surface of 36 mm2 (p 0.0001).
On the basis of these results, we determined pathological values for both examinations. For CS this was a decline in the ability to detect by one less target than the average value in the control group, at least in three frequencies in one eye with simultaneous decline of two frequencies also in the other eye (essentially this always concerned frequency of 3 c/deg). We recorded this finding in 6 patients. For SD-OCT an increase in the content of the examined volume above the average value of the control group, with allowance for the standard deviation always counted as a pathological value. This finding was detected in 21 eyes. At the same time they were accompanied by increased values of average retinal thickness in 20 eyes. These values were manifested in a relative deepening of the foveola (fig. 2) in 4 eyes (patients no. 1, 2), or conversely its softening (fig. 3) also in 4 eyes (patients no. 6, 15). Altogether we recorded the above-stated pathological changes on CS and SD-OCT in 8 eyes (20%) of

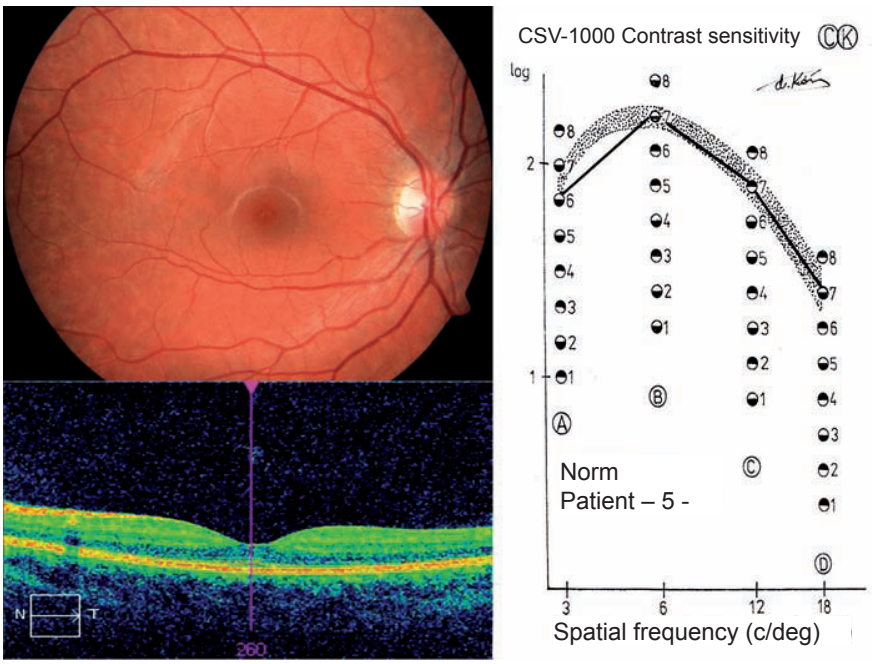

Fig. 1 Patient no. 5. Photo of fundus in right eye, CS: reduction 3 c/deg, SD-OCT: depth of foveola and volume of perimacular cube within norm.

Table 3 Statistical values of CS, SD-OCT and also HbA1c (mmol/mol according to IFCC).

	Patients with DM 1		**Control group**		**T-test**	**Result**
	Average value	Standard deviation	Average value	Standard deviation	Two-sample	Value
CS (number of targets)						
3 c/deg	6.1	0.3	6.4	0.4	p 0.047	**(differs)**
6 c/deg	6.7	0.5	7.1	0.3	p 0.0001	**differs**
12 c/deg	6.9	0.5	7.1	0.5	p 0.0497	**(differs)**
18 c/deg	6.6	0.5	7.2	0.6	p 0.0001	**differs**
SD-OCT (μm)						
Central thickness	249.4	15.7	249.5	16.2	p 0.9614	**does not differ**
Average thickness	289.4	12.1	279.8	12.2	p 0.0008	**differs**
Depth of foveola	38.8	10.8	31.9	13.7	p 0.0153	**differs**
Volume of retina (mm3)	10.4	0.5	9.9	0.4	p 0.0001	**differs**
HbA1c	**Patients with DpR**		**Patients without DpR**			
	Average value	Standard deviation	Average value	Standard deviation		
Current value	82.25	20.1	76.3	11.9	p 0.4908	**does not differ**
Year-round value	81.35	17.7	73.5	8.4	p 0.3014	**does not differ**

five patients, as functional and structural changes conditioning the diagnosis of preretinopathy (DpR) DM 1, in contrast with physiological findings (fig. 1) upon examination by CS and SD-OCT in 12 eyes (30%). In half of the examined eyes we demonstrated certain non-specific changes, but also more serious findings, primarily the incidence of hard scattered exudates (patients 6, 13, 17, 20). Pathological changes were also found on SD-OCT in the first three patients of this sub-group, whereas at the time of examination CS was within the norm. It was also possible to classify this finding into the group of DpR without functional changes. We classified the last stated patient (no. 20) amongst preretinopathy on the basis of pathological values of CS.

At the time of current examination, the value of HbA1c in the patients we observed with DM 1 was within the range from 55 to 125 mmol/mol (7.2% to 13.6% according to DCCT), and the average one year profile curve showed values between 55.5 and 114.7 mmol/mol (7.2% to 12.6% according to DCCT). The values of the individual patients are presented in table 1. We conducted a comparison of the HbA1c values in the patients with signs of functional-structural DpR and an accompanying finding of hard scattered deposits, i.e. in eight of the above patients together with twelve observed without symptoms of DpR. Both values (current examination and year-round profile) were higher in the patients with DpR in comparison with the group of patients without these symptoms. Nevertheless, the average values and standard deviations were without any statistically significant difference (table 3).

DISCUSSION

Upon detection of incipient changes, the basis used is the pathophysiology of diabetic changes. The first demonstrable sign of diabetic retinopathy is a breakdown of the hemato-ocular barrier, which can be assessed e.g. by fluorophotometry. This theory refers to an oxidation stress accompanying hyperglycaemia, in which the resulting pseudohypoxia may breach the cellular membrane of the vascular endothelium, or refers to damage to the pericytes due to a paradoxical glucose deficiency, which leads to their apoptosis (30).

Our group of patients was designated if possible by the lowest age of the onset of the metabolic disorder, maximally up to the age when spatial vision is still

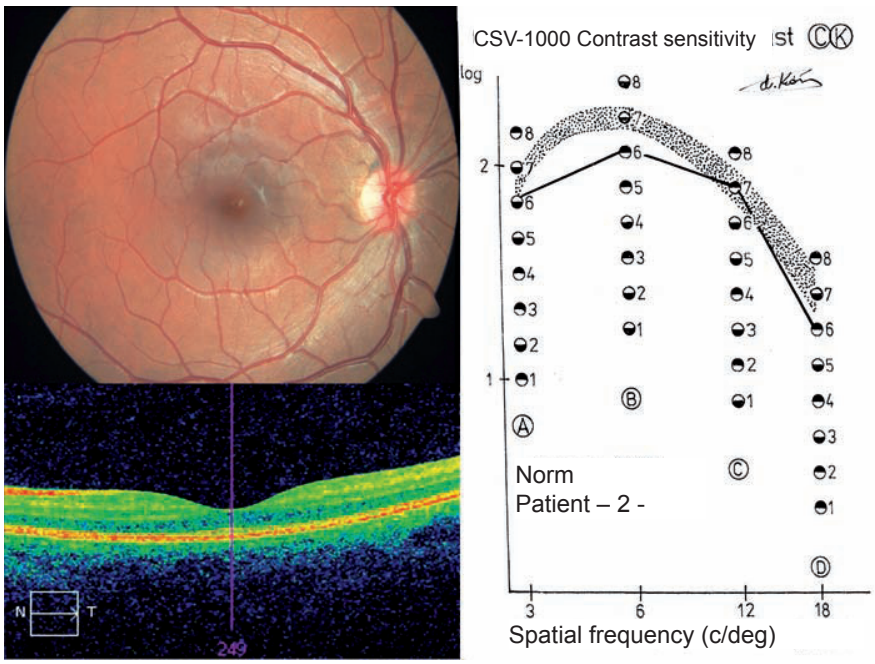

Fig. 2 Patient no. 2. Photo of fundus in right eye, CS: reduction 3, 6, 18 c/deg, SD--OCT: deepening of foveola and volume of perimacular cube increased.

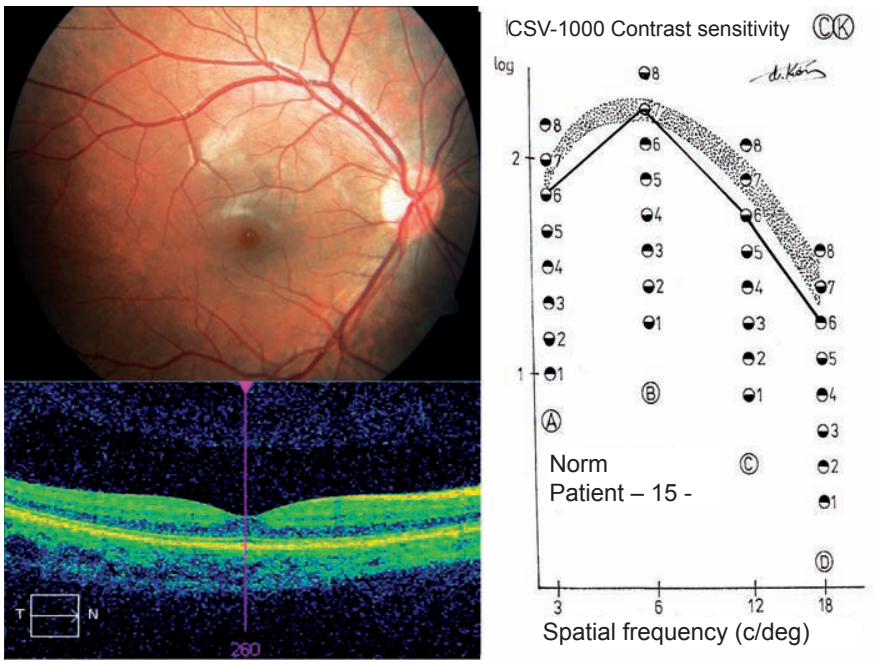

Fig. 3 Patient no. 15. Photo of fundus in right eye, CS: reduction 3, 12, 18 c/deg, SD-OCT: softening of foveola and volume of perimacular cube increased.

becoming fixed and the development of sight is still malleable. The period of duration was set at above ten years, in which incipient changes of the sketch of the retina in the central region had already been regularly detected (18, 20). The first functional abnormalities included defective function of the amacrine and bipolar cells of the photoreceptors, which precedes the onset of diabetic retinopathy (31). A basis forms for a pathological response upon electro-physiological examinations, as well as a reduction of CS. In diabetics without diabetic retinopathy, a decrease in the amplitude of visually evoked potentials was recorded, together with a reduction of CS (23) or abnormalities before the onset of retinopathy with normal visual acuity (24). CS has been used for screening changes for a quarter of a century (10, 29). A reduction of CS has been recorded in the case of DM 1 without manifestations of diabetic retinopathy only in certain spa-

tial frequencies (10, 14, 18, 20, 29, 33, 35, 36), whereas in the case of diabetic retinopathy changes of CS have been markedly abnormal. A significant dependency of the decrease of CS on an increase of HbA1c has been found (18, 20, 29, 36), though this fact has not been confirmed by other authors (10, 35). We took observation of the decrease in the ability to detect individual targets for previous studies of IGA as the basis of our evaluation of CS (22). The value of the decrease in sensitivity by at least two targets attested to DpR, whereas a decrease of four targets or more was more typical of DR (20, 22). We have already demonstrated previously that incipient changes in the transparency of lenses in the anterior and posterior subcapsular layer, detectable using a Schemipflug camera (Pentacam) have no fundamental influence on CS (21).

OCT has been used regularly for over ten years in order to assess the thickness of the macular region in the case of cystoids macular edema for evaluation of the frequency of changes (17) and also for correlation of therapy (13). Assessment of the actual thickness of the foveola has demonstrated a significant difference between the healthy population and patients with proliferative and non-proliferative form of retinopathy using a Zeiss-Humphrey instrument (28). Further research in recent years has not demonstrated differences of central thickness in the region of the foveola between the healthy population and patients with diabetes without signs of diabetic retinopathy (7, 11, 14), but a difference between a control group without DM 1 and non-proliferative form of retinopathy has been repeatedly confirmed (17, 24).

In our own observation we did not find a significant difference in the central thickness of the retina between the control group and the group of patients with DM1. We included a further parameter in the evaluation, namely the actual depth of the foveola, which ensues from the difference between central thickness and average thickness of the retina in the observed perimacular cube, and this helped specify their mutual relationship. This value appeared in an evaluation of the foveolar region in achromatopsia, where deepening of the foveola occurred on a basis of degeneration of the photoreceptors with a progressive character (34), which could also be manifested within the framework of further neuro-degenerations. Modern SD-OCT instruments working on the principle of three-dimensional patterns have enabled better detection of the construction of the actual retina and its individual layers, which has led to the consideration that this represents neuro-degenerative changes in the early stages of diabetes (1, 6, 37, 38). A thinning of the internal retinal layers has been recorded (37, 38), as well as a thickening of the plexiform and nuclear layer, whilst at the same time specific changes have been described in the layer of the ganglion cells (38). In our study we did not conduct an evaluation of the individual layers of the retina, because we did not have special software available. There are differences in the results in the evaluation of the thickness of the retinal layers between contemporary modern instruments, which upon an incorrect correlation may lead to an erroneous interpretation (2). For each study it is necessary to use the same instrument, stipulate own norms within the ratio to the healthy population, and the examination should not be conducted by more than one person.

Another diagnostic procedure in the evaluation of the development of retinopathy and other changes is fluorescence angiography, which assists in the indication of applicable laser treatment (30). In our group of adolescent patients we did not perform this, since the clinical picture of the vascular preretinopathic changes did not constitute a reason for a complex examination. We had already demonstrated clear ophthalmoscopic dilation and tortuosity of the end capillaries, including this contrast examination (19) in the last study using a digital camera. The previous procedure of examination using a mechanically controlled camera with a 15 degree visual field and coarse grain RS2 X-ray films was capable of demonstrating also the fine proportionate content of the capillaries, e.g. the heads of the optic nerve (12), which is difficult for the method of digital technology.

An analysis of the metabolic observation using HbA1c, primarily in its one-year profile, demonstrated higher values in the case of DpR than in individuals with DM 1 even without these functional-structural changes. In our entire study cohort, decompensation of DM 1 had an influence on higher values of HbA1c from the average in two patients during the course of the last year of observation. On the other hand, both patients had a pronounced ophthalmoscopic finding as against the physiological interface, which was a manifestation also of their year-round profile of pathological values of HbA1c. Compensation of diabetes evaluated by the level of HbA1c can be considered optimal at a value of up to 58 mmol/mol (7.5% according to DCCT), suboptimal up to 75 mmol/mol (9% according to DCCT), and higher values are highly risk-laden (27). This relationship is in accordance with the assertion that the long-term pathological values of HbA1c have an influence on the development not only of diabetic retinopathy, but also of diabetic macular edema (15, 16). With regard to compensation of DM 1, puberty represents a significant risk factor. A role in this is played by both biological factors (increase of insulin resistance) and psycho-social factors (worsened compliance with treatment) (4, 25). As a result, compensation of metabolic disorder is of fundamental importance in the prevention of the development of DR (32), which is substantiated also by the last DCCT (Diabetes Control and Complications Trial) (5) and EDIC (Epidemiology of Diabetes Intervention and Complications) (8) studies.

CONCLUSION

Using a complex diagnostic procedure, which combined a functional examination of the retina using CS and an evaluation of the structural changes of the retina using SD-OCT, we defined preretinopathy in the central region in five observed adolescents with type 1 diabetes. Another three patients were included in the group of DpR on the basis of pathological SD-OCT values with the presence of isolated hard scattered deposits. This pilot study was originally intended as the initial communication of the research project IGA NT/14490, which was not accepted. In future, in order to confirm our theory of preretinopathic changes it shall be necessary to extend the study cohort with further patients in other age groups, to retrospectively supplement the period of evaluation of HbA1c and include further parameters of metabolic observation. It is necessary to define the structural changes in more detail, with manifestations in the individual retinal layers, also with regard to the possible development of diabetic macular edema.

Thanks to V. Mezerová from the Faculty of Biomedical Engineering of the Czech Technical University in Prague for performing the statistical calculations.

LITERATURE

1. **Asefzadeh, B., Fisch, B.M. et al.:** Macular Thickness and Systemic Markers for Diabetes in Individuals with No or Mild Diabetic Retinopathy. Clin Experiment Ophthalmol, 36, 2008: 455–463.
2. **Chalam, K.V., Bressler, S.B. et al.:** Retinal Thickness in People with Diabetes and Minimal or No Diabetic Retinopathy: Heidelberg Spectralis Optical Coherence Tomography. Invest. Ophhalmol. Vis Sci, 53, 2012: 8154–8161.
3. **Cinek, O, Kulich, M. et. al.:** The Incidence of Type 1 Diabetes in Young Czech Children Stopped Rising. Pediatric Diabetes, 13, 2012: 559–563.
4. **Danne, t., Mortensen, H.B. et al.:** Persistent Differences Amog Centers over 3 Yeaers in Study of 3.805 Chilren and Adolescents with Type 1 Diabetes from the HvidØre Study Group. Diabetes Care, 24, 2001: 1342–47.
5. **Diabetes Control and Complications Trial Research Group (DCCCT):** The Efffect of Intensive Treatment of Diabetes on tht Development and Progression of Long Term Complications in Insulin-dependent Diabetes Mellitus. N.Engl. J. Med., 329, 1993: 977–986.
6. **DeBuc, C.D., Somfai, G.M.:** Early Detection of Retinal Thickness Changes in Diabetes Using Optical Coherence Tomography. Med. Sci. Monit., 16, 2010: 15–21.
7. **Demir, M., Oba, E. et al.:** Central Makulare Thickness in Pacients with Type 2 Diabetes Mellitus without Clinical Retinopathy. BMC Ophtalmol., 13, 2013, April, doi.:10.1186
8. **Epidemiology of Diabetes Interventions and Complications (EDIC):** Design, Implementation, and Preliminary Results of a Long-term Follow-up of the Diabewtes Control and Complications Trials Cohort., Dabeties Care, 22, 1999: 99–111.
9. **EURODIAB Study Group (ed. Patterson C.C.et al.):** Incidence Trend for Childhood Type 1 Diabetes in Europe during 1989 – 2003 and Predicted New Cases 2005 – 20: Multicentre Prospective Registration Study. Lancet, 373, 2009: 2027–33.
10. **Georgakopoulos, C.D., Eliopoulou, M.I. et. al.:** Decreased Contrast Sensitivity in Children and Adolescents with Type 1 Diabetes Mellitus. J. Pediatr. Ophthalmol. Strabismus, 48, 2011: 92–97.
11. **Hee, M.R., Puliafito, C.A. et al.:** Topography of Diabetic Macular Edema with Optical Coherence Tomography. Ophthalmology, 105, 1998: 360–370.
12. **Karel, I., Peleška, M.:** Fluorescenční angiografie u pseudoedému zrakového nervu. Čs Oftal, 32, 1976: 275–281.
13. **Kalvodová, B., Záhlava, J.:** Výsledky vitrektomie u cystoidního makulárního edému v obrazu OCT. Čes a Slov Oftal, 58, 2002: 224–232.
14. **Katz, G., Levkovitch-Verbin, H. et al.:** Mesoptic Foveolal Contrast Sensitivity Is Impaired in Diabetic Patients without Retinopathy. Graefes Arch. Clin. Exp. Ophthalmol., 248, 2010: 1699–1703.
15. **Klein, R., Lee, K.E. et al.:** The 25-Year Incidence of Visual Impairment in Type 1 Diabetes Mellitus: The Wisconsin Epidemiologic Study of Diabetic Retinopathy. Ophthalmology, 116, 2010: 63–70.
16. **Klein, R., Knudtson, M.D. et al.:** The Wisconsin Epidemiologic Study of Diabetic Retinopathy: XXIII the Twenty-five-year Incidence of Macular Edema in Person with Type 1 Diabetes. Ophthalmology, 116, 2010: 497–503.
17. **Koleva-Georgieva, D.N., Sivkova, N.P.:** Optical Coherence Tomography for the Detection of Early Macular Edema in Diabetic Patients with Retinopathy. Folia Med, 52, 2010: 40–48.
18. **Krásný, J., Brunnerová, R. et. al.:** Test citlivosti na kontrast v časné detekci očních změn u dětí, dospívajících a mladých dospělých s diabetes mellitus I. typu. Čes a Slov Oftal, 62, 2006: 381–392.
19. **Krásný, J., Cihelková, I. et al.:** Citlivost na kontrast a fluorescenční angiografie při hodnocení očních změn v rámci posouzení kompenzace diabetes mellitus I. typu u mladých dospělých pacientů. Čes a Slov Oftal, 63, 2007: 17–27.
20. **Krasny, J., Andel, M. et al.:** The Contrast Sensitivity Test in Early Detection of Ocular Changes in the Relation to the Type 1 Diabetes Mellitus Compensation in Children, Teenagers, and Young Adults. Recent Pat. Inflamm. Alergy Drug Discov, 1, 2007: 232–236.
21. **Krásný, J., Vyplašilová, E., et al.:** Změna transparence čoček u dětí, mladistvých a mladých dospělých s diabetes mellitus 1. typu. Čes a Slov Oftal, 62, 2006: 304–314.
22. **Krásný, J.:** Závěrečná zpráva IGA NR/7952 : „Časná detekce klasifikace očních změn u dětí, dospívajících a mladých dospělých s diabetes mellitus 1. typu".
23. **Lopes de Faria, J.M., Katsumi, O. et al.:** Neurovisual Abnormalities Preceding the Retinopathy in Patients with Long Term Type 1 Diabetes Mellitus. Graefes Arch Clin Exp Ophthalmol, 239, 2001: 643–648.
24. **Martinelli, V., Lacerenza, M. et al.:** The Objective Assessment of Visual Contrast Sensivity by Pattern Reversal Visual Evoked Potential in Diabetes. J Diabet Complications, 2, 1988: 44–46.
25. **Mortensen, H.B., Hougaard, P.:** Comparison of Metabolic Control in a Cross-section Study of 2.873 Children and Adolescents with IDDM from 18 Countries. The HvidØre Study Group on Childhood Diabetes. Diabetes Care, 20, 1997: 714–720.
26. **Pelikánová, T.:** Novinky v diabetologii. Sborník abstrakt XII. Sympozia: Diabetes mellitus – oční komplikace, UP Olomouc, 2011: 5.
27. **Rewers, M., Pihoker, C. et al.:** ISPAD Clinical Practice Consensus Guidelines 2009 Compendium Assessment and Monitoring of Glycemic Control in Children and Adolescents with Diabetes. Pediatr Diabet, 10, 2009, (Suppl. No. 12): 71–81.
28. **Sánchez-Tocino, H., Alvarez-Vidal, A. et al.:** Retinal Thickness Study with Optical Coherence Tomography in Pacient with Diabetes. Invest. Ophthalmol Vis Sci, 43, 2002: 1588–1594.
29. **Sokol, S., Moskowitz, A. et al.:** Contrast Sensivity in Diabetics with and without Background Retinopathy. Arch Ophthalmol, 103, 1985: 51–54.
30. **Sosna, T., Bouček, P., Fišer, I.:** Diabetická retinopatie, J. Cendelín, 2001, Praha, 255 s.
31. **Sosna, T., Švancarová, R.:** Zpráva z EASDec 2011. Sborník abstrakt XII. Sympozia: Diabetes mellitus – oční komplikace, UP Olomouc, 2011: 19–26.
32. **Sosna, T., Veith, M.:** Zpráva z EASDec 2013. Sborník abstrakt XIV. Sympozia: Diabetes mellitus – oční komplikace, UP Olomouc, 2013: 20–26.
33. **Sun, T.S., Zhang, M.N.:** Characters of Contrast Sensitivity in Diabetic Patients without Diabetic Retinopathy. Zhonghua Yan Ke Za Zhi. 48, 2012: 41–46.
34. **Thomas, M..G., Kumar, A. et al.:** Hight Resolution in Vivo Imaging in Achromatopsia. Ophthalmology, 118, 2011: 882–887.
35. **Urban, B., Bakunowicz-Lazarcyk, A. et al.:** The Evaluation of Contrast Sensivity in Children and Adolescents with Insulin-Dependent Diabetes Mellitus. Klin. Oczna. 101, 1999: 111 – 114.
36. **Verrotti, A., Lobefalo, L. et al.:** Relationship bettween Contrast Sensivity and Metabolic Control in Diabetics with and without Retinopathy. Ann Med, 30, 1998: 369–374.
37. **van Dijk, H.W., Kok, P.H. et al.:** Selective Loss of Inner Retinal Layer Thickness in Type 1 Diabetic Patients with Minimal Diabetic Retinopathy. Invest. Ophthalmol Vis Sci, 50, 2009: 3404–3409.
38. **Vujosevic, S., Midena, E.:** Retinal Layers Changes in Human Preclinical and Early Clinical Diabetic Retinopathy Support Early Neuronal and Müller Cell Alterations.. J. Diabetes Res, 2013, Juny, doi.: 10.1155.

ORIGINAL ARTICLE

Cytomegalovirus Infection (CMV) in Patients with Acquired Immune Deficiency Syndrome

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SUMMARY

Cytomegalovirus infection (CMV) in patients with acquired immunodeficiency syndrome (Acquired Immune Deficiency Syndrome, AIDS) is the most common opportunistic infection. This infection is harmless for healthy individuals, but for weakened individuals cause disease. The most common form of CMV-infection in patients with AIDS is cytomegalovirus retinitis, which occurs in 15% to 40% of cases. We report the case of a man twenty-five year old, treated for CMV retinitis and retinal vasculitis vessels. Prescribed Valcyte 900 mg tbl. twice daily for 21 days with a good therapeutic effect. In patients with AIDS and decreased visual acuity is need be primarily thinking about the possible presence of CMV-infection and in time to start treatment..

Key words: Cytomegalovirus (CMV) retinitis, AIDS, valganciklovir
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INTRODUCTION

Human cytomegalovirus (CMV) belongs to the group of herpes viruses (Human Herpesvirus 5). Primary infection by human CMV takes place during life: perinatally (either in the womb or via the mother's milk), in childhood upon close contact, in adulthood through contact transmission in sexual intercourse and also through blood transfusions or blood derivates (11, 22).
Upon an increase in sexual activity in the adolescent and adult population, there is a marked increase in the number of persons with IgG antibodies against CMV (22). We know that as many as 95% of homosexual men and almost all homosexual men infected with HIV (human immunodeficiency virus) are CMV-seropositive (9, 10, 32, 41).
After the primary infection, CMV enters the latent phase, and subsequent reactivation (secondary infection) occurs depending on changes in relationships between the host and the virus (pregnancy, serious illness, stress, immunosuppressive therapy, AIDS).
Reactivation of CMV is most commonly manifested in the following: retinitis (approximately 80% of cases) and gastrointestinal disorders (colitis, esophagitis, gastritis) in approximately 15% of cases (19). Cytomegalovirus retinitis is the most common cause of loss of vision in patients with AIDS (1, 38). Less common infection manifestations of AIDS are encephalitis, pneumonia, polyradiculopathy, sclerotic cholangitis and hepatitis (11, 19, 33).

CASE REPORT

A twenty three year old man was sent to the infections clinic of the University Hospital in Hradec Králové for an eye consultation due to deteriorated vision in the right eye. He was hospitalised in the infections clinic due to clinical and laboratory progression of the base disease (infection with HIV since 2012) as a result of lack of co-operation in therapy. The quantity of CD4+ lymphocytes was equal to 80 bb/mm3L (method of flow cytometry), the viral charge of HIV 531 000 copies/ml. Data from personal anamnesis: homosexual, intravenous drug user (methamphetamine), molluscum contagiosum of the skin, candidiasis of the oropharynx and upper respiratory tracts, wasting syndrome, image of encephalitis according to magnetic resonance imaging. Overall therapy: Truvada tbl. (200 mg emtricitabine and 245 mg tenofovir disoproxil), Reyataz tbl. (atazanavir 100 mg), Norvir (ritonavir 100 mg), Mycomax tbl. (fluconazole 100 mg), Cotrimoxazol AL Forte (sulphamethoxazole 800 mg and trimethroprim 160 mg). The patient had experienced ocular complaints for 1 month as a blind spot in the visual field of the right eye, which was progressively enlarging. Visual acuity in the right eye was 2/50, correction best here, visual acuity in left eye was 6/5 naturally. Finding on the perimeter (Humphrey, test 30-2): in the right eye (fig. 1) there is absolute scotoma throughout the entire scope of the visual field, mean deviation (MD) = -33.17 dB, in the left eye (fig. 2) we find points of reduced sensitivity in the periphery, centre preserved, MD = -15.92 dB. Objectively in the right eye we find numerous precipitates on the corneal endothelium, in the anterior chamber isolated cells, turbidities in the vitreous body BIO 1+ (binocular indirect ophthalmoscopy), papilla is bordered (paler colours), on the retina there are perceptible yellowish-white areas of retinal necrosis with grainy edges, with manifestations of haemorrhage in zones 1 and 2, coarse dysgrouping of pigment, accompanying streaks along capillaries, long sections of capillaries with signs of severe vasculitis, end of vascular channel of lower temporal arcade, irregularity of vascular lumen (fig. 3). In the left eye there is a finding on the anterior segment commensurate to age, cotton wool spots predominate on the ocular fundus in the central periphery, otherwise no pathological intraocular finding (fig. 4).
Optical coherence tomography (OCT) (Zeiss Cirrus) of the central region of the right eye (fig. 5) shows reduced thickness of the neuroretina, which displays signs of necrosis and atrophy, the tissue has a hyperreflexive structure, central thickness is 76 µm.
On the basis of the objective condition, a preliminary diagnosis of CMV--retinitis was determined in the right eye, with a finding of cotton wool

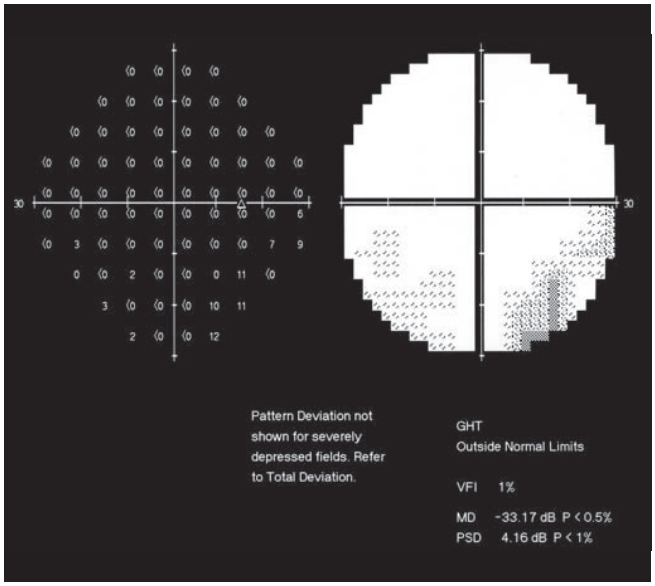

Fig. 1 Perimeter of right eye. Absolute scotoma throughout entire visual field.

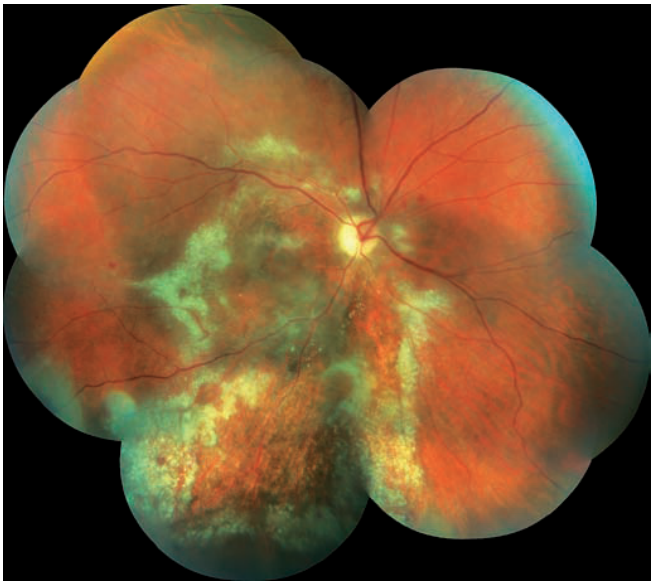

Fig. 2 Perimeter of left eye. Tunnel vision, peripheral scotoma, centre preserved.

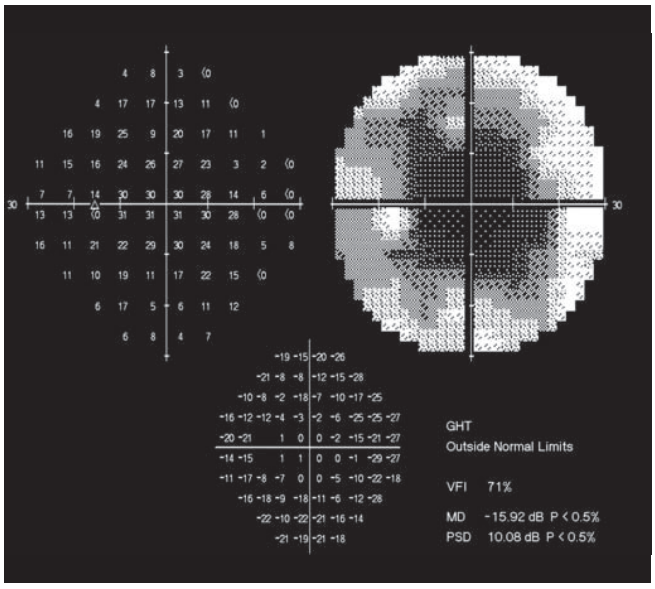

Fig. 3 Right eye. Current stage of CMV-retinitis, yellowish--white areas of retinal necrosis.

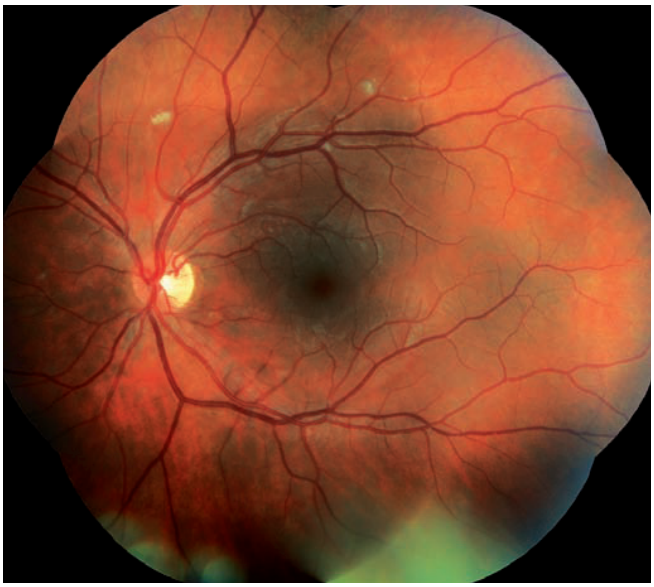

Fig. 4 Left eye. Finding of cotton wool spots.

spots on the retina of the left eye.
On the basis of the results of the laboratory values, and with regard to the finding on the ocular fundus, peroral antiviral treatment was commenced using valganciclovir (Valcyte tbl.) in a dose of 900 mg twice daily for a period of 21 days. During the following eye examinations, the clinical finding in the anterior segment of the right eye and on the ocular fundus bilaterally progressively improved (fig. 6).
At a follow-up examination at an interval of 5 weeks after the commencement of antiviral therapy, the precipitates on the endothelium and Tyndallisation in the anterior chamber of the right eye had disappeared, on the ocular fundus there was pigment hypertrophy on the border of the deposits of chorioretinal atrophy, yellowish-white retinal necroses were receding, cotton wool spots in the left eye had been absorbed (fig. 7, 8). However, atrophy of the retina is perceptible on auto-fluorescence photography of the fundus of the right eye, including the central region – coarse dysgrouping of pigment in the affected area (fig. 9). There was no improvement of central visual acuity in the right eye. The finding on the perimeter of the right eye remained without changes, with only a slight improvement in the left eye (fig. 10, 11).
With regard to the stabilisation of the finding of CMV-retinitis, the patient was transferred onto a maintenance dose of Valcyte 900 mg mg perorally once per day for a period of 3 months.

DISCUSSION

Retinitis is a serious manifestation of cytomegaloviral infection, and the most common cause of loss of sight in patients with AIDS (1, 36, 37, 38). At the time when there was not yet any specific treatment of CMV-retinitis, fears of blindness represented the most common reason for suicides amongst patients with AIDS (8).
It had previously been determined that a low quantity of CD4+ lymphocytes (T-lymphocytes, “helpers”) is a

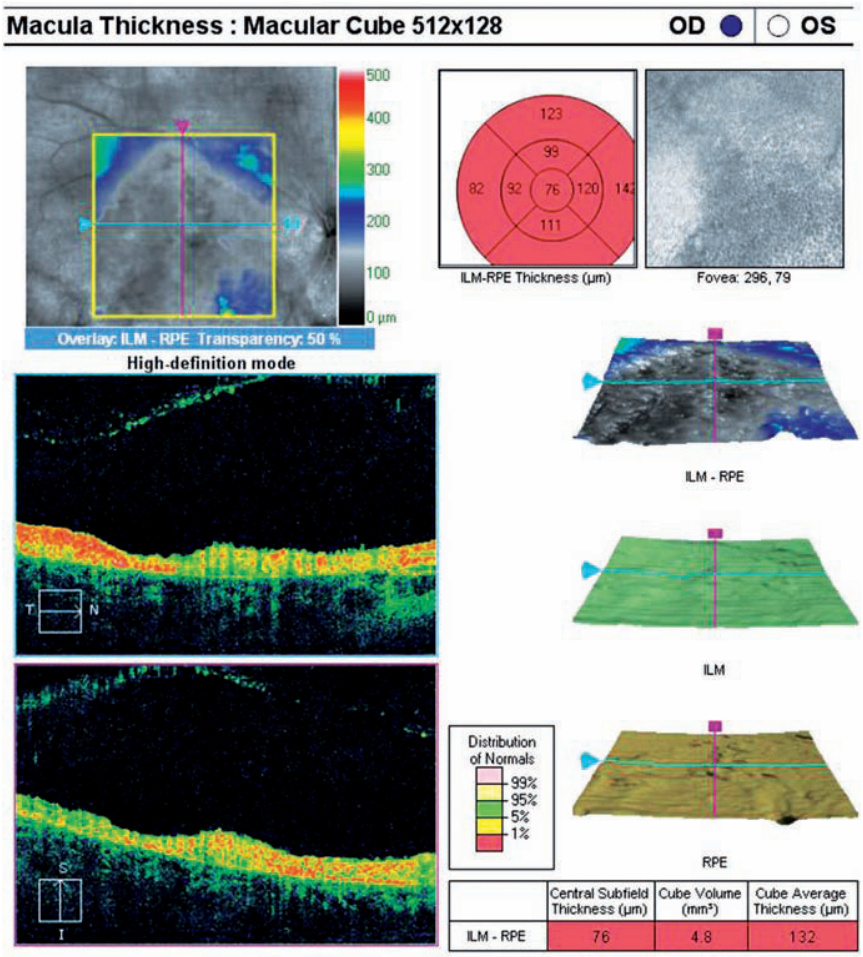

Fig. 5 OCT of right eye: reduced thickness of neuroretina, CT = 76 µm, which manifests signs of necrosis and atrophy, has hyperreflexive structure

significant risk factor for the incidence of CMV-retinitis (38, 41). A large study from the Multicenter AIDS group in the United States demonstrated that a quantity of CD4+ lymphocytes lower than 100 bb/mm3L, HIV seropositivity and homosexuality represent significant risk factors for the development of CMV retinitis (9, 19).

In the case of a reduction of CD4+ beneath the level of 100 bb/mm3L, CMV-retinita occurs in 14% of patients. If CD4+ falls beneath 50 bb/mm3L, CMV-retinitis occurs in 24% of patients (19, 45). In our case the patient had a number of CD4+ lymphocytes of 80 bb/mm3L.

Hodge et al. published a study evaluating various predictors of CMV-retinitis in patients with AIDS (23). The two most significant are previous extraocular manifestations of CMV infection (odds ratio (OR) = 82.99) and photopsia or floating turbidities in the vitreous area (OR = 11.42). Less significant predictors included experience of mycobacterial infection (OR = 3.41), finding of cotton wool spots on the retina (OR = 2.90) and homosexuality (OR = 2.83).

We determine a diagnosis of CMV-retinitis on the basis of an examination of the ocular fundus, most typically there is a presence of yellowish-white areas of retinal necrosis with grainy edges (6, 42). CMV-retinitis is spread through direct contact of the infected tissue with the adjacent healthy retina, often in the form of a “bush fire” (12). Inflammatory

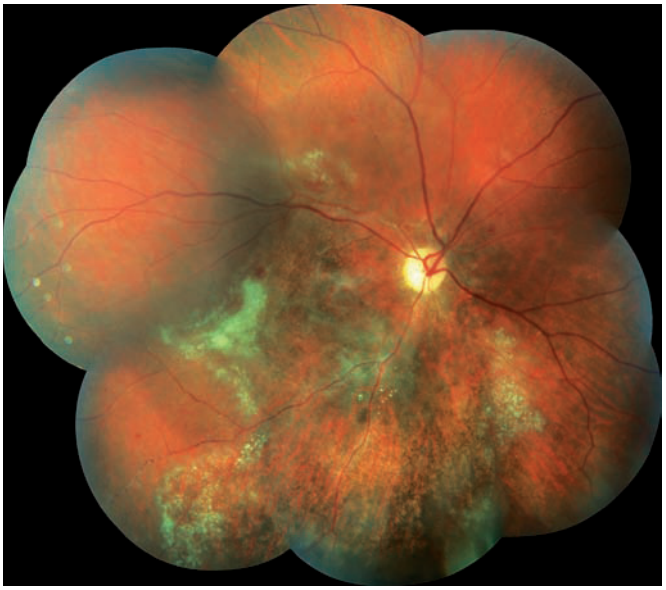

Fig. 6 Right eye. Condition 3 weeks after commencement of treatment with Valcyte. Improvement of finding.

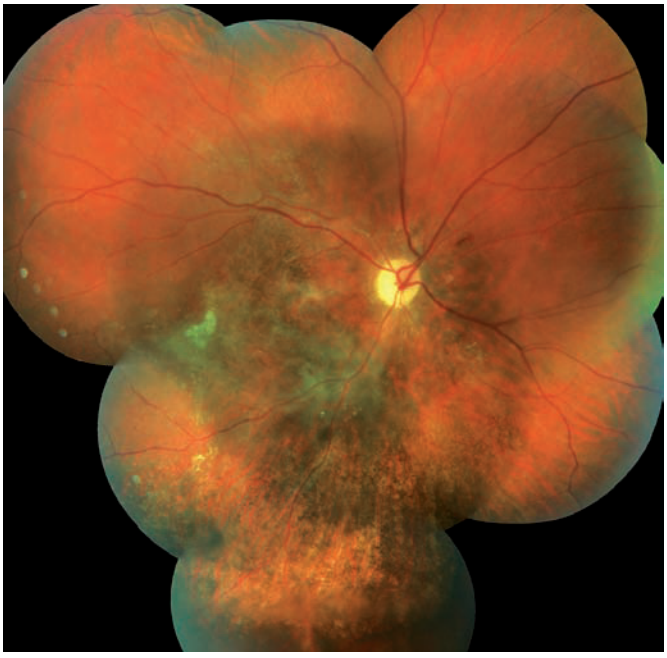

Fig. 7 Right eye. Condition 5 weeks after commencement of treatment with Valcyte. Pigment hypertrophy on border of deposit of chorioretinal atrophy and depigmentation elsewhere.

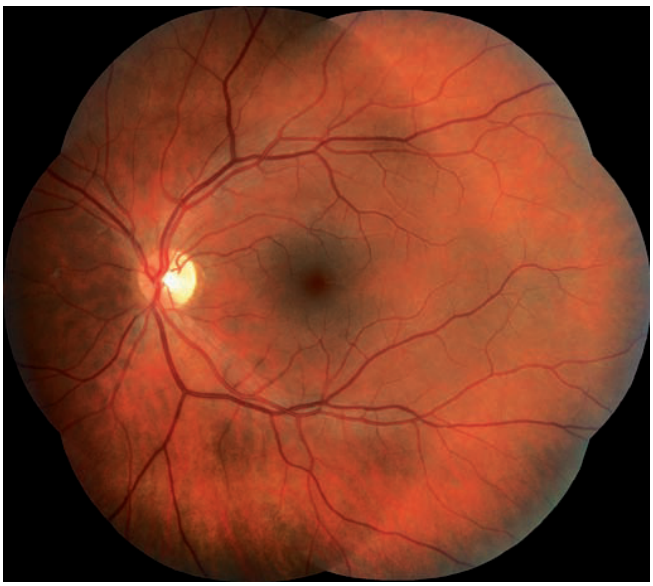

Fig. 8 Left eye. Condition 5 weeks after commencement of treatment with Valcyte. Cotton wool deposits have disappeared.

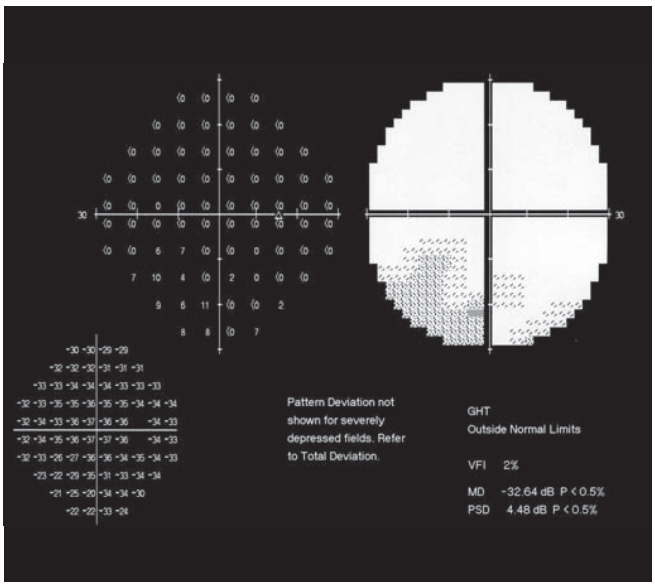

Fig. 10 Perimeter of right eye. Condition 5 weeks after commencement of treatment with Valcyte.

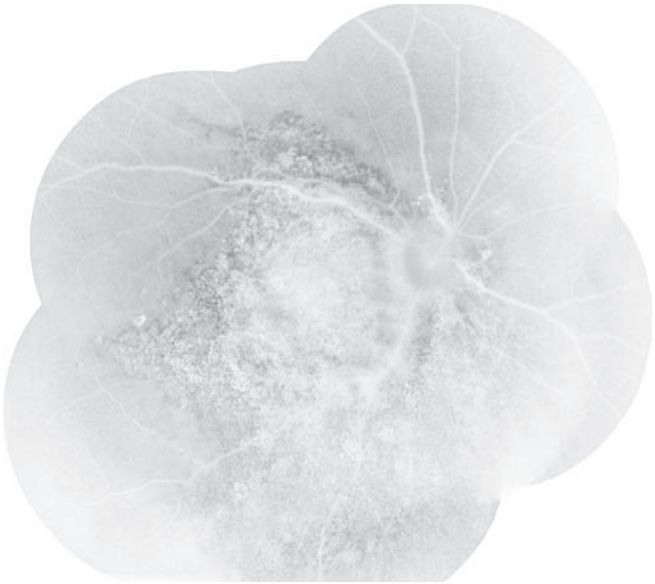

Fig. 9 Auto-fluorescence photograph of ocular fundus of right eye 5 weeks after commencement of treatment with Valcyte. Atrophy of retina including central region – coarse dysgrouping of pigment in affected area.

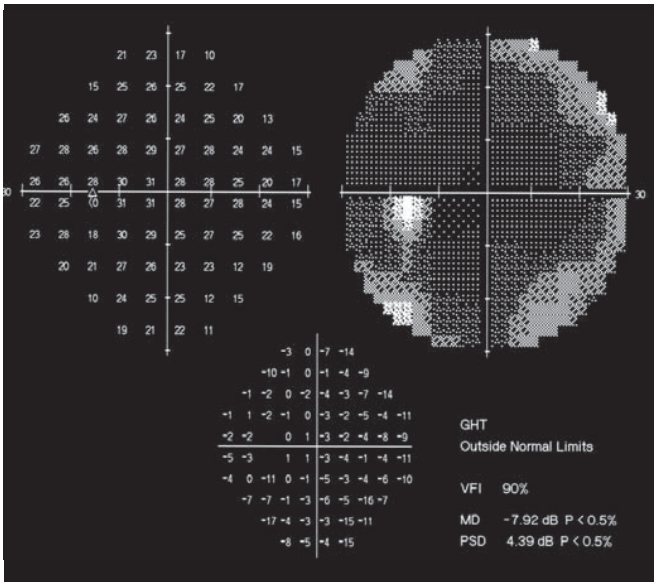

Fig. 11 Perimeter of left eye. Condition 5 weeks after commencement of treatment with Valcyte. Points of reduced sensitivity in periphery, centre preserved.

changes on the retina pass into atrophy with permanent damage to visual functions, which we recorded in our patient.

According to localisation of manifestations of CMV-retinitis on the ocular fundus, we divided the retina into 3 zones according to Lee et al. (40). Zone 1 is located 3 000 μm from the centre of the fovea and 1 500 μm from the disc of the optic nerve. Zone 2 continues from the periphery of zone 1 to the ampular vein. Zone 3 incorporates all the areas from the vortex ampullae to the ora serrata (25). The worst visual prognosis is in the case of affliction of zone 1, because this covers both the fovea and the optic nerve.

CMV-retinitis occurs most frequently in zones 2 and 3 (11, 20). Although signs of inflammation on the anterior segment of the eye and vitreous area are common, they are not so pronounced as to cause reddening, pain, anterior synechia or to impair the view of the retina, even if vitreous turbidities may progress over time (24).

Without the commencement of treatment of CMV-retinitis, a complete destruction of the retina takes place within 6 months (24). The speed of progression is relatively slow, on average 24 μm/day (27). It is important to be aware that the pathology may progress without a manifest finding of whitish deposits of necrosis on the retina, and as a result it is necessary to regularly check the ocular fundus (mainly the condition of the RPE) with applicable photo documentation.

Afflicted patients may see white stains and blind spots in the visual field, photopsia (depending on the area of the afflicted retina), floating flakes (in the case of presence of vitreous turbidities) or deteriorated visual acuity (12, 21).

There are a number of pathological con-

ditions which we have observed within the framework of differential diagnostics, accompanying the finding of CMV--retinitis in our patient. These include HIV-retinopathy, toxoplasmic chorioretinitis, acute retinal necrosis (ARN) and progressive exterior retinal necrosis.

HIV-retinopathy is characterised by a larger quantity of cotton wool spots on the retina. The prevalence of the pathology increases with the decreasing number of CD4+ cells, particularly if the value is less than 50 bb/mm3L (19, 45). It has also been demonstrated that an increased plasmatic HIV-RNA viral charge is a predictor of the development of HIV-retinopathy (17). The cause of HIV-retinopathy remains unknown, although a number of factors have been described which have an influence on the abnormal through flow of blood to the retina upon HIV-retinopathy (2, 14). Small, yellowish-white cotton wool spots appear on the ocular fundus, which represent infarctions of the layer of nerve cells in the retina and may cause the occurrence of a temporary small scotoma in the visual field, but mostly do not cause loss of sight. They are usually located in the retina around the disc of the optic nerve. Occasionally cotton wool spots may be linked to mild intraretinal haemorrhage, which leads to errors in the diagnosis of incipient CMV--retinitis. In such a case it is necessary to check the patient over the course of the following 4 weeks, when it shall be possible to distinguish HIV-retinopathy (spontaneous regression of cotton wool spots) and CMV-retinopathy (spread of original infectious deposit on the retina). In the case of HIV-retinopathy there are no manifestations of inflammation in the anterior chamber or vitreous cavity. By contrast with CMV-retinitis, the deposit of the inflammation may spontaneously disappear within a few weeks even without the commencement of therapy.

Toxoplasmic chorioretinitis may have a similar finding on the retina as CMV-retinitis (18). The only differing symptom is the degree of inflammation. In the case of toxoplasmosis, dense, opaque retinal necrosis with pronounced vitritis appear on the retina. Vitritis differs from CMV-retinitis, which usually shows minimal inflammatory manifestations in the vitreous cavity and has a dry, "granulated" border (13, 26).

Acute retinal necrosis is a rapidly progressing, vaso-occlusive, necrotising retinitis, which spreads around the perimeter of the retina. In contrast with CMV retinitis, ARN is linked with significant inflammation of the anterior segment of the eye and the vitreous cavity and occurs in both immunocompetent and immunocompromised patients. Further symptoms include neuritis of the disc of the optic nerve, scleritis and pain (28). HIV-positive patients with ARN mostly have a lower number of CD4+ cells than 100 bb/mm3L, and the ocular finding is frequently preceded by herpetic dermatological manifestations (4).

In the case of severe immunodeficiency in the later stage of AIDS, there may be secondary occurrence of progressive exterior retinal necrosis (15). However, this finding has also been reported in patients with severe immunodeficiency without HIV infection (5, 16). With the widespread use of highly active antiretroviral therapy, the number of patients with AIDS who have CD4+ T-cells lower than 50 bb/mm3L has been reduced. As a consequence of this, there has been a marked reduction in the number of new cases of opportunistic infections, including CMV-retinitis (7, 29).

Furthermore, the long-term results of CMV-retinitis point to a markedly lower risk of damage to sight upon the use of highly active antiretroviral therapy (30, 31, 35).

At present there are a number of options for treatment of CMV-retinitis that have been approved by The United States Food and Drug Administration (40). This concerns: ganciclovir administered intravenously (Cytovene-IV; Roche Laboratories Inc.), ganciclovir administered perorally (Cytovene; Roche Laboratories Inc.), foscarnet for intravenous application (Foscavir; AstraZeneca LP, Wilmington, Delaware), cidofovir administered intravenously (Vistide; Gilead Sciences, Foster City, California) and valganciclovir for peroral use (Valcyte, Roche Laboratories Inc.). There is also the option of intravitreal application of an implant which progressively releases a medicinal substance: ganciclovir (Vitrasert, Bausch & Lomb Inc., Surgical Division, Irvine, CA) and fomivirsen (Vitravene, Novartis Ophthalmics, Duluth, GA).

As an induction therapy, administration of high doses of anti-CMV drugs (antiviral drugs) is used for a period of 2-4 weeks. With regard to the fact that although the replication of CMV is suppressed, the virus is not completely eliminated from the body, further maintenance virostatic therapy in a lower dose is subsequently necessary in order to prevent the further progression of retinitis. In our case we chose valganciclovir as the induction and maintenance treatment of CMV-retinitis.

Valcanciclovir (L-valine ester ganciclovir) has good biological accessibility for peroral administration and is metabolised into ganciclovir rapidly following absorption through the intestinal wall. Peroral administration provides a similar concentration of the drug in serum as upon intravenous application of ganciclovir (34, 44).

In comparison with perorally administered ganciclovir, valganciclovir has approximately ten times higher peroral biological accessibility and provides a higher plasmatic concentration (3, 34). Valganciclovir as an induction therapy is a good alternative to intravenous administration of ganciclovir for the majority of patients with newly diagnosed CMV-retinitis, and also substitutes ganciclovir for maintenance therapy upon peroral administration (11). The most common adverse effects in connection with treatment by valganciclovir include diarrhoea (35%), nausea (23%), fever (18%), anaemia (haemoglobin < 8.0g/l) (12%) and neutropaenia (absolute number of neutrophiles < 500 cells/μL) (10%) (39). We did not record any adverse effects in our patient.

CONCLUSION

CMV-retinitis is the most common ocular manifestation of opportunistic infection in patients with AIDS.

Without timely commencement of treatment, the pathology gradually progresses and causes total necrosis of the retina and loss of sight within 6 months. Valganciclovir in peroral administration is currently a good alternative to intravenous administration of ganciclovir as both induction and maintenance therapy of CMV-retinitis.

LITERATURE

1. **Anonymous:** Foscarnet-ganciclovir cytomegalovirus retinitis trial: 5. Clinical features of cytomegalovirus retinitis at diagnosis. Studies of Ocular Complications of AIDS Research Group in collaboration with the AIDS Clinical Trials Group. Am J Ophthalmol, 1997; 124: 141–157.
2. **Anonymous:** Foscarnet-ganciclovir cytomegalovirus retinitis trial. 4. Visual outcomes. Studies of Ocular Complications of AIDS Research Group in collaboration with the AIDS Clinical Trials Group. Ophthalmology, 1994; 101: 1250–1261.
3. **Anonymous:** Valganciclovir. U.S. Department of Health and Human Servi-

ces. AIDSinfo, 2004, 1–5. http://aidsinfo.nih.gov/drugs/ htmldrug_tech.asp?int_id_0271. Last accessed February 22, 2006.
4. **Batisse, D., Eliaszewicz, M., Zazoun, L., et al.:** Acute retinal necrosis in the course of AIDS: study of 26 cases., AIDS, 1996; 10: 55–60.
5. **Benz, MS., Glaser, JS., Davis, JL.:** Progressive outer retinal necrosis in immunocompetent patients treated initially for optic neuropathy with systemic corticosteroids. Am J Ophthalmol, 2003; 135: 551–553.
6. **Bloom, JN., Palestine, AG.:** The diagnosis of cytomegalovirus retinitis. Ann Intern Med, 1988, 109: 963-969.
7. **Casado, JL., Arrizabalaga, J., Montes, M., et al.:** Incidence and risk factors for developing cytomegalovirus retinitis in HIV-infected patients receiving protease inhibitor therapy. Spanish CMV-AIDS Study Group. AIDS, 1999; 13: 1497–1502.
8. **Culbertson,WW.:** Infections of the retina in AIDS. Int Ophthalmol Clin, 1989; 29: 108-118.
9. **Drew, WL., Mintz, L., Miner, RC., et al.:** Prevalence of cytomegalovirus infection in homosexual men. J Infect Dis, 1981; 143: 188–192.
10. **Drew, WL.:** Cytomegalovirus infection in patients with AIDS. J Infect Dis, 1988; 158: 449–456.
11. **Dunn, JP., Martin DF.:** Treatment of cytomegalovirus (CMV) retinitis in the era of highly active antiretroviral therapy., Elmwood Park, NJ: Medscape from WebMD, 2003. http://www.medscape.com/viewprogram/663. Last accessed February 22, 2006.
12. **Egbert, PR., Pollard, RB., Gallagher, JG., et al.:** Cytomegalovirus retinitis in immunosupressed hosts. II. Ocular manifestations. Ann Intern Med, 1980; 93: 664–670.
13. **Elkins, BS., Holland, GN., Opremcak, EM., et al.:** Ocular toxoplasmosis misdiagnosed as cytomegalovirus retinopathy in immunocompromised patients. Ophthalmology, 1994, 101: 499-507.
14. **Engstrom, RE, Jr., Holland, GN., Hardy, WD., et al.:** Hemorheologic abnormalities in patients with human immunodeficiency virus infection and ophthalmic microvasculopathy. Am J Ophthalmol, 1990; 109: 153–161.
15. **Engstrom, RE, Jr., Holland, GN., Margolis, TP., et al.:** The progressive outer retinal necrosis syndrome. A variant of necrotizing herpetic retinopathy in patients with AIDS. Ophthalmology, 1994; 101: 1488–1502.
16. **Foster, RE., Petersen, MR., Neuss, MN., et al.:** Progressive outer retinal necrosis syndrome in a lymphoma patient with good visual outcome., Am J Ophthalmol, 2001; 132: 117–120.
17. **Furrer, H., Barloggio, A., Egger, M., et al.:** The Swiss HIV Cohort Study. Retinal microangiopathy in human immunodeficiency virus infection is related to higher human immunodeficiency virus-1 load in plasma., Ophthalmology, 2003; 110: 432–436.
18. **Gagliuso, DJ., Teich, SA., Friedman, AH., et al.:** Ocular toxoplasmosis in AIDS patients. Trans Am Ophthalmol Soc, 1990; 88: 63–86.
19. **Gallant, JE., Moore, RD., Richman, DD., et al.:** Incidence and natural history of cytomegalovirus disease in patients with advanced human immunodeficiency virus disease treated with zidovudine. The Zidovudine Epidemiology Study Group. J Infect Dis, 1992; 166: 1223–1227.
20. **Henderly, DE., Freeman, WR., Causey, DM., et al.:** Cytomegalovirus retinitis and response to therapy with ganciclovir. Ophthalmology, 1987; 94: 425–434.
21. **Hennis, HL., Scott, AA., Apple, DJ.:** Cytomegalovirus retinitis. Surv Ophthalmol, 1989; 34: 193–203.
22. **Ho, M.:** Epidemiology of cytomegalovirus infections. Rev Infect Dis, 1990; 12(7): 701–710.
23. **Hodge, WG., Boivin, JF., Shapiro, SH., et al.:** Clinical risk factors for cytomegalovirus retinitis in patients with AIDS. Ophthalmology, 2004; 111: 1326–1333.
24. **Holland, G., Tufail, A., Jordan, MC.:** Cytomegalovirus diseases. In Pepose, JS., Holland, GN., Wilhelmus, KR., et al.: Ocular Infection & Immunity, New York, Mosby, 1996, s. 1088–1129.
25. **Holland, GN., Buhles, WC Jr, Mastre, B., et al.:** A controlled retrospective study of ganciclovir treatment for cytomegalovirus retinopathy. Use of a standardized system for the assessment of disease outcome. The UCLA CMV Retinopathy Study Group., Arch Ophthalmol, 1989; 107: 1759–1766.
26. **Holland, GN., Engstrom, RE, Jr., Glasgow, BJ., et al.:** Ocular toxoplasmosis in patients with acquired immunodeficiency syndrome. Am J Ophthalmol, 1988; 106: 653–667.
27. **Holland, GN., Shuler, JD.:** Progression rates of cytomegalovirus retinopathy in ganciclovir-treated and untreated patients. Arch Ophthalmol, 1992; 110: 1435–1442.
28. **Holland, GN.:** Standard diagnostic criteria for the acute retinal necrosis syndrome. Executive Committee of the American Uveitis Society. Am J Ophthalmol, 1994; 117: 663–666.
29. **Jabs, DA., Bartlett, JG.:** AIDS and ophthalmology: a period of transition. Am J Ophthalmol, 1997, 124: 227-233.
30. **Jabs, DA., Van, Natta, ML., Thorne, JE., et al.:** Course of cytomegalovirus retinitis in the era of highly active antiretroviral therapy: 1. Retinitis progression. Ophthalmology, 2004; 111: 2224–2231.
31. **Jabs, DA., Van, Natta, ML., Thorne, JE., et al.:** Course of cytomegalovirus retinitis in the era of highly active antiretroviral therapy: 2. Second eye involvement and retinal detachment. Ophthalmology, 2004; 111: 2232–2239.
32. **Jackson, JB., Erice, A., Englund, JA., et al.:** Prevalence of cytomegalovirus antibody in hemophiliacs and homosexuals infected with human immunodeficiency virus type 1., Transfusion, 1988; 28: 187–189.
33. **Jacobson, MA., Mills, J.:** Serious cytomegalovirus disease in the acquired immunodeficiency syndrome (AIDS). Clinical findings, diagnosis, and treatment., Ann Intern Med, 1988; 108: 585–594.
34. **Jung, D., Dorr, A.:** Single-dose pharmacokinetics of valganciclovir in HIV- -and CMV-seropositive subjects. J Clin Pharmacol, 1999; 39: 800–804.
35. **Kempen, JH., Jabs, DA., Wilson, LA., et al.:** Risk of vision loss in patients with cytomegalovirus retinitis and the acquired immunodeficiency syndrome. Arch Ophthalmol, 2003; 121: 466–476.
36. **Kožner, P., Rozsypal, H., Machala, L., et al:** Dva typy cytomegalovirového postižení oka u pacientů s HIV infekcí., Klin mikrobiol inf lék, 2009, 15(5): 180-182.
37. **Kožner, P., Machala, L., Rozsypal, H., et al:** HIV retinopatie. Klin mikrobiol inf lék, 2009, 15(5): 183-184.
38. **Kuppermann, BD., Petty, JG., Richman, DD., et al.:** Correlation between CD4+ counts and prevalence of cytomegalovirus retinitis and human immunodeficiency virus-related noninfectious retinal vasculopathy in patients with acquired immunodeficiency syndrome. Am J Ophthalmol, 1993; 115: 575–582.
39. **Lalezari, J., Lindley, J., Walmsley, S., et al.:** A safety study of oral valganciclovir maintenance treatment of cytomegalovirus retinitis. The Roche Valganciclovir Study Group. JAIDS J Acquir Immune Defic Syndr, 2002; 30: 392–400.
40. **Lee, CH., Bright, DC., Ferrucci, S.:** Treatment of cytomegalovirus retinitis with oral valganciclovir in an acquired immunodeficiency syndrome patient unresponsive to combination antiretroviral therapy. Optometry, 2006; 77: 167–176.
41. **Mintz, L., Drew, WL., Miner, RC., et al.:** Cytomegalovirus infections in homosexual men. An epidemiological study. Ann Intern Med, 1983; 99: 326–329.
42. **Palestine, AG., Rodrigues, MM., Macher, AM., et al.:** Ophthalmic involvement in acquired immunodeficiency syndrome. Ophthalmology, 1984; 91: 1092–1099.
43. **Pertel, P., Hirschtick, R., Phair, J., et al.:** Risk of developing cytomegalovirus retinitis in persons infected with the human immunodeficiency virus. J Acquir Immune Defic Syndr, 1992; 5(11): 1069–1074.
44. **Pescovitz, MD., Rabkin, J., Merion, RM., et al.:** Valganciclovir results in improved oral absorption of ganciclovir in liver transplant recipients. Antimicrob Agents Chemother, 2000; 44: 2811-2815.
45. **Sterling, TR., Chaisson, RE.:** General clinical manifestation of human immunodeficiency virus infection (including the acute retroviral syndrome and oral, cutaneous, renal, ocular, and cardiac diseases). In: Mandell, GL., Bennett, JE., Dolin, R.: Mandell, Douglas and Bennett's principles and practice of infectious diseases., 6th ed., Philadelphia, Elsevier Churchill Livingstone, 2005, s. 1546–1566.

ORIGINAL ARTICLE

Clinical Findings in a Family with Aniridia due to PAX6 Mutation

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SUMMARY

Background: inborn isolated aniridia is rare bilateral impairment of several eye structures manifesting mainly by absence of iris, photophobia and decreased visual acuity. There are also others ocular symptoms associated with aniridia such as nystagmus, strabismus, eyelid ptosis, amblyopia, serious refractive errors, anisometropia, corneal changes, impairment of the lens, chamber angle dysgenesis, optic nerve and macular hypoplasia and congenital or secondary glaucoma. The most frequent aetiology of this eye dysgenesis is mutation in PAX6. Aim of this report is to describe ocular findings in the family with familial aniridia (MIM #106210), to debate their severity, prognosis and therapy options.
Material and methods: assessment of previous medical history and actual ophthalmological findings in 4 persons of 3 generation family with aniridia. According to the compliance, the patients underwent these tests: assessment of the visual acuity, intraocular pressure, refraction test, slit-lamp examination and biomicroscopy, pachymetry test and OCT examination. The genetic counselling was performed with subsequent PAX6 mutation analysis.
Results: all of the examined aniridia family members showed severe symptoms of the disease, the aniridia and photophobia were present. Positive age related correlation showed progressive visual acuity decrease to the practical blindness due to aniridia-associated keratopathy, secondary glaucoma and cataract. DNA analysis revealed presence of p.Gln180X PAX6 mutation in all of the affected persons. The mutation leads to shortened and therefore non-functional protein.
Conclusions: PAX6 mutations leading to premature termination of protein translation are frequently associated with severe symptoms of aniridia and small intrafamilial variability of ocular impairment. This fact is also well demonstrated in members of family described by this report, the symptoms are severe and progressing with age. Therapy is difficult and often with partial success, such in case of secondary glaucoma in young girl from this family. Any eye surgery must be individually judged due to risk of several post-operative complications. And more, the poor vision in aniridia patients is progressively worsening in time to practical blindness.

Key words: aniridia, PAX6, macular hypoplasia, glaucoma

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INTRODUCTION

Isolated congenital aniridia is a rare pathology, the estimated incidence is approximately 1/65000 – 1/96000 (11). Two thirds of cases of aniridia occur familiarly, one third occur sporadically (19). Isolated congenital aniridia represents a panocular anomaly with bilateral occurrence, in which complete or partial absence of the iris and further anomalies predominate as a consequence of defective development of the chamber angle, cornea, lens, retina and optic nerve. Patients have reduced vision, photophobia, with frequent occurrence also of nystagmus and strabismus (25).
Aniridia is the result of abnormal interaction of the neuroectoderm in the region of the prosencophalon and adjacent ectoderm, as a consequence of mutations in PAX6. The protein PAX6 belongs to the family of transcriptional regulators, and has an important role in the development of the eye, but may also influence the development of the brain and pancreas. Its expression takes place in the neurorectoderm of the prosencophalon at the time of induction of the optic disc. After its division into two parts, with the formation of ocular capsules, the expression of PAX6 is restricted to the anterior edge of these capsules. PAX6 then indicates the formation of a placoid lens in the ectoderm adjacent to the ocular capsule. The continuing expression of PAX6 acting together with other transcription factors has an important role also in the development of the lens (29). The expression of PAX6 is also present in the cornea, the conjunctiva, ciliary epithelium and retina, and continues also postnatally (20).
Isolated aniridia as a consequence of the affliction of PAX6 shows autosomally dominant heredity, the risk of incidence of this pathology for descendants of an affected person is theoretically 50%. PAX6 is located on chromosome no. 11, within the region of 11p13. Various types of mutations may occur in this gene, as a consequence of which a formation of a shorter protein or a protein with a different sequence of amino acids takes place. These lead to an impairment of the function or formation of the PAX6 pro-

tein (7). The defective product of the gene leads to an impairment of the morphogenesis especially of the iris. People with congenital aniridia have only one functional PAX6 allele, the second allele is (partially or completely) disabled in its function by the mutation – this therefore concerns a heterozygote constitution. On the basis of case reports describing families in which aniridia was present in both parents, it is assumed that the presence of 2 mutations in PAX6 (homozygote, compound heterozygote) is not viable (12). Penetration of the pathology is full, expression however is variable, and the consequence of this is variable clinical manifestations, sometimes even within the same family. Aniridia (total or partial) occurs in patients, in addition to further connected ocular symptoms, or only other affiliated ocular abnormalities. We differentiate total aniridia from partial aniridia. Even if minimally, the rudiments of iris tissue are gonioscopically mostly perceptible even in the case of total aniridia.

In the case of total aniridia, a wide pupil is formed, reaching all the way to the edge of the cornea, as a result of which the entire equator of the lens is visible, as is sometimes also the suspension apparatus with the ciliary projections. Pathologically-anatomically we mostly find rudimentary tissue of the iris, missing muscles, the pigment epithelium is ectropic and the ciliary projections are generally shorter (21, 25).

The incidence of glaucoma in the case of aniridia is approximately 6-75%. It usually appears at pre-adolescent or early adolescent age (19). Glaucoma develops as a consequence of abnormalities in the drainage pathways of the chamber angle, which prevent the drainage of the chamber fluid via the Schlemm's canal. Margo et al. (15) described abnormalities of the irido--corneal angle in patients with aniridia who also had presence of glaucoma, in which the peripheral residues of the iris in the majority of these patients overlapped the trabecular meshwork. In the case of congenital glaucoma, the drainage pathways are obturated by a membrane containing capillaries originating from the rudiments of the iris. In secondary glaucoma the drainage pathways are blocked by the residual iris tissue. The Schlemm's canal may be missing in both types of glaucomas (4, 8, 25).

In patients with aniridia, the cornea measures 100 μm more than the average thickness of the cornea (3). A typical manifestation is the presence of secondary corneal changes at an early age (the first changes appear in the first decade of life). PAX6 mutation results in changes in the corneal expression of cytokeratin, cellular adhesion and the expression of glycoconjugate. This, together with a deficit of limbal stem cells, contributes to the fragility of the cornea and to aniridia associated keratopathy (AAK). The incidence of keratopathy is stated at approximately 20% (4, 27). In aniridia the production of tears is normal, but tears lack normal viscosity and easier rupture of the lacrimal film occurs. The protective value of the lacrimal meniscus is reduced, thus opening a pathway for irritation processes on the surface of the eye (4, 11, 14, 25).

There are sometimes residues of the vascular tunic on the lens (tunica vasculosa lentis), as well as a persistent pupillary membrane and isolated coloboma of the lens, and ectropia of the lens, subluxation and dislocation of the lens occurs in a low percentage of cases. Present opacities in the lens may occur congenitally. Cataracts develop in more than one half of patients (50-85%) (19). This may typically begin in childhood or afflict young adults. The impaired function of the retina may be contributed to by its defective development during embryogenesis and subsequently during life by a phototoxic effect. A typical manifestation is hypoplasia of the macula, which is histologically characterised by a substitution of the structure of the fovea by a continuous layer of ganglion cells. Foveal hypoplasia is manifested in reduced foveal reflex, macular hyperpigmentation, abnormal vascular remodelling and reduction of the avascular zone. OCT findings identify central foveal thickening and a smaller macular volume, and the foveal depression is generally not preserved (11, 13, 17, 22, 26, 31).

Nystagmus is not a universal finding, but occurs in approximately 85-92% of patients with aniridia. The cause of its occurrence is the hypoplasia of the macula itself, with congenitally caused reduction in vision (25).

MATERIAL AND METHOD

4 persons from one family were examined with the incidence of aniridia in three generations (fig. 1). This concerned the following persons: brother and sister, their mother and the mother's father. The phenotype of the pathology is severely expressed in this family. These persons underwent ophthalmological and genetic examinations.

Within the framework of the genetic examination, the family was provided

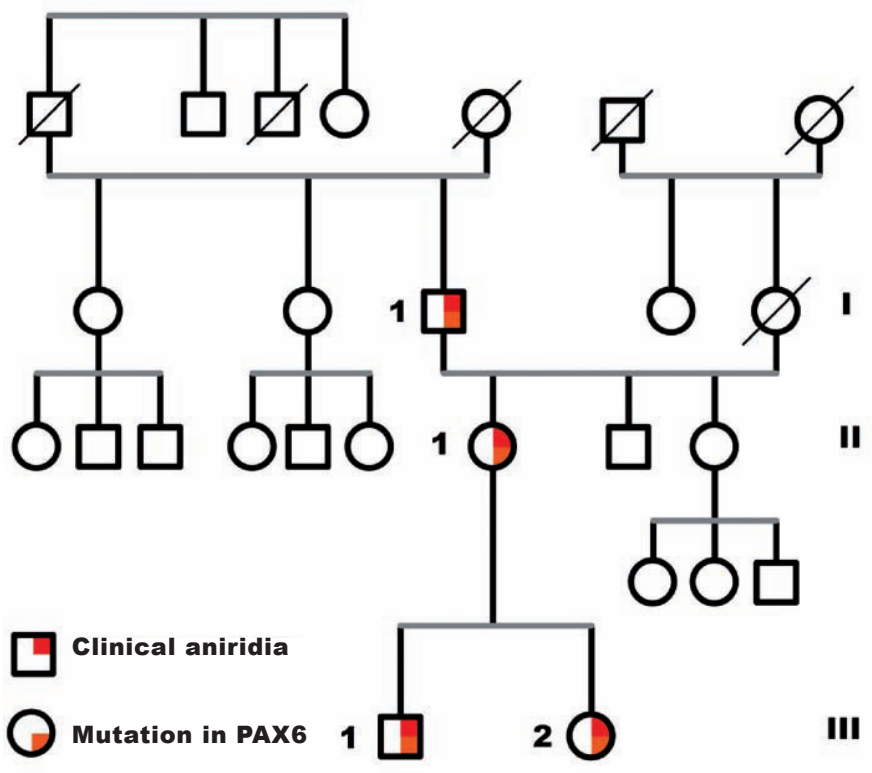

Fig. 1 Genealogy of family with isolated congenital aniridia and PAX6 mutation.

with a genetic consultation, and subsequently also a DNA analysis on the basis of informed consent – an analysis of the presence of mutation in PAX6 with the help of direct sequencing.
The usual examination procedure was used in the examination by the ophthalmologist. First of all visual acuity was determined without correction, then with correction, and measurement of refraction was performed using an auto refractometer. Subsequently measurement of intraocular tension was conducted on the patients using a slit lamp. Pachymetry was conducted on the younger female patient with congenital aniridia. It was possible to obtain photo documentation of the fundus for both the youngest patients with a transparent cornea. Examination by OCT succeeded with difficulties only in the youngest female patient (sister), within the framework of an attempt to examine the papillas of the optic nerve and the macula.

RESULTS

DNA analysis detected the presence of a mutation c. 538C>T/p.Gln180X in PAX6, located in exon no. 8. This mutation was found in all 4 persons with the incidence of congenital aniridia. Extraocular affliction of the individuals in this family was not anamnestically recorded.
The seventy five year old man (patient I-1, fig. 1) is continuously medicated for congenital total aniridia, slight horizontal nystagmus, secondary glaucoma and fully developed AAK with vascular leukoma (fig. 2). Upon examination by slit lamp it is possible only to guess at rudiments of iris tissue due to the non-transparent cornea. According to the anamnesis and documentation, the patient is aphakic. Vision in the right eye with aphakic correction and without correction is 0.5 m fingers in the right eye, in the left eye the patient is blind. It was not possible to assess further symptomatology in the patient either objectively or according to the documentation.
Significant convergent strabismus is present in the thirty five year old mother (patient II-1, fig. 1) with congenital bilateral aniridia with photophobia, and also with horizontal nystagmus and a higher degree of AAK (fig. 3). The cornea has had reduced transparency and vascularisation for several years, and as such does not enable examination of the intraocular structure and the ocular fundus. Visual acuity in the right and left eye is stable at 0.03 and 0.02. It was not possible to measure refraction and noncontact tonometry. Intraocular tension according to Schiötz was 3.5 / 5.5 and 3.5 / 5.5. According to the older documentation and according to the patient's own glasses correction, a myopic refractive error was determined in the patient (-3.75dsf. bilaterally, no data about refraction was found in the older available documentation). According to the documentation, incipient turbidity of the lens appeared in the patient at the age of sixteen, at the age of seventeen local antiglaucomatous therapy was commenced with beta blockers for secondary glaucoma. The last mention of the ocular fundus is in the older documentation from 1998 (when the patient was twenty years old), when a white disc of the optic nerve was found. Further evaluation of the ocular fundus was not possible at this time.
The eight year old boy (patient III-1, fig. 1) had a diagnosis of congenital total bilateral aniridia with photophobia immediately after birth. The presence of severe myopia with myopic astigmatism (refraction of right eye -5.25 D s. - 1.75 D cyl. ax. 22°, refraction of left eye -7-0 D sf. -2.0 D cyl. ax. 173°) was determined also in this patient, as well as objectively slight convergent strabismus. Visual acuity with correction is bilaterally 0.15. Upon examination by slit lamp, the absence of an iris was determined, and a fine vascular sketch was present on the cornea perilimbally, 1 capillary in the right eye permeated through the limbus in the direction toward the centre (fig. 4). Using the slit lamp, isolated opacities were also found in the lens of the right eye, a zonular cataract was indicated in the left eye. Papilla with central minor excavation is present on the ocular fundus bilaterally – without perceptible glaucoma changes, a deficiency of a foveal reflex and thinning of the pigment epithelium of the retina (RPE) mainly in the periphery (fig. 6). Ocular tension is stable – around 15 Torrs (it was not possible to perform gonioscopy, OCT, pachymetry due to poor co-operation and concentration together with poor fixation).
The seven year old girl (patient III-2, fig. 1) was born from the mother's 2nd physiological pregnancy, birth within the term per S.C. from the ocular indication of the mother. An ocular defect was determined immediately after birth and during infant age the patient was examined under general anaesthesia. At the time of measurement total bilateral congenital aniridia was present, with photophobia, refractive error (myopia gravis with myopic astigmatism) and incipient posterior polar cataract more in the right eye (fig. 5). Vision in the right eye was 0.1 bilaterally with and without correction, refractive error in right eye: -7.25 D sf. - 1.5 D cyl. ax. 52° and in left eye: -9.0 D sf. -2.0 D cyl. ax. 125°. Examination by slit lamp demonstrates incipient perilimbal greying on the cornea and mild vascularisation of the periphery of the cornea within the framework of AAK.
According to the documentation, at the age of six years secondary glaucoma was diagnosed in the patient and local monotherapy with beta blockers was commenced. Due to unsatisfactory intraocular tension, a fixed dual combination was selected after six months (brinzolamide with timolol). With regard to the thickness of the patient's corneas (678-680 µm according to pachymetry), intraocular tension is within the range of 20-25 torrs. OCT scans of the papillas, obtained with difficulty (poor fixation of patient), detected smaller papillas (1.23 mm2 in right eye and 1.17 mm2 in left eye), values of the neuroretinal rim and cup/disc ratio testify to advanced glaucoma changes with reduced RNFL. With regard to poor fixation and the quality of the scans, the validity of the examination itself was poorer. According to the OCT scans of the macula, foveal thickening was determined at 279 µm in the right eye and 233 µm in the left eye, macular volume 6.56 mm3 in the right eye and 11.08 mm3 in the left eye. Upon biomicroscopic examination there is a bilateral finding of excavation 0.8-0.9 with residual neuroretinal rim especially in the left eye, the macula is without foveal reflex and the retina is thinned mainly in the periphery with showing sketch of choroid (fig. 7, see photo of fundus). Gonioscopic examination was not performed due to photophobia and difficult co-operation.

DISCUSSION

Mutations in PAX6 are responsible for the incidence of isolated aniridia, in isolated cases this may concern a mutation in the ultra-conserved region of the gene ELP4, 150 kb distally from PAX6 (2). At present over 286 PAX6 mutations are known, in which

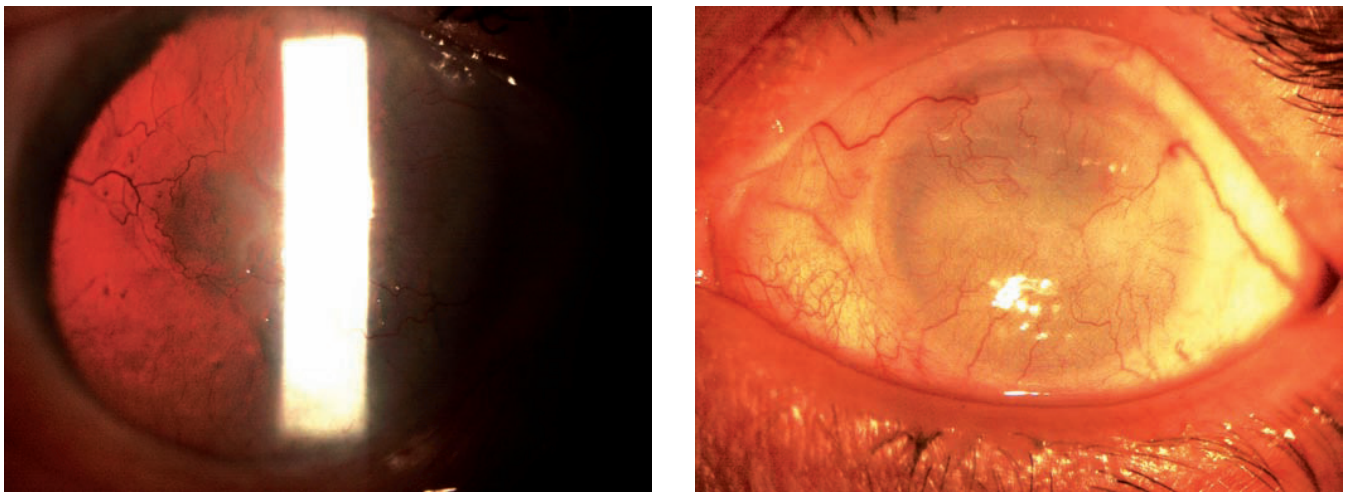

Fig. 2 Patient I-1: right eye (A) and left (B), aniridia, high degree of AAK.

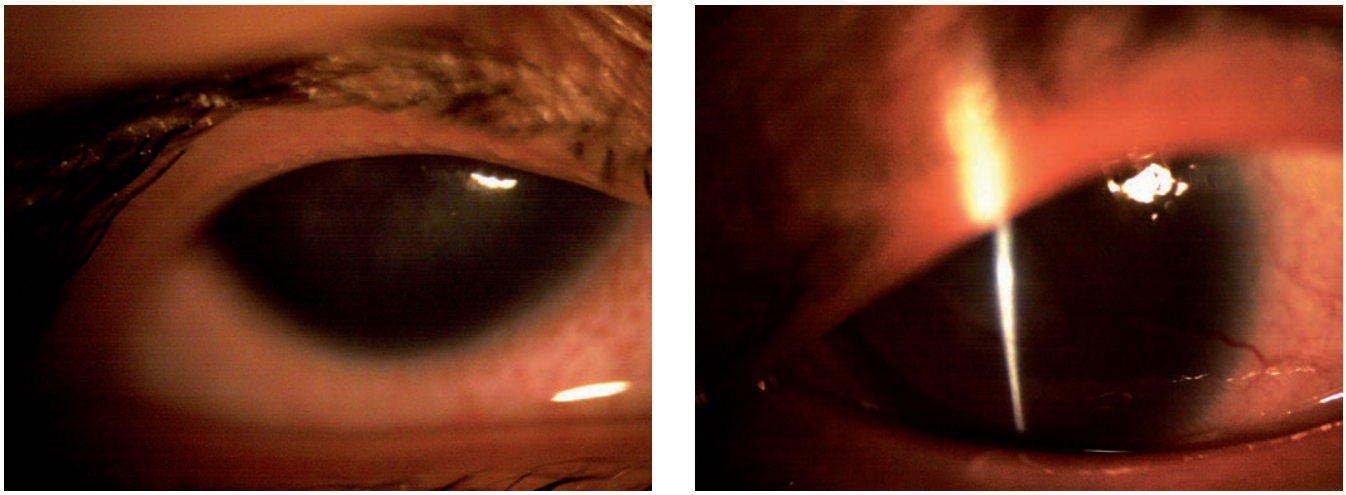

Fig. 3 Patient II-1: right eye (A) and left eye (B), aniridia, higher degree of AAK.

Fig. 4 Patient III-1: right eye (A) and left eye (B), aniridia, visible equator of lens, minimal corneal changes.

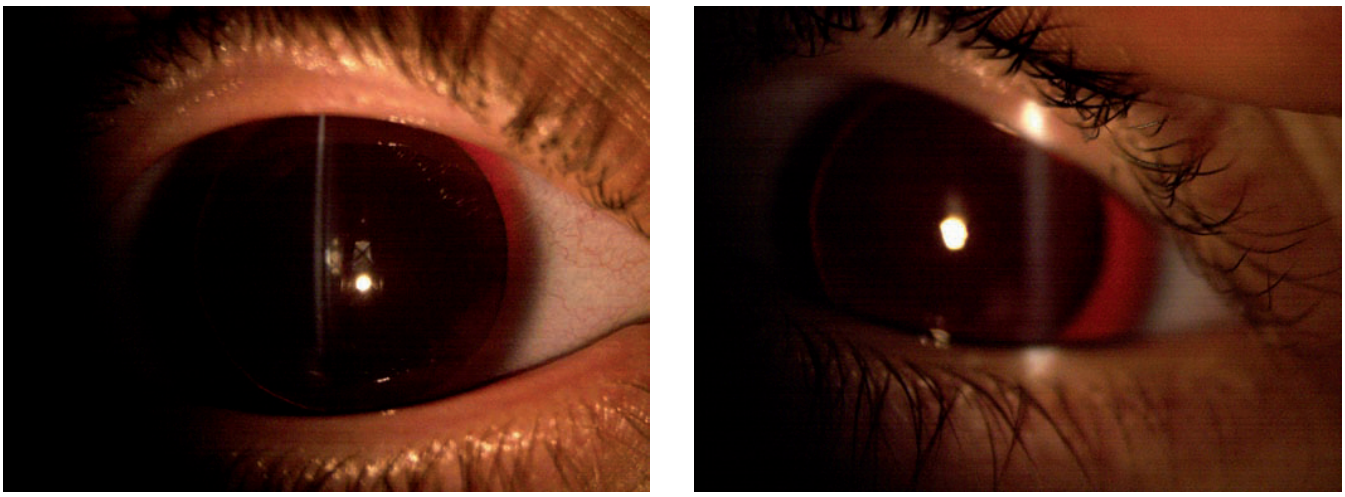

Fig. 5 Patient III-2: right eye (A) and left eye (B), identified aniridia, visible equator of lens, minimal corneal changes, identified cataract in right eye

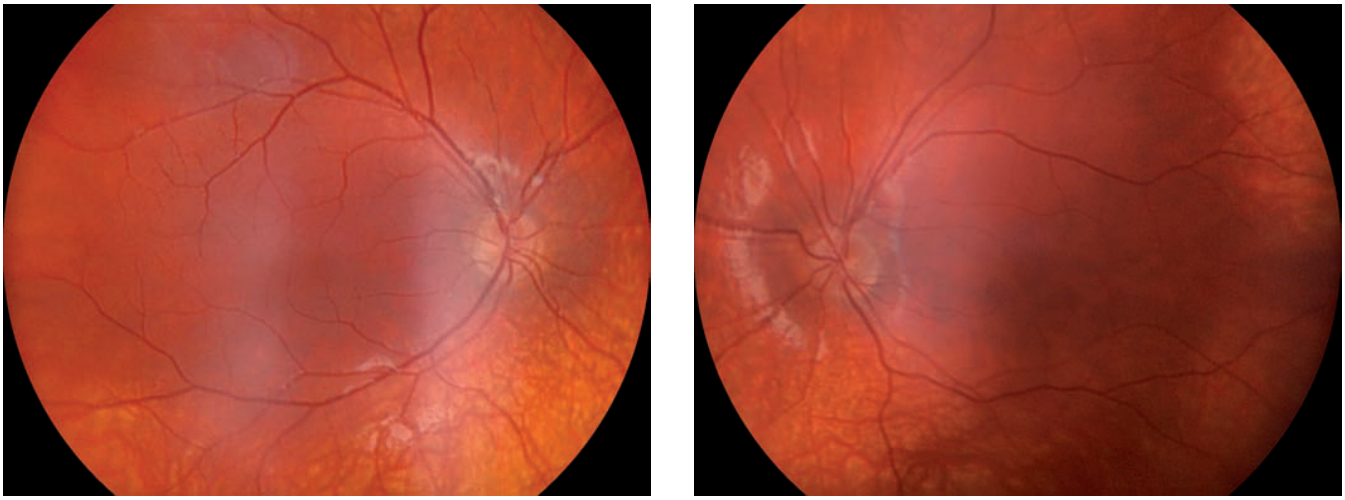

Fig. 6 Ocular fundus in patient III-1: right eye (A) and left eye (B); perceptible thinning of RPE (retinal pigment epithelium) in periphery.

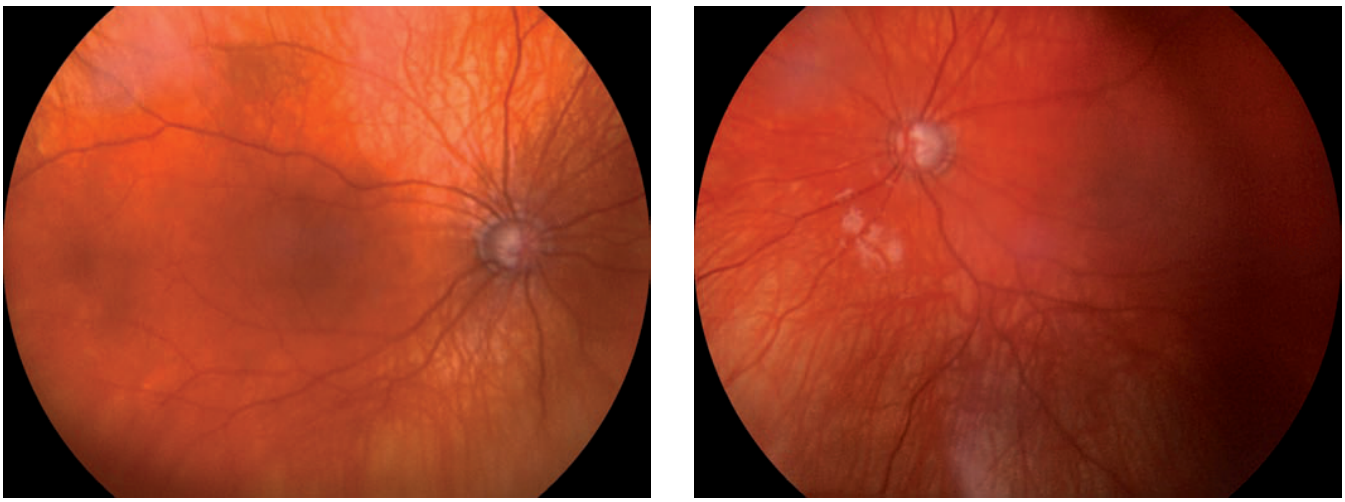

Fig. 7 Ocular fundus in patient III-2: right eye (A) and left eye (B), glaucoma changes of papilla n.II, thinning of RPE.

approx. 90% of these were linked to the incidence of isolated aniridia or connected phenotypes. Mutations may occur within the scope of the entire PAX6, the most frequently occurring are "non-sense" mutations, as a consequence of which premature termination of protein formation takes place. More than one half of these non-sense mutations occur in exons 8-11. In total almost 90% of PAX6 mutations leading to isolated aniridia are of the character of non-sense or alter the sensory framework (28).

Non-sense mutations are very often the cause of severe affliction with lower variability of the phenotype. In this family mutation c.538C>T was found in exon no. 8, as a result of which glycine in position 180 is recorded as a Stop codon, therefore protein formation is terminated immediately. This mutation has been described only once previously in a family with the incidence of aniridia (16). In the family we describe, this is linked to a severe phenotype impact, with imperceptible variability of symptoms, in accordance with the character of non-sense mutations in PAX6.

In many families with aniridia, visual acuity is less than 20/60 (0.33) and lower than 20/200 (0.1) in more than 60% of cases within the framework of congenital foveal hypoplasia or secondary glaucoma (25). The refractive errors found in aniridia include myopia, hypermetropia, astigmatism, and frequently also anisometropia. Haploinsufficiency as a consequence of heterozygote mutation of PAX6 is frequently linked to a high refractive error, especially short-sightedness (10). In the members of this family with total aniridia described in this study, visual acuity is within the range from blindness to best vision with correction of 0.2. There is a prognosis of deterioration of visual acuity especially in patients from families with severely expressed symptomatology (phenotype expression) of the pathology.

Keratopathy associated with aniridia is identified in the individual stages of the pathology according to the age of patients in all of the persons we examined with aniridia. In this family, AAK occurs at an earlier age, and the course of the pathology with progressive deterioration of vision takes place more rapidly.

OCT of the papillas and maculas in the youngest patient testified to glaucoma affliction of the papillas. foveal thickening of 279 μm in the right eye and 233 μm in the left eye was also determined, macular volume was 6.56 mm3 in the right eye and 11.08 mm3 in the left eye. Holström et al. (13) tested the suitability of OCT examination in the diagnosis of foveal hypoplasia in children with albinism and with aniridia (as well as specially selected and obtained scans in the case of difficult fixation and nystagmus). In these patients they demonstrated central foveal thickening by means of OCT (expressed by median foveal minimum of 259 μm) and smaller macular volume (median macular volume 7.00-7.01 mm3) as against a control group of children (median of foveal minimum: median 167 μm, median macular volume 7.10 mm3). They stated that the confirmation of foveal hypoplasia could be of assistance in the diagnosis of isolated congenital aniridia in patients with a mildly expressed phenotype. In a comparison of the only usable OCT finding (the youngest patient of the family we describe) with the above study, central foveal thickening is comparable, in which macular volume is even smaller.

With regard to the fact that pathological changes of the cornea frequently occur, the application of local therapy using drops without preservative agents is important. Even any surgical procedure brings with it the risk of "fibrous syndrome" in aniridia, and worsening of keratopathy. Aniridic fibrosis syndrome is characterised by the occurrence of extensive progressive retrolenticular and retrocorneal fibrous membranes. In decision making and choice of therapeutic procedure, it is therefore necessary to keep in mind all the risks, in order to attain the best vision for the longest possible time. Wearing special contact lenses may also worsen the finding on the cornea. In the case of advanced changes on the cornea, the selection of keratoplasty in order to improve vision is a question for discussion from the long-term perspective, with regard to the potential recurrence of the finding. It is also necessary to take into consideration the incidence of more poorly compensated glaucoma and thus a progressively worsening visual field and vision. A moment of surprise may be the finding on the optic nerve and the retina upon examination of the ocular fundus following renewal of transparency of the optic media after successful keratoplasty or cataract surgery.

In the thirty five year old mother there is a difficult balance in the selection of the procedure to renew transparency of the cornea. This patient had previously be diagnosed with secondary glaucoma, but the ocular fundus has not been examined since the age of 20 years. It is merely a question of what visual acuity we shall attain by the procedure, and for how long. AAK itself also often worsens following surgical procedures on the limbus or following the application of local antimetabolites in the treatment of aniridia-associated glaucoma (19). As a result, from a long-term perspective the benefit of keratoplasty or transplantation of limbal cells is uncertain for this patient.

CONCLUSION

Aniridia represents a severe panocular pathology. Seventy percent of diagnosed individuals with isolated aniridia have afflicted parents. In isolated familial aniridia there is a 50% risk of the occurrence of this pathology in the children of an afflicted parent. In exceptional cases, non-ocular sensory manifestations may also appear in these patients, as well as neurological abnormalities or affliction of the pancreas. A reduction of the sense of smell has been described in patients with isolated aniridia. Behavioural disorders and retarded development are rare occurrences (1, 5, 6, 11, 18, 24, 30). With regard to the potential more pronounced variability of the phenotype, genetic examination is important, and its importance is multiplied especially at early childhood age, when it helps to differentiate children with WAGR (Wilms' tumour, aniridia, genital abnormalities, mental retardation) syndrome, manifested so far only through aniridia, from patients with mutation in PAX6. This concerns a progressing pathology, therapy is frequently arduous especially in patients with severely expressed symptomatology. In these patients it is necessary to consider carefully the selection of ocular procedures with regard to the increased risk of subsequent complications. Even if it is stated that symptoms in aniridia are irreversible and progress with advancing age, studies under way at present can nevertheless provide certain hope for those afflicted. Aniridia is linked to insufficient production of PAX6 (as a consequence of haploinsufficiency of PAX6), and as a result Gregory-Evans et al. (9) attempted to influence the dose of PAX6 postnatally in a model in mice. Specific topical administration of ataluren and gentamicin not only halted the progression of the pathology as a consequence of PAX6 mutation, but to a certain degree even reversed certain changes on the cornea, lens and retina.

LITERATURE

1. **Bamiou, D.E., Free, S.L., Sisodiya, S.M.:** Auditory interhemispheric transfer deficits, hearing difficulties, and brain magnetic resonance imaging abnormalities in children with congenital aniridia due to PAX6 mutations. Arch Pediatr Adolesc Med, 2007; 161(5): 463–469.
2. **Bhatia, S., Bengani, H., Fish, M. et al.:** Disruption of autoregulatory feedback by a mutation in a remote, ultraconserved PAX6 enhancer causes aniridia. Am J Hum Genet, 93: 1126-1134, 2013.
3. **Brandt, J.D., Casuso, L.A., Budenz, D.L.:** Markedly increased central corneal thickeness: an uncrecognized finding in congenital aniridia. AM J. Ophthalmol, 2004; 137(2): 348–350.
4. **Socialstyrelsen.** Congenital aniridia. [online]. 2012-06-19 [citováno 2014-05-15]. Dostupné z: < http://www.socialstyrelsen.se/rarediseases/congenitalaniridia>.
5. **Ellison-Wright, Z., Heyman, I., Frampton, I. et al.:** Heterozygous PAX6 mutation, adult brain structure and fronto-striatothalamic function in a human family. Eur J Neurosci, 2004; 19(6): 1505–1512.
6. **Free, S.L., Mitchell, T.N., Williamson, K.A. et al.:** Quantitative MR image analysis in subjects with defects in the PAX6 gene. Neuroimage, 2003; 20(5): 2281–2290.
7. **Glaser, T., Jepeal, L., Edwards, J.G. et al.:** PAX6 gene dosage effect in a family with congenital cataracts, aniridia, anophthalmia and central nervous system defects. Nat Genet, 1994; 7(4): 463–471.
8. **Grant, W.M. and Walton, D.S.:** Progressive changes in the angle in congenital aniridia, with development of glaucoma. Trans Am Ophthalmol Soc, 1974; 72: 207–228.
9. **Gregory-Evans, C.Y., Wang, X., Wasan, K.M.:** Postnatal manipulation of Pax6 dosage reverses congenital tissue malformation defects. J Clin Invest, 2014; 124(1): 111–116.
10. **Hewitt, A.W., Kearns, L.S., Jamieson, R.V. et al.:** PAX6 mutations may be associated with high myopia. Ophthalmic Genet, 2007; 28(3): 179–182.
11. **Hingorani, M., Hanson, I., van Heyningen V.:** Aniridia. Eur J Hum Genet, 2012; 20(10): 1011–1017.
12. **Hodgson, S.V, Saunders, K.E.:** A probable case of the homozygous condition of the aniridia gene. J Med Genet, 1980; 17(6): 478–480.
13. **Holmström, G., Eriksson, U., Hellgren, K.:** Optical coherence tomography is helpful in the diagnosis od foveal hypoplasia. Acta Ophthalmol, 2010; 88(4): 439–442.
14. **Jastaneiah, S., Al-Rajhi, A.A.:** Association of aniridia and dry eyes. Ophthalmology, 2005; 112(9): 1535–1540.
15. **Margo, C.E.:** Congenital aniridia: a histopathologic study of the anterior segment in children. J Pediatr Ophthalmol Strabismus, 1983; 20(5): 192–198.
16. **Martha, A., Strong, L.C., Ferrell, R.E. et al.:** Three novel aniridia mutations in the human PAX6 gene. Hum Mutat, 1995; 6(1): 44–49.
17. **McGuire, D., Weinreb, R., Goldbaum, M.:** Foveal hypoplasia demonstrated in vivo with optical coherence tomography. Am J Ophthal, 2003; 135(1): 112–114.
18. **Mitchell, T.N., Free, S.L., Williamson, K.A. et al.:** Polymicrogyria and absence of pineal gland due to PAX6 mutation. Ann Neurol, 2003; 53: 658–663.
19. **Nelson, L.B., Spaeth, G.L., Nowinski, T.S. et al.:** Aniridia. A review. Surv Ophthalmol, 1984; 28(6): 621–642.
20. **Nishina, S., Kohsaka, S., Yamaguchi, Y. et al.:** PAX6 expression in the developing human eye. Br J Ophthalmol, 1999; 83(6): 723–727.
21. **Rush, C.C.:** Congenital Aniridia. Trans Am Ophthalmol Soc. 1926;24:332-341.
22. **Seefelder, R.:** Die aniridie als eine entwicklungshemmung der retina. Albrecht Von Graefes Arch Klin Exp Ophthalmol, 1909; 70(1): 65–87.
23. **Shaw M.W., Falls H.F., Neel J.V.:** Congenital Aniridia. Am J Hum Genet, 1960; 12(4): 389–415.
24. **Sisodiya, S.M., Free, S.L., Williamson, K.A. et al.:** PAX6 haploinsufficiency causes cerebral malformation and olfactory dysfunction in humans. Nat Genet, 2001; 28(3): 214–216.
25. **Traboulsi, E.I.:** Genetic diseases of the eye. 2nd ed. (Oxford Monographs on Medical Genetics). China: Oxford University Press Inc., 2012. 923 s. ISBN 978-0195326147.
26. **Tremblay, F., Gupta, S.K., De Becker, I. et al.:** Effects of PAX6 mutations on retinal function: an electroretinographic study. Am J Ophthalmol, 1998; 126(2): 211–218.
27. **Tseng, S.C., Prabhasawat, P., Barton, K. et al:** Amniotic membrane transplantation with or without limbal allografts for corneal surface reconstruction in patients with limbal stem cell deficiency. Arch Ophthalmol, 1998; 116(4): 431-441.
28. **Tzoulaki, I., White, I.M., Hanson, I.M.:** PAX6 mutations: genotype-phenotype correlations. BMC Genet, 2005; 6: 27.
29. **Vacek, Z.:** Embryologie.1. vyd. Praha: Grada Publishing, 2006. 256 s. ISBN 978-80-247-1267-3. Kapitola 8.8, Vývoj smyslových orgánů, s. 231–238.
30. **Valenzuela, A., Cline, R.A.:** Ocular and nonocular findings in patients with aniridia. Can J Ophthalmol, 2004; 39(6): 632–638.
31. **Wu, L., Ma, Q., Chen, Y. et al.:** Abnormalities of ERG in congenital aniridia. Yan Ke Xue Bao, 1991; 7(3): 151–152.

ORIGINAL ARTICLE

Supracor, Laser Correction of Presbyopia: One-year Follow-up Outcomes

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SUMMARY

Purpose: Evaluation of one-year postoperative results after Supracor laser procedure.
Methods: The study group consisted of 8 hyperopic patients (16 eyes) seeking alternatives to wearing glasses for both distance and near vision. These patients underwent Supracor refractive surgery in the Department of Ophthalmology of The University Hospital Brno in the time period from July 2012 to February 2013. The patient mean age at operation was 57,2 years (±4,6), mean binocular uncorrected near visual acuity was Jaeger No. 13, distance visual acuity 0,5 (±0,19). Average follow-up period was 15,5 months (±2,4).We evaluated binocular uncorrected and best corrected distance visual acuity (UDVA, BCDVA), near visual acuity (UNVA, BCNVA), and intermediate visual acuity (UIVA, BCIVA) 12 months after surgery, further the stability of visual acuity, objective refraction, mesopic contrast sensitivity, occurrence of complications, and patient satisfaction.
Results: In all 8 patients, binocular UDVA of at least 1,0 was achieved. The mean spherical equivalent was +0,25 D (±0,64). UNVA was Jaeger No. 3 or better in 7 patients (68,7 %), 5 of them could read Jaeger No. 1. UIVA Jaeger No. 1 was achieved in 5 patients (62,5 %), Jaeger No. 2 in 2 patients (25 %), and Jaeger No. 4 in 1 patient (12,5 %). In all eyes, preoperative mesopic contrast sensitivity was within the normal range for the given age. Postoperatively it remained within the normal range in 11 eyes (68,7 %). In 5 eyes (31,3 %) we found a decrease below the lower limit in higher spatial frequencies (12 and 18 cycles/degree) during the entire follow-up period. According to the patient questionnaire, 7 patients (87,5 %) were fully satisfied with the outcome of the surgery and they felt independent of wearing glasses, 1 patient was dissatisfied. 7 patients (87,5 %) did not report the presence of photic phenomena (halo, glare), 1 patient suffered from these problems.We did not encounter any intraoperative or postoperative complications.
Conlusion: According to our first experience, good distance, near, and intermediate visual acuity and a rather high patient satisfaction can be achieved with the use of the Supracor procedure. Supracor seems to be a suitable method of presbyopia correction in motivated, adaptable patients who meet strict indication criteria. With regard to the small number of patients in our study group, a greater number of patient evaluations will be required in the future, and long-term results will be of interest as well.

Key words: Supracor, presbyopia, LASIK, refractive surgery

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INTRODUCTION

Presbyopia – feeble sightedness – is a physiological, age-related process, during which there is a progressive loss of accommodation and as such the pathology afflicts the predominant majority of the population (8, 14, 15). Accommodation represents a dynamic process, which is difficult to influence surgically (12).
At present patient demands are increasing for vision, and there is a growing number of presbyopic patients who are not satisfied with glasses correction or contact lenses. The possibilities of surgical correction are diverse, each has its advantages but also drawbacks, and may not always provide patients with a satisfactory solution to the problem. With the development of refractive surgery, new possibilities for resolving presbyopia are emerging. Nevertheless, this issue remains a challenge for ophthalmologists (16).
Surgical methods for solving presbyopia incorporate firstly intraocular procedures – refractive replacement of the lens with implantation of multifocal lenses, accommodative lenses as monovision and also scleral surgery (scleral implants). The second group is represented by corneal procedures – the use of corneal implants and for completeness conductive keratoplasty. A further group is laser procedures using monovision techniques (PRK – photoreactive keratectomy, LASIK – laser in situ keratomileusis), bifocal corneal profile (PresbyLASIK), multifocal corneal profile (Supracor) or

intrastromal effect of a femtosecond laser (Intracor) (5, 13).
Supracor (Technolas Perfect Vision) represents a relatively new method for the solution of presbyopia, approved in May 2011 (3). It concerns a bilateral procedure. The principle is first of all the creation of a corneal lamella as with the LASIK method, with the help of a microkeratome or femtosecond laser, and subsequently the creation of a multifocal profile of the cornea with progressive transition between the zone for near vision and to distance vision with the help of an excimer laser (9, 11, 12).

MATERIAL AND METHOD

The study cohort included 8 presbyopic hypermetropic patients (16 eyes), comprising 5 men and 3 women. The patients were seeking an alternative to wearing glasses or contact lenses, and stated a preference for a less invasive procedure above an intraocular operation. They underwent a Supracor laser procedure at the Department of Ophthalmology at the University Hospital Brno in the period from July 2012 to February 2013.
The average age in the study cohort at the time of surgery was 57.2 years (±4.6).
A complete examination was conducted preoperatively before the refractive procedure. The examination incorporated inter alia simulation of postoperative distance vision. An addition of +0.5 Dsf was placed on the non-dominant eye of the patients for full correction. Such a generated reduction of visual acuity should be subjectively tolerated by the patients according to the recommended procedures. All the patients met the relatively narrow indication criteria (table 1).
Patients engaged in professions with high demands for distance vision, vision at night or in sharp light (truck drivers, watchmakers, dressmakers), as well as perfectionists, patients with unrealistic expectations or with intolerance of multifocal glasses were not indicated for the procedure.
The procedure was performed under local anaesthesia. The corneal lamella was created by a microkeratome Zyoptix XP. The thickness of the corneal lamella was 110-120 µm, the diameter of the lamella was > 9mm. A TECHNOLAS® Excimer Workstation 217P excimer laser was used for the subsequent photoablation. The optical zone with a diameter of 6 mm co-

Table 1 Supracor – indication criteria

Presbyopia and hypermetropia +0.75 to +4.0 D
Addition above 1.75 D
Astigmatism below 2.0 Dcyl
Difference between subjective and cycloplegic refraction up to 0.75 inclusive
Value of keratometry 41 to 45 D
Best corrected distance visual acuity above 0.8
Width of cornea 3 mm to 6 mm
Age above 46 years
Clear optical media
Eye without previous operations (exception is artephakia, condition after LASIK)
Sufficient corneal thickness (above 500 µm)
Tolerance to addition + 0.5 Dsf (as simulation of distance vision after procedure)

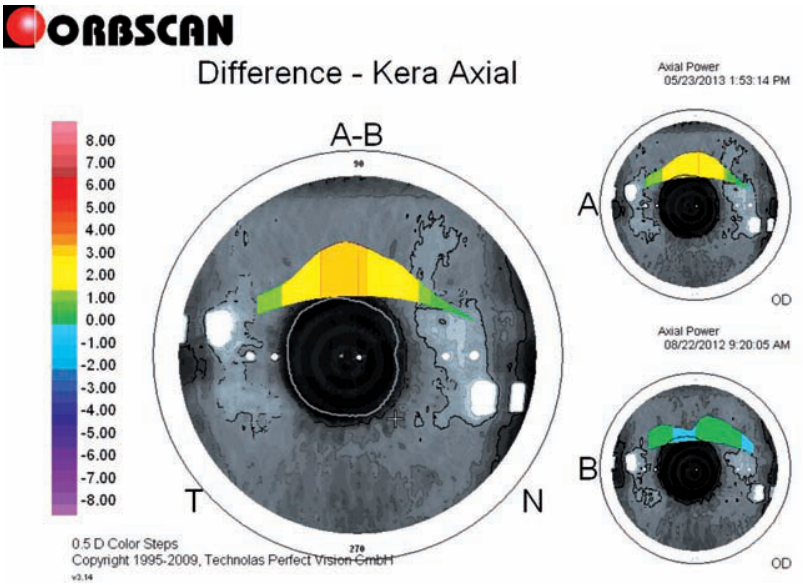


Fig. 1 Elevation in central part of cornea.

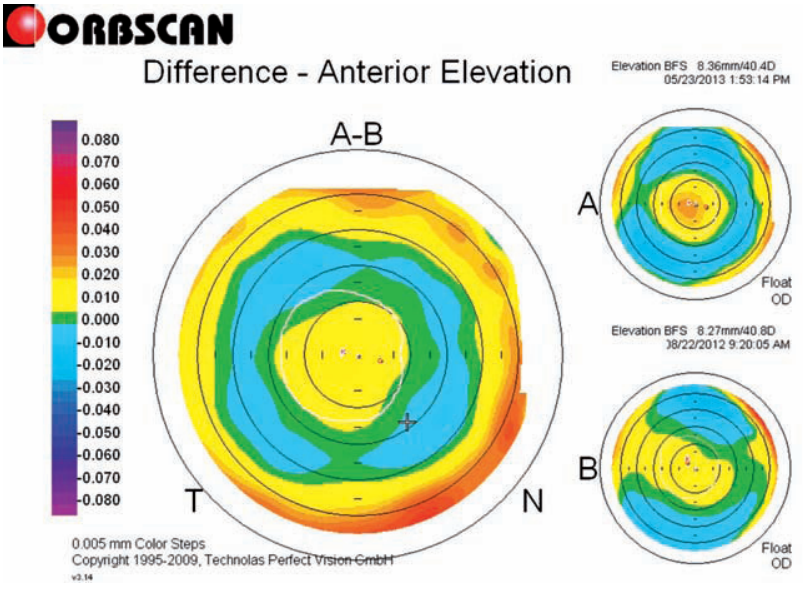


Fig. 2 Orbscan, elevation in central part of cornea.

vered a zone for distance vision with progressive transition in the central three millimetre region for near vision, where elevation and arching were created using 2000 pulses (fig. 1 and 2) (7). The procedure was performed always simultaneously at our clinic in "bilateral asymmetrical regime": the non-dominant eye was targeted to the value of -0.5 Dsf, the dominant eye to emmetropia.

Local antibiotics (levoflaxin) were applied postoperatively for a period of 1 week, and from the 2nd postoperative day also local steroids (fluorometholone) for a period of 2-3 with gradual discontinuation.

A control examination was conducted on the 1st day, 1 month, 3, 6 and 12 months after the procedure, and subsequently at intervals of 6 months. The average observation period was 15.5 months (±2.4). We observed non-corrected (UVA) and best corrected visual acuity (BCVA) for distance, near and intermediate vision, as well as the stability of visual acuity, objective refraction, contrast sensitivity under mesopic conditions, the incidence of perioperative and postoperative complications, and patient satisfaction using a questionnaire.

We examined visual acuity using Snellen's optotypes on an LCD optotype NIDEK SC 1600, refraction on an automatic refractometer NIDEK ARK-530, contrast sensitivity under mesopic conditions using sinus bands on a CSV-1000E (VectorVision) chart. The satisfaction questionnaire contained 14 questions, we evaluated overall patient satisfaction with the result of the procedure, perception of glare and the halo phenomenon, and independence of wearing glasses.

The results are presented in the first year following the procedure.

RESULTS

Average binocular UNVA preoperatively reached the value of J. no. 13, average BCNVA J1 with an average addition of 2.16 D (±0.26). Average binocular UDVA preoperatively reached 0.5 (±0.19), average BCDVA 1.0. The average value of spherical equivalent of manifest refraction preoperatively was +2.27 (±0.48).

UDVA binocularly 12 months after the procedure reached minimally 1.0 in all patients, of whom 1 patient had 1.2 and 1 patient 1.5 (table 2, graph 1). No patient suffered a loss of rows of BCVA. The average value of spherical equivalent postoperatively was +0.25 (±0.64).

Table 2 Binocular uncorrected distance visual acuity

Patient	UVA preoperatively	UVA 1 month	UVA 3 months	UVA 6 months	UVA 12 months
1	0.7	1.0	1.0	1.0	1.5
2	0.2	0.9	1.0	1.0	1.0
3	0.8	0.9	0.9	0.9	1.2
4	0.4	1.0	1.0	1.0	1.0
5	0.6	1.0	1.0	1.0	1.0
6	0.3	1.0	1.0	1.0	1.0
7	0.5	1.0	1.0	1.0	1.0
8	0.5	1.0	1.0	1.0	1.0

Table 3 Binocular uncorrected near visual acuity

Patient	UVA preoperatively	UVA 1 month	UVA 3 months	UVA 6 months	UVA 12 months
1	>J. no. 13	J no. 2	J. no. 1	J. no. 2	J. no. 2
2	>J. no. 13	J no. 1	J. no. 3	J. no. 2	J. no. 1
3	>J. no. 13	J. no. 1	J. no. 1	J. no. 2	J. no. 3
4	J. no. 11	J. no. 4	J. no. 4	J. no. 7	J. no. 7
5	>J. no. 13	J. no. 2	J. no. 1	J. no. 1	J. no. 1
6	>J. no. 13	J. no. 1	J. no. 1	J. no. 1	J. no. 1
7	>J. no. 13	J. no. 1	J. no. 1	J. no. 1	J. no. 1
8	>J. no. 13	J. no. 1	J. no. 1	J. no. 1	J. no. 1

In 7 patients (87.5%) UNVA of J. no. 3 and better was attained, in which 5 patients read J. no. 1 (table 3, graph 2). BCNVA binocularly was J. no. 1 in all patients with average addition of +0.59 (±0.64).

UIVA was J. no. 1 in 5 patients (62.5%), J. no. 2 in 2 patients (25%) and J. no. 4 in 1 patient (12.5%) (table 4). BCIVA was J1 in all patients.

We recorded a relatively long fluctuation of visual acuity following the procedure, stabilisation took place in the majority of patients after 6 months, nevertheless discrete changes occurred in 2 patients also between the 6th and 12th month following the operation.

Contrast sensitivity tested under mesopic conditions was preoperatively within the physiological range for the given age group in all 16 eyes, postoperatively it remained within the norm in 11 eyes (68.7%), in 5 eyes (31.3%) we observed a reduction beneath the lower limit of the norm in higher spatial frequencies (12 and 18 c/deg).

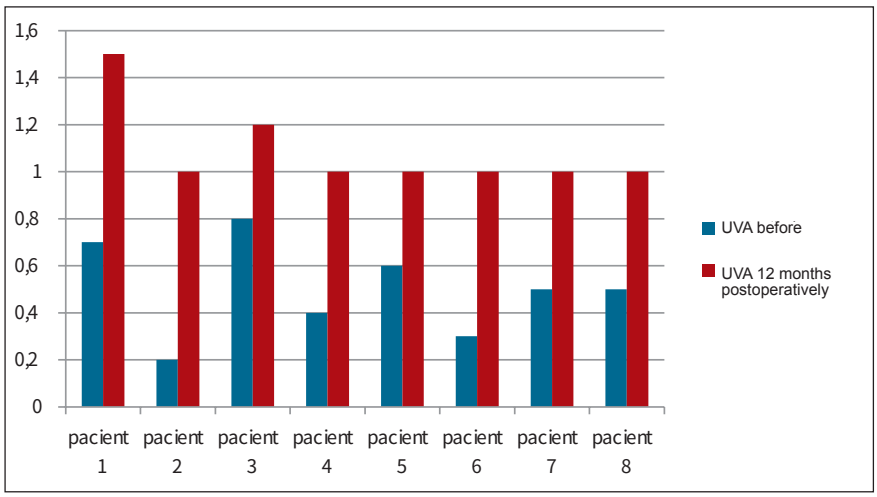
Certain publications evaluating postoperative results following LASIK also point to a similar reduction in contrast sensitivity (10, 17). According to the satisfaction questionnaire, 7 patients (87.5%) stated absolute satisfaction with the result of the procedure, felt independence of wearing glasses and would recommend the procedure to a good friend. 7 patients (87.5%) did not state the presence of photic phenomena (halo, glare), 1 patient had these complaints, nevertheless a further 2 patients also responded to a direct question that they also experienced more frequent dazzling by sunlight, but stated that they did not perceive this as problematic.

One patient was dissatisfied and remained dependent on glasses correction for near vision. Postoperatively we recorded the formation of a nuclear cataract in this patient.

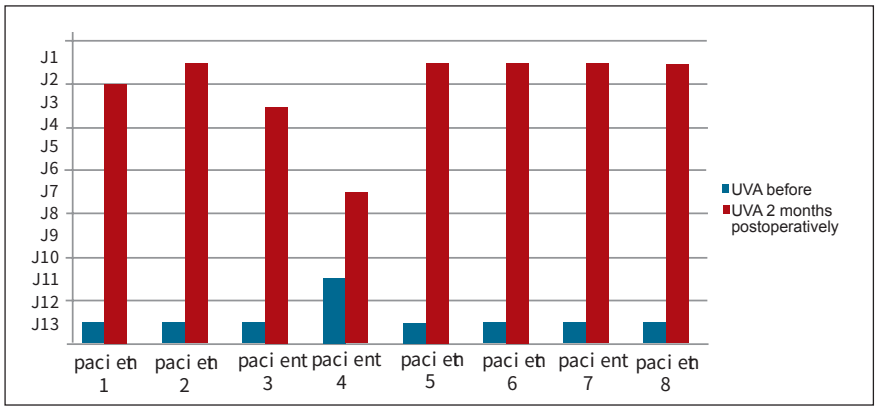
We did not record any complications

Table 4 Binocular uncorrected intermediate visual acuity.

Patient	UVA, 12 months
1	J. no. 2
2	J. no. 1
3	J. no. 2
4	J. no. 4
5	J. no. 1
6	J. no. 1
7	J. no. 1
8	J. no. 1



Graph 1 Binocular uncorrected distance visual acuity before and 12 months after Supracor



Graph 2 Binocular uncorrected near visual acuity before and 12 months after Supracor

either perioperatively or postoperatively, but in the above-stated patient the postoperative results were significantly influenced by the formation and development of a cataract.

DISCUSSION

Corneal procedures to resolve presbyopia are suitable especially due to their minimally invasive nature. Intraocular surgical procedures present serious risks even in the hands of an experienced surgeon, including endophthalmitis (4, 5). The longest used corneal approach is monovision (6) – photoreactive keratectomy (PRK) or laser in situ keratomileusis (LASIK), other corneal methods include conductive keratoplasty, corneal implants and also laser methods using a bifocal corneal profile (presbyLASIK) or multifocal corneal profile (Intracor and Supracor) (6, 13).

Initially PRK was used for the correction of presbyopia by the monovision method, and later LASIK. Monovision means correction of one eye for distance vision and the second eye corrected for near vision (2). The attendant anisometropia results in a reduction of visual acuity, impaired spatial vision and a reduction of contrast sensitivity. A precondition for a satisfactory result is a good ability to suppress the blurred image of one eye, good spatial vision, anisometropia less than 2.5D, satisfactory visual acuity of the eye corrected for distance vision, motivation of the patient and the ability of neuroadaptation (13).

LASIK has become a well known and popular refractive method. Its benefits indisputably include the possibility of further laser correction in case of requirement. In addition it is not burdened by the compensatory effect of regenerating epithelium as in the case of PRK, and it thus represents a method which is easier to monitor, especially for the multifocal profile of the cornea (13). Procedures for correction of presbyopia use the principle of the LASIK method. Following the creation and folding of the lamella, photoablation is performed in bifocal or if applicable multifocal profile. In the case of presbyopic LASIK, we differentiate two approaches – central presbyopic LASIK (central area for near vision, peripheral area for distance vision) and peripheral presbyopic LASIK (central zone for distance vision, paracentral for near). A bifocal corneal profile is thus formed. However, authors dealing with central presbyopic LASIK described deteriorated quality of vision, a reduction of BCDVA in 28% of cases, a reduction in contrast sensitivity in spatial frequencies of 3, 6, 12 and 18 c/deg and an increase in higher order aberrations (1). Transitional zones which may be generated in bifocal procedures increase adverse aberrations and reduce visual acuity (12). In the case of the Supracor method, a multifocal corneal profile is created using an excimer laser with progressive transition between the zone for near and distance vision. The profile is progressive and the incidence of aberrations is thus minimised (12).

According to our first results, it is possible to attain good visual acuity using a Supracor procedure for distance, near and intermediate vision, and patient satisfaction is relatively high.

A disadvantage is the narrow indication criteria and up to now the impossibility of use on emmetropic or myopic presbyopic patients (at present it does not have a CE certificate, it shall be possible to perform Supracor on myopic presbyopic patients from May 2014). An advantage is the speed of the procedure, the option of additional laser correction and the fact that this represents a superficial procedure. We recorded an unsatisfactory result of the operation in 1 patient, in whom binocular UNVA was J. no. 7 and UDVA 1.0, and who was dissatisfied with the result of the procedure and requires glasses correction for near vision. We explain the lack of success partially with reference to the atypical reparation reaction of the corneal tissue, with slight flattening of the corneal profile according to the topography, and partially with reference to incipient changes of the lens – incipient nuclear cataract. Preoperatively the patient met the indication criteria, including clear optic media, perioperatively and postoperatively the course was without complications. Upon progression of the cataract we shall proceed to surgery with implantation of a monofocal artificial intraocular lens.

We compared our results with the foreign literature, nevertheless the comparison is merely referential due to the small number of our patients. The size of our study cohort ensues from the fact that this represents a new method (used at our clinic since July 2012), which is furthermore financially costly and has narrow indication criteria.

Ryan et al. described the results of 23 patients (46 eyes) following a Supracor procedure. The indication criteria were virtually identical to our criteria (table 1). Binocular UDVA of 0.2 logMAR (i.e. 0.63) and better, and UNVA of N8 (corresponding approximately to J. no. 3) was attained in 91% of patients. The average value of the spherical equivalent was -0.69 (±0.71). The authors also evaluated the stability of visual acuity: in 24% of eyes there was a statistically significant change in the spherical equivalent of manifest refraction between the 3rd and 6th month after surgery. In this study aberrations were also observed. There was no significant increase in aberrations with the exception of the central quadratic error of higher order aberrations, which increased from 0.3 μm preoperatively to 0.43 μm postoperatively (12). Twenty two percent of patients underwent additional laser correction on the dominant eye for improvement of UDVA. Satisfaction with the procedure was stated by 96% of patients. Ryan et al. describe that following an evaluation of the first results they introduced a bilateral asymmetrical regime (dominant eye now targeted to emmetropia and non-dominant eye to -0.5 D, this regime is used at our clinic), and this brought about an improvement of the results of distance visual acuity. The original target value was 0.5 D bilaterally (12).

Cosar et al. evaluated the results of 68 patients (123 eyes) 6 months after a Supracor procedure. The indication criteria in this study were less strict (addition +1.5 D and more, hypermetropia to +4.0 D, astigmatism to 2 D, corneal thickness above 500 μm, BCDVA 20/20 and more), they did not include values of keratometry, age, difference between manifest and cyclplegic refraction and pupillometry. The first 55 patients underwent a bilateral procedure, a further 13 patients underwent laser treatment only on the non-dominant eye (the authors do not state a more detailed method and target refraction). UDVA reached 20/20 and better in 22% of eyes, 20/25 in 36.6% of eyes. 28.5% of eyes lost 1 row and 10.6% 2 rows of BCDVA. UNVA was 20/20 and better in 77.2% of eyes and 20/25 in 89% of eyes, average spherical equivalent was -0.33 (±0.10). Stability of visual acuity was not evaluated, follow-up examinations took place on the 1st day, 1 month and 6 months after the procedure (3).

In our study cohort, the patients attained better distance visual acuity and comparable near visual acuity in comparison with the above-stated studies. The validity of the results is however reduced by the very small number of patients in the group.

With regard to the fact that this represents a relatively new method, the number of available published studies is limited.

CONCLUSION

Correction of presbyopia remains a large challenge for ophthalmologists. The spectrum of potential surgical solutions is wide. An interview with the patient, careful examination and an individual approach are of key importance for the choice of a suitable type of procedure.

According to our first experiences, it is possible to attain good visual acuity by the Supracor method for distance, near and intermediate vision, as well as relatively high patient satisfaction. Supracor appears to be a suitable method for the correction of presbyopia in motivated and flexible patients, who meet the strict indication criteria. With regard to the small size of the study cohort, however, it shall be necessary in future to evaluate a larger number of patients, in addition to which the long-term results shall be of interest.

LITERATURE

1. **Alió, J. L., Chaubard, J. J., Caliz, A. et al.:** Correction of presbyopia by technovision central multifocal LASIK (presbyLASIK). J Refract Surg, 2006; 22: 453–460.
2. **Barišić, A., Barić, N., Dekaris, I. et al.:** Comparison of Different Presbyopia Treatments: Refractive Lens Exchange with Multifocal Intraocular Lens Implantation Versus LASIK Monovision. Coll Antropol, 34,2010; 2: 95–98.
3. **Cosar, C. B., Sener, A. B.:** Supracor hyperopia and presbyopia correction: 6-month results. Eur J Ophthalmol, 24, 2014; 3: 325–329.
4. **Fintelmann, R. E., Naseri, A.:** Prophylaxis of postoperative endophthalmitis following cataract surgery: current status and future directions. Drugs, 70, 2010; 11: 1395–1409.
5. **Holzer, M. P., Knorz, M. C., Tomalla, M. et al.:** Intrastromal femtosecond laser presbyopia correction: 1-year results of a multicenter study. J Refracr Surg, 28, 2012, 3: 182–188.
6. **Holzer, M. P., Mannsfeld, A., Ehmer, A. et al.:** Early outcomes of INTRACOR femtosecond laser treatment for presbyopia. J Refract Surg, 25, 2009; 10: 855–861.
7. **Krader, Ch. G., Ryan, A.:** LASIK treats hyperopic presbyopia, Ophthalmology Times, 37, 2012; 22: 20.
8. **Novák, P.:** Refrakční vady a jejich řešení. In Kuchynka, P., Oční lékařství, Praha, Grada Publishing, a.s., 2007, s. 115.
9. **Machačová, H., Vlková, E., Pirnerová, L. et al.:** Supracor – moderní metoda řešení presbyopie, XXI. Výroční sjezd České oftalmologické společnosti ČLS JEP, sborník abstrakt, Brno, 19. - 21. 9. 2013, Hradec Králové: HANZO Production, spol. s r.o., 2013, s 119. ISBN 970-80-905115-4-5.
10. **Montés-Micó, R., Rodriguéz-Galietero, A., Alió, J. L. et al.:** Contrast sensitivity after LASIK flap creation with a femtosecond laser and a mechanical microkeratome. J Refract Surg., 23, 2007; 2: 188– 192.
11. **Pirnerová, L., Vlková, E., Trnková, V. et al.:** Supracor – výsledky ročního sledovacího období, XXI.Výroční sjezd České oftalmologické společnosti ČLS JEP, sborník abstrakt, Brno, 19. – 21. 9. 2013, Hradec Králové: HANZO Production, spol. s r.o., 2013, s 109. 2013. ISBN 970-80-905115-4-5.
12. **Ryan, A., O´Keefe, M.:** Corneal approach to hyperopic presbyopia treatment: Six-month outcomes of a new multifocal excimer laser in situ keratomileusis procedure. J Cataract Refract Surg, 39, 2013, 8: 1226–1233.
13. **Torricelli, A. M., Junior, J. B., Santhiago, M. R. et al.:** Surgical management of presbyopia. Clinical Ophthalmology, 2012, 6: 1459–1466.
14. **Truscott, R. J., Zhu, W.:** Presbyopia and cataract: A question of heat and time, Prog Retin Eye Res, 29, 2010, 6: 487–499.
15. **Urminský, J., Rozsíval, P., Lorencová, V. et al.:** Použití akomodačních čoček k chirurgické korekci presbyopie metodou Prelex. Čes a slov Oftal, 2006, 5: 324–333.
16. **Warung, G. O., Berry, D. E.:** Advances in the surgical correction of presbyopia. Int Ophthalmol Clin, 53, 2013, 1: 129–152.
17. **Zhao, L. Q., Zhu, H.:** Contrast sensitivity after zyoptix tissue saving LASIK and standard LASIK for myopia with 6-month followup. J Ophthalmol, 2011, dostupné z: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3090770/.

ORIGINAL ARTICLE

Lymphangioma of the Orbitopalpebral Area

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SUMMARY

Aim: The authors refer about five patients with different types of lymphangioma, who were followedup at the Department of Ophthalmology, Faculty Hospital Královské Vinohrady (King's Vinegards), Charles University, Prague, Czech Republic, E.U., during the period 1995 – 2013; the follow-up period lasted from 5 to 17 years. The lymphangioma of the orbitopalpebral area is discussed according to the evaluation of the tumor development, histological verification, treatment, and its results.
Methods: In four boys, the first signs of tumor were eyeball protrusion (exophthalmos) and bleeding into the conjunctiva or palpebral skin before the age of 5 years. In all four patients, the histological confirmation of the orbital lymphangioma was performed in the beginning of the disease. In three cases, it was the orbital type, and the fourth one was frontal type with bilateral orbital lymphangiomatosis. In one girl, there were present conjunctival changes only, appearing as one-sided hyperplastic changes. For these changes, she was followed-up since her 13 years of age under the false diagnosis of chronic conjunctivitis. The definite histological confirmation of only conjunctival lymphangioma was done from the diagnostic probatory biopsy not until ten years of symptoms and unsatisfactory treatment.
Results: In the girl with superficial conjunctival lymphangioma and in the patient with lymphangiomatosis, the follow-up was recommended only. In two patients with extraconal type of orbital tumor, the total or sub-total resection was performed. In the years of the follow-up, the remission of the disease was observed. In the patient with mostly intraconal type of the tumor, causing decrease of the visual acuity according to the optic nerve neuropathy and macular cystoid edema, the focused actinotherapy by means of linear accelerator treatment with the dose of 30 Gy after previous evacuation of chocolate cysts under ultrasound control. The regression of the tumor and normalized visual functions lasted for 17 years.
Conclusion: As method of treatment of extraconal lymphangiomas, it seems, it is its resection, and in the intraconal localization of the tumor it is the focused actinotherapy by means of linear accelerator..

Key words: orbital lymphangioma, conjunctival lymphangioma, lymphangiomatosis, tumor resection, linear accelerator actinotherapy

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INTRODUCTION

Lymphangioma occurs on a congenital basis and together with haemangioma is ranked with harmatomas. Their incidence is bound to tissues which are normally present within the orbit, which does not apply in general for lymphatic blood vessels. Due to its cellular structure lymphangioma ranks amongst benign tumours, but from a clinical perspective it does not merit this indication. It does not have a capsule, it grows diffusely and infiltrates the surrounding tissues. Microscopically lymphangioma is formed by a network of lymphatic areas covered with the endothelium and thin stroma with a varying degree of lymphocyte infiltration, within the framework of which lymphoid follicles may be formed. The local behaviour in the orbit is thus similar to that of malignant tumours, but unlike these it does not metastasise (39). Three fundamental clinical forms are distinguished. Superficial lymphangioma afflicts only the conjunctiva, manifested as cystoid edema, after which there is the separate orbital type which afflicts the orbit postseptally. The combined form is manifested pre and postseptally in the oral cavity and in the cheek (13). It grows slowly and persistently without a tendency towards spontaneous involution, thankfully in adulthood growth usually stabilises. It is generally unilateral and presents both cosmetic problems and functional disorders such as strabismus and amblyopia on the same side. A basic symptom includes asymmetry of the face and eye apertures, namely fuller cheeks and bulging of the eyelids on the afflicted side. The conjunctiva is afflicted in any part initially in the form of vitreous chemosis, which passes into a cystic character with lymphangiectasia. The clinical picture is completed with enlargement of the bone part of the eye socket. Protrusion and dislocation of the bulb is variable depending on sudden massive haemorrhage into the tumour, which may represent a severe complication endangering the eyeball itself. It is manifested on the conjunctiva through repeated suffusions. Vision is then endangered by the protraction of the optics or compression of its nutritive blood vessels (39).

OWN OBSERVATION

Lymphangioma was not included in the previous ten-year study (28) on pathologies of the orbit in adulthood at the Department of Ophthalmology at the Královské Vinohrady University Hospital in the years 1998-2007, since the occurrence of this disorder in all the five patients stated at the time was in childhood. As a result, we decided to recall the issue of lymphangioma from a clinical, histological and therapeutic perspective within the framework of long-term observation from 1996-2013, i.e. over the course of 17 years.

Patient no. 1

The first observation of our cohort was the only patient to be observed, even if irregularly, throughout the entire period of our study. In 2013 the patient was aged 32 years. The beginning of his symptoms date from the age of five (1985), when there was protrusion of the eyeball on the right side, accompanied by haemorrhage into the conjunctiva. A complex examination was conducted at a regional centre outside of Prague. Probatory excision was performed, which detected a diagnosis of lymphangioma. After three years there followed actinotherapy of the entire orbital cavity in a dose of 16.7 Gy/focus The condition did not progress further for several years, but haemorrhage into the conjunctiva was repeated. During the course of 1995 progressive protrusion again gradually appeared. In September 1995 CT demonstrated an irregular formation with a diameter of approximately 17 mm with a density of 50 HU. intraorbitally nasally, which was only slightly pronounced postcontrast. The retrobulbar space in the direction of the apex of the orbit was also filed with non-homogeneous masses of the same density as the formation nasally. The condition continued to progress, and as a result the patient was admitted to the Department of Ophthalmology of the KVUH in September 1996 for a decision on the further procedure. The right eyeball was in 5 mm protrusion and deviated outwardly by 10 mm, with limited motility in adduction. A perfused tumour, which was reductible in the direction towards the orbit was evaginated beneath the conjunctiva nasally in the internal corner (fig. 1a). The other finding on the anterior segment was physiological. Cystoid macular edema (CME) was perceptible on the ocular fundus, as well as slightly constricted blood vessels, and the papilla of the optic nerve was bordered (fig. 1c). VOR (= visus of the right exe) 0.33 with + 1.0, J. no. 6 best correction. (In left eye extraocular and intraocular finding was physiological, VOL (visus of the left eye) 1.0 nat., J. no. 1 nat.). A transocular B scan of the eye socket showed a tumour of a cystic character

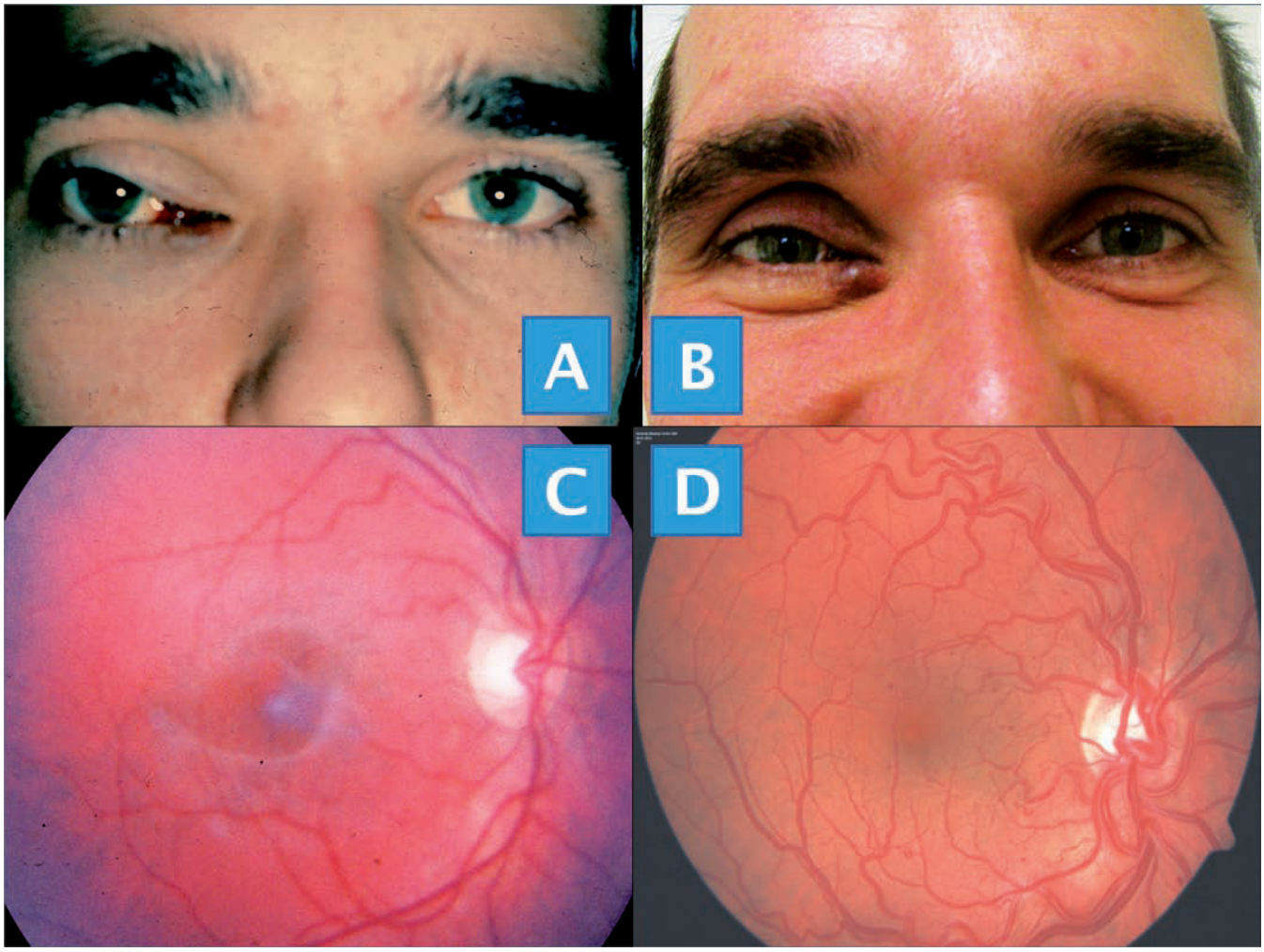

Fig. 1 Patient no. 1
a – clinical picture before aspiration of chocolate cysts and actinotherapy.
b – clinical picture following treatment after 17 years.
c – finding on fundus of right eye with CME before treatment.
d – finding on fundus of right eye after 17 years.

with echographic features typical of lymphangioma: irregular internal structure, high reflectivity of the septa and low reflectivity of the interseptal space filled by the stroma. One of these larger cysts manifested higher internal reflectivity in comparison with the others. This concerned a "chocolate cyst", which was filled with what was probably colliquated blood (fig. 2c). After consultation with the radiotherapists we decided upon repeat actinotherapy. Due to its higher efficacy, it was first of all necessary to drain the content of the cyst, with the aim of relieving the pressure on the optic nerve. We performed partial aspiration of the content of the cyst under general anaesthesia under the control of ultrasound (fig. 2a, b). The subsequent regression of the protrusion and outward deviation by several mm was accompanied also by a change of the image on the transocular B scan, where a reduction of the diameter of the cyst was visible (fig. 2d). Subsequent actinotherapy was performed in April 1996 in fifteen sessions of 2 Gy/1focus (total dose 30 Gy/focus). A linear accelerator was used, and the planning scheme covered the retrobulbar space up to the apex of the eye socket in the form of a "bowl base". Three months after the end of irradiation treatment the protrusion had been reduced to 1 mm and the outward deviation had decreased to a mere 3 mm. Motility of the bulb was now entirely free. The finding on the anterior segment of the eye was physiological, on the ocular fundus the finding in the area of the macular region had been normalised, VOR 0.66 nat., J. no. 2. Three years after actinotherapy of the right orbit, normophthalmos was demonstrated with only a suggestion of outward deviation. The finding on the anterior segment was physiological up to the incipient manifestation of a post-radiation cataract in the posterior cortex in the form of a disciform 2-3 mm cataract. The finding on the ocular fundus was physiological. VOR 0.66 with +2.5/80 and J. no. 2 obt. with +2.5/80. The patient was recommended regular eye checks in his place of residence, which he however did not undergo, upon his own decision. He did not report to our clinic again until January 2013, upon the request of a doctor from a psychiatric institution, where the patient had been monitored due to a schizophrenic disorder. During his residence at the institution, repeated haemorrhage into the patient's conjunctiva occurred repeatedly in the internal corner on the afflicted side. Upon an examination at our clinic, the configuration and position of the bulb in the right eye were entirely physiological, motility free and spatial vision was also within the norm (stereoscope, Randot). Suffusion of the tumescent conjunctiva was perceptible in the internal corner (fig. 1b), in a further finding on the anterior segment only irregular opacities were perceptible in the

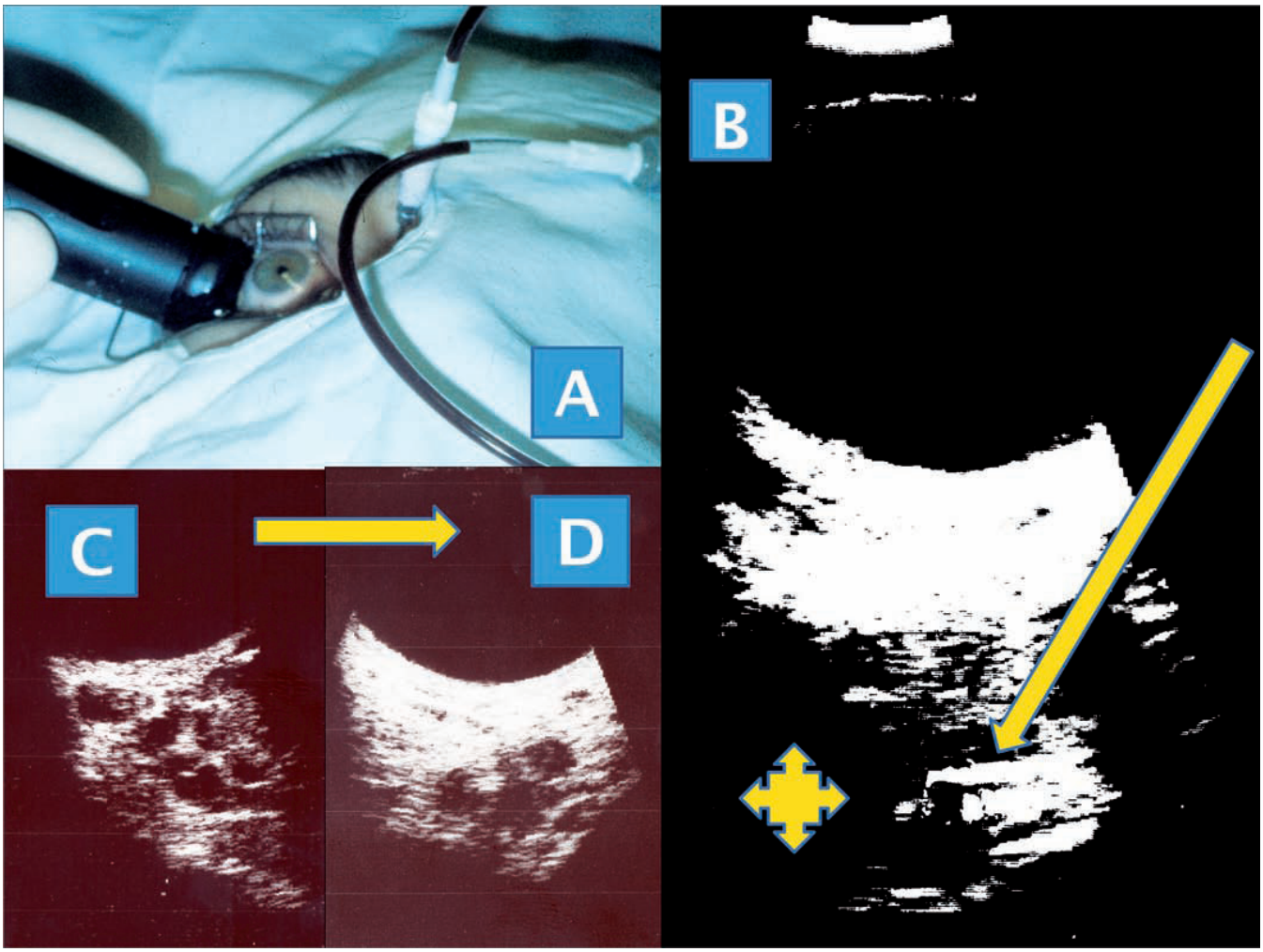

Fig. 2 Patient no. 1
a – aspiration of chocolate cysts under ultrasound control.
b – ultrasound image of aspiration: needle with acoustic shadow by chocolate cyst (arrow).
c – orbital cavity with chocolate cysts.
d – regression of finding after aspiration.

lens beneath the posterior capsule. On the fundus the papilla of the optic nerve was overall paler than on the left side, the macular region now had physiological contours, the blood vessels were of a normal calibre and slightly more coiled (fig. 1 d). VOR 0.8 – 0.9 with +0.5/50 and J. no. 2 with + 0.5/50. Contrast sensitivity was on the borderline of the norm at 3, 6 and 12 c/deg, and only slightly diminished at 18 c/deg (CSV 1000). This change of reduction was confirmed by normal contrast sensitivity in the left eye, where the finding was entirely physiological VOL 1.0 nat., J. no. 1.

Overall the condition following actinotherapy was evaluated as highly satisfactory. The finding on the lens following irradiation did not demonstrate progression, since the initial changes had receded, together with practical normalisation of refraction. The slight atrophication of the optic nerve could be the result of a side effect of irradiation, but also an echo of the nutritional disorder of the head of the optic nerve due to a suppression of the nutritive blood vessels by the tumour itself. Perfusion in the internal corner sketched in an image of possible residue of the tumour, since this area had been only minimally irradiated, and this had been within the framework of the first actinotherapy in 1988. The subjective complaints of dryness of the right eye improving following the application of lubrication were linked to KCS following irradiation.

Patients no. 2 and 3

The next two patients were four year old boys in whom there was identical haemorrhage in the conjunctival orbital region due to medical care, without anamnesis of trauma. The complete laboratory examination in both boys, including sedimentation of erythrocytes, was negative. In patient no. 2 the parents initially noticed haemorrhage beneath the conjunctiva in the internal corner in the right eye, which gradually transformed into an enlarging formation which displaced the eye outwardly. At the first examination at our centre in 1998, there was protrusion of the right eyeball of 2 mm with outward deviation by 5 mm (fig. 3a) after a three week anamnesis. Motility of the bulb was free without diplopia. The further finding on the anterior segment of the eye and the intraocular finding was physiological bilaterally, VOR = VOL 1.0 nat. An ultrasound examination demonstrated a pre-equatorially, irregularly bordered, lobed formation 6 x 5 x 6 mm in the internal part of the orbit, localised retroseptally. With regard to the character of the clinical picture and the speed of onset of the symptoms, we proceeded to a probatory excision for the purpose of histological verification of the nature of the tumour. Due to the minimal haemorrhaging during the course of the operation we performed partial resection of the formation.

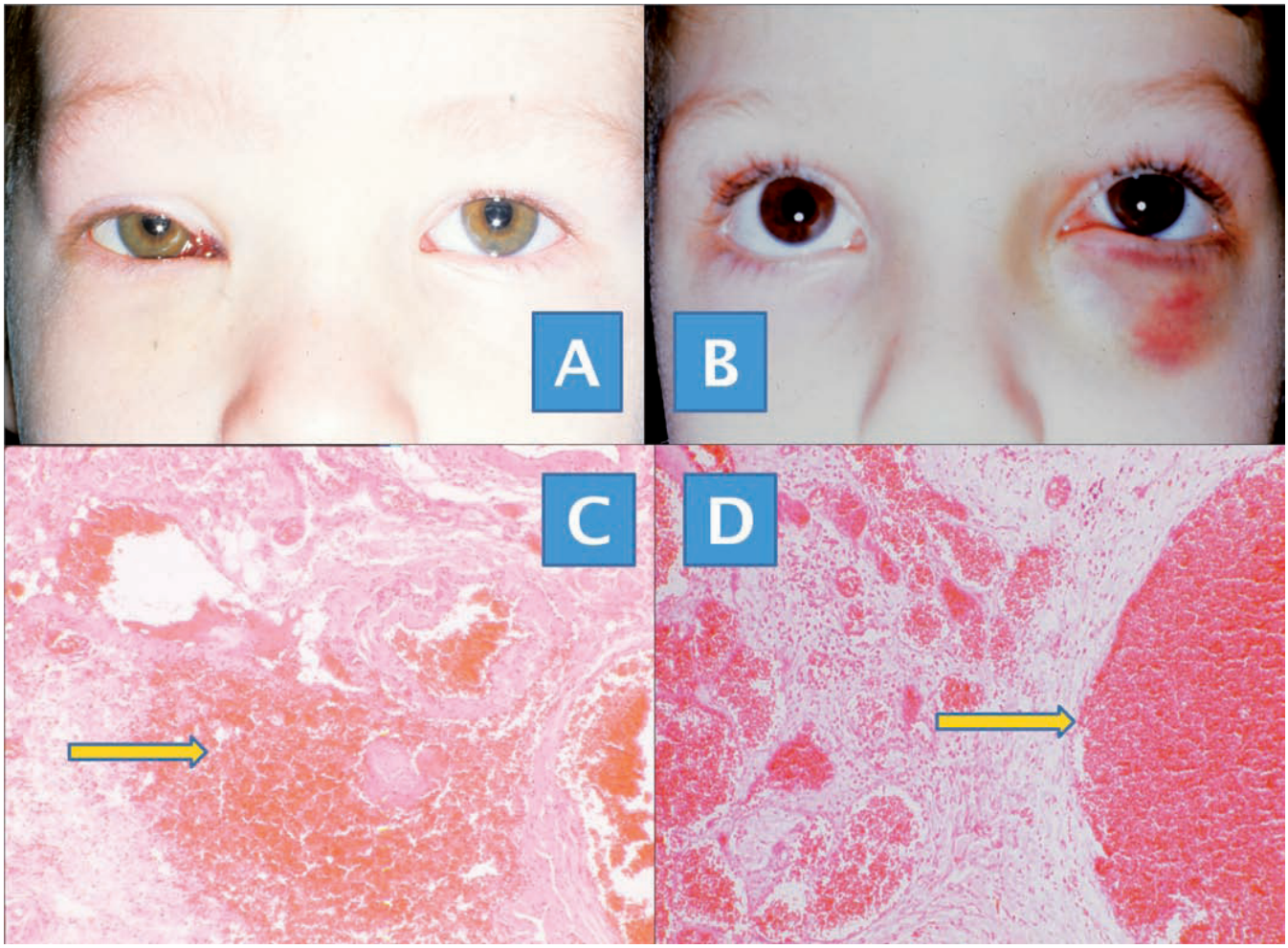

Fig. 3 Patient no. 2 in right eye and patient 3 in left eye
a – clinical picture of lymphangioma in right eye.
b – clinical picture of lymphangioma in left eye.
c – histological verification of right-sided lymphangioma of orbit (blood staining of tumour – arrow).
d - histological verification of left-sided lymphangioma of orbit (haemorrhagic dilated cavity, future chocolate cyst – arrow).

In addition to the haemorrhagic structures of lymphangioma, scarry regression and fibrous conversions of certain parts of the tumour with its microscopic residues were determined in the histological image (fig. 3c). After six months the configuration and position of the right eyeball had been normalised, and no relapse of the tumour was manifested over the further course of several year observation. The reason for hospitalisation of the third patient in our group, also in 1998 was haematoma persisting for ten days in the area of the lower eyelid of the left eye, with dislocation of the bulb upwards and outwardly by 2 mm (fig. 3b). Its motility was free, without diplopia. The further finding on the anterior segment of the eye and the intraocular finding was physiological bilaterally, VOR = VOL 1.0 nat. Ultrasound examination in the internal part of the orbit demonstrated a relatively well bordered oval formation of an irregular internal structure and size of 5 x 4 x 3 mm lying retroseptally and pre-equatorially. From the nasal orbitotomy we performed total resection of the tumour, in which small cystic formations with brownish fluid were perceptible perioperatively, which were evaluated as "chocolate cysts". Lymphangioma was verified histologically, in which especially perfusion of the areas of the tumour predominated amongst the secondary changes, leading in places to their thrombotic content, which in their secondary changes caused the formation of "chocolate cysts" (fig. 3d). The postoperative period of actual healing was accompanied only by epiphora. After six months the orbit was without clinically manifest pathological resistance. As soon as in 1998, monocanalicular dacryocystorhinostomy was subsequently performed on the left eye following interception of the lacrimal pathways within the framework of prior resection of lymphangioma. A control histological sample of scarry tissue was taken, which demonstrated a scarred residue of the angiomatous tumour formed by very dense fibrous tissue. Despite two repeat operations during the further course, attempts to ensure through flow of the lacrimal pathways did not succeed in a fully satisfactory manner up to 2003. The orbital finding did not change. Ultrasound examination did not demonstrate any manifest pathological resistance. In 2006 MRI was indicated for the purpose of repeat verification of the therapeutic result of resection of the tumour, which excluded lymphangiomatic infiltration. The boy continues to be monitored in his place of residence.

Patient no. 4

The fourth case included in our study cohort concerned a twenty three year old woman who had been sent from the district in 1996 due to only unilateral chronic hyperplastic conjunctivitis with recurrences of saturation and perfusion. Anamnestically the complaints had persisted for more than ten years. Subconjunctive tumescence was of a gelatinous charac-

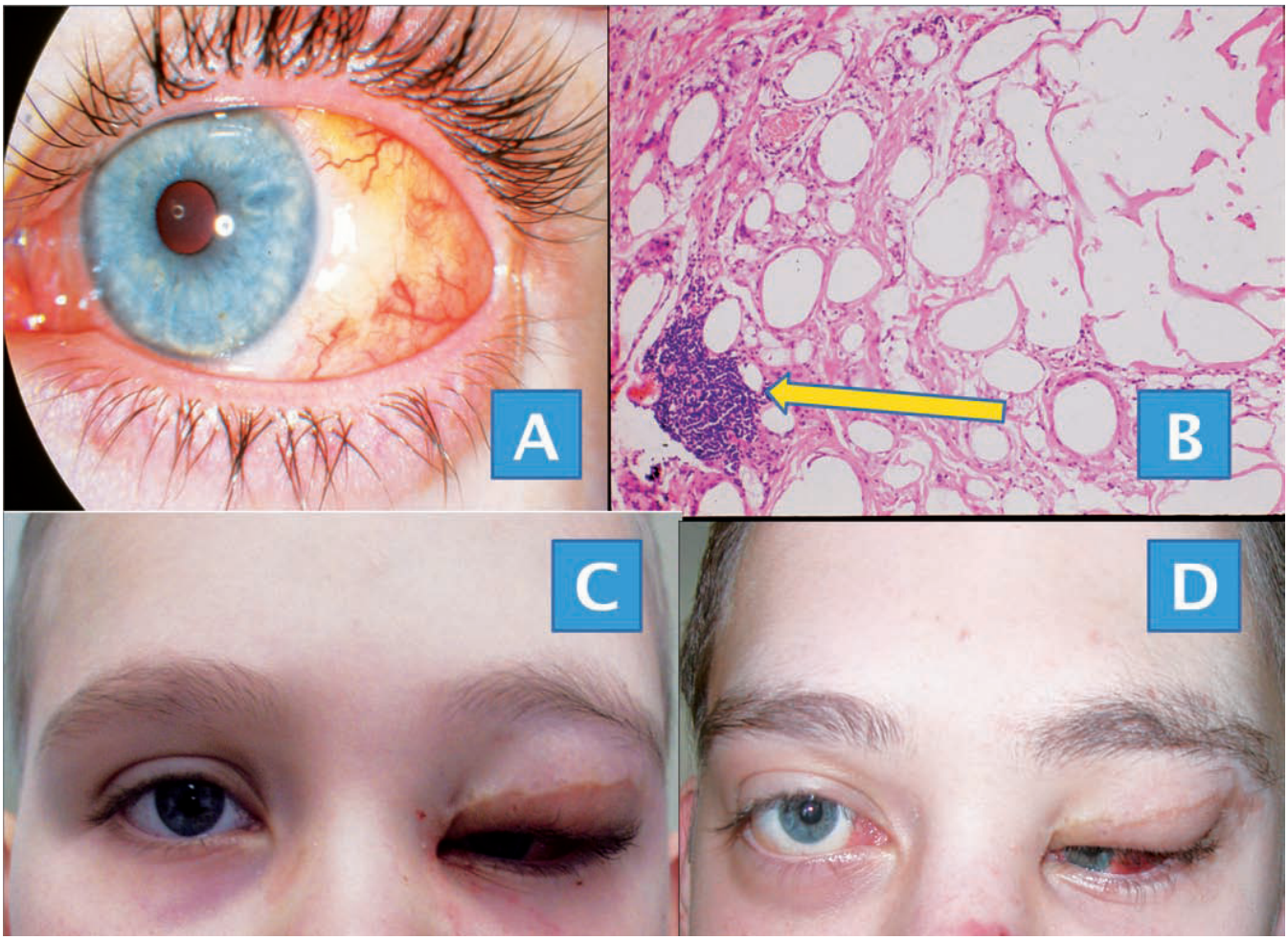

Fig. 4 Patient no. 4 above and patient no. 5 below
a – separate lymphangioma of bulbar conjunctiva.
b - histological verification of conjunctival lymphangioma (lymphatic tissue – arrow).
c – left-sided lymphangioma of orbitopalpebral area in 2006.
d – lymphangiomatosis now afflicting both orbits in 2011.

ter with dilation of the capillaries, and reached from the limbus to the depth of both fornices (fig. 4a). We decided in favour of probatory excision of the subconjunctival tumorous tissue. Histological verification demonstrated chronic inflammatory infiltration, as well as structures characteristic of lymphangioma (fig. 4b), namely a system of interconnected lymphatic cavities, focally in their edges with islands of lymphatic tissue. We did not indicate a radical surgical procedure and recommended solution of the condition at a time of pronounced secondary conjunctival injection with local application of collyrium, namely a combination of antihistamines with adstringent effect and fluorometholone.

Patient no. 5

The last patient in our cohort, sent for consultation in 2006, was a ten year old boy (fig. 4c). The first symptoms of tumorous pathology of the left orbit appeared at the age of three years. Subsequent partial resection of the tumour in the area of the upper eyelid in an attempt to alleviate pseudoptosis at a non-ophthalmological centre did not have the envisaged cosmetic effect. A histological examination verified lymphangioma. This represented a combined type of lymphangioma of the orbit, which also infiltrated the cheek on the afflicted side and reached into the oral cavity. With regard to the fact that it later spread also to the forehead and the other side of the face, the finding was concluded to be a combined venous lymphatic vascular malformation of the frontal area and both orbits. The scope of affliction was confirmed by an MRI examination in 2011, which demonstrated an analogous cystic formation also in the right orbit. On the left there was infiltrative tumescence of the upper eyelid, and the dorsonasal formation in the orbit was stationary. The examination did not detect any further intracranial changes. The actual clinical finding was manifested in an indication of axial protrusion on the right side and on the conjunctiva in the internal corner by gelatinous tumescence of a salmon pink colour. The actual orbit was without palpable resistance, the eyeball was reductible and freely mobile. On the left side the pronounced tumescence of the upper eyelid restricted its motility and constricted the eye aperture, the eyeball was deviated downwards to 20 degrees of convergence (fig. 4d). The finding on the anterior segment and the intraocular finding were physiological bilaterally. Amblyopia from secondary droop of the upper eyelid due to the tumour contributed to a deterioration of vision in the left eye (VOL 0.3 with correction – 0.75 = +2.0/100, VOR 1.0 naturally). We did not recommend a further intervention with regard to the above diagnosis, and the patient remains under our observation.

DISCUSSION

Since the publication of the monograph by Dr. J. Otradovec (1986) (39), we have not recorded any more extensive work on this congenital harmatoma of the orbit in the Czech and Slovak literature. Nevertheless, the observations here remain a literary source, also due to the quality of writing. In addition, the number of works on the theme of oncological orbitology since the breakup of Czechoslovakia in the founding period of both ophthalmological societies in the subsequent two decades has not reached an average of even one article per year. Only nine studies have been devoted to the issue of paediatric ophthalmology (1, 9, 10, 11, 12, 24, 26, 40, 43), three studies are focused on an adult age clinic (19, 28, 29), four articles state separate operation of the orbit (20, 25, 27, 33), and two works focus on special diagnostics (34, 42). The last diversely extensive overviews of lymphangioma are presented in a Slovak monograph by Professor A. Gerinec (2005) (13), and also in chapters 16 and 17 of the Czech attestation textbook Ophthalmology by Professor P. Kuchynka et al. (2007) (30). Lymphangioma ranks amongst tumours of childhood age with manifestations of a finding in adulthood, and is relatively rare, with frequency stated between 1-5% (39). A summary evaluation of the literary citations of the main paediatric ophthalmological works at the end of the 20th century stated its frequency at up to 3% (26). The most comprehensive retrospective study in all age categories over the last thirty years, which assesses 1264 patients with orbital tumours and stimulating conditions, states a 4% frequency of lymphangioma (46). In the Asian population the incidence of vascular tumours in childhood age is higher, a twenty one year study of 244 cases of orbital tumours demonstrated an 11% representation of lymphangioma at the age of up to 9 years (37). One of the first assessments of orbital tumours in children is a cohort of 174 patients from the period 1923-1977, in which lymphangioma was diagnosed in ten children (5.7%) (21). In two Slovak eight year studies, which are to date the only studies on childhood orbital tumours within our region, lymphangioma was diagnosed twice in each study, on 78 paediatric patients from 1987-1994 (10) and 130 paediatric patients from 1992-1999 (12) respectively. Our observation of paediatric patients with lymphangioma in the orbitopalpebral region links to a previous ten-year configuration of orbital tumours in 87 adult patients, since all five patients were observed for varying periods at the time of this study (28). The low frequency of occurrence of this tumour in the population and the unclear symptomatology from the inception of the pathology must always lead to a differential diagnostic consideration. An acute beginning of manifestation of lymphangioma caused by intratumorous haemorrhage is manifested in sudden exophthalmos, which may belong to a picture of a malignant process. At childhood age mainly malignant rhabdomyosarcoma of the orbit is considered in differential diagnostics. Manifestations of initial perfusion of the skin of the eyelids and conjunctiva may also be for example the first manifestation of metastases of Ewing's sarcoma of the bone and above all neuroblastoma, where increased sedimentation of erythrocytes indicates this possibility (26). The issue of orbital cysts in children is extensive, in which processes primarily without cysts, which may however have a cystic component, are more difficult to distinguish. These included adenoidal cystic carcinoma, as well as rhabdomyosarcoma and also lymphangioma. Examination using CT and MRI helps their differentiation, but is not pathognomonic (49).The presence of "chocolate cysts", which form through the colliquation of blood in haematomas generated through haemorrhage into the tumour (8), helps to differentiate lymphangioma from capillary haemangioma. The paramagnetic property of haemoglobin helps in the detection of haemorrhagic cysts in tumours well supplied with blood vessels, in the phenomenon of "through flow" (4). In the method of MRI with high resolution a surface spool is used, which improves the diagnostic information in vascular tumours (cavernous haemangioma, lymphangioma, varices or AV malformation), whereas differentiation in solid tumours is more arduous and histological verification is essential (32).

Ultrasound examination of the orbital cavity ranks amongst the methods of examination which have a fundamental influence from the perspective of differential diagnostics and treatment of orbital afflictions. This represents a non-invasive, quickly available examination which can be performed in outpatient care, in hospitalisation and in the operating theatre. With regard to the undemanding nature

of the examination for the patient and its harmlessness to the human organism, it is possible to conduct this examination repeatedly, not only on adults but also on children. In the case of lymphangioma of the orbit, the display of the echographic image in both B and A is sufficiently characteristic that it mostly does not cause diagnostic difficulties. This represents a soft, compressible tumour without signs of internal vascularisation. With regard to the infiltrative character, the tumour is less well bordered on the B scan, and its surface is generally irregular. Lymphangioma of the eye socket is formed by a network of lymphatic cavities filled with a thin stroma with lymphocyte infiltration. This is corresponded to echographically by the irregular internal structure and reflectivity in the B scan, alternation of higher echoes (septa) and lower echoes (stroma) in the A scan. In the case of haemorrhage into the tumour we record increased reflectivity of the interseptal spaces (2). A modern method is the pulse Doppler technique for the description of intratumorous vascularity, which is of significance for differentiating haemangioma and lymphangioma (7). The result of ultrasound examination in our two four year old boys (patients 2 and 3) was sufficiently conclusive for surgical intervention that the children were operated on on the third day after admittance. The cohort of our five patients included one patient with lymphangioma of the conjunctiva, in which a radial solution was not possible because the lymphangioma had infiltrated the entire bulbar conjunctiva up to both fornices. Isolated observation of lymphangiomatosis, stated as congenital venous and lymphatic malformations in two localities on the skull (6) in our patient primarily afflicted the orbit and upper eyelid on the left side, fro which it passed over the frontal region of the skin of the skull into the right orbit. To date no complex solution is known for this pathology. The main complication is described unconnected vascular malformations (23), which thankfully were not confirmed in our patient. A combination of orbital lymphangioma with cranial arteriovenous malformation and thrombocytopenia, indicated as Kasabach-Merritt syndrome, was described (50). In differential diagnostics it is also necessary to consider the etiology of cavernous haemangioma in the case of these lymphatic-venous malformations. It has similar symptoms of ptosis, pathological deterioration of vision and deviation of the eyeball. Unlike the lymphangiomatic etiology of malformation, a well bordered formation is visible on MRI, nevertheless histological verification is necessary for diagnosis (16).

The main representation in the group of five tumours was of orbital lymphangiomas in three boys, in which the result of the therapeutic endeavour was well evident within a short period of time following total and also partial resection of the tumour. A role in the favourable response through the reduction of the tumour after its partial removal was played by the scarry regression and fibrous conversions already before the surgical intervention. The finding was confirmed by a histological examination after one year within the framework of dacrocystorhinostomy and control MRI eight years later. The result of actinotherapy was highly effective, furthermore without any side effect on post-radiation changes in the lens and other intraocular tissues even after 17 years. The basis of the good effect was the use of a linear accelerator, which enabled very precise and targeted focusing of the actinotherapy.

Both methods are fundamental therapeutic procedures (39), but the results of irradiation were still unconvincing during the last century (22). Repeated excision of small lesions with subsequent cryotherapy was also attempted (44). The fundamental procedure still remains surgical solution. Diffuse orbital lymphangioma can be resolved using a Krönlein surgical technique to achieve a good effect with clinical remission using subtotal resection (3). Extraconal lymphangioma can be resected fully effectively without relapse within a short period. In the case of intraocular location of the tumour the possibility is only of a subtotal effect with the danger of neuropathy of the optic nerve (15). Improvement of the success rate of the surgical solution may be assisted by the application of tissue glue to the base of the fibrin in order to increase the effectiveness of resection and prevent relapse (5, 17). General corticosteroid therapy has been used in a number of children only as a supplement to treatment following surgical intervention in the case of lymphangioma of the orbit (48). By contrast, in the case of capillary haemangioma generally administered corticosteroids are the basic therapy (13, 30), sometimes intratumorous application of a depot corticosteroid is also used (9). On the basis of experiences with slerotisation of lymphangiomatic lesions in the region of the throat and face (35, 36) its use has begun in isolated cases also for lymphangioma in the orbital area. Two preparations have been administered generally. The first is OK-432 (commercially Picibanil, Chugai Pharmaceuticals Co.): methanol acetone extract from Streptococcus pyogenes, which activates NK cells, macrophages and T-cytoxic lymphocytes, and regularly serves as an adjuvant to oncological therapy. The second pharmaceutical is the peptide antibiotic Bleomycin (commercially Bleoxane, Bleocin): a product of fungal Streptomyces verticillus, which is a cytostatic interfering through DNA induction of fractures and intercorporation of thymidine, applied in the case of various types of lymphomas. In a comparison of the surgical procedure and application of one of the above preparations, an optimal effect was produced in the case of OK-432 (35). A side effect was a local inflammatory reaction, which however did not lead to damage or scarring of the surrounding tissue, but after an interval of 13 years of sclerotherapy of lymphangioma in children was no longer evaluated as fully effective (36). In the case of simple cysts and macrocysts, the effect of OK-432 was better than surgical intervention, which was itself more effective on microcysts and cavernous formations (38). The use of OK-432 in a dose from 0.02 to 0.05 mg/ml for sclerotisation of lymphangioma of the orbit has so far been only isolated (31, 49, 51). Ptosis appeared transitionally as a side effect (51) and the evaluation was not entirely favourable primarily due to local inflammatory reaction (31). Nevertheless, the method of sclerotisation of this tumour, formerly used primarily in the South-East Asian region due to the higher occurrence of lymphangioma, is continuing to develop. Further options for non-surgical invasive therapy of orbital manifestations of lymphangioma or other vascular lesions are still being assessed. 5% sodium morrhuate solution has been used as cytoreduction of the tumour without significant side effects (47). Chemoablation using ethanol in combination with intracysytic sodium tetradecyl sulphate upon transcutaneous application has been effective as a primary treatment in macrocystic and microcystic lymphomatic malformations of orbits, as well as for relapses following surgical interventions generally in twenty patients (18). Injections of pingyngmycine into lesions of orbital vascular malformations in 13 patients have reduced their vascularity and were accompanied only by a mild inflammatory reaction (52). In therapy of vascular tumours generally administered beta blockers have also been used, e.g. in the case of capillary haemangioma of the orbit (14). They have also been used so far in isolated cases in lymphangioma (40), but according to PUBMED outside of the orbital region, which is in accordance with the paediatric ophthalmological textbook (13).

CONCLUSION

Lymphangioma of the conjunctiva and rare observation of lymphangiomatosis cannot yet be influenced by complex therapy. Extraconal localisation of the tumour was successfully resolved through surgical intervention in two patients through total and subtotal resection respectively. For intraconal lymphangioma with generated neuropathy accompanied by CME a good choice was actinotherapy by linear accelerator, which enabled targeted focus on the tumour without side effects of radiation on the lens and other intraocular structures.

LITERATURE

1. **Anton, M., Holoušová, M., et al.:** Histiocytóza X a dětská očnice. Čs Oftal, 48; 1998: 176–180.
2. **Baráková, D.:** Echografie v oftalmologii. Professional Publishing., Praha, 152 s.
3. **Berthout, A., Jacomet, P.V., et al.:** Surgical treatment of diffuse adult orbital lymphangioma: two case studies. Fr J Ophthalmol, 31; 2008: 1006–1017.
4. **Bond, J.B., Haik, B.G., et al.:** Magnetic resonance imaging of orbital lymphangioma with and without gadolinium contrast enhancement. Ophthalmology, 99; 1992: 1316– 324.
5. **Boulos, P.R., Harissi-Dagher, M., et al.:** Intralesional injection of Tisseel fibrin glue for resection of lymphangiomas and other thin-walled orbital cysts. Ophthal. Plast Reconstr Surg, 21; 2005: 171–176.
6. **Di Emidio, P., Chibbaro, S. et al.:** Double skull lymphangioma. Case report and review of the literature. Orbit, 28; 2009: 293–296.
7. **Dudea, S.M., Seceleanu, A., et al.:** Doppler ultrasound assessment of intraocular and orbital tumors. Oftalmologia, 51; 2007: 87–92.
8. **Eiferman, R.A., Gushard, R.H.:** Chocolate cyst of the orbit. Ann Ophthalmol, 18, 1986: 156–157.
9. **Gerinec, A., Galbavý, Š.:** Efektivnost liečby kapilárného hemangiomu orbity lokálne podávanými kortikosteroidmi. Čs Oftal, 52; 1996: 356–361.
10. **Gerinec, A., Elízová, I., Chynoranský, M.:** Ochorienia orbity u dětí. Čs Oftal, 52; 1996: 279–285.
11. **Gerinec, A., Chynoranský, M., Galbavý, Š.:** Rhabdomyosarkóm orbity. Čs Oftal, 53; 1997: 11–17.
12. **Gerinec, A.:** Diagnostické problémy tumorov orbity u dětí. Folia Strabol Neuroophtalmol, 3; 2000, suppl.I.: 41–43.
13. **Gerinec, A.:** Detská oftalmológia, Osveta, Martin, 2005, s. 204.
14. **Gondová, G., Sejnová, D., et al.:** Liečba kapilárního hemagiomu orbity betablokátormi. Folia Strabol Neuroophthalmol, 10; 2009, suppl. I.: 82–83.
15. **Gündüz, K., Demirel, S., et. al.:** Correlation of surgical outcome with neuroimaging findings in periocular lymphangiomas. Ophtalmology, 113; 2006: 1231–1238.
16. **Gündüz, K., Kurt, R.A.:** Well-circumscribed orbital venous-lymphatic malformations with atypical features in children. Br J Ophthalmol, 93; 2009: 656–659.
17. **Hayasaki, A., Nakamura, H., et al.:** Successful treatment of intraorbital lymphangioma with tissue fibrin glue. Surg Neurol, 72; 2009: 722–724.
18. **Hill, R.H. 3rd., Shields, W.E. 2nd., et al.:** Percutaneous drainage and ablation as first line therapy for macrocystic and microcystic orbital lymphatic malformations. Ophtal Plast Reconstr Surg, 28; 2012: 119–125.
19. **Chynoranský, M., Furdová, A., Olah, Z.:** Exoftalmus podmíněný ochorením slzné žlázy. Čs Oftal, 50; 1994: 48–51.
20. **Chynoranský, M., Furdová, A., Olah, Z.:** Exenteracia očnice. Čs. Oftal., 50, 1994: 92–97.
21. **Illif, V.J.:** Orbital tumors in children. In Jakobiec, F.A.: Ocular and adnexal tumors. Aesculapius, Birmingham, 1978, p. 669–684.
22. **Iliff, V.J., Green, V.R.:** Orbital lymphangiomas. Ophthalmology, 86; 1979: 914–929.
23. **Katz, S.E., Rootman, J., et al.:** Combined venous lymphatic malformation of the orbit (so-called lymfangiomas). Association with noncontiguous intracranial vascular anomalies. Ophthalmology, 105; 1998: 176–184.
24. **Kostolná, J., Gerinec, A., et al.:** Odontogenná dermoidná cysta orbity (kazuistika). Čes a Slov Oftal, 67; 2001: 101–103.
25. **Kozák, J., Pochop, P., Hubáček, M.:** Chirurgické řešení následků onkologické léčby. Čes a Slov Oftal, 59; 2003: 113–118.
26. **Krásný, J.:** Nádory oka a jeho adnex v dětství. III. Orbita. Čes a Slov Oftal, 54; 1998: 50–55.
27. **Krásný, J. Novák, V., Otradovec, J.:** Orbitální protéza po exenteraci očnice se zachováním víček a spojivkového vaku. Čes a Slov Oftal, 62; 2006: 94–99.
28. **Krásný, J., Šach, J., et al.:** Orbitální tumory u dospělých – desetiletá studie. Čes a Slov Oftal, 64, 2008: 219–227.
29. **Krist, P., Plesník, J.:** Maligní lymfom orbity. Čes a Slov Oftal, 58; 2002: 247–253.
30. **Kuchynka, P. a kol.:** Oční lékařství, Grada, Praha, 2007, s. 627 a 650.
31. **Lanuza, G.A., Banon, N.R., et. al.:** Unsuccessful treatment with OK-432 pibanil for orbital lymphangioma. Arch. Soc. Esp Oftalmol, 87; 2012: 17–19.
32. **Lemke, A.J., Kazi, I., et al.:** Differential diagnosis of intraconal orbital masses using High-resolution MRI with surface coils in 78 patients. Rofo, 176; 2004: 1436–1446.
33. **Matoušek, P., Lipina, R. et al.:** Transnazální endoskopická chirurgie nádorů očnice. Čes a Slov Oftal, 68; 2012: 202–206.
34. **Novák, Z., Pábl, L., et al.:** Stereotaktická biopsie nádorů očnice. Čes a Slov Oftal., 53, 1997: 220–222.
35. **Ogita, S., Tsuto, T., et al.:** Treatment of lymphagiomas arising aroud cervico-facial region: Surgery, bleomycin and OK-432 therapy. Nihon Geka Gakkai Zasshi, 90; 1989: 1389–1391.
36. **Ogita, S., Tsuto, T., et al.:** OK-432 therapy in 64 patients with lymphagioma. J Pediatr Surg, 29, 1994: 784–785.
37. Ohtsuka, K., Hashimoto, M., Suzuki, Y.: A review of 244 orbital tumors in Japanese patients during a 21-year period: origins and locations. Jpn J Ophthamol, 49; 2005: 49–55.
38. **Okazaki, T., Iwatani, S., et al.:** Treatment of lymphangioma in children: our experience of 128 cases. J Pediatr Surg, 42; 2007: 386–389.
39. **Otradovec, J.:** Choroby očnice, Avicenum, Praha, 1986, s. 111, 249–251.
40. **Ozeki. M., Kanda, K., et al.:** Propranolol as an Alternative Treatmant Option for Pediatric Lymphatic Malfomation. Tohoku J Exp Med, 229; 2013: 61–66:
41. Pliskvová, J. Pernica, P., Omelková, A.: Rhabdomysarkom orbity. Čs Oftal, 48; 1998: 295–300.
42. **Pochop, P., Čumlivská, E., Kodet, R.:** Využití CT pro lokalizování bioptické jehly při odběru tkáně z očnice. Čes. a Slov. Oftal., 58, 2002: 165–170.
43. **Řehůřek, J., Autrata, R.:** Ke klinické diagnostice rhabdomyosarkomu dětské očnice. Čs. Oftal, 53, 1997: 215–219.
44. **Saraux, H., Laroche, L., et al.:** Lymphangioma of the orbit. J Fr Ophtalmol, 8, 1985: 579–584.
45. **Shields, J.A., Shields, C.L.:** Orbital cysts of childhood-clasification, clinical features, and management. Surv Ophthalmol, 49; 2004: 281–299.
46. **Shields, J.A., Shields, C.L, Scartozzi, R.:** Survey of 1264 patients with orbital tumors and simulating lesions: The 2002 Montgomery Lecture, Part 1., Ophthalmology, 111; 2004: 997–1008.
47. **Schwarcz, R.M., Ben Simon, G.J., et al.:** Sclerosing therapy first line treatment for low flow vascular lesions of the orbit. Am J Ophthamol, 141; 2006: 333–339.
48. **Sires, B.S, Goins, C.R., et al.:** Systematic corticoid use in orbital lymphangioma. Ophtal Plast Reconstr Surg, 17; 2001: 85–90.
49. **Suzuki, Y., Obana, A., et al.:** Management of orbital lymphangioma using intralesional injection of OK-432. Br J Ophtamol, 84; 200: 614–617.
50. **Vachharajani, A., Paes, B.:** Orbital lymphangioma with non-contiguous cerebral arteriovenous malformation, manifesting with trombocytopenia (Kasabach-Merritt syndrome) and intracranial hemorrhage. Acta Paediatr, 91, 2002: 98–99.
51. **Yoon, J.S., Coi, J.B., et al.:** Intralesional injection of OK-432 for vision-threatenig orbital lymphangioma. Graef Arch Clin Exp Ophthalmol, 245; 2007: 1031–1035.
52. **Yue, H., Qian, J., et al:** Treatment of orbital vascular malformations with intralesional injection of pingyagmycin. Br J Ophthalmol, 97; 2013: 739–745.

CASE REPORT

RARE CASE OF PATHOLOGICAL BIOMINERALIZATION OF EYE TISSUE

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SUMMARY

The authors have described the case of pathological biomineralization of ocular posterior chamber. Histological examination of affected eye shells revealed inflammation, oedema, dyscirculatory violations, and signs of dystrophic calcification. Structural-phase and chemical analyses of calcification have revealed that the biomineral consists of hydroxyapatite with relatively small crystallite size and defective crystal lattice. In the described case the formation of pathological biomineral in the vision organ was initiated by penetrating wound. Eye injury might have caused a hemophthalmus and chronic inflammatory reaction in the shells, these processes eventually led to the organ subatrophy and to the development of dystrophic and necrobiotic changes in the tissues. Pathological biomineralization in the affected organ developed as a type of dystrophic calcification..

Key words: eye, pathological biomineralization, hydroxyapatite

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INTRODUCTION

Biominerals formation in the vision organ is a fairly rare pathology. The literature in connection with the processes of biomineralization and calcification described choroidal osteoma, idiopathic choroidal sclerosis, focal tofus-like calcification of the sclera and intraocular osseous metaplasia [1, 2, 3, 4, 5]. Choroidal oseoma is a benign tumor that replaces a choroidal shell by a mature bone. This tumor was first described in 1975. It is more often onesided, but sometimes there is a later affection of the second eye (20%). Etiology of choroidal osteoma is unknown; it is believed that this pathology relates to choristoma [3]. Intraocular osseous metaplasia, according to Vemuganti GK et al (2002), is found in 5.2% of patients who were performed ocular enucleation owing to tuberculosis of eyeball, staphyloma, absolute glaucoma or microphthalmos [5]. The processes of dakrioliths formation in the lacrimal sac were also studied [6]. The reason of the development of pathological mineral formation processes in the vision organ and in the accessory glands has not been determined [1, 2, 6].
The aim of this work is to study the pathological processes of biomineralization in the eye tissues, using pathomorphological methods and applied materials science

MATERIALS AND METHODS

Surgical material was fixed in 10% neutral formalin for further fixation in alcohols at carousel device AT-4M and filling in paraffin blocks, from which 4-6 microns thick sections were manufactured by a rotary microtome "Shandon Finesse 325". Histological sections were stained with hematoxylin-eosin and examined with the use of a light microscope of the company "Carl Zeiss Primo Star". Micrographs were obtained with the help of a digital image output system "SEO Scan ICX 285 AK-F IEE-1394".
Mineral component was isolated by heat treatment at 200 ° C for 1 hour. X-ray diffraction investigations were performed on a diffractometer DRON4 -07 ("Petrel"). Radiation CuK (wavelength 0,154 nm) was used under conditions of Bragg-Brentano focusing (J-2J) (2J – Bragg angle) [7]. The current and the voltage at the X-ray tube were 20 mA and 30 kV, respectively. Samples were shot with the continuous registration (speed 2 °/ min) in the angular range 2J from 10 to 60°. All procedures of the experimental data processing were performed by licensed software support package and processing of the results (DIFWIN-1, TOO "Etalon PTC"). Identification of the crystalline phases was carried out by automatic comparison of the obtained results with the database cards Powder Diffraction File 2 without imposing restrictions on the elemental composition of samples; the software package Crystallographica Search-Match (Oxford Cryosystems) was used in this work.
Scanning electron microscopy with X-ray microanalysis was performed by the instrument REMMA102 (SELMI). In the energy-dispersive microanalysis (EDX) conditions analytical signal accumulated from three fragments of calcifications for 200 seconds at an accelerating voltage of 20 keV and a current probe 3 nA. The sample particles, deposited on a special conductive carbon tape, were coated with a thin conductive film of gold in a vacuum system VUP -5M (SELMI) to prevent the accumulation of electrostatic charge during the investigation of the elemental composition.

CLINICAL CASE

A 61-year-old man was taken to the ophthalmology department of Sumy Regional Hospital in November 2011. The patient complained of cramps, redness, and blindness of the right eye. The pain was observed for 10 days, but he didn't apply for medical

aid. The history tells about a penetrating wound of the right eye in 1965. An ophthalmic status of the right eye: the right eyeball reduced in size, mobility is not limited, visual acuity of 0 (zero), blepharospasmus, lacrimal path is passable. Conjunctiva is hyperemic, there is a mixed injection of the eyeball, and the cornea is edematous and lumpy. The anterior chamber is filled with pus, deeply lying structures aren't visualized. The left eye examination revealed: visual acuity is 0.9, the intraocular pressure is 20 mm, the eyelids are not changed, the lacrimal path is passable, the conjunctiva is pale pink, the cornea is clear, the anterior chamber is depth average, the iris pattern is not changed, the lens has starting opacities in the cortical layers, the vitreous and optic disc are unremarkable. After examination the diagnosis was determined as a penetrating wound exodus (1965), subatrophy, and endophthalmitis of the right eye. The patient had incipient senile cataract of the left eye. Enucleation of the right eye with the formation of the stump was performed.

Pathologic study of surgical material revealed that the eyeball reduced in size to 1,3x0,9 cm, the tone also dramatically reduced. Dirty yellowish color clots were revealed in the anterior chamber. Hardening of the inner lining of the eye, which was almost total, was detected in the posterior chamber. The formation was of a graybrown color, the size was 1,2 x0, 7 cm, had the form of a funnel with a hole of 0.2-0.3 cm diameter at the tapered end, weighing 0,815 g (Fig. 1A). After scorching in a muffle furnace at 200°C for 60 minutes, the formation retained the structure of a funnel and acquired gray-whitish color, the weight was 0.17 g (Fig. 1B).

Histological examination of the sclera revealed the signs of swelling (Fig. 2A), there were signs of intense inflammatory infiltration, congestion of vessels, foci of hemorrhage in choroid (Fig. 2B). Deposits of calcifications, most likely of degenerative genesis, were revealed in the tissue of posterior chamber membranes.

Research of biomineralith using the methods of applied materials science

The decoding of a ray diffraction pattern of the sample of pathological mineral formation of the eye showed a good accordance to the structural data of hydroxyapatite ($Ca_{10}(PO_4)_6(OH)_2$, JCPDS 9-0432). Considerable broadening and overlap of the diffraction peaks shows a low degree of crystallinity of the investigated material, namely the small size of the areas with a regular periodic structure (crystallites or mosaic blocks) and a large proportion of defects, which lead to lattice microstrain. Scherrer's [3] estimating of the crystallites size in the perpendicular direction to the crystallographic plane (002) gave meanings very close to the typical size of bone tissue crystallites (L002=18,2 nm). It should be noted that the line of the longitudinal size of apatite crystals (e.g., 002) are broadened much less than the lines of the transverse size (e.g., 310). It means that the crystals along the hexagonal axis are elongated and have small sizes in their transverse direction. This morphology of biological apatite crystals (like a plate, needle or bar) is typical for bone tissue and similar synthetic biomaterials.

Data of EDX microanalysis showed that the main elements of deposit were calcium and phosphorus in the typical ratio of apatite (Fig. 4, Table 1). Some excess of Ca in the stoichiometric apatite indicates the presence of calcium-fortified non-apatite mineral constituents, which are the source of biocrystal growth, and it is typical for many cases of investigated pathological deposits [8, 9].

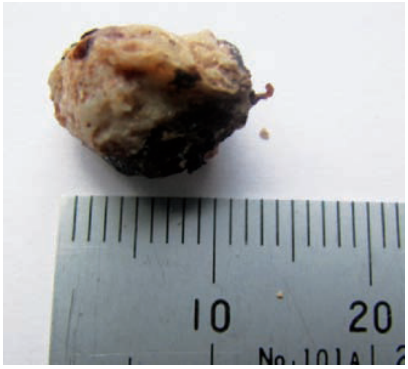

Fig. 1a

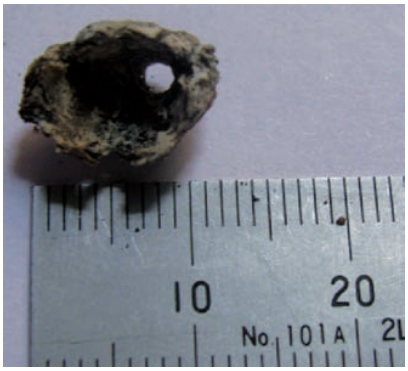

Fig. 1b

Fig. 1 Eye macropreparations after fixation in formalin (Fig. 1A). Biomineral component after heat treatment of the material (Fig. 1B).

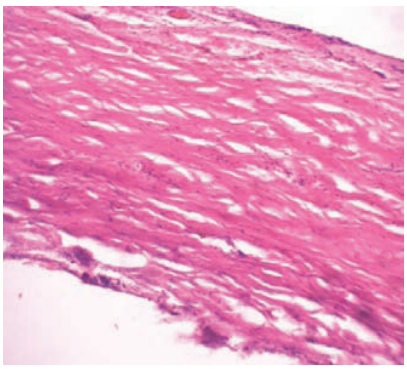

Fig. 2a

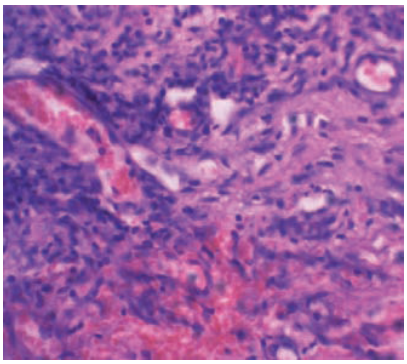

Fig. 2b

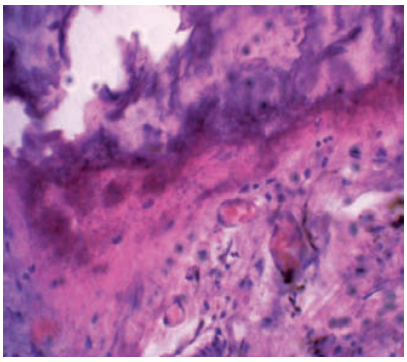

Fig. 2c

Fig. 2 Histological examination of the affected eye. A – the sclera with symptoms of oedema. B – inflammatory infiltration (1), hemorrhage (2) in the choroid . C – calcification in the shells of eye posterior chamber (1). Hematoxylin-eosin. Increase is shown in the lower left corner of each image. Asterisk* points artifact crack, which was formed in the manufacture of sections of mineralized tissue.

DISSCUSION

Chronic inflammation cells, bone morphogenetic proteins, growth factors and stem mesenchymal cells play possible role in the pathogenesis of

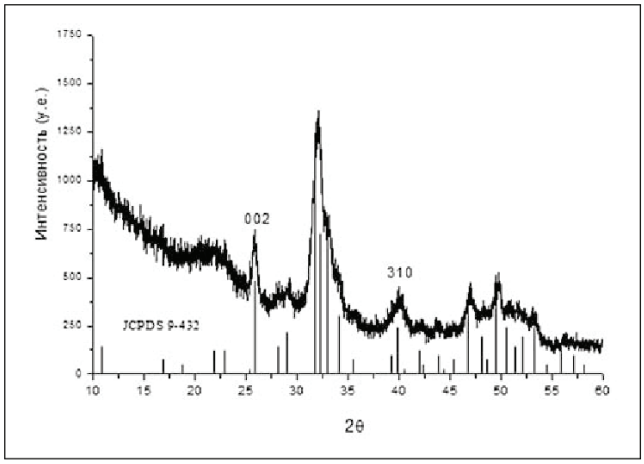

Fig. 3 Original diffraction of mineral material of pathological eye formation, vertical lines indicate the position and intensity of the peaks of hydroxyapatite Ca10(PO4)6(OH)2 according to the reference data JCPDS 9-0432.

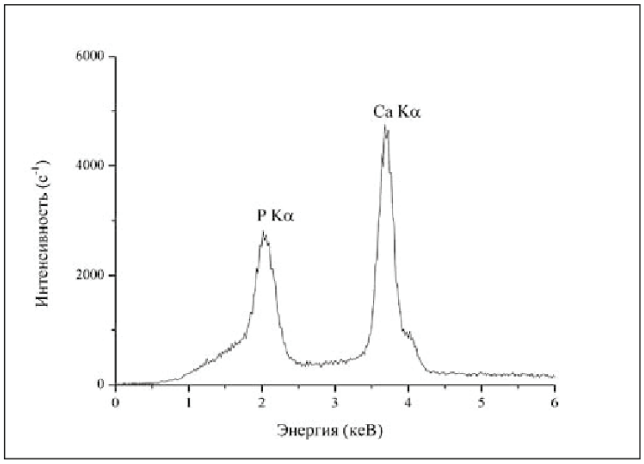

Fig. 4 EDX spectrum of pathological mineral eye formation.

intraocular biomineralization. Under the influence of bone morphogenetic proteins and growth factors mesenchymal stem cells differentiate into osteoblasts. They secrete bone matrix (osteoid), whose regeneration and remodeling lead to the formation of bonelike tissue [10]. In the paper of Rohrbach J. M. et al (1990) it is indicated that intraocular osseous metaplasia is triggered by chronic changes of the Retinal Pigment Epithelium (RPE). The primary lesion (e.g. trauma, inflammation or tumor) plays a minor or no role [11].

The peculiarity of the investigated case is a significant size of ocular lesion and a long period of the process development (46 years). Of course, the process of biomineralization in the tissues grows slowly, but this long period of development inclines us to believe that a key element in the development of ocular calcification is the Retinal Pigment Epithelium (RPE). We believe that the biomineralization processes in the investigated case should not be called intraocular ossification or bone formation, but dystrophic calcification. Firstly, structures characteristic of bone (formation of bone marrow, Havers tubules) have not been observed in the ocular tissues. Secondly, the data about a small size of the crystals of hydroxyapatite and defective crystal lattice, which were received using the methods of applied material science, indicate the development of dystrophic biomineralization, but not bone tissue, in the ocular tissues.

Table 1 The concentrations of Ca and P in three areas of the investigated calcifications according to EDX analysis (wt.%).

Analysis area	P	Ca	Ca/P
1	8.967	22.910	2.55
2	12.018	25.682	2.14
3	11.396	26.202	2.30

CONCLUSION

Detection of pathological biomineralization in the vision organs of patients is a rare event, thus being of great interest. In this case the treatment of the patient in a conservative manner was futile and posed a threat to the healthy eye. Histological examination of the affected eye shells revealed inflammation, oedema, dyscirculatory violations, and signs of dystrophic calcification.

Structure-phase and chemical compositions of biomineral were established using the methods of applied materials science. Presence of apatite crystal phase with relatively small crystallite size and defective lattice in pathological formation is quite typical for many cases of ectopic biomineralization, but in our opinion, for the first time it was described for ocular calcifications and using the methods of applied materials science.

In the described case the pathological biomineral formation in the vision organ was initiated by penetrating wound. Eye injury probably led to hemophthalmus and chronic inflammatory reaction in the shells, these processes eventually led to organ subatrophy, development of dystrophic and necrobiotic changes in the tissues. Pathological biomineralization in the affected vision organ developed as a type of dystrophic calcification. The value of presented work is not only a rare case, but also the opportunity to raise the ophthalmologists' knowledge of morphogenesis of pathological biomineralization in the diseases of vision organ.

LITERATURE

1. Daicker B.: Tophus-like, conglomerated, crystalline calcification of the sclera. Ophthalmologica, 1996; 210(4): 223–8.
2. Chen KH, Li MJ, Cheng WT. et al.: Identification of monoclinic calcium pyrophosphate dihydrate and hydroxyapatite in human sclera using Raman microspectroscopy. Int J Exp Pathol, 2009; Feb;90(1):74–8.
3. Pandey N., Guruprasad A. Choroidal osteoma with choroidal neovascular membrane successful treatment with intravitreal bevacizumab. Clinical Ophtalmology, 2010; 4, :1081–84.
4. Bourcier T, Blain P, Massin P. et al.: Sclerochoroidal calcification associated with Gitelman syndrome. Am J Ophthalmol, 1999; Dec;128(6): 767–8.
5. Vemuganti G.K, Honavar S.G, Jalali S.: Intraocular osseous metaplasia. A clinico-pathological study. Indian J Ophthalmol, 2002 Sep; 50(3): 183–8.
6. Karakeshinian . [Dacrolithiasis]. Noviy armianskiy meditsinskiy jurnal [New Armenian Medical Journal]. 2011; 5 (3):71–72. (In Russ).

7. Klug H.P., Alexander L.E. X-Ray diffraction procedures for polycrystallite and amorphous materials.-New York: JohnWiley and Sons, 1974.-965p.
8. Moskalenko R., Romanyuk A., Danilchenko S. et al.: Morphogenetic aspects of biomineralization on the background of benign prostatic hyperplasia. Georgian medical news, 2013; 214 (1): 54–61.
9. Danilchenko S.N., Kuznetsov V.N., Stanislavov A.S. et al.: The mineral component of human cardiovascular deposits: morphological, structural and crystal-chemical characterization. Crystal research and technology, 2013, 48 (3): 153–62.
10. Munteanu M, Munteanu G, Giuri S, Zolog I, Motoc A.G.: Ossification of the choroid: three clinical cases and literature review of the pathogenesis of intraocular ossification. Rom J Morphol Embryol, 2013;54(3 Suppl): 871–7.
11. Rohrbach J.M, Liesenhoff E, Steuhl K.P.: [Principles of intraocular ossification exemplified by secondary choroid ossification]. Klin Monbl Augenheilkd, 1990 Nov;197(5): 397–403.