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ORIGINAL RESEARCH ARTICLE

Electrophysiological testing as a method of cone–rod and cone dystrophy diagnoses and prediction of disease progression

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Abstract
Purpose To determine the characteristics of patients with cone (CD) and cone–rod dystrophies (CRD) and to evaluate the changes in flash electroretinograms in both groups.
Methods The retrospective study involved 48 patients—34 with CRD and 14 with CD. The patients underwent full ophthalmological examination, including Goldmann perimetry and full-field flash electroretinogram (FERG) within the initial examination. These examinations were then repeated seven, or more, years later. The longest follow-up period was 10 years, with the mean at 8.2 years. During both examinations, we assessed the amplitudes of the b wave in the scotopic ERG test 0.01 (which reflects rod response), the maximal scotopic ERG test 3.0 (which reflects cone and rod response) and the photopic 3.0 ERG test (which reflects cone response). The results were then compared against normal values.
Results The progression over time of ERG b wave amplitudes in the scotopic ERG 0.01, maximal scotopic ERG 3.0 and photopic ERG tests was assessed. There were significant differences in rod, maximal and cone responses, between CD and CRD patients. While rod responses were markedly decreased in CRD patients during their initial examination, the decrease in the rod function in both CD and CRD patients was similar in their follow-up examination ($p = 0.2398$). Moreover, during initial examination, maximal responses were less common amongst CRD patients, over those with CD. Following the observation period, patients suffering from CRD exhibited a significant decrease in both maximal ($p = 0.0125$) and cone ($p = 0.0046$) responses.
Conclusion The clinical course of CRD and CD may vary; however, the latter appears to have a more favourable course than former. Although, at initial examination, the cone function was more diminished in CD patients, the final examinations reveal a more significant drop for CRD patients. Consequently, a differential diagnosis is essential for treating patients and forecasting their disease progression.

Keywords Cone dystrophy · Cone–rod dystrophy · Electroretinogram (ERG)

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Introduction

Cone–rod (CRD) and cone dystrophies (CD) are a heterogeneous group of hereditary, progressive retinal diseases that are characterized by the degeneration of photoreceptors [1, 2]. Histopathology studies have demonstrated cell death, decreasing outer segments of photoreceptors and abnormal photoreceptor synapses

[3]. CRD and CD may have autosomal dominant, autosomal recessive or X-linked inheritance, as several causative genes have been described in the literature [4].

The typical age range for the appearance of first symptoms is between the first and third decades of life [5]. The principal symptoms include bilateral, progressive visual loss. Photophobia, colour vision abnormalities and nystagmus also appear for those suffering from CD. Meanwhile, with CRD, the central vision impairment is followed by nyctalopia. Colour vision abnormalities are also present, as well as, variable degrees of photophobia [1, 6].

Typical eye fundus manifestation in CD sufferers consists of bull's eye maculopathy; however, the fundus in patients with CRD may be within normal limits or only has subtle changes in the macula such as minor atrophy or pigmentation. Retinal changes in CRD cases may also be similar to retinitis pigmentosa with bone spicule-like pigmentation [7–10].

A visual field examination of a CD patient shows central scotoma, while subsequent peripheral ring scotomas appear in CRD cases. A fluorescein angiogram of patients with CRD may reveal a window defect with no leakage [8].

A full-field electroretinogram (ERG) of CD patients shows severe cone function impairments, with normal rod responses, whereas maximal responses reveal an absence of b waves. ERG of CRD patients reveals both cone and rod function impairment [11, 12].

In both CD and CRD cases, the disorder's clinical presentation may vary, which could result in inaccurate diagnoses. Symptoms and signs may be mild to moderate and often nonspecific. In such cases, patients may be misdiagnosed as having other diseases, particularly if only experiencing very mild changes to the macula [7, 9].

Aim

The aim of this study was to determine the characteristics of patients with CD and CRD and to evaluate the changes in flash electroretinogram results in both groups.

Materials and methods

The retrospective study involved 48 patients. Fourteen patients (five female and nine male individuals) had been diagnosed CD, while 34 patients (18 female and 16 male individuals) with CRD. The study included patients who had been diagnosed at our hospital and had been experiencing symptoms for under a year. The primary diagnosis was established upon clinical manifestation of the disease and ERG results. The inclusion criteria for CD were a progressive decline of visual acuity, colour vision disturbances and reduced cone amplitudes on the ERG, with rod responses remaining within normal limits. Similarly, inclusion criteria for CRD were a progressive decline of visual acuity, colour vision disturbances and reduced cone and rod amplitudes on the ERG, where cone responses were more severely reduced than rod responses. This was done in order to exclude patients with rod–cone dystrophy. Exclusion criteria for the study consisted of concomitant eye disorders, such as congenital nystagmus, ocular media opacities, ocular trauma or a history of ocular surgery, as well as systemic disorders.

The patients were observed at the ophthalmology department of the Medical University of Warsaw for at least 7 years. The longest follow-up period was 10 years, with the mean being 8.2 years.

Besides typical clinical examinations, the patients all underwent visual field examinations, fluorescein angiographies and flash ERG. Their best-corrected visual acuity (BCVA) was assessed on Snellen charts. Colour vision was assessed with the use of Panel D-15 tests, while their visual field was assessed by the use of the Humphrey perimeter and FF120 programme. A flash full-field electroretinogram (FERG) was performed following ISCEV standards [14], using the RetiScan RetiPort system (Roland Consult). ERG results were compared with normal values.

The control group consisted of 40 healthy individuals, students and medical professionals, whose age range and collective refractive errors were similar to the examined group. The control group was randomly selected from 200 potential subjects, thereby representing a reference group for electrophysiological testing for our department. The authors assessed amplitudes of b wave in the scotopic ERG test 0.01, maximal scotopic ERG test 3.0 and photopic ERG 3.0 test, during both initial and final examinations. In each patient, the values from the right and left eyes were averaged. Such averaging of variables prevents the correlation of observations. Associations between numerical variables were examined using the Mann–Whitney U test. The significance criterion was set at the level of $p < 0.05$. Statistical analysis was

Table 1 Baseline characteristics of CD and CRD patients

	Cone–rod dystrophy (total number, $n = 34$)	Cone dystrophy (total number, $n = 14$)
Mean age at initial examination, *n* (SD)	29 (11)	32 (12)
Mean age at onset, *n* (SD)	14 (8)	18 (9)
Gender		
Male, *n* (%)	18 (53)	9 (65)
Female, *n* (%)	16 (47)	5 (35)
Refractive error		
>+6 D	1	0
+2/+6 D	4	2
−2/+2 D	17	6
−2/−6 D	10	5
<−6 D	2	1

performed using Stata Statistical Software: Release 10, Stata Corporation LP 2007.

Genotyping was not undertaken

This research was conducted according to the tenets of the Declaration of Helsinki, and all patients had provided full, informed consent following a thorough explanation of all examinations.

Results

A baseline characteristic of all patients is summarized in Table 1. While the initial BCVA ranged from 0.2 to 0.6 on the Snellen chart in the CD group, the CRD group demonstrated a wider range of BCVA, from counting fingers to 1.0 on the Snellen chart. The mean change in BCVA following the observation period was a two-row reduction on the Snellen chart in both groups.

All patients had been observed for at least 7 years. The longest follow-up period was 10 years, with the mean period being 8.2 years (SD 0.85) in CRD patients and 8.3 years (SD 0.95) in the CD patients. There were no significant differences in follow-up period in either the CD or CRD group.

The representative ERG waveforms for patients with CD and CRD, in comparison with normal values, are shown in Fig. 1.

Table 2 shows the clinical findings of the examinations of CD and CRD patients at both the initial and final checkup.

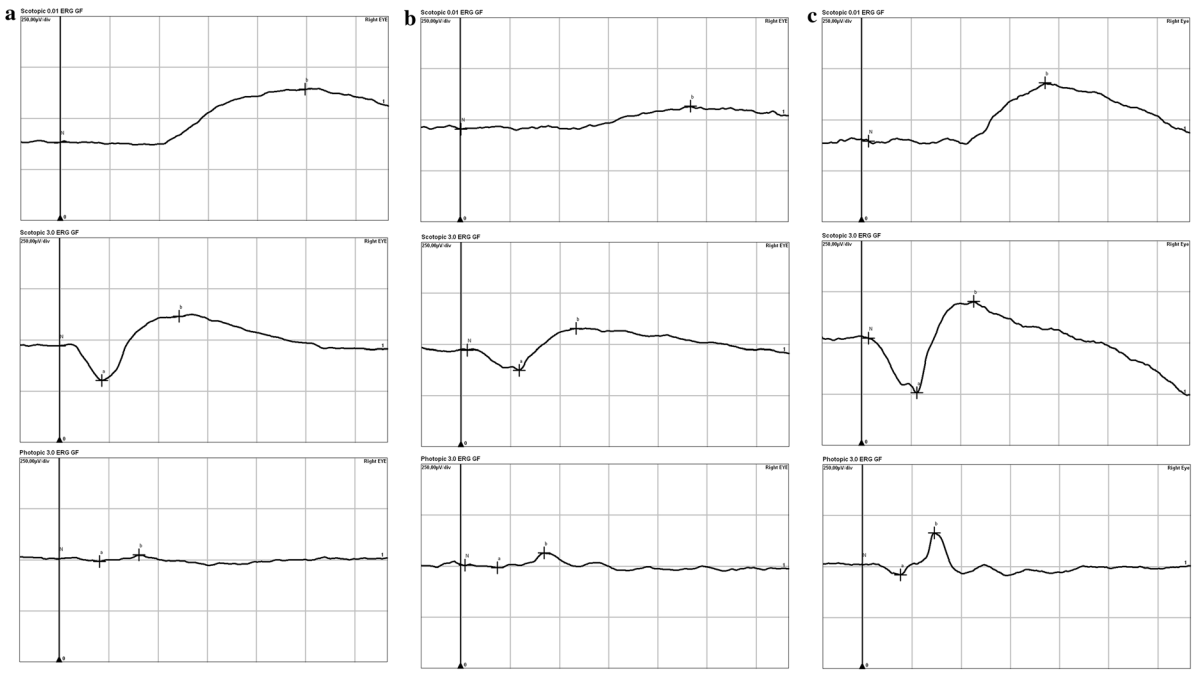

Fig. 1 Representative ERG waveforms (scotopic 0.01, scotopic 3.0 and photopic 3.0) for patients with CD (**a**) and CRD (**b**), in comparison with normal values (**c**)

Table 3 shows b wave amplitudes in scotopic 0.01, maximal scotopic 3.0 and photopic 3.0 tests at the initial and final examinations and their statistical analyses. This is represented in Figs. 2, 3 and 4. Comparing the CD and CRD patients, there were significant differences in rod, maximal and cone responses in both initial and final examinations. We did not observe any changes in ERG responses in the control group during the follow-up period.

Rod responses were markedly decreased in patients with CRD, whereas those with CD were within normal limits. During initial examination of rod responses, the mean b wave amplitude in CRD sufferers was 67.91 μV (±SD 13.18), while for CD patients, it was 180.71 μV (±SD 35.03). During the final rod response examination, the mean b wave amplitude was 54.94 μV (±SD 12.33) and 173.14 μV (±SD 37.83) for CRD and CD patients, respectively. The decrease in the rod function in CD and CRD patients was similar in during the follow-up period ($p = 0.2398$). None of patients with CD developed rod involvement (i.e. amplitudes under the lower limit of normal values) during the observation period.

Maximal responses had dropped more significantly in CRD patients than in those with CD. The maximal responses for all CRD patients were under the lower

Table 2 Clinical findings of CD and CRD patients at the time of initial and final examination

	CRD at initial examination (total number, $n = 34$)	CRD at final examination (total number, $n = 34$)	CD at initial examination (total number, $n = 14$)	CD at final examination (total number, $n = 14$)
Macular appearance, *n*/*N* (%)				
Normal	4/34 (12)	2/14 (6)	1/14 (7)	0/14 (0)
Pigmentary changes	12/34 (35)	14/34 (41)	3/14 (21)	4/14 (28)
Bull's eye	18/34 (53)	18/34 (53)	10/14 (72)	10/14 (72)
Visual field, *n*/*N* (%)				
Central scotoma	32/34 (94)	34/34 (100)	14/14 (100)	14/14 (100)
Peripheral scotoma	21/34 (61)	34/34 (100)		

Table 3 The b wave amplitudes in μV in scotopic 0.01, scotopic 3.0 and photopic 3.0 responses in CRD and CD groups, at initial (IE) and final examination (FE) and the change of values in the observation period (SD = standard deviation, P50 = median)

Variable	*N*	Min	Max	Mean	SD	P50
b Wave scotopic 0.01 CRD IE	34	43.00	90.00	67.91	13.18	69.00
b Wave scotopic 0.01 CRD FE	34	32.00	76.00	54.94	12.33	52.00
Change scotopic 0.01 CRD	34	−24.00	−1.00	−12.97	4.65	−14.00
b Wave scotopic 0.01 CD IE	14	147.00	259.00	180.71	35.03	164.50
b Wave scotopic 0.01 CD FE	14	132.00	247.00	173.14	37.83	163.00
Change scotopic 0.01 CD	14	−22.00	22.00	−7.57	12.48	−12.00
b Wave scotopic 3.0 CRD IE	34	142.00	221.00	179.94	21.68	178.00
b Wave scotopic 3.0 CRD FE	34	122.00	210.00	155.59	22.05	154.00
Change scotopic 3.0 CRD	34	−47.00	−11.00	−24.35	8.85	−22.50
b Wave scotopic 3.0 CD IE	14	235.00	366.00	288.57	38.61	280.00
b Wave scotopic 3.0 CD FE	14	220.00	304.00	270.36	32.53	271.00
Change scotopic 3.0 CD	14	−62.00	8.00	−18.21	16.19	−16.00
b Wave photopic 3.0 CRD IE	34	39.00	82.00	66.09	10.46	66.00
b Wave photopic 3.0 CRD FE	34	32.00	69.00	50.09	10.28	50.00
Change photopic 3.0 CRD	34	−37.00	−6.00	−16.00	6.64	−15.00
b Wave photopic 3.0 CD IE	14	20.00	66.00	43.14	14.02	41.50
b Wave photopic 3.0 CD FE	14	16.00	56.00	32.21	11.28	31.00
Change photopic 3.0 CD	14	−20.00	−4.00	−10.93	4.68	−10.00

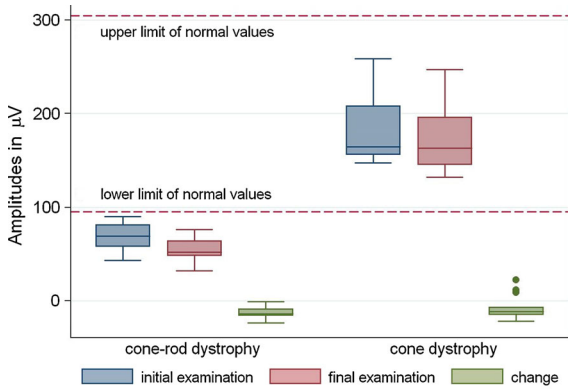

Fig. 2 The b wave amplitude of scotopic 0.01 responses at initial and final examination in the group of CRD ($n = 34$) and CD ($n = 14$) patients, in comparison with normal values, and the change of responses in the observation period

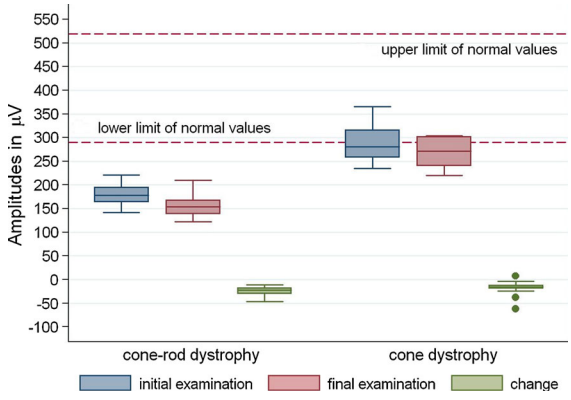

Fig. 3 The b wave amplitude of scotopic 3.0 responses at initial and final examination in the group of CRD ($n = 34$) and CD ($n = 14$) patients, in comparison with normal values, and the change of responses in the observation period

limit during both the initial and final examinations. From the CD patients, six individuals showed maximal responses that were under the lower limit at both the initial and final examinations. At the initial maximal response examinations, the mean b wave amplitude in CRD patients was 179.94 µV (±SD 21.68), while it was 288.57 µV (±SD 38.61) for CD patients. During the final maximal response examination, the mean b wave amplitude was 155.59 µV (±SD 22.05) and 270.36 µV (±SD 32.53) for CRD and CD patients, respectively. The change in maximal responses during the observation period between CRD and CD patients was statistically significant ($p = 0.0125$), with the decrease being more apparent in those with CRD.

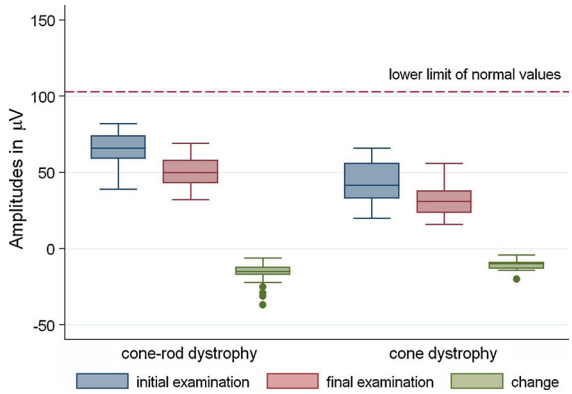

Fig. 4 The b wave amplitude of photopic 3.0 responses at initial and final examination in the group of CRD ($n = 34$) and CD ($n = 14$) patients, in comparison with normal values, and the change of responses in the observation period

Meanwhile, although cone responses were decreased in both groups, this was particularly apparent for patients with CD. At the initial cone response examination, the mean b wave amplitude in CRD patients was 66.09 µV (±SD 10.46), while it was 43.14 µV (±SD 14.02) for those with CD. During the final cone response examination, the mean b wave amplitude in CRD patients was 50.09 µV (±SD 10.28), and 32.21 µV (±SD 11.28) for the CD group. The decrease in cone responses during the observation period was more significant in the CRD group ($p = 0.0046$).

Discussion

For patients with retinal dystrophies, their BCVA depends on underlying genetic mutations, visual field changes (i.e. central scotomas), their age at the onset of the disorder and photoreceptor functional changes. It was the aim of our study to assess the rod and cone functions in CD and CRD patients. The progression of functional changes is particularly important to be able to predict the timing of visual blindness. Patients with CD had a slightly more favourable disease course than those with CRD. The mean age of reaching legal blindness is 48 for patients with CD and 35 for patients with CRD [6, 8]. In CRD patients, the second stage of the disease brings more apparent nyctalopia and progressing ring scotoma, so patients may experience

problems in moving autonomously. In our study group, all CRD patients presented central and peripheral scotomas by the time of their final examination.

The majority of published studies describe only the phenotypes of CD and CRD in relation to underlying genetic mutations [4, 8, 13]. Although some authors have described the progression of the diseases based on visual acuity and visual field changes, there are a limited number of articles concerning the evaluation of ERG in large groups [8, 15].

In our study, rod responses were diminished in the CRD group; however, the decrease in rod function was similar in both the CD and CRD groups during the follow-up period. We did not observe rod involvement in patients with cone dystrophy, as amplitudes were within the normal values during final examination—however, it should be emphasized that the mean follow-up period was 8.2 years. Conversely, the literature demonstrates that when the mean follow-up period becomes 19 years, the observed rod involvement in CD patients becomes 37 % [8]. In the group with RPGR gene mutations, rod function remained unaffected in 24 of 25 patients. Only one patient experienced slightly diminished rod responses [6]. The involvement of rod systems may occur in some cases of cone dystrophy, so regular electrophysiological evaluations should be taken into the consideration. Such patients should be diagnosed as suffering from CRD [10, 16].

The maximal responses in all CRD patients were under the lower limit at both initial and final examinations. Meanwhile, for those with CD, 43 % of cases showed maximal responses that were under the lower limit at both initial and final examinations, and 57 % of cases showed maximal responses that were within normal limits. The decrease in maximal responses during the observation period was more apparent in the CRD group.

Although cone functions were more diminished in the CD group at initial examination, the cone function decrease was more significant in the CRD group during the observation period.

In conclusion, we would like to emphasize that CD and CRD patients have different patterns of retinal function change. The decrease in maximal and cone responses in ERG testing was more apparent in CRD patients. The clinical course of CRD and CD may vary, and consequently, an accurate diagnosis is essential for treating patients and forecasting their disease progression, which appears to have a more favourable course for those with CD rather than CRD.

We believe that in future prospective studies, a large group of patients with a follow-up period of over 20 years should be taken into consideration, in order to describe retinal functional changes more accurately.

Conflict of interest The authors had no financial or proprietary interest in any of the products, materials or methods mentioned.

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