Assessment of macular function by multifocal electroretinogram in age-related macular degeneration before and after photodynamic therapy

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INTRODUCTION

Choroidal neovascularization (CNV) is a leading cause of central vision loss in developed countries [1]. The neovascularization originates from choroidal blood vessels that grow through Bruch’s membrane, usually at multiple sites into the subretinal pigmented epithelial space, and disrupts the anatomy of the retinal pigment epithelium-photorceptor complex [2-4]. While approximately 10%-15% of cases of ARMD involve leaky neovascularization forming in the choroid behind the macula, this neovascularization accounts for roughly 90% of the cases of severe and relatively rapid vision loss attributable to ARMD.

The potentially poor natural history of many subfoveal CNV lesions and the limitations of thermal laser photocoagulation for these lesions have prompted the search for alternative treatment modalities, including photodynamic therapy [5-9], which involves the intravenous injection of a photosensitizer followed by irradiation of the neovascular tissue to be treated by nonthermal light at one of the red or near infrared absorption peaks of the dye. In neovascular tissue, cellular damage to the endothelium can lead to thrombosis within the vessels. Experiments in normal monkey eyes demonstrated minimal damage to photoreceptors and verteporfin dose-dependent reco-

Purpose: To evaluate macular function, before and after photodynamic therapy (PDT) using multifocal electroretinogram (MF-ERG), in eyes suffering from classic subfoveal choroidal neovascularization (CNV) resulting from age-related macular degeneration (ARMD).

Methods: Twenty eyes of 20 patients (10 male, 10 female) with classic subfoveal CNV resulting from ARMD were studied. All received PDT with verteporfin. Fluorescein angiography and MF-ERG were performed in each patient 1 day before and 1 week, 3 months and 6 months after photodynamic therapy.

Results: Before treatment, visual acuity (VA) and electrical retinal response densities (RRD) in the foveal and parafoveal areas were low in all patients. The mean VA (ETDRS chart) before PDT was 24.35 (SD, 15). The mean RRD before PDT in area 1 was 4.39 nV/deg² (SD, 2.59) and in area 2 it was 2.11 (SD, 1.86). Six months after treatment, the mean VA was stable in 70% of the patients. However, the mean RRD after PDT was 2.24 nV/deg² in area 1 (SD, 2.59) and 1.07 nV/deg² in area 2 (SD, 1.59).

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very of the retinal pigment epithelium (RPE) and chorio-
capillaris [10, 11].

Visual function before and after PDT is assessed by
visual acuity (VA) measurement. However, visual acuity
is a single parameter of the impaired visual function re-
sulting from CNV. Visual field testing and contrast sen-
sitivity are other methods to evaluate the results of PDT.

The purpose of this study was to record the electro-
retinographic changes of the foveal and perifoveal
areas by means of MF-ERG in eyes with classic subfoveal
CNV resulting from ARMD, before and after photody-
amic therapy, and to assess its efficacy objectively.
MF-ERG, introduced by Sutter and Tran [12], provides
the simultaneous derivation of 61 or 103 local ERG si-
gals in a central visual field 30° in diameter around the
fovea. This technique allows functional mapping of the
retina and contributes to the detailed evaluation of re-
tinal function, especially in regional disorders of the in-
ner retinal layers.

## PATIENTS AND METHODS

### Population

Twenty eyes of 20 patients with classic subfoveal CNV
resulting from ARMD were studied. These cases were
selected among 145 patients receiving photodynamic
therapy during the period 2000-2002. In the 20 eyes
included in our study, a remission of CNV leakage was
observed 6 months after treatment. In the remaining
125 eyes, a CNV leakage was still present 6 months
after treatment and these patients were excluded from
the study.

Choroidal neovascularization was judged to be asso-
ciated with ARMD when the patient was older than
50 years and drusen or other retinal pigment epithelial
abnormalities commonly associated with ARMD were
seen on the photographic documentation.

Ten patients were male and ten female, between
53 and 80 years old (mean age, 69.05 years) and the
best-corrected visual acuity measured on an ETDRS
chart at screening ranged preoperatively from 4 to
49 (table I).

The study protocol was approved by the local institu-
tional review board of our hospital. All patients signed
a written consent statement before entering the study.

Best-corrected visual acuity was measured on a Bailey-
Lovio chart (ETDRS, number of letters) and the Snellen
equivalent is shown in table II. In addition, a standardiz-
ed refraction protocol was carried out at baseline and at 1
week, 3 months and 6 months. Patients were included
when it was determined by fluorescein angiography that
they had specific characteristics of CNV. The CNV lesion
had to be no greater than nine macular photocoagula-
tion study disc areas and showed a pattern of classic
CNV, with no evidence of occult CNV.

All patients received verteporfin therapy, at a dose of
6mg/m² of body surface, administered via intravenous
infusion of 30ml over 10 min. Fifteen minutes after start-
ing the infusion, a 689-nm nonthermal (diode) laser
light delivered 50 J/cm² at an intensity of 600mW/cm²
for 83 s using a spot size with a diameter 1000µm lar-
ger than the greatest linear dimension of CNV.

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* ETDRS charts.
All patients were examined by MF-ERG on the same day before PDT treatment and 1 week, 3 months and 6 months after PDT treatment. Patients with other ocular diseases such as glaucoma, generalized retinal degeneration and cataract were excluded from our study, so that the first-order responses of MF-ERG recording would not be influenced by the above-mentioned diseases.

**Multifocal ERG recording**

The VERIS III (Visual Evoked Response Imaging System, Tomey, Nagoya Japan) was used to record the multifocal ERG. The stimulus matrix consisted of 61 hexagon elements displayed on a cathode ray tube, a color monitor driven at a frame of 75Hz. These hexagons elicit approximately equal signal amplitude at all locations on a normal retina.

Each hexagon was independently alternated between black and white at a rate of 75Hz and this stimulation technique provided a retinal response for each stimulus. The luminance of the stimulus for white was 200cd/m² and the contrast was 99.3%. The radius of the stimulus array subtended approximately 20° high and 25° wide. The bandwidth of the amplifier was 10-300Hz and the amplification 10,000×.

The subjects pupils were dilated by means of Tropicamide 0.5% and phenylephrine 5% and the eyes optically corrected for near vision. Because all patients had poor central vision, a spoke-shaped (filled cross) fixation aid was used and each patient was instructed to fixate at the intersection of the spokes. For signal derivation, a bipolar contact lens was used in which the reactive and the reference electrodes were incorporated in the contact lens. The ground electrode was attached to the ear lobe. The opposite eye was closed and the duration of data acquisition was 4 min divided into eight 30-s sessions.

The response density (amplitude per unit of retinal area, nV/deg²) of each local response was estimated as the dot product between the normalized response template and each local response. (fig. 1) shows a MF-ERG recorded from a normal subject. The normal ranges for the amplitudes were defined by calculating the median and the 95% confidence intervals in one eye of 30 volunteers aged 35-50 years (mean age, 38.8 years). MF-ERG stimuli location and anatomical areas correspond roughly as follows: ring 1 to the fovea, ring 2 to the parafovea, ring 3 to the perifovea, ring 4 to the near periphery and ring 5 to the central part of the middle periphery. The amplitude of each group is scaled to reflect the angular size of the stimulus hexagon, which produces the response. These averages give a more accurate view of the relative response density of each group. The average relative response density of the retinal area corresponding to ring 1 (fovea) is roughly 20.03 nV/deg²; to ring 2, (parafovea) 15.04 nV/deg²; to ring 3, 12.8 nV/deg²; to ring 4, 10.1 nV/deg²; and to ring 5, roughly 10.0 nV/deg². The retinal response density decreases with eccentricity, although there is no further decrease from ring 4 to ring 5.

### Results

One week after treatment, fluorescein angiography showed the same decrease in CNV leakage in all patients. Three months after PDT, minimal leakage was still observed in five cases and the PDT was repeated. Six months after PDT, a complete absence of CNV leakage was observed in all cases included in the study.

The initial visual acuity at screening was 30-49 in seven eyes (35%), 15-29 in four eyes (20%) and 13 or less in nine eyes (45%) (table I). The final visual acuity 6 months after treatment ranged from 1 to 64. More precisely, visual acuity was 30-42 in four eyes (20%), 15-29 in four eyes (20%) and 14 or less in 11 eyes (55%). In only one eye (case no. 16), visual acuity was 64 (5%).

Figure 2 shows the distribution of eyes with improvement, stability or worsening of visual acuity after treatment. In one eye (case 16), the final visual acuity was higher than before treatment. In 14 eyes (70%), visual acuity remained the same as before treatment.
and in the remaining five eyes (25%), visual acuity showed a decrease after treatment.

**MF-ERG results**

The results of MF-ERG are summarized in table III, which shows that before treatment the mean retinal response density in area 1, representing the fovea, was 4.39 nV/deg² (SD, 2.59), and in area 2, representing the parafoveal area, 2.11 nV/deg² (SD, 1.86).

One week after the PDT, these values decreased to 1.97 nV/deg² (SD, 1.35) for area 1 and 1.08 nV/deg² (SD, 1.39) for area 2.

Three months after PDT, the mean retinal response density in area 1 was 2.36 nV/deg² (SD, 1.81) and in area 2 it was 1.22 nV/deg² (SD, 1.5).

At the end of follow-up, 6 months after PDT, the mean value of retinal response density in area 1 was 2.24 nV/deg² (SD, 2.59) and in area 2 1.07 nV/deg² (SD, 1.59) (fig. 3).

*Figure 4* shows schematically that the retinal response densities decreased 1 week after PDT. Three months later, although there was an improvement in the electrophysiological values, these remain lower than before PDT. At the end of the follow-up, 6 months after PDT, the mean retinal response density remained lower than before treatment.

**DISCUSSION**

Various studies have claimed that PDT reduces the risk of severe vision loss in patients with classic subfoveal CNV resulting from ARMD [6-9]. The overall beneficial outcome with verteporfin therapy has until now been...
Table III
Retinal response densities of MF-ERG before and after PDT in eyes with classic subfoveal CNV resulting from ARMD.

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based on visual acuity, contrast sensitivity, visual fields and fluorescein angiography. However, the first three parameters are subjective and the last one provides anatomical support to visual acuity results, based on the extent of the retinal pigment epithelium lesions and its atrophy [7, 8].

Although some patients show a satisfactory recovery or stability of visual acuity after treatment, the majority continues to complain of abnormal color vision, metamorphopsia and light sensation, symptoms equally important to the amelioration of visual acuity. This means that despite the improvement or the stability of visual acuity, the overall central retinal vision remains abnormal and other types of visual function test should be adopted for the interpretation of macular function.

In our study, the first-order response of MF-ERG was recorded before and after successful PDT in eyes with CNV resulting from ARMD. With this method, the electrical activity of the photoreceptors and bipolar cells of the foveal and parafoveal area is assessed and an objective evaluation of central retinal function is obtained.

Our results show that in these patients, despite the improvement in visual acuity in one eye and the stability of visual acuity in 70% of the eyes after treatment, the mean visual acuity postoperatively was lower than before treatment. In other words, the degree of the decrease in visual acuity, in the eyes where this occurs, is greater than the degree of improvement or stability.

Furthermore, the electrophysiological study of the cases included shows the same results. The mean retinal response density of area 1 and 2 after PDT was lower than before treatment.

According to previous studies, the MF-ERG responses over the affected area of the CNV complex showed moderate to high statistical correlation with visual acuity [7]. Our study observed a discrepancy between the MF-ERG findings and the best-corrected visual acuity. This could be because the retinal response densities in areas 1 and 2 reflect not only the integrity of the photoreceptors of the fovea, which are few in number and responsible for visual acuity, but also the electrical responses of the bipolar and the Muller cells of the inner layers of the parafoveal area [13]. Therefore, in eyes where the CNV affects the juxtapfoveal region more than the fovea, despite higher visual acuity, the recorded MF-ERG is worse. This suggests that the retinal response densities also depend on the extent and the site of the lesion and not only on visual acuity.

The decrease in the electrical activity of the fovea and parafovea after PDT in eyes with ARMD cannot be easily explained. It could be attributed to severe damage to the photoreceptors or bipolar cells of these areas caused by PDT. This hypothesis is in contradiction with other experimental studies, which demonstrate that in normal monkeys with no abnormalities of the macula, only minimal damage to photoreceptors and verteporfin dose-dependent recovery of retinal pigment epithelium (RPE) and choriocapillaris occurs [10]. However, in this study the eyes included already suffer from ARMD: the choroidal blood flow and volume in the macular area is already reduced [14, 15] and the choriocapillaris dysfunction increases the vitality of photoreceptors. This could therefore support the idea that PDT may cause greater damage to the photoreceptors or bipolar cells.

The second hypothesis for the loss of the electrical activity of the foveal and parafoveal areas is related to the anoxia of these retinal layers. According to Ross et al. [16], poor perfusion of the choroid in the macula plays an important role in the development of choroidal neovascularization in patients with ARMD. The decrease in electrical activity in the foveal and parafoveal areas after PDT may result from the substantial impairment of choroidal perfusion in the macula and the greater dysfunction of the regional photoreceptors.
In conclusion, MF-ERG provides objective measures of retinal dysfunction. With MF-ERG it is possible to monitor the macular function of patients with classic CNV resulting from ARMD who underwent PDT treatment. In addition, the electrophysiological data from our preliminary investigation poses, for the first time, a number of questions on the possible damage to the photoreceptors and bipolar cells caused by PDT. For this reason, without ignoring the therapeutic value of PDT, further investigation on a greater number of patients with a longer follow-up is in progress in our department.

REFERENCES