Unusual rod electroretinogram (ERG) intensity-response functions were recorded from three female patients with retinal degeneration who had visual acuities of 20/200, retinal arteriolar narrowing, and diffuse granularity of the retinal pigment epithelium. All three patients had rod b-waves that were profoundly subnormal in amplitude and markedly delayed in implicit time to dim stimuli, but normal or supernormal in amplitude and minimally delayed in implicit time to bright stimuli. Rod a-wave slopes were reduced 50% below normal, indicating photoreceptor involvement. These unusual rod ERG intensity-response functions are similar to those previously reported for the isolated cat eye with elevated retinal cyclic guanosine monophosphate (cGMP) after perfusion with isobutylmethylxanthine. This finding supports the idea that these three patients may have an elevation of retinal cGMP. Their rod ERG intensity-response functions are contrasted with those recorded from some patients with retinitis pigmentosa. Invest. Ophthalmol. Vis. Sci. 31:2283-2287, 1990

Unusual electroretinogram (ERG) stimulus intensity-response functions have been reported for five patients with retinal degeneration. Specifically, b-wave amplitudes were reduced below normal at low stimulus intensities, but normal or supernormal at high stimulus intensities. B-wave implicit times (ie, time to peak) were more delayed at low than at high stimulus intensities. Elevations in retinal cyclic guanosine monophosphate (cGMP) have been considered as a possible pathophysiologic mechanism leading to photoreceptor cell death in these patients.

We present rod ERG intensity-response functions from three additional patients who appear to have the same form of retinal degeneration as that reported previously. This report compares these functions with those reported for the isolated cat eye perfused with isobutylmethylxanthine (IBMX), a phosphodiesterase inhibitor, that results in elevated retinal cGMP. Rod ERG intensity-response functions from these three patients were also compared with those recorded from selected patients with retinitis pigmentosa to see if the two groups share a common abnormality in rod function.

Materials and Methods

A review of full-field ERGs of 3000 patients with retinal degeneration in the data bank of the Berman-Gund Laboratory revealed three unrelated female patients (ages 10, 39, and 39 yr) who had subnormal and delayed or nondetectable rod responses to 0.5-Hz flashes of dim blue light, normal or supernormal cone and rod responses to 0.5-Hz flashes of white light, and subnormal and delayed cone responses to 30-Hz flashes of the same white light (Fig. 1). These three patients (designated as A, B, and C) had corrected visual acuities of 20/200 in both eyes, retinal arteriolar attenuation, and diffuse granularity of the retinal pigment epithelium (ie, fine hyperpigmentation and depigmentation) without signs of intraretinal bone-spicule pigmentation in the periphery. All had clear media. All three reported progressive loss of vision by history, but none had any family history of retinal degeneration. Patients B and C returned 3 and 14 yr, respectively, after their initial evaluation to determine if they had lost additional retinal function.

Eleven patients with generalized retinitis pigmentosa (ages 14-53 yr) were studied for comparison.
The ERGs were elicited with 10-μsec full-field flashes presented at 0.2 Hz in a Ganzfeld dome. An intensity series of blue light (λ<sub>max</sub> = 440 nm, 90 nm half bandwidth) was used to obtain rod-isolated responses at low intensities and cone and rod responses at high intensities. An intensity series of red light (λ<sub>50% cut-on</sub> = 605 nm) was used to obtain cone and rod responses for all intensities that elicited a cone component. Responses to dim blue light were then subtracted from responses to scotopically matched red light to derive cone ERGs, based on the principle of independence of rod and cone components in the ERG.<sup>5-7</sup> These derived cone ERGs to red light were then subtracted from responses to photopically matched blue light to derive rod ERGs, similar to a procedure described previously.<sup>8</sup> This computer subtraction procedure was done to insure that these derived rod responses did not have a cone component that would affect amplitude and implicit time.

Rod b-wave amplitude versus retinal illumination data were then fitted by a least-squares method to $V/V_{\text{max}} = P'(P' + \sigma^n)$ to estimate $V_{\text{max}}$ (the asymptotic peak amplitude), $\sigma$ (the semisaturation retinal illuminance), and $n$ (a measure of response homogeneity).<sup>9</sup> B-wave implicit time versus retinal illumination data were assessed by linear regression to derive slopes and intercepts. At our three highest stimulus intensities, an alteration of the b-wave was apparent in the digitally subtracted responses of normal subjects (Fig. 2). This alteration was considered to be due to subtraction of a cone b-wave to red light from a sublinear summation of cone and rod b-waves to blue light; responses to these intensities were not used for b-wave analyses. Mean b-wave $V_{\text{max}}$, $\sigma$, $n$, and slope of implicit time versus retinal illuminance for patients A, B, and C, the normal subjects, and the patients with retinitis pigmentosa were compared with student's t-test. Slope ($\mu V/msec$) of the rod a-wave to a bright flash, quantified by linear regression,<sup>4</sup> was also compared among the three groups.

Full-field rod ERGs from patients A, B, and C were also compared with those from isolated cat eyes perfused with 0.1-1.0 mM IBMX taken from a previous report.<sup>4</sup> At a dose of 1.0 mM IBMX, retinal cGMP had been found to be elevated twofold.<sup>4</sup> The rod ERGs from these cat eyes were elicited in the dark with 5-msec white flashes from a Ganzfeld dome at 0.2 Hz. Responses were quantified and analyzed with the same procedures as presently used for patients.

**Results**

Patients A, B, and C had rod b-wave amplitude versus stimulus retinal illumination functions that showed profoundly reduced amplitudes to low retinal
illuminances, but normal or supernormal amplitudes to high retinal illuminances (Fig. 3). Mean $V_{\text{max}}$, $\sigma$, and $n$ were significantly elevated above normal for these patients, designated as RD in Table 1. B-wave implicit times were more delayed to low than to high retinal illuminances (Fig. 3) with steeper-than-normal slopes (Table 1). These patients had $a$-wave slopes that were 50% of normal (Table 1). Patient B, reassessed 3 yr after her initial recordings, showed a 28% decline in b-wave amplitude to 0.5-Hz flashes of white light. Patient C, reassessed 14 yr after her initial recordings, showed a 33% reduction in b-wave amplitude to 0.5-Hz flashes of white light. Assuming an exponential rate of decline, these two patients lost, on average, 7% per yr of remaining ERG amplitude to white light.

The rod ERGs of patients A, B, and C resembled those of the isolated cat eye with elevated retinal cGMP after perfusion with IBMX (Fig. 4). In the cat, rod b-waves were profoundly subnormal in amplitude and markedly delayed in implicit time to a dim flash, but normal or supernormal in amplitude and less delayed in implicit time to a bright flash. Rod b-wave sensitivity ($1/\sigma$) was reduced from 0.5–0.8 log unit. In addition, rod $a$-wave slopes to bright flashes were subnormal, indicative of rod photoreceptor involvement.

The patients with retinitis pigmentosa had rod b-waves that were subnormal in amplitude and, in most cases, delayed in implicit time; the amplitude reductions and delays were comparable for both dim and bright flashes (Fig. 5). On average, their $V_{\text{max}}$ was smaller than that of patients A, B, and C and their implicit time versus log retinal illuminance slope was normal (Table 1). Their $\sigma$ fell between the values for patients A, B, and C and the normal subjects.

**Discussion**

The rod ERGs of patients A, B, and C correspond closely with those of the isolated cat eye with elevated retinal cGMP after perfusion with IBMX, lending support to the designation of the human condition as an elevated cGMP-type retinal degeneration. The large and slow rod responses to bright flashes, which make this condition remarkable, could be explained by an elevation in rod cGMP levels, which increases the maximum size and the implicit time of the rod photoreceptor response by increasing the number of molecules of cGMP that must be hydrolyzed to generate rod photoreceptor excitation. However, this designation is provisional as cGMP levels have not been measured from human postmortem eyes with this retinal degeneration and because this designation could change if a molecular genetic defect is revealed.
Table 1. ROD ERG function of patients with retinal degeneration (RD) versus retinitis pigmentosa (RP) and normal subjects (N)

<table>
<thead>
<tr>
<th></th>
<th>RD (n = 3)</th>
<th>RP (n = 11)</th>
<th>N (n = 17)</th>
<th>P§</th>
<th>P§</th>
<th>P§</th>
</tr>
</thead>
<tbody>
<tr>
<td>log b-wave V max*</td>
<td>2.69 ± 0.04</td>
<td>2.06 ± 0.09</td>
<td>2.59 ± 0.01</td>
<td>0.004</td>
<td>0.14</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>log b-wave a*</td>
<td>1.02 ± 0.09</td>
<td>0.60 ± 0.11</td>
<td>0.14 ± 0.08</td>
<td>NS</td>
<td>&lt;0.001</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>b-wave n*</td>
<td>1.22 ± 0.23</td>
<td>0.85 ± 0.13</td>
<td>0.84 ± 0.04</td>
<td>NS</td>
<td>&lt;0.006</td>
<td>NS</td>
</tr>
<tr>
<td>b-wave Δimp/Δlog ret illum</td>
<td>-51.8 ± 5.4</td>
<td>-24.8 ± 2.1</td>
<td>-24.1 ± 1.0</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>NS</td>
</tr>
<tr>
<td>a-wave slope‡</td>
<td>-6.5 ± 1.1</td>
<td>-3.7 ± 0.9</td>
<td>-12.3 ± 0.6</td>
<td>NS</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Values are mean ± SEM.
* Derived from modified least-squares regression for V/V max = 1/(1 + e^n);
V max is in μV and a is in scot. td.-sec.
† Derived from least-squares regression based on responses to flashes of 0.2 to 2.1 log scot. td.-sec.; slope in msec/log scot. td.-sec.
‡ Linear regression on a-wave for 3.0 log scot. td.-sec.; slope in μV/msec.
§ Student’s t-test (2-tailed) between RD and RP.
¶ Student’s t-test (2-tailed) between RD and N.
** Student’s t-test (2-tailed) between RP and N.

In contrast, the rod intensity-response functions of the patients with retinitis pigmentosa evaluated in this study did not resemble those seen in an elevated cGMP-type retinal degeneration. These patients with retinitis pigmentosa showed subnormal b-wave amplitudes to both dim and bright stimuli with b-wave implicit time delays that did not depend on stimulus intensity. Their intensity-response functions could be explained by a combination of cell loss to account for the reduced V max and outer segment shortening (ie, decreased optical density) to account for the elevated a. These findings suggest that some patients with retinitis pigmentosa lose photoreceptor function by a pathophysiologic mechanism not involving elevated retinal cGMP.

It is not known whether patients with an elevated cGMP-type retinal degeneration will eventually show the losses of peripheral field and profound reductions in ERG amplitude characteristic of patients with retinitis pigmentosa. Two lines of evidence suggest that they have a milder form of disease. First, based on the available follow-up visits, two of the patients showed a rate of progression averaging 7% per yr according to an exponential model compared with an average 16% per yr exponential rate of loss of ERG function seen in retinitis pigmentosa. Second, one of the patients (patient A) was nearly two decades younger than the other two patients at the time of testing, but she had ERGs comparable to those of the older patients. Therefore, patients with these unusual waveforms appear to have only mildly progressive disease. However, we cannot exclude the possibility that others presenting with more severe retinal degeneration have a more advanced stage of an elevated cGMP-type condition, with rod b-wave amplitudes even to bright flashes so reduced that their rod intensity-re-
response functions cannot be fully evaluated. This question may be resolved when patients with retinal degeneration, regardless of stage at initial evaluation, can be typed through molecular genetic analysis of leukocyte DNA.

Key words: retina, retinal degeneration, rod, cone, cyclic GMP, retinitis pigmentosa, electroretinogram

References