Influence of Retinal Degeneration Stages on RGC Threshold under epiretinal electrical stimulation: A Modeling Study

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Abstract. Retinal prosthesis is an effective treatment to restore partial functional vision for degeneration diseases such as retinitis pigmentosa (RP). The persistent degeneration of the retina can influence the effect of retina prosthesis. However, how the different stages of retinal degeneration influence the retinal ganglion cell (RGC) responses to external electric stimulation remains unclear. In our present study, we established the multilayer retinal model of normal mouse and retinal degeneration (rd1) mouse in different degeneration stages (early, middle and late stage). Meanwhile, the morphology-realistic RGC models with membrane dynamics in different degeneration stages were also established. The effects of retinal structure, ganglion cell morphology and Na+ conductance on RGC threshold in response to electrical stimulation were explored. The simulation results showed that the RGC threshold of early degeneration stage was basically the same as that of the normal retina. While the thresholds of middle and late degeneration stages were both higher than that of the early stage, and the late stage showed a higher RGC threshold than the middle stage. This study would provide theoretical support for personalizing design of visual prostheses to treat different degeneration stages of RP patients.

1. Introduction
Retinitis pigmentosa (RP) is one of the causes of untreatable human visual impairment. Because of the progressive apoptosis of retinal photoreceptor cells start from rod, RP patient has the following clinical manifestations: visual loss and night blindness in the early stage, tubular visual field in the middle stage and severe visual loss or even complete blindness in the late stage [1]. Even in the late stage of RP, the inner neurons in the retina still partially remain survive. The retinal prosthesis, a device that stimulates the patient's residual retinal neurons using electrical stimulation, is an effective treatment for the RP patients. Retina prostheses have made great progress in recent years, and have commercial equipment such as Argus II (Second Sight Medical Products) and Alpha AMS (Retina Implant AG). These devices have helped patients identify words and read slowly, locate light sources, detect movement, recognize objects and recover long-term visual functions [2][3]. However, at present, all RP patients implanted with retinal prosthesis are advanced RP patients who have experienced years of blind life. In order to make more blind people with earlier stage retinal degeneration benefit from retinal prosthesis, we explored in this study the effects of different degeneration stages on the retinal ganglion cell (RGC) responses to electrical stimulation in different stages of RP that undergoing continuous retinal structure and neuronal changes.

The retinal degeneration stages of human and rd1 mouse are reported to have the similar pathogenesis. In this study, multi-layer retinal models at normal and different degeneration stages were built using a finite element (FEM) software (COMSOL Multiphysics). Then, based on Coombs et al ‘s study of...
mouse RGCs [6], we established a morphologically realistic RGC model using the NEURON software. The differences between the response thresholds of RGCs in three stages of retinal degeneration (early, middle and late stage) were explored. Further, the influence of changes of the retinal tissue structure, the RGC morphology and the electrical conductivity of Na⁺ channel on the RGC response threshold were investigated. These results would provide the theoretical support for the design of visual prostheses at different stages of degeneration.

2. Methods

2.1. Multi-layer retina model in different stages of retinal degeneration

The retinal multi-layer models at normal and different degradation stages were built using a finite element (FEM) software (COMSOL Multiphysics 4.4, Inc. Palo Alto, CA, USA). Figure 1(a) & (b) shows the retinal multi-layer electrical stimulation model in the normal rd1 mouse. The relationship between electrode and retina tissue was shown in Figure 1(a). The multi-layered retina can be divided into vitreous body (VB), nerve fiber layer (NFL), ganglion cell layer (GCL), inner plexiform layer (IPL), inner nuclear layer (INL), outer plexiform layer (OPL), outer nuclear layer (ONL), outer segments of photoreceptor (OS) and retinal pigment epithelium layer (RPE) (Figure 1(b)). The process of human and rd1 retinal degeneration can be roughly divided into three stages [7-10] according to the progress of neuronal apoptosis as shown in Figure 1(c). Particularly, in the early stage of retinal degeneration, photoreceptors were stressed and further its outer segment was shortened; In the middle stage, photoreceptors underwent apoptosis and the visible scars were formed; In the late stage, RGCs underwent apoptosis and complex neuronal remodeling [11]. The thickness and electrical conductivity of each layer in each stage of degeneration was determined based on the research of Pennesi ME et al [12] and Boshuo Wang et al [13] as shown in Table 1. The platinum disk electrode was placed into VB as the stimulation electrode. And the outer surface of the sclera was set as the distal return for the electrical stimulation. In this model, the diameter of the electrode (Diam) was set at 50μm and the distance between the electrode and the lower surface NFL layer (ERD) of the retina was set at 25μm.

2.2. RGC model in different stages of retinal degeneration

The single RGC model was built using the NEURON software, which had great advantages in simulating neurons with complex geometry and rich ion channels. Based on the research of Coombs et al [6], a morphologically realistic RGC model was constructed. A simplified schematic diagram of RGC model was shown in Figure 1 (d). Its morphology consisted of three parts: the dendrite, the soma and the axon including the high-density sodium channel band (SOCB). The axon also included the initial, the narrow and the distal axon. The length of SOCB section was set about 30μm. Furthermore, each node of the RGC model was built based on the Fohlmeister–Coleman–Miller (FCM) model. In our previous studies [14], this FCM model contained five ion channels: a sodium current (I_{Na}), a calcium current (I_{Ca}), a delayed rectifier (I_{DR}), a potassium type A (I_{K}), a calcium-activated potassium current (I_{KCa}) and Leak current (I_{L}). In different stages of retinal degeneration, the changes in RGC morphology and the electrical conductivity of Na⁺ channel were shown in Table 2 [15][16].
Figure 1. Multi-layer retina and electrical stimulation model and morphology based RGC model. (a), Schematic diagram of epi-retina electrical stimulation: dark gray filled: electrode, light gray filled: base, white filled: layered structure of vitreous body and nerve fiber layer. (b), Multilayer model of normal retina and simplified RGC model. (c), The stage of retinal degeneration in human RP and rd1 mouse [7-10]; Phase 1, phase 2, and phase 3 represented the early, middle, and late stages of degradation, respectively. “?” mean that there was no exact point in time between phase 1 and phase 2. (d), The morphology based RGC model constructed in NEURON software.

Table 1. Thickness and conductivity at normal and different stages of degeneration retinal [12][13]

| Structure | Thickness(µm) / Conductivity(S/m) |
|-----------|----------------------------------|
|            | Normal (µm / S/m) | Early (µm / S/m) | Middle (µm / S/m) | Late (µm / S/m) |
| NFL       | 12 / 1.095        | 12 / 1.095       | 12 / 1.095        | 12 / 1.115      |
| GCL       | 17 / 1.242        | 17 / 1.242       | 17 / 1.276        | 16 / 1.300      |
| IPL       | 96 / 0.658        | 96 / 0.658       | 96 / 0.855        | 94 / 0.881      |
| INL       | 65 / 0.373        | 65 / 0.373       | 65 / 0.687        | 59 / 0.781      |
| OPL       | 18 / 0.316        | 18 / 0.316       | 9 / 0.316         | -               |
| ONL       | 71 / 0.299        | 71 / 0.299       | 21 / 0.299        | -               |
| IS        | 31 / 0.639        | 31 / 0.639       | -                 | -               |
| OS        | 34 / 1.101        | 17 / 1.101       | -                 | -               |
| SCAR      | -                  | -                | 20 / 0.009        | 35 / 0.009      |
| RPE       | 11 / 0.093        | 11 / 0.093       | 11 / 0.093        | 14 / 0.092      |

Table 2. Changes in RGC morphology and Na⁺ channel conductivity [15][16]

| Structure | Dendrites (diameter) | Soma (diameter) | Na⁺ (conductivity) |
|-----------|----------------------|-----------------|--------------------|
| NFL       | -                    | -               | -                  |
| GCL       | -                    | -               | -                  |
| IPL       | -                    | -               | -                  |
| INL       | -                    | -               | -                  |
| OPL       | -                    | -               | -                  |
| ONL       | -                    | -               | -                  |
| IS        | -                    | -               | -                  |
| OS        | -                    | -               | -                  |
| SCAR      | -                    | -               | -                  |
| RPE       | -                    | -               | -                  |
3. Results

3.1. Electrical potential analysis of the retina in different degeneration stages
We analysed and compared the changes of retinal electric potential distribution at different stages of degeneration in rd1 retina, as shown in Figure 2. Figure 2 (a) was the retinal potential distribution diagram of normal retina, and Figure 2 (b), (c) and (d) were the retinal potential distribution diagram of the early, middle and late stages of degeneration. As shown in (e), the normal retinal potential was basically the same as that in the early stage of degeneration, but lower than that in the middle and late stage of degeneration. Especially, the middle stage of retinal degeneration showed the highest potential.

### Table 1: Retinal Potential Distribution

| Stage    | Early | Middle | Late |
|----------|-------|--------|------|
| Potential| 100%  | 100%   | 100% |
|          | 80%   | 95%    | 81%  |
|          | 70%   | 88%    | 74%  |

Figure 2. Retinal potential distribution in rd1 mouse at different stages of degeneration. (a), (b), (c) and (d) represented retinal potential distribution in the normal mouse and in the early, middle and late degeneration stage of rd1 mouse. (e) showed the potential curve in the lower surface of NFL (the normal was similar to late).

3.2. Threshold analysis of RGC in different degradation stages
The threshold of RGC was influenced by different parameters in simulation. Firstly, we analysed the RGC threshold variation when only RGC’s morphology and ion channel parameters were varied during the early, middle and late stages of degeneration (as mentioned in Method). The control variable method was used to analyse the influence of these parameters, i.e., only one parameter (dendrites field area, soma size or Na+ concentration) was varied while other conditions were kept the same in different simulations in order to find the influence of the specific parameter variation. In these cases, the electric field distribution was calculated under normal retina model. We changed the position of the stimulation electrode relative to the RGC soma (step size: 20micrometer), calculated the threshold of RGC along the axon direction, and obtained the threshold curve of each degradation stage. Figure 3 (b), (c) and (d) showed the influence of dendrite field area, the soma size and Na+ channel conductivity changes on the activation threshold. It can be seen that the decrease of RGC dendrite field area will increase the threshold current of the activation site located in the dendrite. The change of soma size had little influence on RGC with activation site in the soma. The decrease of Na+ channel electrical conductivity will increase the RGC threshold, especially in SOCB segment.
We then analysed the overall influences when all the variations of retinal tissue and RGC properties in different degeneration stages were taken into account. The electric potential was calculated in the corresponding multi-layer retinal models with different degeneration stages (as shown in Fig. 4(a)). Figure 4(a) shows the electric potential curve along y at z = 0 plane which was the junction of NFL and GCL at normal retina and three stages of degeneration retina. Figure 4 shows the threshold curves of RGC in different degeneration stages when the position of stimulation electrode was changed as in Figure 3. It can be seen that the minimum threshold gradually increased in the early, middle and late stages.
4. Discussion and conclusion

In this study, we investigated the electrical stimulation threshold of RGC in different stages of retinal degeneration and its influencing factors. Firstly, the stratified retinal tissue model of normal and early, middle and late degeneration stages was established in COMSOL, respectively, which was used to investigate the effect of retinal tissue structure changes on the spatial potential distribution during retinal degeneration. Secondly, we used the Neuron software to establish the RGC model and explored the influence of dendrite, soma and electrical conductivity of Na⁺ channel changes on RGC electrical stimulation threshold during degeneration. Our results showed that the thresholds were influenced by the RGC morphology, Na⁺ ion channel parameters and retina tissue structure.

On the macro level, as shown in Figure 2, the changes in the tissue structure during the retinal degeneration have a certain impact on the spatial potential distribution of the retina. Compared with the normal retina, the denaturing retinal potential was the highest in the middle stage, followed by the later stage, and the difference between the early stage and the normal stage was not significant as shown in Figure 2. The difference was likely caused by the influence of scar tissue structure in the middle stage, further apoptosis in the outer layer of the retina and changes in electrical conductivity caused by the reconstruction of neurons in the later stage. On the micro level, as shown in Figure 3 the reduction of dendrite field in the degradation process increases the response threshold of neurons located near the dendrite. The change of soma had little effect on the threshold of neurons. The decrease of Na⁺ concentration also led to the increase of RGC electrical stimulation threshold. Finally, as shown in Figure 4, retinal degeneration led to an increase in RGC electrical stimulation threshold, which was consistent with the results of electrophysiological experiments [17]. Through the simulation results, it can be seen that the RGC electrical stimulation threshold increases gradually as the retinal degeneration becomes more serious.

In conclusion, these results would provide the theoretical basis for the study of retinal degeneration and help design the personalized visual prosthesis for patients with different stages of degeneration.

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