Arab J Sci Eng (2018) 43:2889–2898
https://doi.org/10.1007/s13369-017-2918-z

RESEARCH ARTICLE - ELECTRICAL ENGINEERING

Comparison of Monophasic and Biphasic Electrical Stimulation by Using Temporal Analysis for Different Inter-electrode Spacings in the Hexagonal Arrays

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Received: 22 March 2017 / Accepted: 24 October 2017 / Published online: 13 November 2017

Abstract Recent multidisciplinary studies in the field of visual prosthesis rely on electrically stimulating retinal tissue by placing electrode array into different part of it, bypassing the nerve cells of the visual pathway which have lost their functionality due to various degenerative diseases. The visual prosthesis systems could be more efficient by mimicking physiologically natural electrical signals. In this study, the responses of the retina to biphasic and monophasic electrical stimulations were compared temporally using in-vitro rabbit retina. The retinal tissue was electrically stimulated with charge-balanced biphasic and monophasic pulses. When temporal diversity was comparatively analyzed, spike activity was observed to intensify in the first 20 ms after the moment of stimulation for biphasic stimulation. On the other hand, the spike activity was observed to form in a way that it can be classified into two categories, the primary one forming in the first 20 ms and the secondary delayed one forming between 80 and 100 ms for monophasic stimulation. Moreover, the effect of retina stimulation amplitude was also analyzed. The spike distribution was observed to cumulate mostly in the post-stimulation time interval of the first 20 ms as the stimulation amplitude for biphasic stimulation increased. In monophasic stimulation, secondary delayed spike activity was observed over 80 μA. The obtained experimental findings suggested that lower amplitude charge-balanced short biphasic waveforms could better elicit the precise and proper stimulation patterns necessary to mimic the neural activity of retinal circuitry that resembles the natural ones when it was compared with monophasic stimulation.

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Keywords Retinal ganglion cells · In vitro experiment · Cognitive systems · Biphasic stimulation · Monophasic stimulation · Temporal analysis · Hexagonal microelectrode array

1 Introduction

Retinitis Pigmentosa (RP) and Age-Related Macular Degeneration (AMD) are the most common degenerative retinal diseases which cause blindness. Affecting more than 1.5 billion people in the world, there is no known and applied cure [1–3]. These diseases damage the photoreceptors at the outermost layer of the retina. The dysfunction of the photoreceptors, which are responsible for converting light into electrical signals, interrupts the progress of the electrical signals in the visual pathway, causing blindness [4]. It has been reported that despite serious damage to the photoreceptors, the nerve cells and retinal ganglion cells in the innermost layer of the retina remain morphologically intact and functionally viable, even in patients who have been blind for a long time [5,6].

Recently, advances in various areas such as electronics, miniaturization, materials and packaging made the development of retinal prostheses possible. Retinal prostheses aim to elicit the visual perception with the help of electrical stimulation of the remaining tissue in the brain, bypassing the nerve cells of the visual pathway which have lost their functionality due to various diseases [7–10]. The electrical stimulation is performed with the delivering of stimulation patterns to the retinal tissue via a microelectrode arrays placed on targeted retina region. This emerges an important hope for blind patients with the regaining of significant visual information.

The literature is reviewed to see what extent vision can be restored by visual prostheses and the state of art in research.

It is basically seen that there are two important conditions for retinal prosthesis to be supposed as an option [11]. Firstly, even if the photoreceptors in the outermost layer of the retina are damaged, the remaining nerve cells must be mostly functional and viable. Secondly, depending on the placement location of the device, either a device which mimics the function of the damaged part is placed or the damaged area is bypassed and functional nerve cells are electrically stimulated. Thus, the retinal prostheses are divided into 3 main categories depending on the targeted retinal area to which the microelectrode arrays are implanted [1]. Firstly, in epiretinal approach, the electrode array is implanted on top of the retina. The aim is to stimulate the retinal ganglion cells, nerve fibers, which form optic disk. It has the longest follow-up data from both animal experiments and acute or chronic trials with human subjects [12–15]. Clinical studies performed with human subjects showed that recognizing and distinguishing objects, reading letters with advanced correctness are possible [16–18]. Studies conducted recently have focused on increasing the resolution by using more electrodes which are restricted by electrode impedance that rises as the electrode size decreases. While some researchers have been studying on different electrode materials to remove the impedance, others are focused on electrode coatings for next-generation neural interfaces instead of metal electrodes [19,20]. Second, subretinal prostheses are implanted under the retina, between the retinal pigment epithelium layer and the photoreceptors. It has been reported that passive implants which use the incident light in order to activate the photodiodes on the silicon disk fail to generate the necessary stimulation current [21–23]. A circuitry has been added to raise the photodiode current to sufficient levels. Chronic and acute trials demonstrated that reading large letters and object detection could be successively achieved [24–27]. Although several researchers are studied on nanorods to provide wireless electronic system, some other studies carried out recently are based on improving indirect, network mediated, stimulation which uses bipolar cells and photoreceptors for stimulation [28,29]. Thus, retinal responses resemble real physiological signal because contributions of each neuron type are included to overall response. Moreover, different types of neurons are physiologically investigated to determine unique responses of each neuron type [30,31]. Determining optimal stimulation pattern is also researched to minimize the charge required for neuron activation by morphologically investigating different retinal ganglion cell clusters [32,33].

Lastly, suprachoroidal prostheses are based on the electrical stimulation of the area between the sclera and choroid. Considering remarkable surgical and safety advantages, some researchers aim to develop less invasive approach by using suprachoroidal space. It is reported that this approach show better results in terms of surgical procedure, tissue detachment and fixation [34–36]. Additionally, phosphene perception is also provided in clinical trials with human patients [27].

The visual prosthesis systems developed until now offer such low spatial resolution that the distinction of objects and faces are impossible, which is quite far from the planned situation. The resolution needs to be increased in order to achieve this. The increase in resolution is possible by increasing the number of electrodes and using effective stimulation methods. Some biological and technical difficulties and limitations are involved with increasing the number of electrodes. The safe stimulation of the retinal tissue requires the application of currents within certain amplitudes, which protects the retina tissue from damage due to high charge and prevents electrode corrosion. Technical difficulties consist of important difficulties regarding camera, image and video processing, electrode arrays, integrated circuits and telemetry systems, all functions which require careful engineering and innovative approaches [6]. On the other hand, in order to stimulate the retina with the most effective stimulation method, one needs to understand the retina's reaction to electrical stimulation. Findings of the experimental studies to be performed in related fields are quite valuable to reach this aim. The stimulation limits of the retina, electrodes and the retinal response to stimulation parameters need to be determined based on the findings of the experimental studies for obtaining a stimulation within the safe stimulation limits.

In a healthy person, seeing initiates with the light coming to the eye. Light is converted into electrical signals as it reaches the photoreceptor layer. Subjected to various spatial and temporal processing steps in the following sections of the retina, these signals encode the information from the outside world and are delivered to the visual cortex of the brain. There are different types of neurons which are responsible for processing the signal along visual pathway. The retinal ganglion cells are the last processing units in the retina before the optic disk. Reported to exist in approximately 12 different forms, these cells form unique spiking patterns in cortical and non-cortical areas [37,38]. When spatially distinguishable light points are ensured with electrical stimulation at the size of a nerve cell, then high resolution becomes possible. Temporal resolution presents for spike trains at different frequencies which encode the contrast and depth information. Examining the effects of stimulation patterns which have high temporal acuity on the retina by mimicking the light elicited spike trains in the healthy eye and analyzing the stimulation elicited spike trains are significantly important for designing high performance retinal prostheses.

Physiologically natural spikes which resemble the signals delivered to the brain in a healthy eye could be formed by better understanding temporal analysis of recorded retinal responses to electrical stimulation. Thus, an efficient system needs to be developed with the lowest power consumption and hardware by mimicking the optimum stimulation

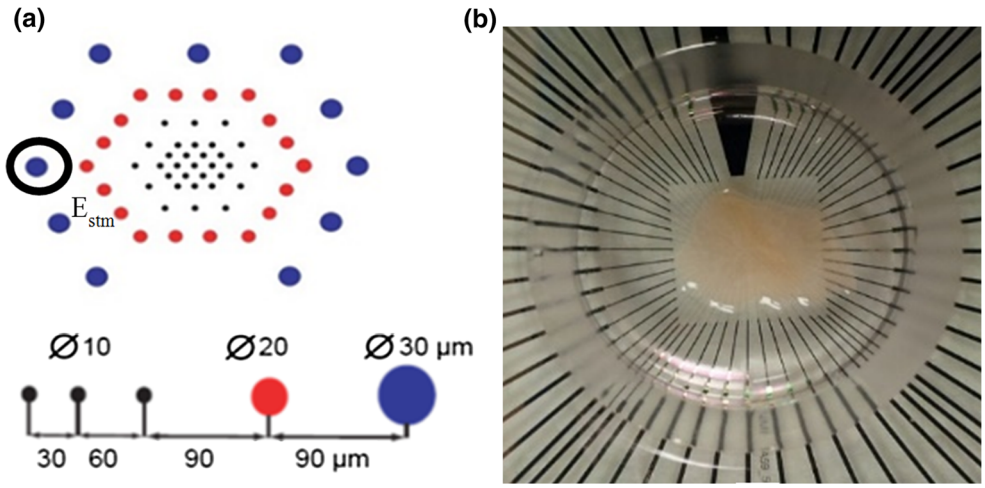

Fig. 1 Microelectrode array layout and retina-electrode interfacing. **a** Electrode layout. **b** Retinal tissue placed microelectrode array. E_{stm} indicates stimulation electrode

that is closest to the real one. In addition, the ability to increase the performance of a retinal prosthesis is dependent on better understanding of the neural mechanisms of the retina during electrical stimulation and how these mechanisms could be controlled in order to obtain useful vision. In this study, we focused on the comparison of different stimulation patterns, biphasic and monophasic, by temporal analysis of retinal responses. The temporal effects of the biphasic and monophasic stimulation pulses with varying pulse amplitudes were comparatively analyzed via in vitro studies using rabbit retinas. In this context, following electrical stimulation the retinal responses were analyzed based on the post-stimulus time intervals (PSTI), which were 0–20, 20–40, 40–60, 60–80 and 80–100 ms.

2 Experimental Methods

The methods of the study are the retinal preparation phase, electrical stimulation and the analysis of the neural activity with recording systems. The retinal preparation phase signifies the extraction of retinal tissue and its carriage onto the microelectrode array system. The electrical stimulation and recording system represent the system components used in the process of perceiving and recording the neural signals. The analysis of the neural activity is consistent with the work necessary to convert the recorded neural activity into interpretable data for analyses.

2.1 Preparation of the Retina

GATA Animal Experiments Ethics Board Guidelines were fulfilled in the use of experiment animals. White, New Zealand type female rabbit subjects that were provided with adaptation to dark and weighed 2–3 kg were sedated with Ketamine (40 mg/kg) and Xylazine (10 mg/kg) with intramuscular injections [5–7]. They were sacrificed with intravascular high dose sodium pentobarbital. Following the enucleation of the eyeball, the front portion of the eye and vitreous were thrown away. Divided into segments of approximately 4 mm x 4 mm, the eyeball was put into Ames' medium (Sigma Chemical Co.). The time interval between sacrificing and putting the eyeball into the Ames' medium is 4–9 min. Ames' medium was buffered with sodium bicarbonate and equilibrated with a gas mixture of 95% oxygen and 5% carbon dioxide [5].

2.2 Retina-Electrode Interfacing

Following the extraction of the retina, sclera and the retinal pigment epithelium and the inner limiting membrane were isolated from the vitreous and other parts by removal with tweezers. The retina pieces and a small amount of solution were taken into the microelectrode array chamber. The retinal ganglion cells were placed in such a way that nerve cells faced to electrodes while the solution was pulled back. Thus, complete adhesion of the retina to the microelectrode arrays was ensured. After this, the solution, adjusted between 35 ± 1 °C, was perfused with a perfusion system. These processes were performed under dim red light. Standard perfusate (solutes in mM: 100 NaCl, 30 $NaHCO_3$, 50 glucose, 6 KCl, 2 $MgSO_4$, 1 $CaCl_2$, 1 $NaHPO_4$) bubbled with 95% O_2, 5%CO_2 with a pH of 7.5 ± 0.2 and a temperature of 33 ± 1 C. The retina placed on the microelectrode array is shown in Fig. 1. Before the recording process initiated, the retina tissue was quickly enucleated, placed to the microelectrode array that facing retinal ganglion cells and Ames solution was applied using perfusion system to keep the tissue alive. Next, whether the

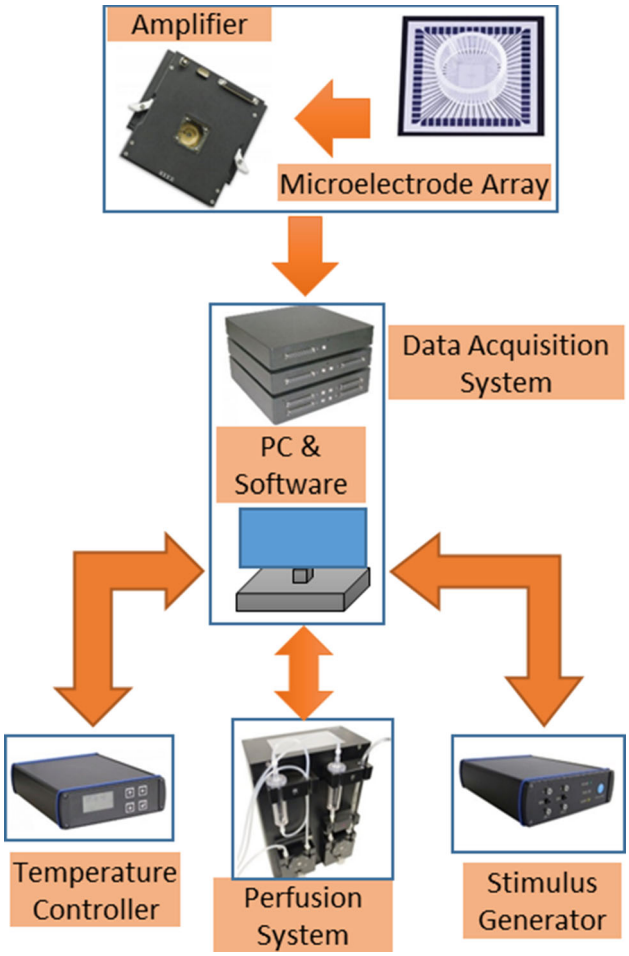

Fig. 2 Experimental system block diagram

retina tissue was viable or not was checked using light stimulation. The tissue in the microelectrode array that was placed to the amplifier was stimulated with a typical light source. It was checked that recording channels showed extra neural activity related to its aliveness when the light source was on. If the tissue was viable after this procedure, it could be used to examine electrical stimulation effects.

2.3 Electrical Stimulation and Recording of Retinal Responses

The recording system includes several main components. These are microelectrode array (60HexaMEA-Ti, Multi Channel Systems GmbH, Germany), amplifier (MEA1060-Amplifier, Multi Channel Systems GmbH, Germany), data acquisition computer and software, stimulation generator (STG4002, Multi Channel Systems GmbH, Germany), temperature controller and the peristaltic perfusion system. The system block diagram is presented in Fig. 2.

The electrode array is made up of 60 hexagonally arranged electrodes. While electrode diameters vary between 10, 20, 30 μm, inter-electrode distances vary between 30, 60, 90 μm. The amplifier of the system which stimulates and records the retinal activity with 60 electrodes at bandwidths between 1 and 5000 Hz with gain of 1000 is protected with what resembles a Faraday cage and grounded. In the recording processes, the sampling frequency is 25 kHz.

Stimulation trains with different amplitudes and pulse widths were adjusted. The amplitudes of the spike trains were 10, 50, 80, and 120 μA, and the pulse widths were 100 and 200 μs. Biphasic and monophasic pulses were used as stimulation patterns, as shown in Fig. 3. The stimulation current trains produced by the stimulus generator were delivered to selected electrodes of the hexagonal microelectrode array, and other electrodes were used as recording ones. Charge-balanced anodic-first biphasic and monophasic pulses were used.

There were 20 successive stimulations that were applied for each stimulation type, including four different amplitudes and two different pulse widths. It was taken into consideration that there was a need for sufficient trials to compare the results for each stimulation types. This procedure was repeated to obtain comparable results until each stimulation pattern ended. Before analysis phase, most active recording electrodes, which showed discernible spike activity, were selected. Some recordings were excluded because of large amounts of signal distortion, low tissue-electrode contact, residual layers on electrodes. We made preparations to handle these problems by cleaning the microelectrode arrays carefully using ultrasonic cleaners, using a poly-lysine coating on the electrodes to elicit the adhesiveness for the retina and a grounding process with a cage similar to Faraday's cage. However, all electrodes that could not successfully record neural activity were excluded. This was a common situation that has also been noted in the literature.

2.4 Analysis of the Neural Activity

The neural activity originating in the electrical stimulation was recorded with microelectrode arrays. The raw neural waveforms were taken into the MATLAB environment following the recording process and analyzed with custom written codes. The data were filtered at 500 Hz high pass filter and digitized at 25 kHz sampling frequency. Neural waveform which was filtered and superimposed forms of isolated spike activity relating to a part of the randomly selected neural wave forms are presented in Fig. 4.

Analysis of neural activity required identifying and removing spontaneous activity. In this study, when the retina was in place and recording was started, the signal was only recorded until stability was reached, which was at least 10 s. Then, electrical stimulations began. The stimulation process was completed in such a way that at least 10 s were recorded in a blank, unstimulated area, when the next stimulation

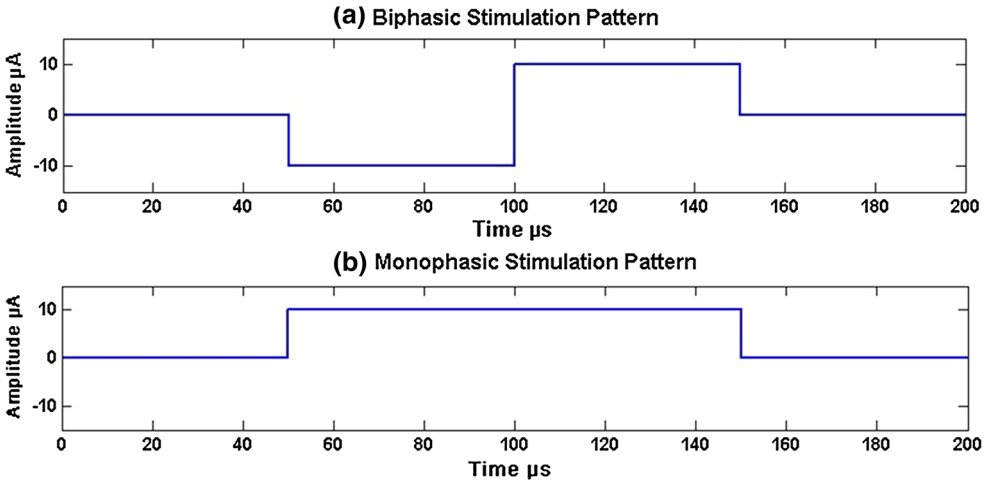

Fig. 3 Different types of stimulation pulses applied to retinal tissue. **a** Biphasic pulses, **b** monophasic pulses

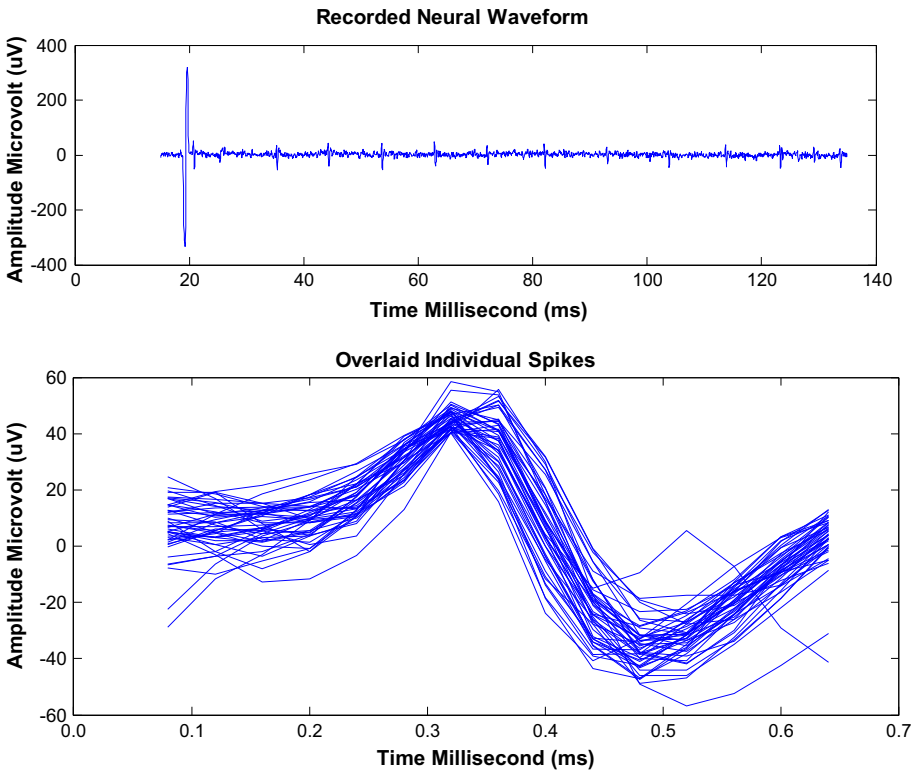

Fig. 4 Analysis of neural activity, (top) neural waveform which is filtered, (bottom) overlapping isolated spikes

amplitudes were set. In the analysis process, to eliminate the misleading effects of spontaneous activity, the unstimulated areas were examined, and average spike counts were determined and used as the 'background activity'. Then, the spike counts were determined for gaps between consecutive stimulation pulses. The spike counts and background activity were compared. If the spike count observed between two successive stimulation pulses was greater than background activity, it was accepted that the stimulation pulse generated spikes. This procedure was repeated for each stimulation trial. To prevent misleading effects that would create a difference between the beginning and the end of the recorded signal, background analysis was performed based on the areas unstimulated for 10 s, which were left for each stimulation amplitude. In this way, misleading effects of spontaneous activity were eliminated.

3 Results

The activity of the retinal ganglion cell layer during the repeatedly applied electrical stimulation was recorded and

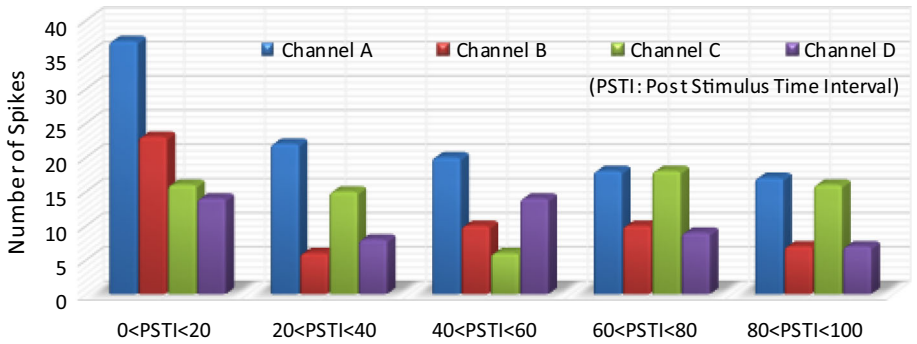

Fig. 5 Number of spikes from selected recording channels through biphasic stimulation

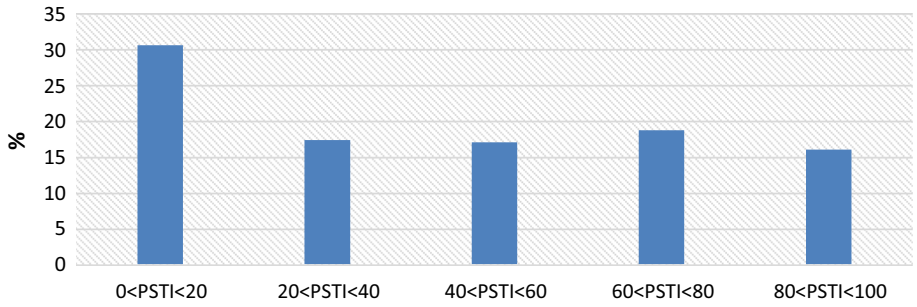

Fig. 6 Isolated spike distribution percentage among post-stimulus time intervals following biphasic stimulation

temporal profiles quantitatively analyzed according to different stimulation patterns. In the analysis of the retinal tissue response to electrical stimulation, the most active electrodes, which showed distinguishable spike formation, were firstly determined and selected for further steps. The electrical stimulation moments applied to these electrodes were identified to determine the spikes coinciding with the post-stimulus time intervals (PSTI). PSTI were arranged in 5 groups with 20 ms intervals, up to 100 ms. The spikes after 100 ms were not included in the analysis. Number of spikes at the PSTI intervals and the spike percentages within these intervals following stimulations at each channel were determined for the temporal analysis of the neural activities from the channels with the best records. In order to eliminate the misleading effects of spontaneous activities, the signals were recorded and sampled for a while before electrical stimulation started. The mean number of action potentials in the sampled signal pieces was subtracted from the number of action potentials in the target area. Thus, the effect of activity not related to electrical stimulation was eliminated.

The results of identified spike numbers in relation to the time intervals following the stimulation of the 4 most active electrodes, determined among the neural activities due to the application of biphasic stimulation pulses onto the retinal tissue are presented in Fig. 5. An important part of the identified spikes were observed to occur in the first 20 ms following the stimulations of each electrode.

The temporal distribution of the identified spikes at the designated recording electrodes following the stimulations is shown in Fig. 6. As a result of direct stimulation of the retinal ganglion cells and the adaptation of the applied charge-balanced stimulation on cell physiology, the action potentials were observed to occur in the first 20 ms PSTI. The spike distributions in the first 20 ms interval and at each of the other intervals were determined to be 31 and $17.5 \pm 1.5\%$, respectively.

The number of spikes obtained according to selected channels as a result of monophasic stimulation is shown in Fig. 7. The spike distribution in the channels selected for analysis showed short and long latency properties similar to each other. The short latency spikes were observed in the 20 ms PSTI right after electrical stimulation while the long latency spikes were observed 75 ms later.

When the spikes obtained by monophasic stimulation were evaluated, the same properties of the spikes observed in the recording channels are seen again, Fig. 8. 28% of all spikes were observed in the first 20 ms PSTI. The ratio of spikes seen between 20 and 80 ms was approximately $16 \pm 1\%$ for each interval. The ratio representing the long latency spikes between 80–100 ms PSTI was observed to be 25%.

The results of the comparisons of the spike activities obtained by the application of biphasic and monophasic electrical stimulations are shown in Table 1.

In analyses performed with regards to stimulation amplitudes, the neural activity results obtained from 5 recording electrodes for both biphasic and monophasic stimulation were used. The proportional changes in spike activity with regards to stimulation amplitude were analyzed for biphasic and monophasic stimulations. The percentages of the spikes which correspond to their own time intervals detected depending on applied stimulation pulses with different ampli-

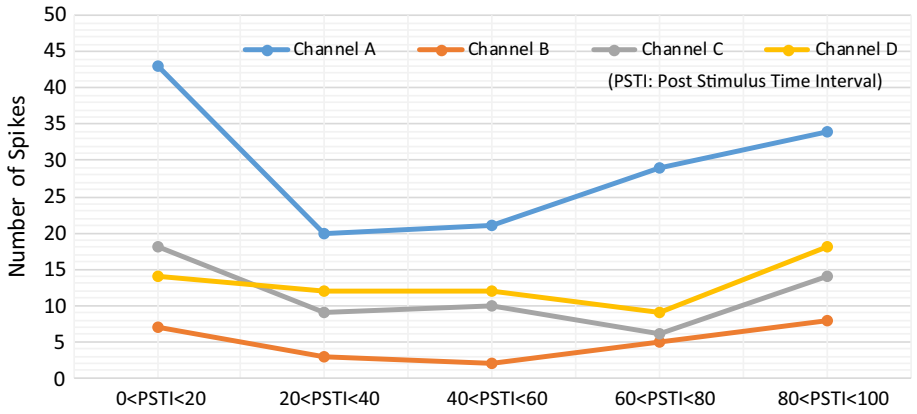

Fig. 7 Distribution of spikes according to selected channels followed by monophasic stimulation

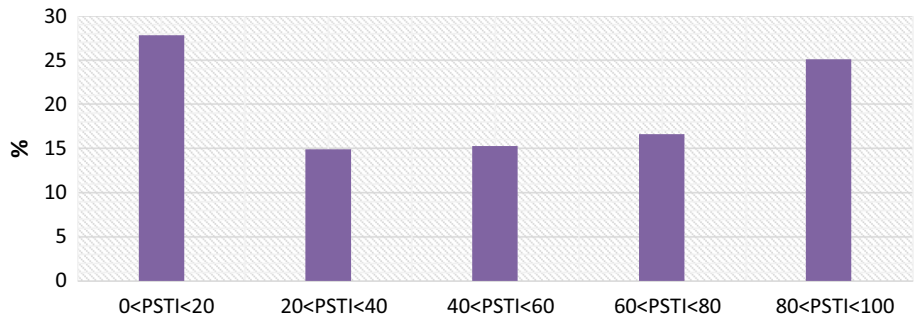

Fig. 8 Isolated spike distribution percentage among post-stimulus time intervals following monophasic stimulation

Table 1 Comparison of analysis results

Post-stimulus time intervals (ms)	Biphasic stimulation		Monophasic stimulation	
	Number of spikes	%	Number of spikes	%
0 < PSTI < 20 ms	90	31	82	28
20 ms < PSTI <40 ms	51	17	44	15
40 ms < PSTI <60 ms	50	17	45	15
60 ms < PSTI <80 ms	55	19	45	15
80 ms < PSTI <100 ms	47	16	79	27

tudes are shown in Table 2. Moreover, when isolated spike numbers were investigated for each amplitude, it was seen that higher current amplitudes elicited more spikes than lower ones.

The distribution of the spike activity obtained by biphasic stimulation was mostly intensified in the first 20 ms PSTI when low stimulation amplitudes such as 10 μA were applied. Although the spike activity moved slightly toward the 20–40 ms and 40–60 ms intervals with stimulation amplitudes of 50 μA and above, no significant change was observed due to increasing amplitudes and intensification within the first 20 ms interval was observed. On the other hand, while the spike activity obtained with monophasic stimulation pulses did not show any significant change in the first, second, third and fourth 20 ms PTSI with increasing amplitudes, a significant increase was seen in the long latency spike activity with amplitudes of and above 80 μA.

The distance between stimulation electrode and the most active recording electrodes were varied. The effect of the distance between electrodes on spike formation was investigated and results are presented in Table 3. It indicated that while closer electrodes generated more spikes for biphasic stimulation, more distanced electrodes detected more spikes for monophasic stimulation.

4 Discussion

Physiologically natural spikes which resemble the signals conducted to the brain in a healthy eye could be evidenced by the temporal analysis of spike trains. The performance of retinal prosthesis is dependent on better understanding of retinal responses to electrical stimulation. The dynamics and underlying neural mechanisms of them play a critical role to elicit useful vision with high resolution. In the previous studies which used temporal analysis, either voltage-focused stimulations or a single type of stimulation in current-dependent stimulations were used [37–39]. Some studies used the patch

Table 2 Comparison of spike activity percentages according to various stimulation amplitudes

Stimulation amplitude (μA)		0 < PSTI < 20 (ms)	20 < PSTI < 40 (ms)	40 < PSTI < 60 (ms)	60 < PSTI < 80 (ms)	80 < PSTI < 100 (ms)
Biphasic stimulation	10	38	12	15	24	12
	50	23	19	14	22	23
	80	23	32	15	13	17
	120	27	11	26	17	20
Monophasic stimulation	10	32	15	17	15	21
	50	33	12	16	20	22
	80	27	17	17	18	22
	120	23	15	13	12	38

Table 3 Number of spikes depending on the distance between stimulation and active recording electrodes

	Ch-1	Ch-2	Ch-3	Ch-4
Biphasic stimulation				
Distance (μm)	135	172	220	255
Number of spikes	56	114	71	52
Monophasic stimulation				
Distance (μm)	135	270	273	272
Number of spikes	25	65	58	147

clamp method instead of the microelectrode arrays with extracellular recordings [25,26,33,39]. In this study, charge-balanced biphasic and monophasic stimulations were used and the records were taken extracellularly. To ensure a more precise analysis of the temporal effects on the retina, the experimental studies were conducted in similar conditions. During in vitro studies, two different types of stimulations were conducted using direct stimulation, which is epiretinal approach. While most of the spike activity was observed to occur in the first 20 ms PSTI upon biphasic stimulation pulses were used, the activity was seen to intensify between 0–20 and 80–100 ms intervals in two parts and as short and long latency activities with the use of monophasic stimulation pulses. The temporal response latencies were observed to be of two types. First part of them was under the 25 ms while the other was in the interval above 80 ms. The findings which were important aiming to prove the relationship between temporal response profiles and the stimulation configurations showed that retinal ganglion cells, whose temporal response profiles change depending on the stimulation configurations were diverse in structure and heterogeneous in themselves. On the other hand, upon examining the effect of the stimulation amplitude on the spike activity as recorded from the retina, it was seen that increasing amplitudes did not cause significant changes in spike activity in biphasic stimulations, but in monophasic stimulations, especially as the amplitude exceeded 80 μA, a significant increase was seen in the formation of long latency spike activity.

The spike activity was formed via two mechanisms at the out of the retinal ganglion cells. The first one was the formation of spikes with the indirect stimulation of retinal ganglion cells through the bipolar amacrine cells due to the stimulation of the retinal network, which was the aim of subretinal studies. In this pathway, the stimulation of the presynaptic excitatory neurons such as the bipolar cells caused depolarization and spike was formed. The amacrine cells were stimulated with two main mechanisms. These were direct electrical stimulation or synaptic stimulation due to stimulation of the electrically activated bipolar cells. The bipolar cells were connected to the amacrine cells to excite. The second method was the formation of spikes through direct stimulation of the retinal ganglion cells. The direct stimulation had some advantages. The response latencies were shorter and fewer temporal deviations were seen. In network stimulation, because the processes were subsequent, the latency tended to be higher. The spikes formed at direct stimulation seemed to be more robust in nature. Direct activation induced spike stimulations generated high temporal precision, however they were usually followed by low temporal precision spikes forming due to longer pulses and network stimulations. Selective stimulation was very difficult due to similar threshold levels. This was why the stimulation of retinal ganglion cells with stimulation currents made up of short current pulses and stimulation parameters comprising variables such as stimulation amplitudes within certain limits, frequencies and pulse widths and epiretinal stimulation might only be an option that excited the retinal ganglion cells and offered high temporal precision. The obtained results were compliant with the study of Tsai et. al. In that study, Tsai et. al claimed that the temporal characteristic of the retinal ganglion response showed different properties which could be classified into several categories by using short pulse subretinal stimulation [39]. Margarit et al, Thoreson and Fried et al showed that the activation of amacrine cells gave rise

to wide inhibitory post-synaptic currents and that these currents ended after 500 ms and suppressed retinal ganglion cell excitability [40]. Another study indicated that retinal responses were more robust if it was stimulated directly [5]. Additionally, it was reported that the retinal ganglion cells are more robust at high frequency stimulations when compared to the stimulations via the retinal network [37,41].

This study showed that the temporal response profiles of retinal ganglion cells were heterogeneous and varied according to stimulation parameters. Moreover, these profiles could be divided into two categories: short and long latency clusters. Early phase spikes occurred due to direct electrical stimulation of the retinal ganglion cells. Late phase spike formation existed due to the complicated interaction of presynaptically connected neurons. Stimulations of short pulse widths, 100 μs, were shown to mildly decrease the presynaptic excitatory effects, therefore decreasing the number of retinal ganglion neurons activated through this way and decreasing the late phase spike components. As the temporal responses following the biphasic stimulation at 100 μs pulse width were evaluated, it was seen that the primary spikes represented the spikes occurring before 20 ms following the stimulation. These were short latency action potentials and occurred due to electrical stimulation rather than presynaptic origins. Although the increasing stimulation current amplitude elicited more spikes than it was compared to lower amplitudes, it was not seen to have a significant effect on bringing together the spikes to primary intervals. The formation of long latency spikes was increased with increasing stimulation amplitudes, especially at 80 μA and above and these were between 80-100 ms. The reason for this increase might be the triggering network stimulation at the deeper units of the retinal layer under the electrodes at increasing current amplitudes.

5 Conclusion

Developments in microelectronic, packaging and biomedical engineering areas and multidisciplinary studies have made significant improvements for the field of visual prosthesis system enabled recently. Although successful results have been obtained on reconstruction of visual restoration, the targeted visual perception have significant shortcomings in terms of resolution, color and phase-depth perceptions. An important part of these shortcomings may be handled by better understanding of the neural mechanisms of the retina during electrical stimulation and how these mechanisms will be controlled in order to mimic useful vision that could allow the formation of physiologically natural spike forms which resemble the electrical signals conducted to the brain. For this, retinal responses are comparatively analyzed depending on the most used stimulation types, charge-balanced biphasic and monophasic, with varying amplitudes. Obtained experimental findings show that the precise and right patterns of stimulation which is necessary to mimic the neural activity that resembles the natural ones are able to generated with low amplitude, charge-balanced, short biphasic wave forms instead of monophasic pulses.

Acknowledgements This work is supported by Turkish Scientific and Technological Research Council (TUBITAK), Project Code: 113E181.

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