Ultrashort Phenomena in Biochemistry and Biological Signaling

Robert Splinter$^{1,2}$

$^1$Splinter Consultants; Graham, NC 27253; USA;
$^2$Physics and Optical Science, University of North Carolina at Charlotte, Charlotte, NC 28223, USA

Email: splinter@consultant.com or rsplinter@gmail.com

Abstract. In biological phenomena there are indications that within the long pulse-length of the action potential on millisecond scale, there is additional ultrashort perturbation encoding that provides the brain with detailed information about the origin (location) and physiological characteristics. The objective is to identify the mechanism-of-action providing the potential for encoding in biological signal propagation. The actual molecular processes involved in the initiation of the action potential have been identified to be in the femtosecond and pico-second scale. The depolarization process of the cellular membrane itself, leading to the onset of the action potential that is transmitted to the brain, however is in the millisecond timeframe. One example of the femtosecond chemical interaction is the photoresponse of bacteriorhodopsin. No clear indication for the spatial encoding has so far been verified. Further research will be required on a cellular signal analysis level to confirm or deny the spatial and physiological encoding in the signal wave-trains of intercellular communications and sensory stimuli. The pathological encoding process for cardiac depolarization is however very pronounced and validated, however this electro-chemical process is in the millisecond amplitude and frequency modulation spectrum.

1. Introduction

In biology there is still insufficient information regarding the encoding of signals with respect to their location of origin. The cellular action potential is fairly similar for all cell, with variability for the specific cells, in particular the cardiac muscle cell [1]. The representative action potential is outlined in Figure 1, the time scale will vary based on a range of physiological factors.

Figure 1. General outline of the human action potential, based on electrochemical mechanisms [1].
Various phenomena and diseases in humans are providing the indication that the central nervous system and the brain have a mechanism of action to identify the location from which a signal originates and what the function of this signal is. When analyzed on a pure macroscopic level the signals are transmitted either by myelinated or unmyelinated nerve cells with primarily the frequency of the pulse train offering the encoding for the magnitude of the signal [1, 2]. The frequency of the pulse-train phenomenon is outlined in Figure 2.

![Figure 2](image.png)

**Figure 2.** Frequency profile of the nerve action potential for various applications. The top pulse-train indicates the changes in magnitude of a single pressure sensor cell stimulated by pressure from the tip of a needle [1, 2].

The nerve cells are linked together to transmit the signal from the sensor or the chemical environment of the organ through a chain of command with divergence to various locations in the spine or the brain for processing. The divergence links the axon of one nerve cell to several dendrites to provide signal amplification by means of greater number of signal paths, hence a stronger signal resulting from superposition. On a practical note, there are several clinical reports that may indicate that there is a location specificity associated with the propagation of nervous impulses. For instance people with reported damage to certain segments of their nervous system have been documented to “learn” to regain control over muscles or process sensory signals through parts of the brain that were not originally associated with those tasks [1]. The fact that the brain can be retrained to overcome physiological and biological interruption in signal transduction can also be mediated by the fact that other sensor information provides the spatial information required for the learning process which is communicated on a chemical level through intra-cellular communications. Other sensors will yield feedback, such as hearing, touch, sight and for instance balance.

In order to describe the potential for high-frequency signal encoding there are documented ultra-short phenomena in physiology that can provide the mechanism-of-action to support such modulation principles [1, 2]. Two important spectroscopic phenomena with respect to biological applications are found in photosynthesis and vision [2, 3, 4, 5, 6, 7, 8]. For vision specifically, the light interaction in the rods of the eye (Rhodopsin) involving the conversion of light into changes in the chemical bond structures under ultra-fast conditions will be described to illustrate the potential for spatial encoding.

One specific example of a long time-scale physiological encoding is found in the depolarization pulse-shape of the cardiac muscle. The depolarization pulse-width for the heart muscle cells are in the order of tens of milliseconds as illustrated in figure 3.
The pulse-shape, pulse-duration, rise- and fall time for the cardiac muscle are influenced by chemical conditions. These chemical conditions include, but are not limited to: biochemical influences from hormones, next to prescription drug as well as recreational drugs, in particular cocaine. Most of these physiological chemical conditions are based on for instance: physiological factors, food or exercise (hormones, pheromones, etc.). For the cardiac muscle the pulse rise and fall time form a mechanism of changing the chemical exchange process, specifically the oxygen supply and absorption and exchange against carbon-dioxide.

2. Background
For vision specifically, the light interaction in the rods of the eye (Rhodopsin) next to the conversion of light into chemical energy and oxygen release in the mammalian skin as well as in the leaves of plants take place on a femtosecond scale.

Vision is one biological phenomenon that has very surprising capabilities [4, 5, 7, 8, 9]. The fluence detection range of the eye covers better than 14 orders of magnitude. The minimum detectable fluence by a single rod is in the order of a single photon. A single molecule of photoactivated rhodopsin (R*) in the cell membrane of the rod catalyzes the activation of up to 1000 transductin molecules, and represents the initial stage in the signal amplification process. The transduction molecules provide various stages of the activation process for depolarization and the formation of the action-potential. The entire cell membrane of the rod has multiple bacteriorhodopsin molecules spread within the membrane.

The cones are more particular about the trigger stimulus and require a significant amount more photons. However, the brain and neural communication mechanisms have filters in place (threshold action-potential, chemical transduction in synapses etc.) to trigger a conscious response when at least, depending on personal acuity, about five to nine action-potentials arrive within less than 100 ms. In the case that single photon detection were possible there would be a continuous stream of visual "noise" under dim illumination, hence this filter is a necessity, not a weakness.

On experimental level, a single rod photoreceptor cell from the eye of a frog has proven to operate as an extremely sensitive detector that can respond to individual photons and provide an electro-chemical reaction that can be registered [10, 11, 12, 13]. The frog rod can also be used establish the coherence of extremely weak pulses of light. The sensitivity of rhodopsin, the photosensitive chemical of the rods (and cones; rod as radiance indicators only, no spectral resolution required), is highest at 568-nm.

Neural filtering provides the biological means of implementing a threshold before the brain will register a sensory input. Neural convergence is the physical joining of several axons or dendrites leading to and from a multitude of nerve cells involved in the signal transduction for a single sensor cell. In the retina the ganglion cells are connected on a one to one order to the rod cells as well as for the cone cells in the fovea. Neural convergence is responsible for increasing sensitivity of the rod (radiance vision; “intensity”), however reduces the cone vision sensitivity.
2.1. Theoretical Background
The mechanism of the chemical response of bacteriorhodopsin to light is described.

In biological cells there are vibrational mode transitions, specifically in amino-acids, that take place on femtosecond or shorter scale [4, 14]. Two important spectroscopic phenomena in biological functionality in the ultrashort timeframe are found in photosynthesis and vision [4, 5]. The light interaction in the cones of the eye (receptor protein: Rhodopsin, chromophore derived from vitamin-A) take place on a femtosecond scale. Rhodopsin is present in both rods and also found in certain cones for vision. In vision the quantum states of the excited molecules yields information on the energy transfer. One significant detectable interaction is $C_{11} = C_{12}$ isomerization.

2.2. Mechanism
The activation of bacteriorhodopsin forms the basis of the action-potential leading to neural recognition.

The energy of the photon response is provided by the cis-trans C11 torsion potential of rhodopsin. By reducing the ‘twisting’ force in the configuration of the C11 position in rhodopsin the twisting angle of the C11=C12 bond is reduced in the negative direction to about -80 degrees [15, 16]. The photoisomerization relaxation energy obtained by this change amounts to an order of 10 kcal/mol. The energy storage base for bathorhodopsin consists approximately 40% of charge separation, with the remaining 60% of conformational distortion, the primary energy carrier mechanism. From rhodopsin, bacteriorhodopsin can be experimentally observed within a 2.3 picoseconds [7, 16] with a quantum yield of 0.67 [7, 11].

3. Theoretical Analysis
The light interaction in the rods and cones of the eye (receptor protein: Rhodopsin, chromophore derived from vitamin-A) take place on a femtosecond scale. Rhodopsin is present in both rods and cones for vision. In vision the quantum states of the excited molecules yields information on the energy transfer. One significant detectable interaction is $C_{11} = C_{12}$ isomerization, which reveals itself through torsional vibrations; specifically the low vibration frequency in bathorhodopsin that shows anomalous behavior. The first chemical process is associated with a transition defined by the Franck-Condon principle, which has an approximate lifetime of 200 fs. [1, 5, 6, 7, 8]. The Frank Condon principle is defined by the Franck-Condon factor (FCF) that relies on the energy eigenfunctions overlap for the two eigenconditions (associated with the Franck-Condon states, energy status based on minimal variability in coordinates within the system) with their respective waveform: $FCF = \int_{-\infty}^{\infty} \Psi_n^\dagger(\alpha)\Psi_m(\alpha) d\alpha$. Initially, there is no change in the molecular geometry associated with the electronic transition, due to the short transaction time, only a vertical transition in the electronic potential. The primary chemical event in rhodopsin is an II-cis to II-trans photoisomerization, a conformational change that involves rearrangement of a large fraction of the retinyl polyene [7, 8]. The photon interaction process and time-line with bacteriorhodopsin is illustrated in Figure 4.
Figure 4. Temporal and spectral profile of energy transfer processes in Rhodopsin - Bathorhodopsin. The photon interaction takes place in the first 50 fs (at the far right of the graph; Franck-Condon event); ‘a’ is Raman emission and ‘b’ a broad-band energetic restructuring fs event. Bathorhodopsin is formed \( \approx 1 - 2 \) ps into the process. The vibrational molecular interaction is most pronounced for 568nm [5, 6], data from Yan et al 1991.

The spectrophotometric interaction for Rhodopsin is illustrated in Figure 5. The predominant absorption of 568 nm is illustrated, representing the Cis-Trans absorption spectrum for the energy transfer in the torsional state of the C11=C12 bond.

**Molecular absorptivity: Rhodopsin**

**Chromophore: C11 cis-trans exchange Within 10fs**

The energy of the photon response is provided by the cis-trans C11 torsion potential of rhodopsin:

10 kcal/mol

Figure 5. Spectral absorption profile for Rhodopsin, identifying the photo-chemical response of a single photon within the 10 fs interaction time frame [8, 9, 10].
After the interaction with the first photon the absorption band of the rhodopsin derivative in the rod of the eye switches its spectral profile as outlined in Figure 6.

**Figure 6.** Time-resolved spectral attenuation profile (x-axis; wavelength: nm) for bacteriorhodopsin as a function of time (y-axis) after the seed photon absorption 1000 fs into the process. Batorhodopsin formed ~1ps after excitation [4, 5, 9, 10, 11], data from Birge 1990.

### 3.1. Comments
Additional research is required on a cellular level to determine the shape of the propagating exact depolarization wavefront along the path of the transported signal. This process will require in-situ non-invasive high temporal resolution measurement of the electro-chemical processes. External electrodes are insufficient to provide this kind of detail since they average the biphasic and triphasic aspects of the depolarization process based on volume conduction and superposition.

### 4. Conclusions
No conclusive evidence for signal encoding has so far been established. No supporting information is currently available to validate the hypothesis of location specific biological communications based on encoding. However on an electro-chemical level there are mechanisms available that can potentially support encoding at the initial stage of pulse generation providing a modulation process.

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