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OPTICAL DATA PROCESSING WITH BACTERIORHODOPSIN AND ITS GENETICALLY MODIFIED VARIANTS

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1. Abstract

With Bacteriorhodopsin (BR) it will be demonstrated that biological systems - optimized by nature under evolutionary conditions - can be applied with great success to solve material requirements in technical systems. The possibility of genetic engineering further increases the technical applicability of these systems. The application of BR and its variants as a molecular processing unit in optical data processing will be shown in several examples.

2. Introduction

Nature has optimized biological systems on a 'molecular device' level by 'trial and error' during a long period of evolution. The first aim of this article is to examine these materials - and especially Bacteriorhodopsin (BR) - from a technical point of view and describe how advantage can be taken of these naturally optimized systems in various technical applications. The second aim is to examine the idea of using conventional mutagenesis or genetic engineering in order to obtain a variety of systems with different properties, where each of them may meet the requirements of a different technical application in a very specific way. This further broadens and optimizes the potential applications of the biological systems.

It should be emphasized that with this direction of research a new approach in materials science has been introduced [1,2]. In one sense this approach is related to the goal of supramolecular chemistry; however, whereas in supramolecular chemistry the chemist tries to synthesize highly organized artificial systems often employing principles of living nature, the above approach uses nature directly as a "supramolecular" chemist.

3. Structure and Function of Bacteriorhodopsin

BR from *Halobacterium halobium* [3] is embedded as a twodimensional crystallin lattice of BR-trimers in the lipid bilayer of the cell membrane. BR consists of a single polypeptide chain of 248 amino acids which is arranged in seven transmembrane α -helices. A retinal molecule bound via a Schiff base to lysine-216 forms the chromophoric group. Under illumination BR creates a proton

gradient across the cell membrane which is used by a membrane-bound ATP-ase for ATP synthesis. Proton transport through the cell membrane is closely connected to the photocycle of BR [4]. Under illumination with light of about 570 nm the photochemical reaction of B→J is induced. From the J-state BR passes through a couple of short-living intermediates to the M-state in about 50 μ s. The M-intermediate has the longest lifetime which is about 10 ms for wildtype BR in suspension. From the M-state BR can relax to the B-state either thermally or photochemically. The photochemical transition from M→B is initiated by the absorption of a photon of, e.g., 413 nm by the M-intermediate.

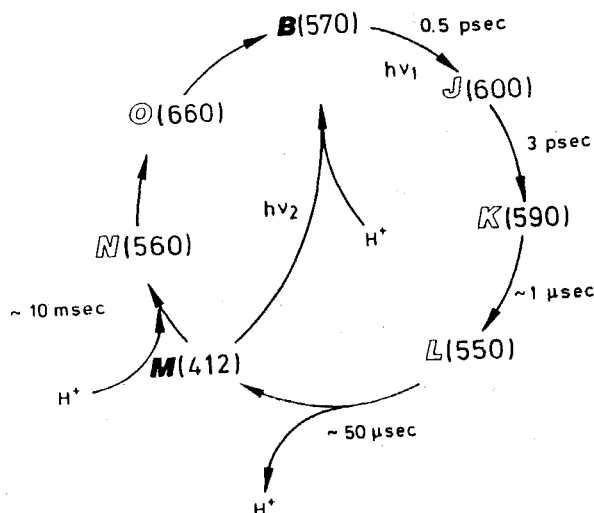


Fig. 1.: Photocycle of Bacteriorhodopsin

4. Holographic Properties of Bacteriorhodopsin and its Mutuated Variants

Dried films of PM-suspension or PM embedded into inert matrices like polyvinylalcohol or polyacrylamide can be used as holographic media. Due to the extreme stability of PM toward photochemical degradation the state of absorption can be changed reversibly at least one million times. For hologram formation and erasure both conversions B→M ("B-type" holograms) and M→B ("M-type" holograms) can be used. Both types of hologram formation induce a local light-dependent population distribution between B and M-state. In B-type recording the information is written with a wavelength inside the absorption band of the initial state B (e.g. 568 nm). M-type recording uses a pumping beam initiating the B→M reaction to achieve a high population of the M-state. The information is recorded with blue light (e.g. 413 nm) which stimulates the M→B reaction. The read-out beam can be simultaneously used as pumping beam; therefore it is not destructive but constructive for the hologram formation [5].

Alternatively the amino-acid sequence of BR can be changed in order to modify the characteristics of the BR-molecule. These modifications can be obtained by conventional mutagenesis of wildtype bacteria and a sophisticated isolation procedure [6]. The variant BR(D96N) which was selected for our experiments differs from the wildtype by the exchange of aspartic acid in position 96 with asparagine. This exchange leads to a loss of the internal proton donor function which results in a strong retardation of the proton dependent thermal relaxation $M \rightarrow B$. Therefore BR(D96N)-films have an about 50% higher recording sensitivity and a two-fold higher diffraction efficiency compared to wildtype films [5].

5. Optical Signal Processing with BR

5.1 Dynamic Optical Filtering with Spatial Light Modulators

Lenses can be used as optical signal processing systems. This is explained in fig. 2a: There the letter "E" is

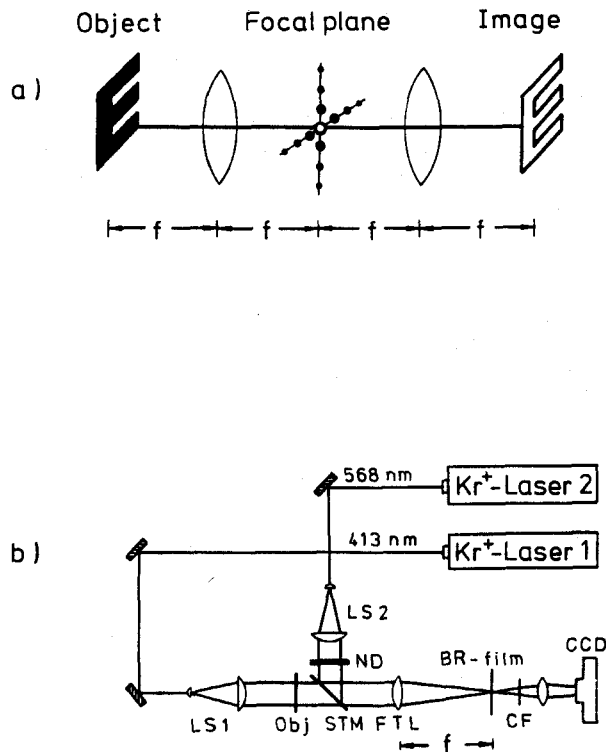


Fig. 2.:

a) Principle of edge-enhancement by suppression of the 0th-order Fourier component.

b) Setup for the dynamical spatial filtering with BR-film.

placed as input object in the front focal plane of the Fourier transform lense FTL1. If the object is illuminated with parallel light, the two-dimensional Fourier transform of the letter "E" is seen in the back focal plane. It is obvious that a second lense FTL2 can retransform the light distribution in the Fourier plane resulting in an identical image of the object. However, weakening or amplifying of selected spatial frequency components by filtering in the Fourier plane will change the output in a definite way. This gives rise to optical signal processing. As an example edge-enhancement of the letter "E" can be obtained by a suppression of the inner part of the Fourier pattern with a mask (high pass filter). However, this can be accomplished with BR and the setup [7] shown in Fig. 2b in a much better and more useful way. In this experiment two lasers operating at wavelength $\lambda_1 = 413$ nm and $\lambda_2 = 568$ nm are used. Let us first follow the beam of laser 1. After expansion it illuminates the object 1 which is a transparency of the letter "E". The Fourier transform pattern is then formed by the first FTL lense and illuminates the BR-film with light of $\lambda_1 = 413$ nm. Since the film is transparent for this light the second FTL lense will finally retransform the Fourier pattern and an unchanged letter "E" will be observed with the CCD-camera. However, when the second beam enters the common light path - assuming for simplicity no object in this beam - the FTL lense will focus (Fourier transform) the beam onto the 0th order Fourier component of the letter "E". Since this beam is of wavelength $\lambda_2 = 568$ nm it will initiate the photoreaction $B \rightarrow M$ in the 0th-order spot and thus diminish or even suppress the light intensity of the 0th-order frequency component. Thus this component is lost for the retransformation by the second FTL lense leading to an edge-enhanced letter "E" on the CCD-camera. For this experiment BR(D96N) was used because of the more efficient population of the M-state, i.e. the stronger suppression of the 0th-order in the Fourier plane. This demonstrates the usefulness of the concept of mutants.

Such experiments are of high interest because they demonstrate that logical operations can be performed between the two inputs at λ_1 and λ_2 . These logical operations are not restricted to subtraction but can in principle involve all kinds of operations in the Fourier plane using proper wavelengths and intensities to manipulate the photocycle of BR. It should be emphasized that the processing operations work in a fully parallel way due to the parallel Fourier transformation of the lense. Further because of the high reversibility of BR a high sequence of consecutive parallel operations can be performed. Finally, it is obvious that in all these (and the following) examples BR acts in the central part of the system representing the molecular processor unit.

5.2 Optical Pattern Recognition

Real-time pattern recognition is of fast growing interest for many practical applications where the identification and localization of objects out of a complex background is necessary, e.g. in robot vision or automatic inspection of large data bases like those of satellite or medical imagery. Whereas digital computers are relatively slow because of the large computational capacities required optical techniques are much faster due to their inherent fully parallel processing. The lower flexibility of optical implementations may be overcome by the use of new designs of correlators and the further development of fast input modulators for rapidly changing the object and filter patterns.

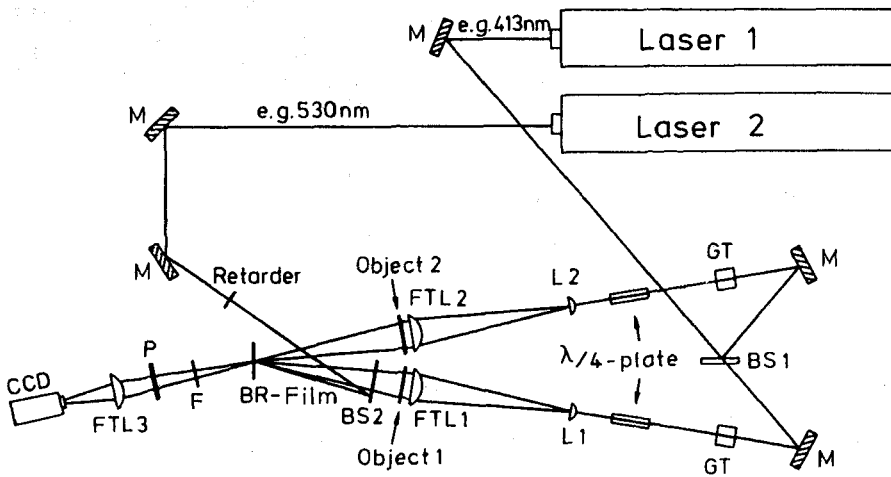


Fig. 3.: A dual-axis joint-Fourier-transform correlator for pattern recognition with a BR-film as processing medium.

In Fig. 3 an optical setup [8] for real-time pattern recognition with a dual-axis joint-Fourier-transform correlator (DA-JFT) is shown. Since again BR will work as a processing medium two lasers are used with $\lambda_1 = 413$ nm and $\lambda_2 = 530$ nm. With the split beam of λ_1 two optical axis are installed containing object 1 (master pattern) and object 2 (filter pattern). Both objects are post-lense Fourier-transformed into the same Fourier plane and recorded into the BR-film. Since the light in both axis is mutually coherent overlapping parts of the Fourier spectra will form a hologram. These overlapping parts identify common patterns of both objects. The hologram (M-type) is read out with the beam of λ_2 of the second laser. The retransformation with lens FTL3 leads to light spots on the frame of the CCD camera indicating the location and the degree of correlation between common patterns of object 1 and 2. Fig 4 shows one of our typical experimental results [9].



Fig. 4.: Holographic pattern recognition with BR-films. On the left side the "master" pattern is shown, on the right side the correlation signal together with the "search" pattern.

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