

## Integrated Optical Switching Based on the Protein Bacteriorhodopsin<sup>†</sup>

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### ABSTRACT

According to our earlier pioneering study, a dry film containing native bacteriorhodopsin (bR) shows unique nonlinear optical properties (refractive index change, controllable by light of different colors, greater than  $2 \times 10^{-3}$ ) that are in many respects superior to those of the materials presently applied in integrated optics. Here, we report on the first integrated optical application based on a miniature Mach–Zehnder interferometer (see Figs. 1 and 2) demonstrating a real switching effect by bR (efficiency higher than 90%) due to the M-state. Our results also imply that the refractive index change of the K-state ( $9 \times 10^{-4}$ ) is high enough for fast switching.

### INTRODUCTION

Since the beginnings of integrated electronics, development has been well described by “Moore’s law”: the density (hence performance) of integrated electronic circuits doubles about every 1.8 years. While this “law” has remained valid for a remarkable period of 30 years, on physical grounds it is hard to see how it can continue much longer without revolutionary new principles. Presently, all possible candidates are being explored in the search for new routes. Molecular electronics combined with optical data processing is regarded as being among the most promising emerging technologies alternative to traditional semiconductor-based electronics.

The key element of nearly every optical application is a thin film of photoactive material (“adlayer”) on a proper solid support (usually glass, waveguide or electrode). Given that theory for optical data processing has already been developed, the basic problem is to find new photomaterials that are more efficient in terms of speed, sensitivity and refractive index variation than the traditional ones (1). Tremendous efforts are concentrated on synthesizing photosensitive dyes with the required properties; however, up to now, natural photomaterials of biological origin seem to be superior in many respects. Among the several biological molecules under consideration for use in computer hardware, the bacterial chromoprotein bacteriorhodopsin (bR) has generated the most interest, because of its favorable nonlinear optical properties, sensitivity

to light polarization, photoelectric activity, repeatability, *etc.* (2–9). Due to these remarkable and modifiable properties, bR can be regarded as a programmable protein.

In this paper, we report on all-optical switching experiments carried out using an integrated optical Mach–Zehnder (M–Z) interferometer. Integrated optics is a rapidly emerging discipline of engineering optics, aiming at integrating miniature active and passive optical elements on the surface of a single substrate (usually of silicon or glass) and, thereby, representing the optical analog of conventional integrated electronics. Passive elements are various waveguide structures confining and guiding the information carrying light *via* total internal reflection, while active elements (*e.g.* modulators and switches), playing the role of transistors in integrated optical circuits, utilize the nonlinear optical properties of an adlayer attached to the waveguide surface. In our case, a simple M–Z interferometer (see Figs. 1 and 2) served as a passive waveguide part of the switch, while a bR film adlayer played the role of the light-sensitive, nonlinear optical material. A highly efficient all-optical switching based on this structure is demonstrated, and possible practical implications of the results are discussed.

### MATERIALS AND METHODS

The waveguide structure was constructed of polymerized NOA81 (Norland Product, Inc.) optical adhesive. First, a 9  $\mu\text{m}$  thick layer of the NOA81 was spin coated onto a glass coverslip. The layer on the coverslip was then taken to the motorized stage of an inverse microscope (Axiovert200, Zeiss). The beam of a Kr-ion laser (Innova 304, Coherent; 407 nm, 10 mW) was focused onto the layer by means of a 10 $\times$  microscope objective. A computer-controlled shutter and the stepper motors of the microscope stage controlled the writing of a M–Z structure into the resin film. After the writing process was completed, the nonexposed part (which is not hardened and stuck to the coverslip glass) was removed by acetone/ethanol 3:1 mixture. The width of the hardened lines was 10  $\mu\text{m}$ .

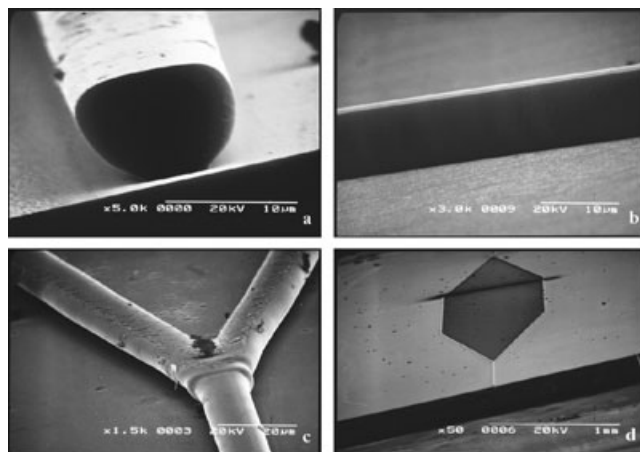
The physical dimensions of the structure allowed incoupling of the light directly from a single-mode optical fiber core (5  $\mu\text{m}$  in diameter). For this reason, the optical fiber was placed and fixed close to the end face of the stripe; finally all parts were fixed to a regular microscope slide that was used as a base plate of the structures (Fig. 1a–d).

bR-containing purple membrane fragments (pm) were prepared according to the standard procedures (10). A water suspension of pm (pH 6 and OD 35) was layered on both arms of the M–Z interferometer and dried at 25°C under a laminar flow for 3 h. The relative humidity of the films was adjusted to be 50%. The transfer function of the interferometer was measured under a microscope (see above). An optical fiber was fixed at the output of the interferometer

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**Figure 1.** Electron micrograph images of the photopolymeric Mach-Zehnder interferometer (two oppositely joined Y-junctions, symmetrically dividing and rejoining the beams, respectively). (a) Cross-sectional view of a single strip. (b) Side view of a single strip. (c) View of a Y-branch. (d) View of the whole Mach-Zehnder structure (shaded inside).

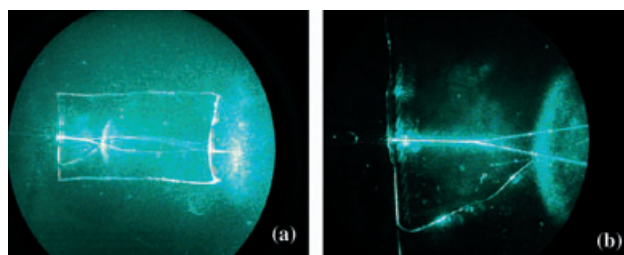
(Fig. 2) in order to guide the transmitted red light to a photomultiplier (Hamamatsu), and its signal was recorded by a digital storage oscilloscope (Le Croy). The beam of an Ar-ion laser (Stabilite2016, Spectraphysics) was used to excite the bR ( $\lambda = 488$  nm, 110 mW), while the measuring light was that of a diode laser ( $\lambda = 676$  nm, 8 mW).

## RESULTS AND DISCUSSION

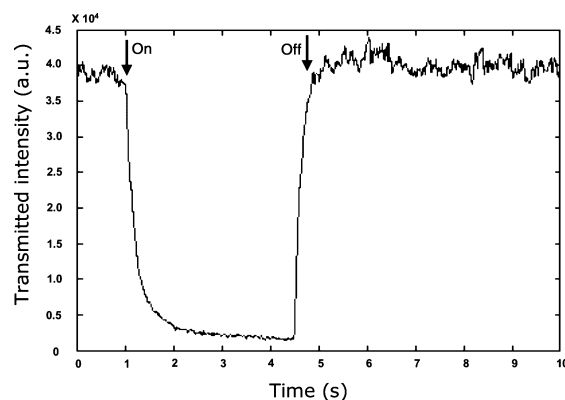
The principle of switching is based on a reversible change of the refractive index of the bR film on one of the arms of the interferometer. The interference of the light at the output can thereby be changed from constructive to destructive or the reverse. Without exposure, the optical environment and the effective refractive index stays the same in both arms. When illuminating only one of the covered arms, a variation in the index of refraction leads to a phase imbalance in the two arms, and interference at the output of the interferometer.

Figure 3 demonstrates the high efficiency of bR-based all-optical integrated optical switching. The exciting light intensity was adjusted to show the maximal effect (25 mW, 90% intensity modulation, respectively). After switching on the exciting light, the steady state is reached within a few seconds, while the decay reflects the relaxation of the refractive index of the bR film after the excitation was switched off.

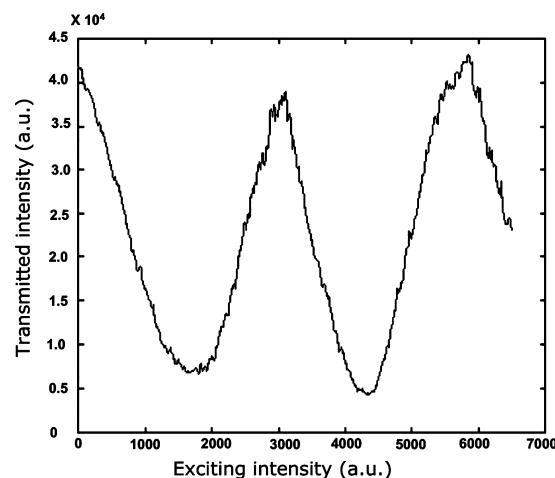
Figure 4 shows the steady-state light intensity measured at the output of the M-Z interferometer as a function of the



**Figure 2.** Photographs of the Mach-Zehnder interferometer guiding green light (488 nm line of an Ar-ion laser).



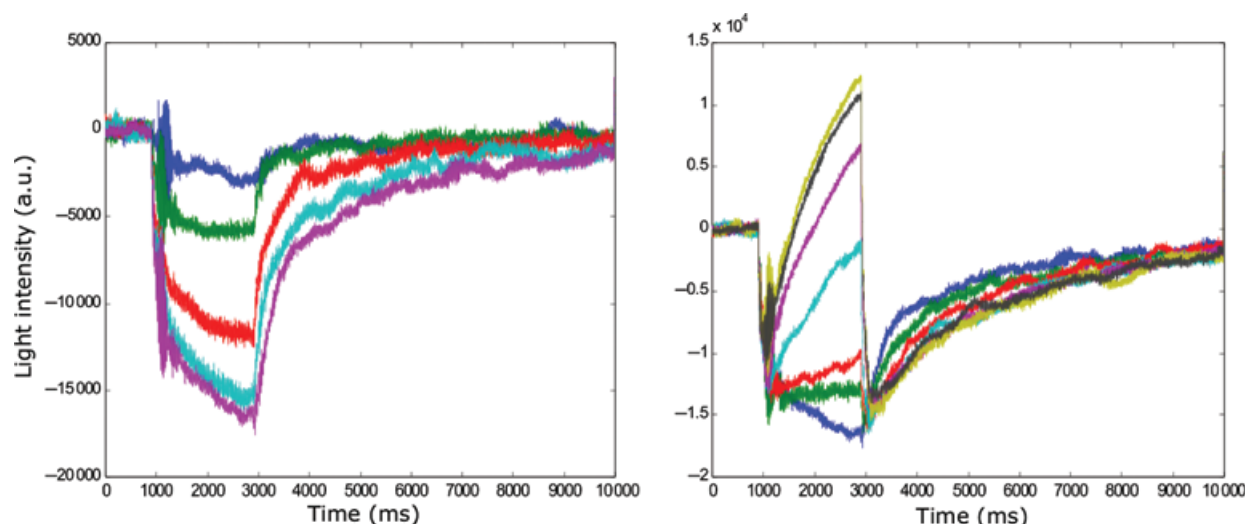
**Figure 3.** Demonstration of the all-optical switching effect by the photopolymeric Mach-Zehnder interferometer covered with a bR film as an adlayer. Guided light: diode laser, 670 nm, 8 mW; Exciting light: 488 nm Ar-ion laser, 25 mW.



**Figure 4.** Sinusoidal modulation of the transmitted red light as a function of exciting light intensity (sample as before, maximal power: 110 mW).

exciting light intensity. In the investigated range, two minima and two maxima were observed, however, no saturation of the effect was found despite the high-intensity excitation.

In order to reveal the reasons for this unexpected finding, analogous experiments were performed on a single-striped waveguide structure. Here, contrary to the M-Z structure (where the observed effects could be attributed to light-induced refractive index changes) only the absorption changes of the bR film were observed. Traces in Fig. 5a,b represent intensity changes of the measuring light passing through the single bR-coated waveguide stripe. Upon excitation by green light, due to its well-known photochromic effect, the protein containing adlayer undergoes absorption changes giving rise to modification of the intensity of the measuring light. The effects are interpreted according to a simplified model describing the bR photocycle at moderate humidities, where the N and O forms are missing and only the K, L and M intermediates are present (Fig. 6). The red light intensity increase at lower levels of excitation (Fig. 5a) is due to the accumulation of the M form in steady-state, where the longest-living intermediate is present with the



**Figure 5.** Changes in the output intensity of red light guided by a single photopolymeric stripe covered with a bR film upon changing the intensity of exciting green light by 10 mW increments (distinguished by different colors). Note that the signal increases first (power range: 10–50 mW, a), then undergoes a turnover phase (power range: 50–110 mW, b).

highest weight. Under conditions where the concentration of M is maximal (at about 45 mW exciting light, corresponding to the second maximum in Fig. 4), the size of the refractive index change (an actual refractive index *decrease*) of the film is in the range of  $2 \times 10^{-3}$ , as observed previously upon flash excitation (11) by the optical waveguide lightmode spectroscopy (OWLS) technique (12). The subsequent absorption increase at 676 nm shows that another photoproduct is accumulated at higher light intensities that is spectrally redshifted compared with the ground-state bR. Note that after switching off the excitation, the absorption always returns to a minimum value corresponding to M-saturation before final relaxation. This finding strongly suggests that the redshifted intermediate returns to the ground state (bR) via M. Taking into account the well-known fact that light excitation of both the L and M intermediates drives them back to the ground state (13,14), we suggest that the implicated redshifted intermediate must be K or of its type.

As M is spectrally blueshifted, while K is redshifted as compared with the ground state, their contribution to the refractive index change at this wavelength must be of opposite sign. At a certain exciting light intensity (corresponding to the third maximum in Fig. 4), these contributions compensate each other and the original output level of the M–Z interferometer is restored. At the highest applied light intensity, the

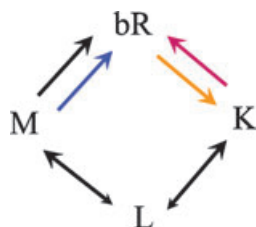
contribution of K dominates the absorption change, consequently the refractive index change of the film must be positive, and was estimated to be  $9 \times 10^{-4}$ . This high value, which is comparable with that accompanying the BR–M transition but with opposite sign, should be interpreted by the anomalous refractive index of K having an absorption maximum close to the wavelength of the measuring light.

Our results imply that the K intermediate has remarkable nonlinear optical properties that look even more favorable than those of M due to the picosecond kinetics of the refractive index change and may readily be utilized in integrated optics or other nonlinear optical applications.

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## REFERENCES

- Service, R. F. (1995) 2 steps for light-altering polymers. *Science* **268**, 1570.
- Takei, H. and N. Shimizu (1994) Nonlinear optical properties of a bacteriorhodopsin film in a Fabry-Perot cavity. *Opt. Lett.* **19**, 248–250.
- Birge, R. R. (1995) Protein-based computers. *Sci. Am.* **272**, 90–95.
- Vsevolodov, N. N. (1998) *Biomolecular Electronics: An Introduction via Photosensitive Proteins*, pp. 71–217. Birkhauser, Boston.
- Korchemskaya, E. Y., D. A. Stepanchikov, A. B. Druzhenko and T. V. Dyukova (1999) Mechanism of nonlinear photoinduced anisotropy in bacteriorhodopsin and its derivatives. *J. Biol. Phys.* **24**, 201–215.
- Hampp, N. (2000) Bacteriorhodopsin as a photochromic retinal protein for optical memories. *Chem. Rev.* **100**, 1755–1776.
- Váró, G. and L. Keszthelyi (1983) Photoelectric signals from dried oriented purple membranes of *Halobacterium-halobium*. *Biophys. J.* **43**, 47–51.
- Dér, A., P. Hargittai and J. Simon (1985) Time-resolved photoelectric and absorption signals from oriented purple membranes immobilized in gel. *J. Biochem. Biophys. Methods* **10**, 295–300.
- Dér, A. and L. Keszthelyi (2001) *Bioelectronic Applications of Photochromic Pigments*, pp. 6–224. IOS Press, Szeged, Hungary, Amsterdam.



**Figure 6.** A simplified scheme of the photocycle of dried bacteriorhodopsin. Black arrows represent thermal, while colored ones light-driven reactions. The color of the arrows correspond to the color of the exciting light, respectively, and the size of the arrow heads indicate the relative probability of the transition.

10. Oesterhelt, D. and W. Stoerkenius (1974) Isolation of the cell membrane of *Halobacterium halobium* and its fractionation into red and purple membrane. *Methods Enzymol.* **31**, 667–678.
11. Ormos, P., L. Fábrián, L. Oroszi, E. K. Wolff, J. J. Ramsden and A. Dér (2002) Protein-based integrated optical switching and modulation. *Appl. Phys. Lett.* **80**, 4060–4062.
12. Ramsden, J. J. (1994) Experimental methods for investigating protein adsorption-kinetics at surfaces. *Quart. Rev. Biophys.* **27**, 41–105.
13. Balashov, S. P. (1995) Photoreactions of the photointermediates of bacteriorhodopsin. *Isr. J. Chem.* **35**, 415–428.
14. Ormos, P., Z. Dancsházy and L. Keszthelyi (1980) Electric response of a back photoreaction in the bacteriorhodopsin photocycle. *Biophys. J.* **31**, 207–213.