trolled elution was developed for the second time range. From data in this time domaine the blocking effect of tetrodotoxin was found to occur at K $_{\rm app}$ 5x10 $^{-0}$ M. Simple theoretical models are used to estimate the influence of molecular transport parameters on experimental data from efflux assays.

Spectroscopical studies of the ASTACUS visual pigment

K.J. Hamacher and K.-D. Kohl ++

*Institut für Biotechnologie und **Institut für Neurobiologie der KFA, D-5170 Jülich, FR Germany

The rhabdoms of a single dark adapted (20 h) crayfish retina were isolated by density gradient centrifugation. The quantity of isolated and purified rhabdoms, which was entirely free from screening pigments, was sufficient to allow measurements of the extinction spectra of the membrane-bound chromoprotein obtained from a single retina.

Extinction spectra of the membrane-bound chromoprotein show that at 0°C and pH 7.0 the system is stable and photoreversible for several days. The maximum of the extinction spectrum of ASTACUS rhodopsin (0°C, pH 7) is $\lambda_{\rm max}$ = 530 nm. Rhodopsin (R) is isomerized nearly quantitatively to metarhodopsin (M) ($\lambda_{\rm max}$ = 500 nm) by irradiation at λ = 630-640 nm.

Kinetic studies of the conversion from R to M and vice versa were performed by flash photolysis. The resolution of flash-induced transmission changes of the rhabdom suspension allowed single shot experiments (pulse length of the exciting light 6 ns). From our measurements we postulated the following temporary reaction scheme:

$$R \underset{R'}{\swarrow} M' \xrightarrow{M} M$$

Both reaction pathways, $R \rightarrow M$ and $M \rightarrow R$, consist of a photoreaction and at least one dark reaction; additional dark reactions exist presumably in the time range < 10 μs . The kinetic data for both dark reactions are investigated in the temperature range from 0° to 20° C. Increasing the temperature from 0° to 20° C the half times for the dark reactions decrease from 3.9 ms to 0.24 ms in the case of $M' \rightarrow M$ and from 16 ms to 1 ms in the case of $R' \rightarrow R$.

The activation energies were calculated from the slope of the Arrhenius plot and seem to be the same for both reactions, $\Delta E_{M' \to M} = \Delta E_{R' \to R} = 22.5$ kcal mol⁻¹.

Flash-spectroscopic measurements in the wavelength range from 430 to 650 nm show that the difference spectra of M-R' and M-R have the same maximum at 495 nm and the same minimum at 575 nm. This suggests that the absorption maxima of R and R' are similar, but differ in the molar extinction coefficient.