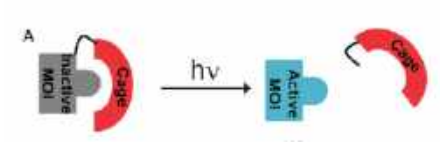
13. Photo-control of biological function

1. Photocaging
2. Azobenzene photoswitching
3. Photoswitching applications

1

Photocontrol of biological activity

Photocaging



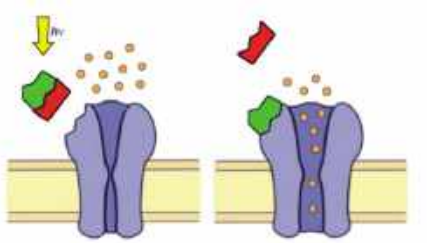


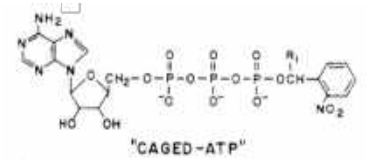
Fig. 2 Activation of an ionotropic receptor by photorelease of a neurotransmitter.

2

Photocontrol of biological activity — photocaging

Rapid Photolytic Release of Adenosine 5′-Triphosphate from a Protected Analogue: Utilization by the Na:K Pump of Human Red Blood Cell Ghosts†

Jack H. Kaplan, Bliss Forbush III, and Joseph F. Hoffman*



These "caged ATP" compounds released adenosine 5′-triphosphate on illumination at **340 nm** in aqueous solution and P3-1-(2-nitro)phenyl- ethyl-ATP gave about a **70% yield** in **under 30 s**.

Hoffman *Biochemistry* **1978**, *17*, 1929

3

Photocontrol of biological activity — photocaging

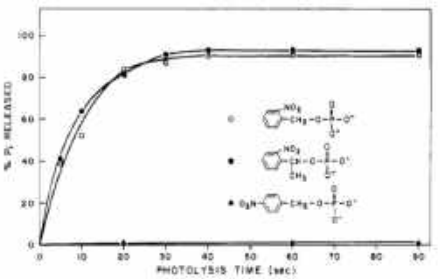
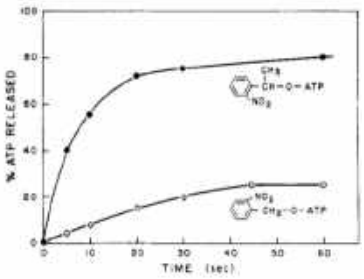
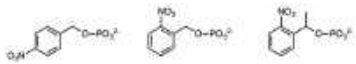


FIGURE 3: Comparison of photorelease of P_i from phosphate esters. Aqueous solutions (2×10^{-4} M) of the phosphate esters were photolyzed as described in Materials and Methods, and the photolyzed solution was then assayed for free P_i.



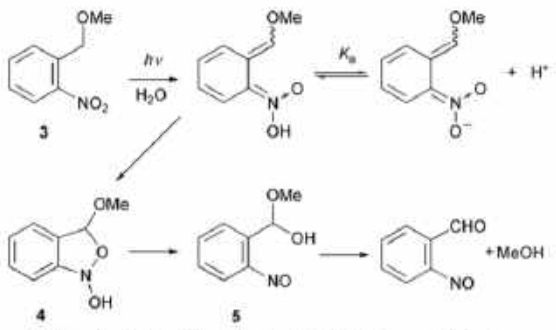
- *Ortho* position is required
- Substituted benzylic position is faster



Hoffman *Biochemistry* **1978**, *17*, 1929

4

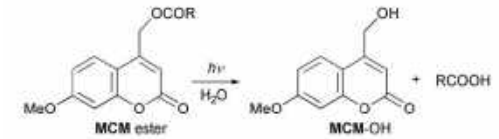
Photocaging mechanism



Scheme 4 Revised[17] mechanism for 2-nitrobenzyl derivatives.

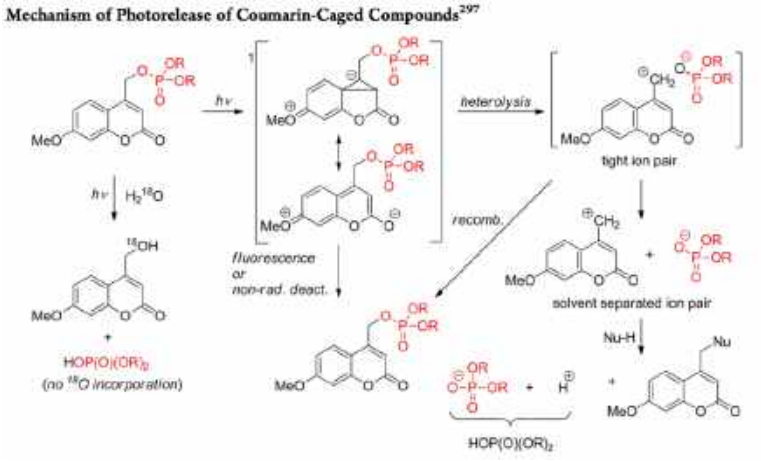
Wirz *Photochem. Photobiol. Sci.* **2002**, 441

Coumarinyl group



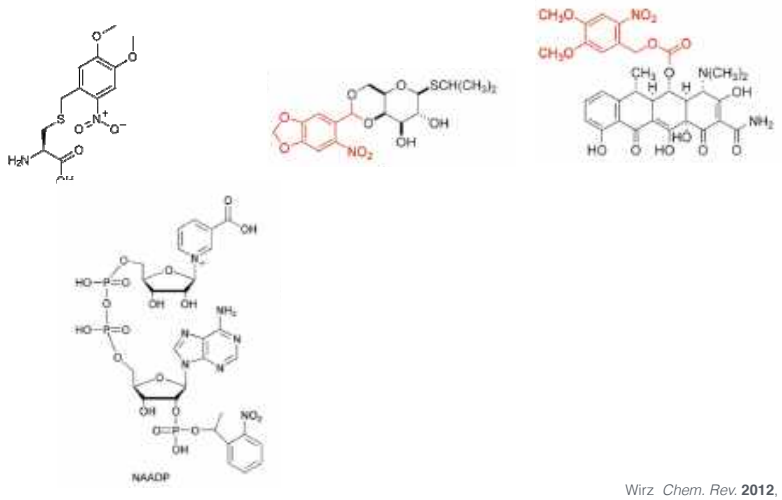
Scheme 11 Photocleavage of MCM esters.

Wirz *Chem. Rev.* **2012**, 119



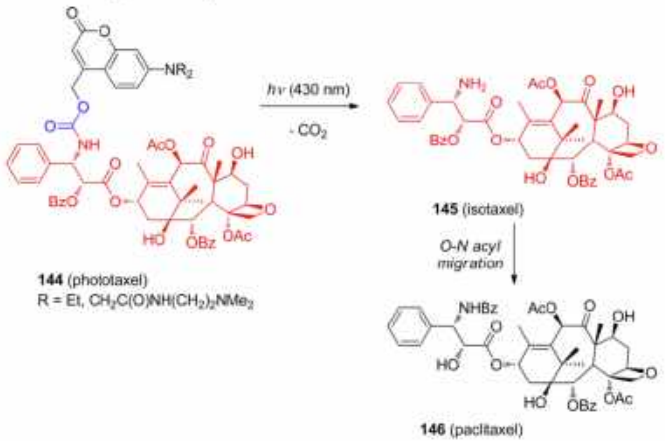
Wirz *Chem. Rev.* **2012**, 119

Examples of Photocaged Molecules



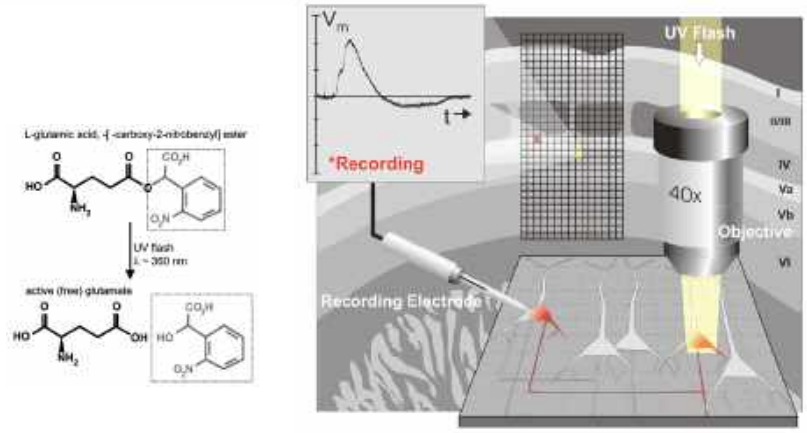
Wirz *Chem. Rev.* **2012**, 119

Activation of Paclitaxel by Visible Light[283]



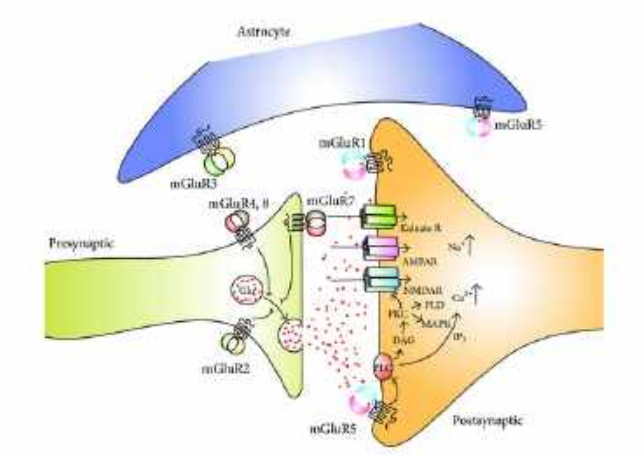
Wirz *Chem. Rev.* **2012**, 119

Optical release of caged glutamate for stimulation of neurons in the *in vitro* slice preparation



Koetter *J. Biomed. Optics* **2005**, 011003

ionotropic glutamate receptor (iGluR)



Koetter *J. Biomed. Optics* **2005**, 011003

Photocaged Glutamate to trigger action action potentials in neurons

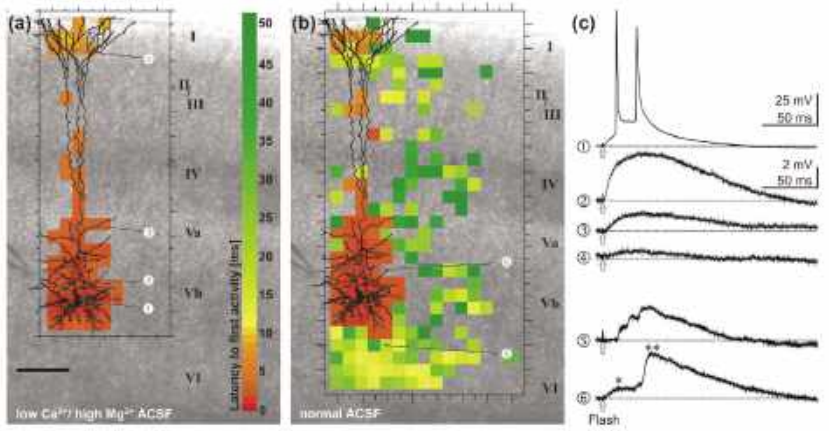
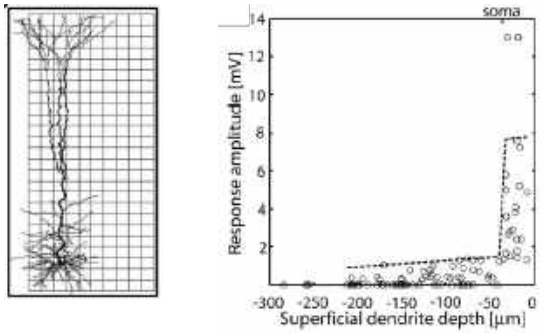


Fig. 4 Topographical maps of activity induced by flash photolysis of caged glutamate for an IB pyramidal neuron from layer Vb (cf. Ref. 10). (a)

Koetter *J. Biomed. Optics* **2005**, 011003

Photocaged Glutamate to trigger action action potentials in neurons



Morphology, peak response amplitudes, depth of most superficial neuronal structure, and relationship between response amplitude and depth for a layer Vb IB cell.

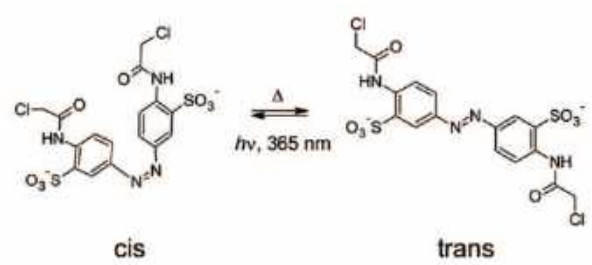
Koetter *J. Biomed. Optics* **2005**, 011003

13. Photo-control of biological function

1. Photocaging
2. Azobenzene photoswitching
3. Photoswitching applications

Photoswitching

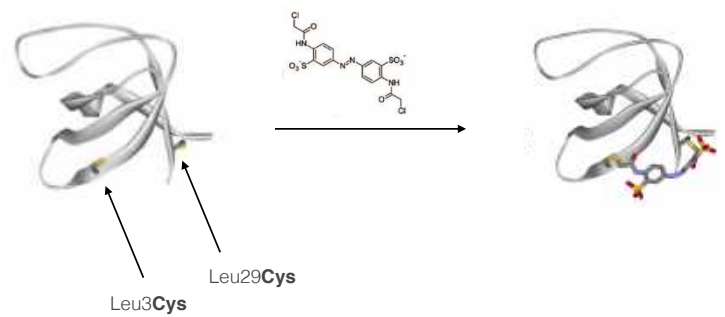
Chemical structure of the **thiol-reactive** BSBCA linker in *cis* and *trans* forms.



BSBCA: 3,3′-bis(sulfonato)-4,4′-bis(chloroacetamide) **azobenzene**

Woolley *J. Am. Chem. Soc.* **2009**, *131*, 2283

Photoswitching

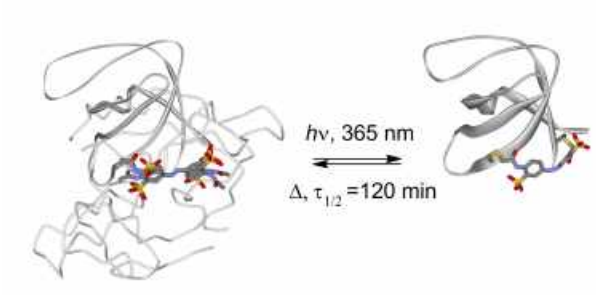


model: FynSH3 domain
(Fyn-tyrosin kinase)

free energy of folding of wild-type FynSH3 is ~**4.2 kcal/mol**

Woolley *J. Am. Chem. Soc.* **2009**, *131*, 2283

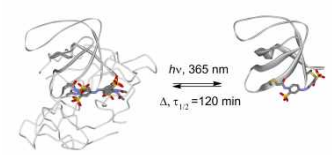
Photoswitching

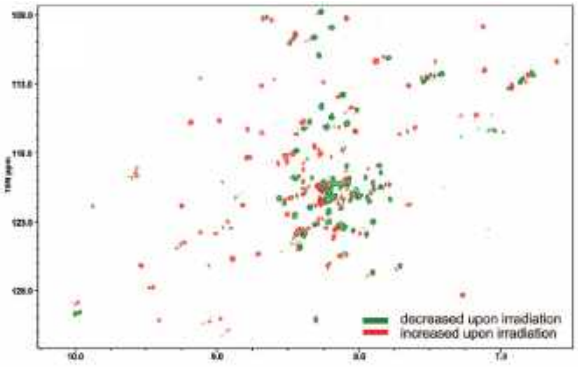


The energy available from azobenzene isomerization (~**10 kcal/mol**) is larger than the folding free energies of many proteins. Thus photo-control of protein function via control of folding should be possible.

Woolley *J. Am. Chem. Soc.* **2009**, *131*, 2283

Conformational changes measured by NMR



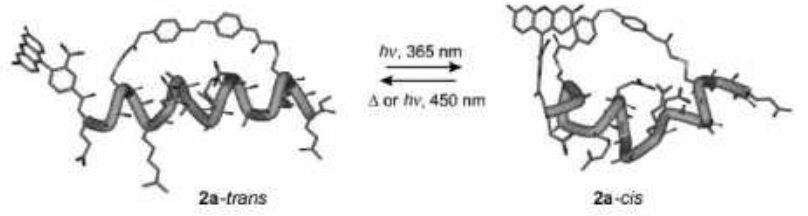


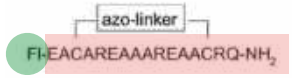
Difference (^{1}H,^{15}N) **HMQC spectrum** of X-L3C-L29C-T47AFynSH3.

Signals that increased upon irradiation are shown in red; signals that decreased are shown in green.

Woolley *J. Am. Chem. Soc.* **2009**, *131*, 2283

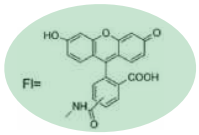
Irradiation at 365 nm of an azobenzene-cross-linked peptide, causes ***trans-to-cis*** isomerization, which enhances helicity, favors a coiled-coil formation.





Irradiation in the UV range (350–390) causes ***trans*-to-*cis* isomerization** of the photoswitch as well as excitation of **fluorescein**. Irradiation with blue light (440–490) causes ***cis*-to-*trans* isomerization** as well as fluorescein excitation.

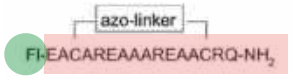
• The *cis* isomer of the peptide has a lower quantum yield for fluorescence than the *trans* isomer. Irradiation of the reporter peptide with UV light produces a time-dependent fluorescence decrease. Conversely, irradiation of *cis* isomer with blue light causes a time-dependent fluorescence increase.

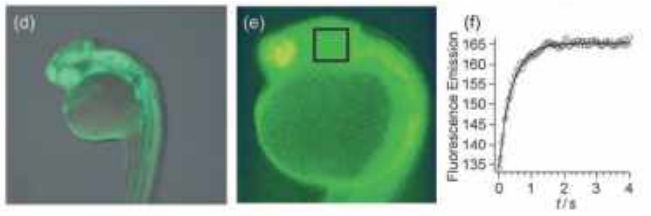


Woolley *Angew. Chem. Int. Ed.* **2011**, 1325

Fluorescence Imaging of Azobenzene Photoswitching In Vivo

(proof of concept)





The **D-peptide** analog of **2a** (chosen to minimize the possibility of protease cleavage) was microinjected into zebrafish embryos at the 1–2 cell stage. Zebrafish are optically transparent while young. Embryos were incubated at 28 °C and imaged using fluorescence microscopes. Illumination with a UV bandpass filter was used to drive ***trans-to-cis* isomerization**.

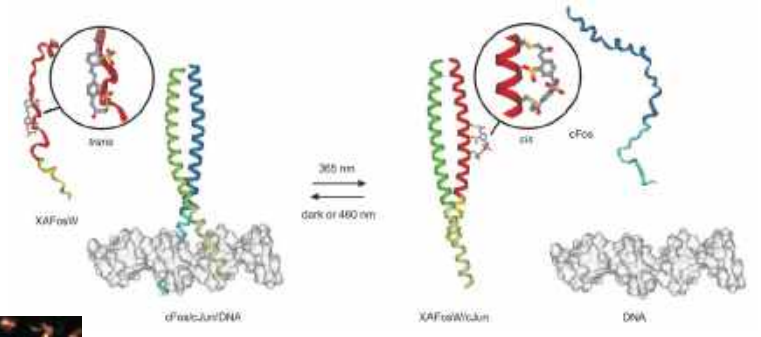
Woolley *Angew. Chem. Int. Ed.* **2011**, 1325

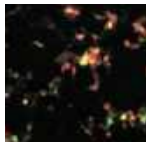
Light-Switchable Proteins

Photocontrol of Coiled-Coil Proteins in Living Cells**

Fuzhong Zhang, Katharina A. Tinn, Katja M. Arndt, and G. Andrew Woolley**

FosW inhibition of the DNA binding of homodimeric cJun/cJun. Coiled coils in the form of basic leucine zipper(bZIP) domains are common in transcription factors that control gene expression.





Irradiation at 365 nm of an azobenzene-cross-linked AP-1 dominant negative peptide, XAFosW, causes trans-to-cis isomerization, which enhances helicity, favors a coiled-coil formation, and thereby leads to the inhibition of DNA binding.

Woolley *Angew. Chem. Int. Ed.* **2010**, 3943

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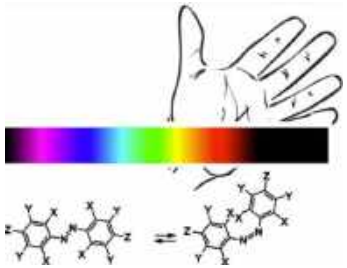
ACCOUNTS of chemical research

Red-Shifting Azobenzene Photoswitches for in Vivo Use

Mingxin Dong,† Amirhossein Babalhavaeji,† Subhas Samanta,†,‡ Andrew A. Beharry,†,§ and G. Andrew Woolley*,†



Woolley *Acc. Chem. Res.* **2015**, 2662

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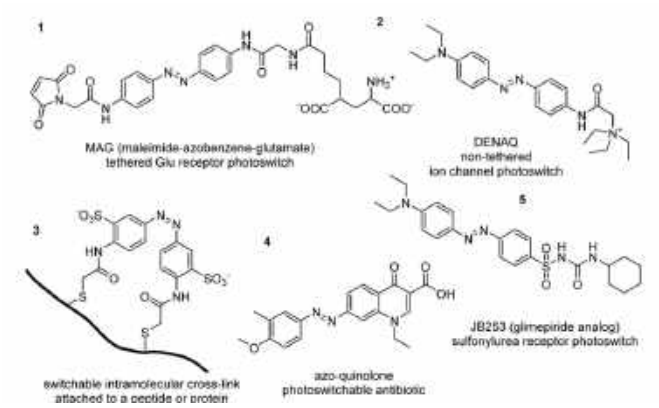
Red-Shifting Azobenzene Photoswitches for in Vivo Use

Mingxin Dong,† Amirhossein Babalhavaeji,† Subhas Samanta,†,‡ Andrew A. Beharry,†,§ and G. Andrew Woolley*,†



Woolley *Acc. Chem. Res.* **2015**, 2662

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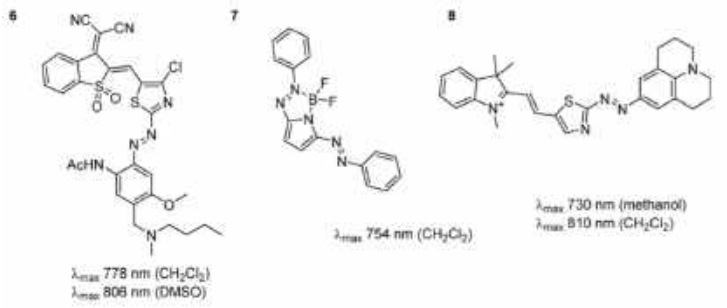
Red-Shifting Azobenzene Photoswitches for in Vivo Use

Mingxin Dong,† Amirhossein Babalhavaeji,† Subhas Samanta,†,‡ Andrew A. Beharry,†,§ and G. Andrew Woolley*,†

Some long wavelength azo dyes



Bruce *Proc. Amer. Soc. Mass Spec.* **2014**

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13. Photo-control of biological function

1. Photocaging
2. Azobenzene photoswitching
3. Photoswitching applications

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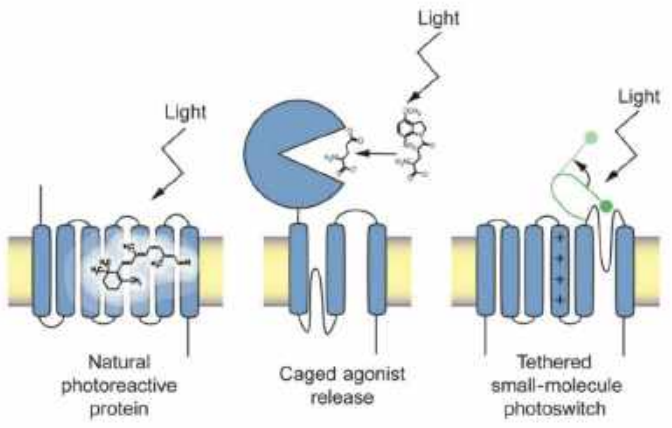
nature chemical biology

Photochemical tools for remote control of ion channels in excitable cells

Richard H Kramer, James J Chambers & Dirk Trauner



Trauner *Nature Chem. Biol.* **2005**, 360

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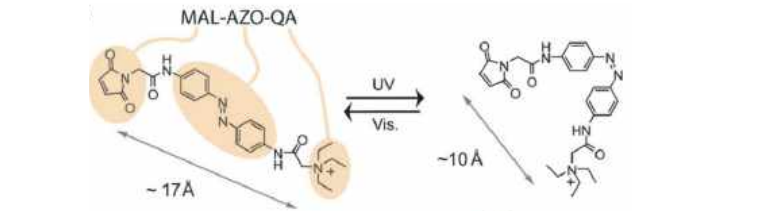
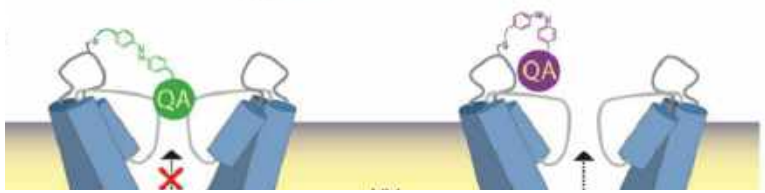
nature chemical biology

Photochemical tools for remote control of ion channels in excitable cells





Trauner *Nature Chem. Biol.* **2005**, 360

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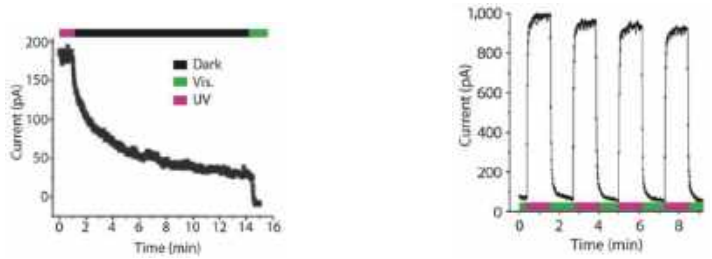
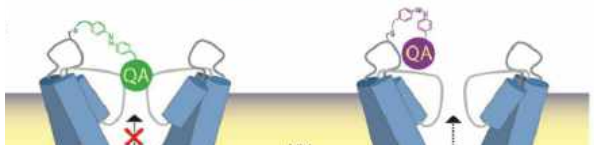
nature chemical biology

Photochemical tools for remote control of ion channels in excitable cells





Trauner *Nature Chem. Biol.* **2005**, 360

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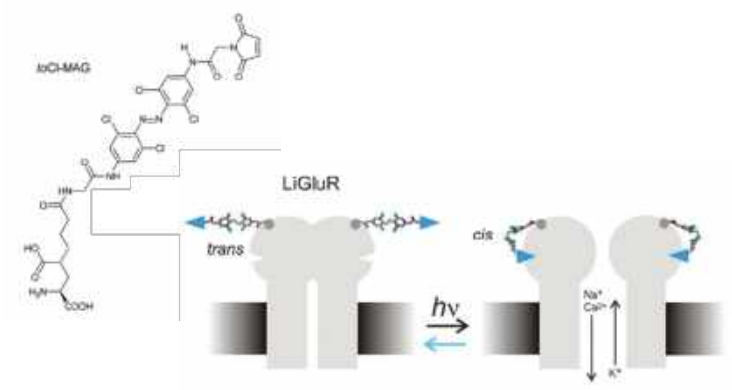
Long wavelength optical control of glutamate receptor ion channels using a tetra-*ortho*-substituted azobenzene derivative†

A. Rullo,‡[a] A. Reiner,‡[b] A. Reiter,[c] D. Trauner,[c] E. Y. Isacoff[b] and G. A. Woolley*[a]



Wolley, Trauner *Chem. Comm.* **2014**, 14613

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Long wavelength optical control of glutamate receptor ion channels using a tetra-*ortho*-substituted azobenzene derivative†

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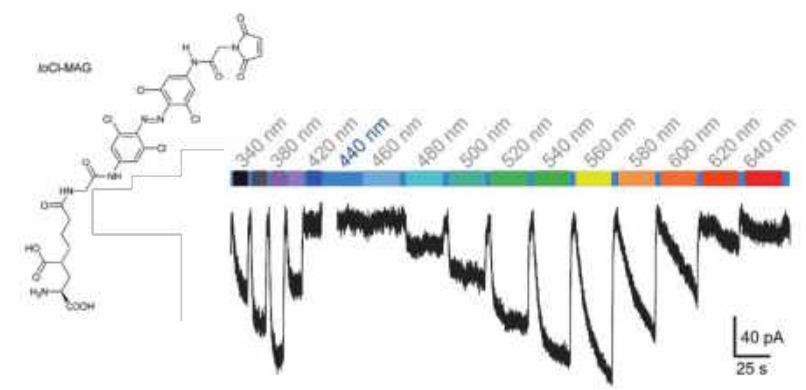

- Maximum channel current in the yellow range (560 nm) — **open**
- No current at all in the blue range (440 nm) — **closed**

Wolley, Trauner *Chem. Comm.* **2014**, 14613

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Long wavelength optical control of glutamate receptor ion channels using a tetra-*ortho*-substituted azobenzene derivative†

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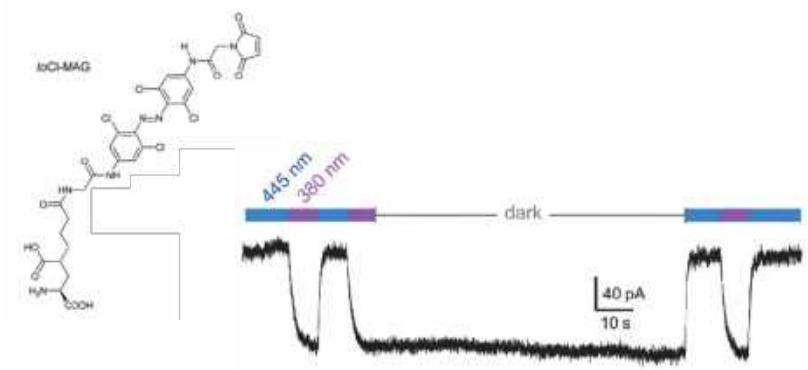

- **Open** *and* **Closed** conformations are stable

Wolley, Trauner *Chem. Comm.* **2014**, 14613

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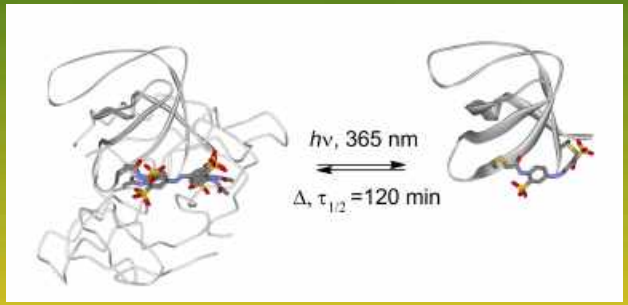
13. Photo-control of biological function



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