

Photoactivatable ("Caged") Derivatives of Glutamic Acid and NMDA

Introduction

Light activation of "caged" neurotransmitters allows controlled delivery of physiological stimuli with spatial and temporal precision that far exceeds that of microinjection or perfusion (Figure 1).¹ Molecular Probes offers several structurally distinct caged derivatives of the excitatory neurotransmitter L-glutamic acid, with different activity suppression and photochemical release properties (Table 1). Glutamate activates a family of ionotropic receptors that includes the kainate, AMPA and NMDA subtypes. We also offer a caged derivative of the subtype-specific ligand NMDA (*N*-methyl-D-aspartic acid) — β -(DNBH-caged) NMDA (M-13652). Attachment of the DNBH (2,2' dinitrobenzhydryl) caging group yields a derivative that is completely inactive prior to photolysis and is moderately water-soluble. Photolysis occurs on the microsecond timescale with quantum yield of 0.18.²

Materials

Caged glutamate derivatives C-7122, G-2504 and G-7055 are all supplied as solids in units of 5 mg. β -(DNBH-caged) NMDA (M-13652) is supplied as a solid in units of 1 mg. All these products should be stored frozen at -20°C, desiccated and PROTECTED FROM LIGHT.

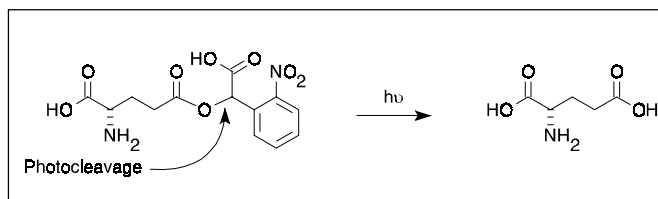


Figure 1. Ultraviolet photolysis of γ -(CNB-caged) L-glutamic acid (G-7055) rapidly releases the excitatory neurotransmitter L-glutamic acid.

Stock Solutions

Stock solutions (≥ 1 mM) may be prepared in the solvents listed below.

- C-7122 aqueous buffer
- G-2504 aqueous buffer
- G-7055 aqueous buffer, DMSO

Stock solutions (≥ 1 mM) of β -(DNBH-caged) NMDA (M-13652) may be prepared in DMSO. This compound is also moderately soluble (~ 0.25 mM) in aqueous buffers.²

Table 1. Molecular Probes' caged glutamate derivatives.

Catalog #	Caging Group	Caging Position	Notes
G-7055	CNB ¹	γ -carboxyl	Rapid photolysis with high quantum efficiency ²
C-7122	CNB ¹	α -amino	Highly resistant to spontaneous hydrolysis in water ^{2,3}
G-2504	DMNB ⁴	α -carboxyl	Slow photolysis with low quantum efficiency

1. α -carboxy-2-nitrobenzyl; 2. Proc Natl Acad Sci USA 91, 8752 (1994); 3. Science 286, 110 (1999); 4. 4,5-dimethoxy-2-nitrobenzyl.

Photoactivation

Photoactivation (uncaging) of these compounds is accomplished by exposing them to ultraviolet light (wavelengths

≤360 nm). Suitable light sources include lasers,³ flashlamps and suitably-equipped fluorescence microscopes.^{4,5} A listing of commercial suppliers of instrumentation specifically designed for photolysis of caged compounds is shown in Table 2.

Table 2. Suppliers of instrumentation for photolysis of caged compounds.

Company	Location	Web site
Cairn Research Ltd.	Faversham, UK	www.cairnweb.com
Intracellular Imaging, Inc.	Cincinnati, OH, USA	www.intracellular.com
Fryer Company, Inc.	Huntlely, IL, USA	www.fryerco.com/prairie
Hi-Tech Scientific	Salisbury, UK	www.hi-techsci.co.uk
Photonic Instruments, Inc.	Arlington Heights, IL, USA	www.photonic-instruments.com
Rapp OptoElectronic	Hamburg, Germany	www.rapp-opto.com
T.I.L.L. Photonics	Martinsried, Germany	www.till-photonics.com
Prairie Technologies, LLC	Middleton, WI	www.prairie-technologies.com

References

1. *Methods in Enzymology*, Volume 291, G. Marriott, ed. Academic Press (1998); 2. *Biochemistry* 38, 3140 (1999); 3. *J Neurosci Methods* 66, 47 (1996); 4. *Intracellular Messengers* (Neuromethods, Volume 20), A. Boulton, G. Baker and C. Taylor, eds., Humana Press (1992) pp 369–396; 5. *Biotechniques* 23, 268 (1997).

Product List Current prices may be obtained from our Web site or from our Customer Service Department.

Cat #	Product Name	Unit Size
G-2504	L-glutamic acid, α -(4,5-dimethoxy-2-nitrobenzyl) ester, hydrochloride (α -(DMNB-caged) L-glutamic acid)	5 mg
G-7055	L-glutamic acid, γ -(α -carboxy-2-nitrobenzyl) ester, trifluoroacetic acid salt (γ -(CNB-caged) L-glutamic acid)	5 mg
C-7122	<i>N</i> -(α -carboxy-2-nitrobenzyl)-L-glutamic acid (<i>N</i> -(CNB-caged) L-glutamic acid)	5 mg
M-13652	<i>N</i> -methyl-D-aspartic acid, β -(2,2'-dinitrobenzhydryl) ester, trifluoroacetic acid salt (β -(DNBH-caged) NMDA)	1 mg

Contact Information

Further information on Molecular Probes' products, including product bibliographies, is available from your local distributor or directly from Molecular Probes. Customers in Europe, Africa and the Middle East should contact our office in Leiden, the Netherlands. All others should contact our Technical Assistance Department in Eugene, Oregon.

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