Understanding the Relationship Between Photolysis Efficiency and Metal Binding Using ArgenCast Photocages

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ABSTRACT

ArgenCast-1 (1), a photocage for silver utilizing acyclic polythioether 3,6,12,15-tetrathia-9-azaheptadecane receptor and 4,5-dimethoxy-2-nitrobenzyl (DMNB) chromophore has been prepared using trimethylsilyl trifluoromethanesulfonate-assisted electrophilic aromatic substitution. Metal binding studies with ArgenCast-1 reveal interactions with Ag⁺, Hg²⁺ and Cu⁺, but only Ag⁺ coordinates in both aqueous and organic solvents. The uncaging mechanism of ArgenCast-1 metal complex involves a photoreaction that converts the nitrobenzydrol into the electron withdrawing nitrosobenzophenone that participates in a resonance interaction with a metal-bound aniline nitrogen atom. The structural change following photolysis decreases availability of the nitrogen lone pair for Ag⁺ coordination, but strong interactions between Ag⁺ and the thioether ligands mitigates metal ion release. A resonance interaction with a key aci-nitro intermediate reduces the photolysis quantum yield of ArgenCast-1, so several naphthyl-based nitrobenzyl groups were screened as alternatives to DMNB. The naphthyl Ag⁺ photocages, Argen-Cast-2 and -3, exhibit nearly identical quantum yield to ArgenCast-1 owing to the dominance of resonance between aci-nitro intermediate and the aniline nitrogen atom.

INTRODUCTION

Initially, photocages were developed to interrogate the biological activity of adenosine-5'-triphosphate (ATP) using light (1). Photocages allow spatial- and temporal-controlled introduction of analytes by blocking biological function until a photoreaction initiates activator release (2). The first photocaged complexes were developed to study Ca²⁺-based signal transduction pathways by Tsien and Zucker (3). The original Ca²⁺ photocaged complexes integrate a nitrobenzhydrol photoactive group into the aniline-derived chelator 1,2-*bis*(*o*-aminophenoxy)ethane-*N*, *N*,*N'*,*N'*-tetraacetic acid (BAPTA). When irradiated, the benzhydrol is converted into a benzophenone. The resulting resonance interaction between the Ca²⁺-bound aniline lone pair and the benzophenone carbonyl decreases the chelators' affinity for the metal ion, which shifts the equilibrium toward unbound Ca²⁺.

In addition to the various photocaged complexes that have elucidated the roles of Ca^{2+} in neurotransmission, we have

developed several tools for controlling the release of Mg^{2+} (4), Fe^{3+} (5,6), Hg^{2+} (7) and Zn^{2+} (8) that utilize analogous uncaging strategies. We have dubbed the class of photocaged complexes based on the nitrobenzhydrol to benzophenone conversion *Cast* complexes, because the metal ion is *cast off* after photolysis. To date, adapting the Cast uncaging strategy for use with soft metal ions has been problematic, so we were interested in further elucidating the scope of applications for this family of photocaged complexes. Soft metal ions are involved in neurotransmission like Cu⁺ (9), neural toxicity like Hg^{2+} (10) and therapeutics like Ag^{+} (11), so there is a wide scope of possible uses for the resulting photocaged complexes.

MATERIALS AND METHODS

General synthetic procedures. All the materials listed below were of a research grade or a spectro-grade in the highest purity commercially available from Acros Organics or TCI America. Dichloromethane (CH_2Cl_2) , toluene $(C_6H_5CH_3)$ and tetrahydrofuran (THF) were sparged with argon and dried by passage through a Seca Solvent Purification System. All chromatography and TLC were performed on silica (230-400 mesh) from Silicycle unless otherwise specified. Activated basic alumina (ca 150 mesh) was obtained from Acros Organics. TLCs were developed with mixtures of ethyl acetate (EtOAc)/hexanes or CH₂Cl₂/methanol (CH₃OH) unless otherwise specified and were visualized with 254 and 365 nm light and I₂ or Br₂ vapor. ¹H and ¹³C NMR spectra were recorded using a Brüker 400 MHz NMR instrument and chemical shifts are reported in ppm on the δ scale relative to tetramethylsilane. IR spectra were recorded on a Nicolet 205 FT-IR instrument and the samples were prepared as KBr pellets. Highresolution mass spectra were recorded at the University of Connecticut mass spectrometry facility using a micromass Q-TOF-2TM mass spectrometer operating in positive ion mode. The instrument was calibrated with Glu-fibrinopeptide B 10 pmol μL^{-1} using 50:50 CH₃CN/H₂O with 0.1% acetic acid (AcOH). N,N-bis(2-((2-(ethylthio)ethyl)thio)ethyl)aniline (1) (12) and 3-nitro-2-naphthaldehyde (13) were synthesized as described.

(4-(Bis(2-((2-(Ethylthio)ethyl)thio)ethyl)amino)phenyl)(4,5-dimethoxy-2-nitrophenyl)methanol (2, ArgenCast-1). To a solution of 1 (390 mg, 0.65 mmol) and ortho-nitroveratraldehyde (161 mg, 0.650 mmol) in CH₂Cl₂ (3.0 mL) was added 2,6-dimethypyridine (0.316 mg, 1.51 mmol) and trimethylsilyl trifluoromethanesulfonate (TMSOTf, 0.289 g, 1.30 mmol). The resulting dark blue/green colored solution was stirred for 16 h at 23°C. The solution was diluted with 50 mL of CH₂Cl₂, washed with brine (50 mL), dried over MgSO₄, filtered and evaporated to dryness. Flash chromatography on silica with a solvent gradient (20 \rightarrow 30% EtOAc in petroleum ether) afforded 0.109 g of TMS-ether protected ArgenCast as a yellow oil ($R_f = 0.39$. 3:1, EtOAc/petroleum ether). The TMS-ether was dided (1.2 mL). After 5 min of stirring, the solvent was removed. Flash chromatography (silica, 1:1 EtOAc/petroleum ether) furnished 2 as

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yellow oil (0.096 g, 16%). $R_f = 0.47$ (1:1 EtOAc/petroleum ether). ¹H-NMR (400 MHz, CDCl₃) δ 7.63 (s, 1 H,), 7.45 (s, 1 H), 7.19 (dd, J = 8 Hz, 2 H), 6.60 (dd, J = 8 Hz, 2 H), 6.48 (s, 1 H), 4.01 (s, 3 H), 3.96 (s, 3 H), 3.53 (m, 4 H), 2.47 (m, 12 H), 2.60 (m, 4 H), 1.28 (m, 6 H). ¹³C-NMR (100 MHz) δ 153.59, 147.93, 146.49, 140.11, 134.87, 130.48, 128.81, 111.87, 110.26, 108.34, 71.53, 56.68, 56.58, 51.80, 32.71, 32.06, 29.65, 26.33, 15.03 ppm. IR (neat) 3434.6, 2963.0, 2925.4, 1611.2, 1517.7, 1330.6, 1271.8, 1062.5. HRMS (+ES1), calculated for MH⁺ 601.1898, observed 601.1824.

(4-(Bis(2-((2-(ethylthio)ethyl)thio)ethyl)amino)phenyl) (4,5dimethoxy-2-nitrosophenyl)methanone (3, ArgenUnc-1). A solution of 2 (0.076 g, 0.127 mmol) in CH₃CN (4.0 mL) sealed in a quartz cuvette was irradiated with a 1000 W source for 5 h. The solvent was removed under reduced pressure, and flash chromatography (silica, 1:9 EtOAc/CH₂Cl₂) furnished 3 (0.039 g, 54.0%) as a yellow oil. $R_f = 0.58$ (1:1 EtOAc/petroleum ether). ¹H-NMR (400 MHz, CDCl₃) δ 7.78 (dd, J = 8 Hz, 2 H), 7.14 (s, 1 H), 6.61 (dd, J = 8 Hz, 2 H), 6.36 (s, 1 H), 4.05 (s, 3 H), 3.93 (s, 3 H), 3.63 (m, 4 H), 2.79–2.73 (m, 12 H), 2.60–2.54 (m, 4 H), 1.28–1.25 (m, 6 H). ¹³C-NMR (100 MHz) δ 193.9, 160.0, 151.2, 139.9, 133.0, 127.7, 110.8, 109.8, 92.0, 57.0, 56.4, 51.7, 32.8, 32.1, 29.6, 26.4, 15.0 ppm. IR (neat) 2913, 2848, 1632, 1562, 1342, 1257, 1043 cm⁻¹. HRMS (+ESI) calculated for MH⁺ 583.1793, observed 583.1764.

(4-(Bis(2-((2-(Ethylthio)ethyl)thio)ethyl)amino)phenyl)(3nitronaphthalen-2-yl)methanol (5, ArgenCast-2). To a solution of 1 (0.186 g, 0.479 mmol) and 3-nitro-2-naphthaldehyde (4, 0.106 g, 0.527 mmol) in CH₂Cl₂ (3.0 mL) was added lutidine (0.077 g, 0.718 mmol) and TMSOTf (0.138 g, 0.623 mmol). The resulting dark blue/green colored solution was stirred for 16 h at 23°C. The solution was diluted with 50 mL of CH₂Cl₂, washed with brine (50 mL), dried over MgSO₄, filtered and the solvent was removed. The resulting crude TMS-ether was dissolved in CH₃CN (3.0 mL) to which 18-crown-6 (0.379 g, 1.43 mmol) and KF (0.278 g, 4.79 mmol), dissolved in minimal amount of water, was added. The resulting reaction mixture was stirred for 12 h at 23°C. The reaction mixture was diluted with water (50 mL) and extracted with CH₂Cl₂. The organic extract was washed with aqueous KCl and dried over MgSO₄. The solvent was removed under vacuum and flash chromatography (silica, 9:1 CH₂Cl₂/EtOAc) furnished 5 an orange oil (0.164 g, 98%). $R_f = 0.40 (9:1 \text{ CH}_2\text{Cl}_2/\text{EtOAc})$. ¹H-NMR (400 MHz, CDCl₃) & 8.49 (s, 1 H,), 8.39 (s, 1 H), 7.98 (m, 2 H), 7.71–7.68 (m, 2 H), 7.65-7.61 (m, 2 H), 7.18 (m, 2 H), 6.61 (m, 2 H), 6.53 (m, 1 H), 3.56-3.52 (m, 4 H), 2.77–2.74 (m, 12 H), 2.60–2.54 (m, 4 H), 1.26 (m, 6). ¹³C-NMR (100 MHz) δ 134.9,134.8, 131.4, 130.3, 129.8, 129.2, 129.1, 128.6, 128.5, 128.1, 125.9, 72.1, 51.8, 32.7, 32.1, 29.7, 26.3, 15.01 ppm. IR (neat) 3415.3, 2964.0, 2910.0, 1610.2, 1519.6, 1349.9, 1176.3. HRMS (+ESI) calculated for MH⁺ 591.1844, observed 591.1802.

(4-(Bis(2-((2-(ethylthio)ethyl)thio)ethyl)amino)phenyl)(1-nitro naphthalen-2-yl)methanol (7, ArgenCast-3). To a solution of 1 (0.204 g, 0.525 mmol) and 1-nitro-2-naphthaldehyde (6, 0.116 g, 0.577 mmol) in CH₂Cl₂ (3.0 mL) was added lutidine (0.084 g, 0.787 mmol) and TMSOTf (0.151 g, 0.682 mmol). The resulting dark green colored solution was stirred for 16 h at 23°C. The solution was diluted with 50 mL of CH2Cl2, washed with brine (50 mL), dried over MgSO₄, filtered and evaporated to dryness. The resulting crude TMSether was dissolved in CH₃CN (3.0 mL) to which 18-crown-6 (0.416 g, 1.57 mmol) and KF (0.305 g, 5.24 mmol) dissolved in minimal amount of water, was added. The resulting reaction mixture was stirred for 12 h at 23°C. The reaction mixture was diluted with water (50 mL), extracted into CH₂Cl₂. The organic extract was washed with aqueous KCl, dried over MgSO₄, and the solvent was removed. Flash chromatography on silica with CH2Cl2 furnished 7 a yellow oil (0.225 g, 72%). $R_f = 0.34 (CH_2Cl_2)$. ¹H-NMR (400 MHz, CDCl₃) δ 7.98 (d, 1 H,), 7.86 (d, 1 H), 7.71 (t, 2 H), 7.63-7.55 (m, 2 H), 7.24 (d, 2 H), 6.60 (d, 2 H), 6.03 (s, 1 H), 3.55–3.51 (m, 4 H), 2.75–2.73 (m, 12 H), 2.56-2.54 (m, 4 H), 1.27 (m, 6). ¹³C-NMR (100 MHz) δ 146.6,146.4, 133.4, 133.2, 131.1, 130.0, 128.8, 128.2, 127.6, 124.5, 124.4, 121.9, 112.0, 71.2,51.8, 32.7, 32.0, 29.64, 26.3, 15.0 ppm. IR (neat) 3407.6, 2964.0, 2910.0, 1610.2, 1519.6, 1357.6, 1267.0, 1176.3. HRMS (+ESI) calculated for MH⁺ 591.1844, observed 591.1802.

General spectroscopic methods. All solutions were prepared with spectrophotometric grade solvents. HEPES (4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid) and KCl (99.5%) were purchased and used as received. Cu⁺ stock solutions were prepared from 99.9% pure (CH₃CN)₄CuPF₆, whereas Ag⁺ and Hg²⁺ were prepared from the

perchlorate salts. Cu⁺ solutions was introduced stabilized in thiourea (0.2 mM). Absorption spectra were recorded on a Cary 50 UV–visible spectrophotometer under the control of a Pentium IV-based PC running the manufacturer supplied software package. Spectra were routinely acquired at 25°C, in 1-cm path length quartz cuvettes with a total volume of 3.0 mL. Analytical photolysis was performed using a 8 W photoreactor at 350 nm. Photolysis reactions were conducted at 25°C, in 1 cm quartz cuvette with a total volume of 3.0 mL. Preparative scale photolysis was carried out using a 1000 W Xe small arc lamp. Solutions of 2 in CH₃CN (3.0 mL) were irradiated for 5 h in a 1 cm quartz cuvette and the conversion checked periodically with TLC. Quantum yields of photolysis were measured using reported procedures at equimolar concentrations of metal ions (7).

Binding constants. Stock solutions of ArgenCast-1 and -1, and the metal ions of Cu^+ , Hg^{2+} and Ag^+ were prepared in mM concentration in spectrophotometric grade CH₃CN. A 25 μ M solution of the ligand was prepared in 3000 μ L CH₃CN and titrated in triplicate with each of the metal stock solutions. Absorbance spectra were corrected for dilution by multiplying the measure absorbance by the following formula:

$$\frac{V_0 - V_{\text{add}}}{V_0} \tag{1}$$

where V_0 is the initial volume and V_{add} is the added volume of the titrant. The conditional dissociation constant (K_d) was calculated by fitting data at *ca* 270 and 350 nm using the XLfit (14) Richards model. The three values of K_d ' found for each cation were averaged and standard deviation was calculated. Using 25 μ M of ArgenUncast-1, the procedure was repeated and the conditional dissociation constant was calculated as described elsewhere at *ca* 350 nm.

RESULTS AND DISCUSSION

Several fluorescent sensors utilize metal ion chelators containing thioether ligands to provide selectivity for Ni²⁺ (15), Pd²⁺ (16), Cu⁺ (17,18), Ag⁺ (19), Cd²⁺ (20) and Hg²⁺ (21,22). Binding selectivity primarily stems from favorable soft-soft acid-base interactions. CrownCast-3, which utilizes a 10phenyl-1,4-dioxa-7,13-dithia-10-azacyclopentadecane (AT₂15 C5) macrocyle, exhibits high affinity for Hg²⁺ similar to analogous fluorescent sensors (23), but only a modest decrease in affinity upon uncaging (7). Strong Hg²⁺-thioether coordination coupled with the structural restrictions of the macrocycle force a Hg²⁺-anilino interaction that inhibits metal ion release. By eliminating macrocyclic effects, we reasoned that a photocage could utilize thioether-heavy metal ion selectivity without compromising uncaging properties.

Many Cu⁺ and Ag⁺ fluorescent sensors derive binding specificity from the acyclic polythioether ligand 3,6,12,15tetrathia-9-azaheptadecane (TTAHD; 17,19,24–30). We envisioned using this chelator to make a photocaged complex for soft, monovalent metal ions using the Cast photocage framework. With a phenyl-derived TTAHD ligand, a nitrobenzhydrol caging group would provide a means to attenuate the electron-donating ability of the aniline lone pair similar to the Tsien Ca²⁺ photocages (Fig. 1).

The desired 9-phenyl-TTAHD (1) was prepared as previously described (12), and the corresponding photocage was synthesized by an electrophilic aromatic substitution reaction with *ortho*-nitroveratraldehyde promoted by TMSOTf (Scheme 1). The photocage ArgenCast-1 was isolated in 63% yield after removal of the silyl group and purification by flash chromatography. The photoproduct, ArgenUnc-1, also was prepared by bulk photolysis for metal binding studies.

The interaction between the photocages and metal ions was assessed spectrophotometrically. Monitoring the disappear-



Figure 1. Expected uncaging action of ArgenCast-1 photocage. The conversion of the nitrobenzhydrol into a benzophenone creates a new resonance interaction between the aniline lone pair and the carbonyl oxygen. The decreased electron density on the aniline nitrogen atom inhibits its ability to engage in bonding; in addition, the introduction of partial positive charge on the aniline helps to repel the positively charged guest metal ion. With thiophilic metal ions, the release is inhibited by strong sulfur-metal interactions, which significantly stabilize the photocage-metal complex.



Scheme 1. Synthesis of ArgenCast-1 and ArgenUnc-1.



Figure 2. (A) Titration of 25 μ M ArgenCast-1 with AgClO₄ in CH₃CN. The absorbance changes were fit to a 1:1 binding isotherm (inset). The error bars represent the variance in the measurements over three trials. (B) Titration of 25 μ M ArgenUnc-1 with AgClO₄ in CH₃CN. The absorbance changes were fit to a 1:1 binding isotherm (inset). The error bars represent the variance in the measurements over three trials.

ance of the aniline-derived absorption band at ca 270 nm upon metal ion addition allows the metal ion affinity of the photocage to be quantified (Fig. 2A). In an analogous manner, the metal ion binding-induced erosion of the charge-transfer

band at *ca* 350 nm for the uncaged ligand allows the binding strength to be addressed (Fig. 2B). As predicted, the soft metal ions Ag^+ , Hg^{2+} and Cu^+ interacted with ArgemCast-1 because of the thioether ligands (Table 1). The binding

M ⁺	СН ₃ СN (µм)			СН ₃ ОН (µм)			Buffer (µм)		
	ArgenCast-1	ArgenUnc-1	$\Delta K_{\rm d}$	ArgenCast-1	ArgenUnc-1	$\Delta K_{\rm d}$	ArgenCast-1	ArgenUnc-1	$\Delta K_{\rm d}$
$\begin{array}{c} Ag^+\\Cu^+\\Hg^{2+}\end{array}$	$\begin{array}{c} 11.4 \ \pm \ 0.4 \\ NB^{\dagger} \\ 7.2 \ \pm \ 0.3 \end{array}$	$\begin{array}{r} 10.2 \ \pm \ 0.1 \\ \mathbf{NB}^{\dagger} \\ 6.9 \ \pm \ 0.1 \end{array}$	0.89 NA [‡] 0.95	$\begin{array}{l} 11.9 \ \pm \ 0.1 \\ 10.2 \ \pm \ 0.2 \\ NB^{\dagger} \end{array}$	7.2 ± 0.4 7.7 ± 0.6 NB [†]	0.61 0.76 NA [‡]	$\begin{array}{l} 9.8\ \pm\ 0.2\\ NA^{\dagger}\\ NB^{\dagger} \end{array}$	$\begin{array}{c} 5.3\ \pm\ 0.3\\ \mathrm{NA}^{\ast}\\ \mathrm{NB}^{\dagger} \end{array}$	0.54 NA [‡] NA [‡]

Table 1. Dissociation constants of ArgenCast-1 and ArgenUnc-1 with metal ions in different solvent systems.

The 1:1 binding constants were calculated with XLfit; \uparrow No binding observed (NB): metal binding was not observed when the ligand was exposed to > 5000 equiv of M^{n+} ; \uparrow Not applicable (NA): the value for ΔK_d could not be calculated, because no K_d value could be measured. Buffer: 20 mm HEPES/dioxane (1:1), pH 7.0; see supporting information for titration data.

chemistry of ArgenCast-1 with Cd^{2+} , Zn^{2+} and Cu^{2+} also was screened, but no interactions were observed when the ligand were exposed to > 3000 equivalents of these metal ions.

The binding properties of ArgenCast-1 were investigated in different solvent systems. In CH₃CN, binding with both Ag⁺ and Hg²⁺ were observed with K_d values of 11.4 and 7.2 μ M respectively, but the ligand could not compete effectively with CH_3CN for Cu^+ binding. In CH_3OH , Cu^+ and Ag^+ interacted with ArgenCast-1 with K_d values of 10.2 and 11.9 μ M respectively, but no binding was observed with Hg²⁺. As the spectrophotometric assay relies on interaction between the aniline nitrogen atom and the metal ion, a thiophilic metal ion like Hg²⁺ can bind tightly to the ligand without significantly perturbing the ligand absorption bands. Binding also was investigated under aqueous conditions (20 mm HEPES/dioxane 1:1, pH 7.0) where dioxane was used to enhance solubility of the photocage. Only Ag⁺ binding could be assessed under these conditions. As Ag^+ was the only metal ion that consistently interacted with the photocage, we adopted the name ArgenCast-1 and ArgenUnc-1 for the photocage 2 and photoproduct 3 respectively. The components of the name reflect the class of photocages (Cast) and the Latin word for silver, Argentum (Argen).

To function as an effective photocage, the chelator should exhibit a strong metal ion affinity that drops significantly upon photolysis. The ΔK_d , which is the ratio of the disassociation constant of the uncaged ligand to the original photocage, ideally should be >10, and larger if possible; however, the $\Delta K_{\rm d} < 1$ suggests that ArgenUnc-1 possesses a higher binding affinity than ArgenCast-1. This anomalous behavior results from the nature of the absorption bands. As demonstrated previously, the ca 350 nm absorption feature of the uncaged ligand is a well-behaved benzophenone charge-transfer band. The absorption band at ca 270 nm of the unphotolyzed photocage appears to be a complicated combination of transitions that do not change linearly with metal ion binding (7). The $\Delta K_d < 1$ measured for ArgenCast instead suggests that there is only a minimal change in binding affinity when uncaged, similar to CrownCast-3. The minimal changes compromise the ability of ArgenCast-1 to function as an effective Ag⁺ photocage. Furthermore, the results suggest that the thioether-metal ion interaction dominates the observed affinity even when not incorporated into a macrocycle.

The conversion of ArgenCast-1 into ArgenUnc can be quantified by simultaneously monitoring the disappearance of the absorption band at $\lambda = -270$ nm and the appearance of the absorption at $\lambda = -350$ nm (Fig. 3). Similar to the binding studies, the quantum yield of photolysis (Φ) was



Figure 3. Photolysis of 25 μ M ArgenCast-1 in CH₃CN using an 8 W source. The solution was photolyzed at $\lambda = 350$ nm.

measured in the three different solvent systems (Table 2). The Φ of apo-ArgenCast-1 was 0.8% in CH₃CN, 1.4% in CH₃OH and 1.4% in buffer/dioxane. The addition of metal ions did not have a statistically significant effect on the Φ , which provides further evidence that the aniline nitrogen atom does not contribute significantly to metal complex stability in ArgenCast-1. In analogous Cast photocages, metal ion binding appears to inhibit an unwanted resonance interaction between the key *aci*-nitro intermediate and the aniline lone pair, which enhances the quantum yield (6). The formation of a stable thioether-metal ion complex would result in a weaker interaction between the aniline nitrogen atom and metal ion that would not prevent the resonance interaction that lowers the Φ (Fig. 4).

Previously, dimethoxylnitrobenzyl (DMNB) group has been replaced with other chromophores in photocages to enhance Φ (31,32). The typical strategy for enhancing photolysis efficiency is to increase the absorption cross section of the chromophore, which means more of the photons introduced into the system instigate photochemical reactions. The 3-nitro-2-naphthyl-substituted photocages (3NN) have Φ as high as 60% (33–35), and we have investigated the 1-nitro-2-naphthyl (1NN) as an alternative (6). Although 3NN requires a multistep synthesis, the aldehyde precursor to make 1NNderived Cast photocages is commercially available. We were interested in comparing the two nitronaphthyl chromophores to determine if there are any quantitative differences in the resulting photocages. ArgenCast-3 (5) was prepared with the TMSOTf promoted reaction between 1 and 3-nitro-2-naph

	ε (ci	$\mathrm{m}^{-1} \mathrm{m}^{-1}, \lambda = 350 \mathrm{n}$	m)	$\Phi_{ m photolysis}$ (%)			
	CH ₃ CN	CH ₃ OH	Buffer	CH ₃ CN	CH ₃ OH	Buffer	
ArgenCast-1	6300	5300	6600	0.77 ± 0.2	1.4 ± 0.3	1.3 ± 0.2	
ArgenCast-2	2600	2400	2600	0.81 ± 0.1	1.3 ± 0.1	1.2 ± 0.3	
ArgenCast-3	2200	2300	2700	0.69 ± 0.3	1.3 ± 0.2	0.39 ± 0.1	
$\left[\operatorname{Ag}(\operatorname{ArgenCast-1})\right]^{+1}$	6000	5700	6300	0.92 ± 0.3	1.5 ± 0.5	1.4 ± 0.4	
[Cu(ArgenCast-1)] ⁺¹	NA	5600	NA	NA	1.5 ± 0.2	NA	
[Ag(ArgenCast-2)] ⁺¹	2400	2500	2400	0.98 ± 0.1	1.4 ± 0.3	1.3 ± 0.2	
[Cu(ArgenCast-2)] ⁺¹	NA	2200	NA	NA	1.4 ± 0.4	NA	
$[Ag(ArgenCast-3)]^{+1}$	1300	1500	NA	0.74 ± 0.4	1.3 ± 0.3	NA	
[Cu(ArgenCast-3)] ⁺¹	NA	1600	NA	NA	1.3 ± 0.1	NA	

Table 2. $\Phi_{\text{photolysis}}$ for ArgenCast ligands and their corresponding metal complexes in various solvents.

NA, metal binding not observed under these conditions; photolysis for 25 μ M photocage using an 8 W reactor with $\lambda = 350$ nm. Buffer: 20 mM HEPES/dioxane (1:1), pH 7.0 (μ M). Complex data was obtained at 25 μ M of Mⁿ⁺; see Supporting Information for photolysis data.

resonance delocalization of the aniline electrons into aci-nitro intermediate



Figure 4. Resonance between the *aci*-nitro intermediate and the aniline nitrogen atom during photolysis of ArgenCast-1. In metal complexes with a strong nitrogen-metal bond, the electron pair less readily participates in resonance with the *aci*-nitro intermediate, which increases the photolysis quantum yield. In ArgenCast, the Ag⁺-thioether interaction appears to dominate complex stability with the aniline lone pair interacting minimally with the metal, which leads to increased participation in resonance interactions and lower quantum yields.



Scheme 2. Synthesis of ArgenCast-2 and -3.

thaldehyde (13), which was prepared as previously described. ArgenCast-4 (7) was synthesized in an analogous manner using 1-nitro-2-naphthaldehyde. The yield of ArgenCast-2 was 58% and ArgenCast-3 was prepared in 72% yield (Scheme 2).

Upon investigation of the photochemistry, no significant variation in Φ was observed for either the apo ligand or the

corresponding metal complexes. Moreover, there is no significant difference between the DMNB caging group and the two naphthyl derivatives, which suggests that the resonance in the Cast photocage framework precludes realizing the advantages of a more strongly absorbing chromophore. Determination of whether 1NN has any advantages or disadvantages over 3NN will require incorporation and analysis in photocages constructed to avoid resonance between the *aci*-nitro intermediate and the receptor.

In summary, three ArgenCast compounds have successively been synthesized and characterized. The thioether-based receptor of the photocages has affinity for Ag⁺, Hg²⁺ and Cu⁺ as predicted by the hard-soft acid-base theory. Argen-Cast-1 interacts with Ag^+ in all solvents tested with a K_d values ranging 9–11 μ M, which demonstrates that solvents had limited effects on the binding affinity. In principle, ArgenUnc-1 has a decreased electron density on the aniline nitrogen atom, and hence a lower affinity for guest cations; however, it retains a relatively high affinity for Ag⁺. These results suggest that while thioether ligands can impart the desired soft metal ion binding, they also compromise metal ion release upon photolysis. Devising strategies to enhance metal ion release and increasing quantum yields in Cast systems are the subject of ongoing efforts to design effective photocages for Ag^+ and other soft metal ions, such as Pd^{2+} , Pt^{2+} and Au^+ that present similar challenges.

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SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article:

Figure S1. (A). Titration of 25 μ M ArgenCast-1 with AgClO₄ in CH₃CN. (B) Difference curve (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S2. (A) Titration of 25 μ M ArgenCast-1 with Hg₂ClO₄ in CH₃CN. (B) Difference curve. (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S3. (A) Titration of 25 μ M ArgenCast-1 with AgClO₄ in MeOH. (B) Difference curve. (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S4. (A) Titration of 25 μ M ArgenCast-1 with (CH₃CN)₄CuPF₆ in MeOH. (B) Difference curve. (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S5. (A) Titration of 25 μ M ArgenCast-1 with AgClO₄ in buffer. (B) Difference curve. (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S6. (A) Titration of 25 μ M ArgenUnc-1 with AgClO₄ in CH₃CN. (B) Difference curve. (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S7. (A) Titration of 25 μ M ArgenUnc-1 with Hg₂ClO₄ in CH₃CN. (B) Difference curve. (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S8. (A) Titration of 25 μ M ArgenUnc-1 with AgClO₄ in MeOH. (B) Difference curve. (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S9. (A) Titration of $25 \,\mu\text{M}$ ArgenUnc-1 with (CH₃CN)₄CuPF₆ in MeOH. (B) Difference curve. (C) Binding

isotherm. Error bars represent the variance in the measurements over three trials.

Figure S10. (A) Titration of 25 μ M ArgenUnc-1 with AgClO₄ in buffer. (B) Difference curve. (C) Binding isotherm. Error bars represent the variance in the measurements over three trials.

Figure S11. Photolysis of 25 μ M ArgenCast-1 in MeCN using a 8 W lamps photo reactor source at 350 nm.

Figure S12. Photolysis of 25 μ M ArgenCast-1 in MeOH using a 8 W lamps photo reactor source at 350 nm.

Figure S13. Photolysis of 25 μ M ArgenCast-1 in buffer using a 8 W lamps photo reactor source at 350 nm.

Figure S14. Photolysis of $25 \,\mu$ M ArgenCast-2 in MeCN using a 8 W lamps photo reactor source at 350 nm.

Figure S15. Photolysis of $25 \,\mu$ M ArgenCast-2 in MeOH using a 8 W lamps photoreactor source at 350 nm.

Figure S16. Photolysis of 25 μ M ArgenCast-2 in buffer using a 8 W lamps photoreactor source at 350 nm.

Figure S17. Photolysis of $25 \,\mu$ M ArgenCast-3 in MeCN using a 8 W lamps photoreactor source at 350 nm.

Figure S18. Photolysis of $25 \,\mu$ M ArgenCast-3 in MeOH using a 8 W lamps photoreactor source at 350 nm.

Figure S19. Photolysis of 25 μ M ArgenCast-3 in buffer using a 8 W lamps photoreactor source at 350 nm.

Figure S20. Photolysis of 25 μ M ArgenCast-1 in the presence of 25 μ M AgClO₄ (1 Eq) in MeCN using a 8 W lamps photoreactor source at 350 nm.

Figure S21. Photolysis of 25 μ M ArgenCast-1 in the presence of 25 μ M AgClO₄ (1 Eq) in MeOH using a 8 W lamps photoreactor source at 350 nm.

Figure S22. Photolysis of 25 μ M ArgenCast-1 in the presence of 25 μ M AgClO₄ (1 Eq) in buffer using a 8 W lamps photoreactor source at 350 nm.

Figure S23. Photolysis of 25 μ M ArgenCast-1 in the presence of 25 μ M (CH₃CN)₄CuPF₆ (1 Eq) in MeOH using a 8 W lamps photoreactor source at 350 nm.

Figure S24. Photolysis of 25 μ M ArgenCast-2 in the presence of 25 μ M AgClO₄ (1 Eq) in MeCN using a 8 W lamps photoreactor source at 350 nm.

Figure S25. Photolysis of 25 μ M ArgenCast-2 in the presence of 25 μ M AgClO₄ (1 Eq) in MeOH using a 8 W lamps photoreactor source at 350 nm.

Figure S26. Photolysis of 25 μ M ArgenCast-2 in the presence of 25 μ M AgClO₄ (1 Eq) in buffer using a 8 W lamps photoreactor source at 350 nm.

Figure S27. Photolysis of 25 μ M ArgenCast-2 in the presence of 25 μ M (CH₃CN)₄CuPF₆ (1 Eq) in MeOH using a 8 W lamps photoreactor source at 350 nm.

Figure S28. Photolysis of 25 μ M ArgenCast-3 in the presence of 25 μ M AgClO₄ (1 Eq) in MeCN using a 8 W lamps photoreactor source at 350 nm.

Figure S29. Photolysis of 25 μ M ArgenCast-3 in the presence of 25 μ M AgClO₄ (1 Eq) in MeOH using a 8 W lamps photoreactor source at 350 nm.

Figure S30. Photolysis of 25 μ M ArgenCast-3 in the presence of 25 μ M (CH₃CN)₄CuPF₆ (1 Eq) in MeOH using a 8 W lamps photoreactor source at 350 nm.

Figure S31. ¹H and ¹³C NMR for ArgenCast-1.

Figure S32. ¹H and ¹³C NMR for ArgenUNC-1.

Figure S33. ¹H and ¹³C NMR for ArgenCast-2.

Figure S34. ¹H and ¹³C NMR for ArgenCast-3.

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