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# Conditional Copper-Catalyzed Azide–Alkyne Cycloaddition by Catalyst Encapsulation\*\*

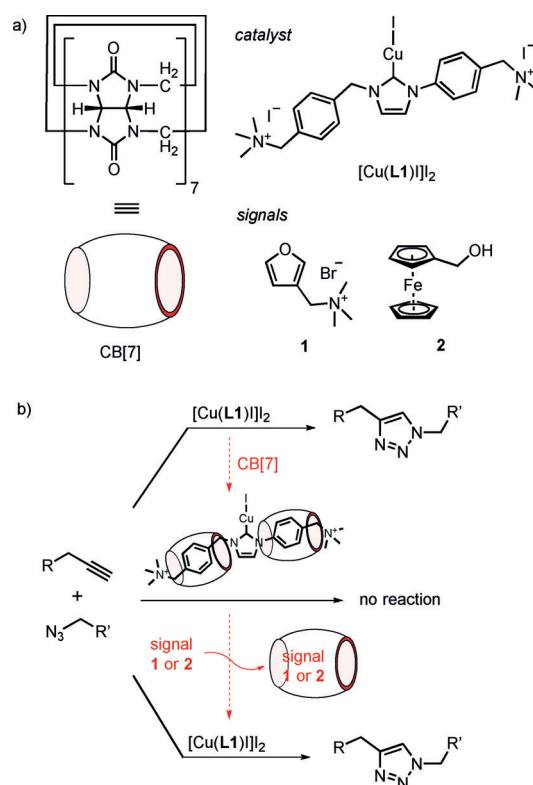
Tobias G. Brevé, Mike Filius, Can Araman, Michelle P. van der Helm, Peter-Leon Hagedoorn, Chirlmin Joo, Sander I. van Kasteren, and Rienk Eelkema\*

**Abstract:** Supramolecular encapsulation is known to alter chemical properties of guest molecules. We have applied this strategy of molecular encapsulation to temporally control the catalytic activity of a stable copper(I)–carbene catalyst. Encapsulation of the copper(I)–carbene catalyst by the supramolecular host cucurbit[7]uril (CB[7]) resulted in the complete inactivation of a copper-catalyzed alkyne–azide cycloaddition (CuAAC) reaction. The addition of a chemical signal achieved the near instantaneous activation of the catalyst, by releasing the catalyst from the inhibited CB[7] catalyst complex. To broaden the scope of our on-demand CuAAC reaction, we demonstrated the protein labeling of vinculin with the copper(I)–carbene catalyst, to inhibit its activity by encapsulation with CB[7] and to initiate labeling at any moment by adding a specific signal molecule. Ultimately, this strategy allows for temporal control over copper-catalyzed click chemistry, on small molecules as well as protein targets.

Herein we describe the use of an external chemical signal to control the rate of copper-catalyzed alkyne–azide cycloaddition (CuAAC) reactions. CuAAC, commonly viewed as one of the primary click reactions,<sup>[1]</sup> is a robust way of “clicking”<sup>[2]</sup> a large range of azide and terminal alkyne functionalized molecules together in biological and material-based environments and has been used in many different settings.<sup>[3]</sup> There is,

however, also an increased need for the spatiotemporal control of these click reactions.<sup>[4]</sup> Such control is usually made possible by light-activated click chemistry, with potential associated problems of phototoxicity and side reactions. In expanding the CuAAC toolbox there is therefore a desire for a general method to spatiotemporally control the catalytic activity of the CuAAC to enable spatiotemporal control over gel formation,<sup>[5]</sup> polymer conjugation, material properties, fluorescence properties, and biomolecule labeling.

Host–guest chemistry can be used to control the chemical properties of encapsulated guests.<sup>[6]</sup> In this study, we use a host–guest system to switch from an inactive to an active catalytic state of the Cu catalyst. A promising class of hosts for encapsulating small-molecule guests in aqueous environments is the cucurbituril family. Cucurbituril is a versatile molecular container (Figure 1a) that is biocompatible, has relatively low toxicity,<sup>[7]</sup> and has been used in many different



**Figure 1.** a) Structure of cucurbit[7]uril (CB[7]) and molecular structure of the Cu<sup>I</sup>–NHC catalyst [Cu(L1)]<sub>2</sub> and signal molecules 1 and 2. b) When [Cu(L1)]<sub>2</sub> is encapsulated by CB[7], catalytic activity in the CuAAC is “switched off”. After the addition of the signal molecule 1 or 2, [Cu(L1)]<sub>2</sub> is released, which “switches on” the catalytic activity.

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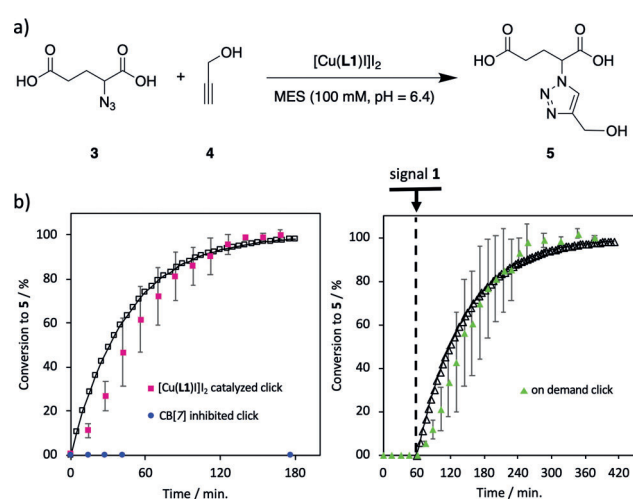
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systems to encapsulate drugs,<sup>[8]</sup> fluorescent dyes,<sup>[9]</sup> and biologically active molecules.<sup>[10]</sup> In some specific cases, cucurbiturils can also act as catalysts themselves, for example in the cycloaddition<sup>[11]</sup> of alkynes and alkyl azides or in promoting acid hydrolysis.<sup>[12]</sup> Furthermore, compartmentalized structures with a cucurbituril barrier can be used to shield a substrate from the catalyst, thus leading to reduced turnover. The addition of a competitive guest then removes the cucurbituril and restores catalytic activity.<sup>[13]</sup> Leigh and co-workers showed using rotaxane-based switchable organo-catalysts that shielding of the catalytic center with a macrocycle is an effective approach to regulate catalytic activity.<sup>[14]</sup> In our study, we wanted to use cucurbiturils to encapsulate a copper catalyst to directly modulate its catalytic activity in CuAAC.

There are reports of triggered CuAAC relying on mechanochemical processes that convert an inactive copper(I) biscarbene catalyst into an active copper(I) monocarbene catalyst.<sup>[15]</sup> Besides mechanochemical triggers, electro-<sup>[16]</sup> and light-based<sup>[17]</sup> triggers can also enable on-demand CuAAC. Another interesting strategy for control over catalytic activity in the CuAAC was reported by Schmittel and co-workers, who prepared a molecular switch that can release a Cu<sup>I</sup> catalyst.<sup>[18]</sup>

In the current study, switching from an “off” state to an “on” state relies on the host–guest chemistry between the catalyst and cucurbit[7]uril (CB[7]). The catalyst was designed in such a way that CB[7] binds with high affinity to the N-heterocyclic carbene (NHC) ligand coordinating to the Cu<sup>I</sup> center. When CB[7] is bound to the Cu<sup>I</sup>–NHC, the Cu<sup>I</sup> center is not catalytically active, most likely because it is not accessible for substrates or is unable to form a catalytically active species. However, when a more strongly binding guest (a chemical signal) is added to the system, the catalyst will be released from CB[7], after which the liberated catalyst can catalyze the click reaction (Figure 1b). We selected copper carbene catalysts because of their high stability in aqueous environments, their high activity in bioconjugation settings,<sup>[19]</sup> and the possibility to modify the NHC ligand in such a way that it binds to CB[7]. The ligand can bind to CB[7] through favorable interactions between its positively charged ammonium groups and the polar portal area of CB[7], as well as interactions between the apolar benzene rings and the apolar cavity of CB[7]. Using this catalyst, we demonstrated temporal control over catalytic activity in copper-catalyzed azide–alkyne cycloaddition in a controlled model reaction and on a biomacromolecule, the protein labeling of which was initiated on demand.

The catalyst was synthesized in five steps to yield the stable Cu<sup>I</sup>–NHC [Cu(L1)I]<sub>2</sub> (see the Supporting Information for details). The complex was soluble in a DMSO/MES buffer solution (100 mM, pH 6.4, 25% v/v DMSO). We used the model reaction shown in Figure 2a to explore the activity of [Cu(L1)I]<sub>2</sub>. Azide **3** and propargyl alcohol (**4**) were selected on the basis of their water solubility and low binding affinity for CB[7] (see Figure SI15 in the Supporting Information). In a typical reaction, azide **3** and propargyl alcohol (**4**) were dissolved in MES buffer, after which the catalyst (2 mol% relative to **3**) dissolved in DMSO was added. Under these



**Figure 2.** a) Model click reaction used to investigate the catalytic activity of [Cu(L1)I]<sub>2</sub>. b) Left: Formation of triazole **5** in MES buffer (100 mM, pH 6.4, 25% v/v DMSO) as catalyzed by [Cu(L1)I]<sub>2</sub> (0.4 mM) ([**3**] = 15 mM, [**4**] = 57 mM). Full conversion was reached after 180 min (magenta). Error bars are standard deviations after *n* = 3 experiments. Open squares represent a pseudo-first-order model fit to experimental data. Encapsulation of [Cu(L1)I]<sub>2</sub> by CB[7] results in a catalyst–CB[7] complex that does not show any catalytic activity (blue). Right: The addition of signal molecule **1** at *t* = 60 min (dashed line) to the inhibited catalyst–CB[7] complex resulted in immediate activation of the copper(I)-catalyzed click reaction (green). Error bars are standard deviations after *n* = 2 experiments. Open triangles represent a pseudo-first-order model based on signal activation of CB[7]-inhibited [Cu(L1)I]<sub>2</sub> (see Figure SI 9). DMSO = dimethyl sulfoxide, MES = 2-morpholino-4-ethanesulfonic acid.

conditions at 25 °C, full conversion into triazole **5** was reached after approximately 180 min (Figure 2b, magenta data points). An estimate of the reaction rate constant gave a value with the same order of magnitude ( $(6 \pm 2) \times 10^4 \text{ M}^{-1} \text{ s}^{-1}$ , concentration refers to Cu, *n* = 3) as that of reported systems using typical Cu<sup>I</sup> catalysts with activating triazole ligands under similar reaction conditions (see the Supporting Information for details).<sup>[20]</sup> When determining the rate constant under pseudo-first-order conditions (see Figure SI 16), a rate constant of  $2 \times 10^4 \text{ M}^{-1} \text{ s}^{-1}$  (concentration refers to Cu) was found, thus confirming the model in Figure 2a.

The binding between CB[7] and [Cu(L1)I]<sub>2</sub> was examined by <sup>1</sup>H NMR spectroscopy and isothermal titration calorimetry (ITC; see Figures SI12–SI14). Using the method of continuous variation (see Figures SI12 and SI13) we determined that the binding stoichiometry between CB[7] and [Cu(L1)I]<sub>2</sub> was 2:1, and the binding constants *K*<sub>a1</sub> and *K*<sub>a2</sub> were  $(1.21 \pm 0.08) \times 10^9$  and  $(3.5 \pm 0.3) \times 10^6 \text{ M}^{-1}$ , respectively (see Figure SI 14). When using catalyst [Cu(L1)I]<sub>2</sub>, which was premixed with 4.6 equivalents of CB[7] (2:1 CB[7]:[Cu(L1)I]<sub>2</sub> complex formation is  $\geq 99.97\%$  in these conditions), no formation of triazole **5** was detected over the course of 7.5 h (Figure 2b, blue, data shown up to 180 min). This result demonstrates that the 2:1 binding of CB[7] to [Cu(L1)I]<sub>2</sub> effectively switches off the catalytic activity of [Cu(L1)I]<sub>2</sub>. The catalytic activity of the 1:1 CB[7]–[Cu(L1)I]<sub>2</sub> complex was also examined and showed conversion of azide **3** into triazole **5** (see Figure SI10; the reaction rate constant was of

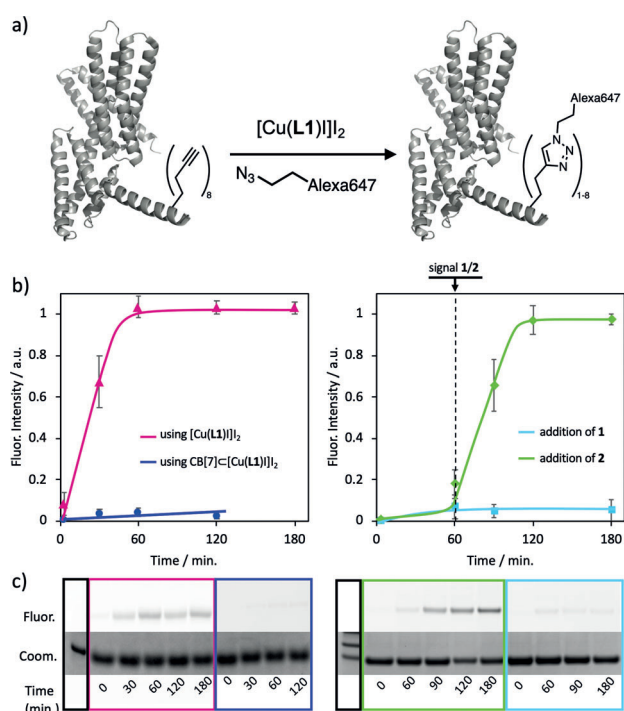
the same order of magnitude as that of the uninhibited experiment).

To demonstrate temporal control of CuAAC, we conducted an experiment in which the signal molecule (3-furylmethyl)trimethylammonium bromide (**1**; Figure 1a) was added (8.8 equiv relative to CB[7], compound **1**:CB[7] complex formation is  $\geq 99.99\%$  under these conditions) to the CB[7]-inhibited catalyst-CB[7] complex. The positively charged signal molecule **1** shows strong interaction ( $K_a = (1.8 \pm 0.2) \times 10^7 \text{ M}^{-1}$ ) with CB[7] in an aqueous environment. We therefore expected that the CB[7]-bound catalyst (inactive) could be displaced from the CB[7] cavity by the addition of **1**, thus activating the catalyst. We added the 2:1 CB[7]:[Cu(L1)]<sub>2</sub> complex to a mixture of **3** and **4** in MES/DMSO to confirm that no reaction took place over the course of 60 min. At  $t = 60$  min we added signal molecule **1**, at which point the click reaction yielding triazole **5** immediately started (Figure 2b, dashed line).

The reaction reached complete conversion approximately 200 min after signal addition, thus showing a reaction rate after activation that was of the same order of magnitude as the rate with the free catalyst (Figure 2b, green line). Combined, these results show that it is possible to deactivate a click catalyst by supramolecular encapsulation, and that it can be reactivated using a chemical signal. We were interested to find out if our method would enable temporal control over protein labeling using a chemical trigger and designed an assay for clicking fluorescent molecules to a protein. We obtained recombinant vinculin<sup>[21]</sup> equipped with multiple alkyne click handles. Vinculin is a cytoskeletal protein with a molecular weight of 116 kDa and has been shown to play a role in cell-matrix and cell-cell adhesion.<sup>[22]</sup> Moreover, it has been shown to be associated with anti-citrullinated protein antibody (ACPA)-positive rheumatoid arthritis as an antigen.<sup>[23]</sup> Using alkyne-modified recombinant vinculin (35.6 kDa), we sought to attach an azide analogue of the Alexa 647 fluorescent probe (Figure 3a). We mixed alkyne-modified vinculin with Alexa 647 in MES buffer (100 mM, pH 6.4, 25% v/v DMSO) and observed that the reaction took place after addition of the catalyst (Figure 3b, magenta line and Figure 3c, magenta square). Furthermore, we observed that the fluorescence labeling of vinculin reached its maximum relative value after 60 min, thus suggesting that the solvent-available alkyne moieties had been labeled.

Next we evaluated whether we could inhibit the catalyst with CB[7]. An attempted labeling reaction with a mixture of CB[7] and [Cu(L1)]<sub>2</sub> (7.4:1 ratio) showed no conversion into the fluorescently labeled protein (Figure 3b, blue line and Figure 3c, blue rectangle). This result agrees with our previous findings in the small-molecule reaction to form triazole **5**. To switch on the catalytic activity and allow for a click reaction to occur, a more strongly binding guest for CB[7] was added as a signal molecule. In the first instance we used signal molecule **1** (Figure 1a), as this triggered the model click reaction immediately after its addition (Figure 2b). However, we found that signal molecule **1** was not able to trigger the protein labeling (Figure 3b,c, cyan).

We tried various high-affinity guests for CB[7], such as amantadine ( $K_a = 4.23 \times 10^{12} \text{ M}^{-1}$ ),<sup>[24]</sup> phenylalanine ( $K_a =$



**Figure 3.** a) [Cu(L1)]<sub>2</sub>-catalyzed fluorescence labeling of alkyne-handle-modified vinculin protein with azide-Alexa 647. The recombinant vinculin protein has eight alkyne groups at various positions in its structure. b) Left: Relative conversion of fluorescence labeling over time. Uninhibited fluorescence labeling reached its maximum value after approximately 60 min (magenta), whereas no significant conversion was observed for CB[7]-inhibited fluorescence labeling over the course of 120 min (blue). Right: Activation of fluorescence labeling using signal molecules. No catalyst activation was observed when signal molecule **1** was added after 60 min (dashed line) to a sample with the CB[7]-inhibited catalyst (cyan). In a similarly inhibited sample, the addition of signal molecule **2** after 60 min led to immediate fluorescence labeling (green). Lines have been drawn to guide the eye. c) Scans of gels stained with Coomassie (Coom.) and analyzed for fluorescence intensity (Fluor.) over time (min). Coomassie bands shown in the black frames indicate the 40 kDa protein marker (recombinant vinculin MW is 35.6 kDa (453–724 amino acid sequence of wild-type vinculin)).

$1.8 \times 10^6 \text{ M}^{-1}$ ),<sup>[10c]</sup> and (dimethylaminomethyl)ferrocene ( $K_a = 10^{12}$ ),<sup>[25]</sup> but none of these guests triggered labeling of the protein. We hypothesized that these positively charged guests might have a strong nonspecific interaction with the protein, owing to a large negatively charged patch on the outside of the protein (see Figure SI 11). This patch may bind the guests, thereby preventing the guests from interacting with the inactive CB[7]-catalyst complex. To test this hypothesis, we used noncharged hydroxymethyl ferrocene **2** ( $K_a = 3.0 \times 10^9 \text{ M}^{-1}$ ;<sup>[26]</sup> Figure 1a). Directly after the addition of signal molecule **2**, protein labeling started (Figure 3b, green line). Absolute fluorescence reached its maximum value 60 min after the addition of the signal molecule, again showing a kinetic profile similar to that of the uninhibited catalyst. This result demonstrates that a competitive guest can act as a signal molecule to activate the Cu<sup>I</sup> catalyst for protein labeling using click chemistry.

To conclude, we synthesized the catalyst  $[\text{Cu}(\text{L1})\text{I}]_2$  and demonstrated that it effectively catalyzed the azide–alkyne click reaction. The catalyst binds to CB[7] in aqueous environments, leading to a complete loss in catalytic activity. A competitive guest for CB[7] can act as a chemical signal, leading to release and activation of the catalyst, as we have demonstrated in both a small-molecule model reaction and a protein-labeling experiment. These results show that host–guest chemistry is a powerful tool to exert temporal control over catalytic activity. For future applications it would be interesting to implement this chemically triggered CuAAC in biological processes that are regulated by alkaloids or other small organic cations, such as choline, as these compounds are potential signal molecules for activation of the CB[7]-inhibited catalyst.<sup>[27]</sup> Cucurbituril complexes are known to facilitate the delivery of a broad variety of cargos across the cell membrane.<sup>[28]</sup> We envision that the strategy presented herein could be used for “on-demand” labeling of biomolecules in the cell, which could have great implications for studying a variety of dynamic biological processes. We are currently looking into enhancing solvent and buffer tolerance to enable these applications in living systems. Finally, this approach could be adopted with different biocompatible metal carbene catalysts, such as NHC–Pd<sup>[29]</sup> and NHC–Ru,<sup>[30]</sup> to regulate their catalytic activity.<sup>[31]</sup>

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## Conflict of interest

The authors declare no conflict of interest.

**Keywords:** catalysis · click chemistry · cucurbit[n]uril · host–guest systems · protein labeling

[1] M. Meldal, C. W. Tornøe, *Chem. Rev.* **2008**, *108*, 2952–3015.

[2] V. V. Rostovtsev, L. G. Green, V. V. Fokin, K. B. Sharpless, *Angew. Chem. Int. Ed.* **2002**, *41*, 2596–2599; *Angew. Chem.* **2002**, *114*, 2708–2711.

[3] a) C. J. Pickens, S. N. Johnson, M. M. Pressnall, M. A. Leon, C. J. Berkland, *Bioconjugate Chem.* **2018**, *29*, 686–701; b) L. Liang, D. Astruc, *Coord. Chem. Rev.* **2011**, *255*, 2933–2945; c) T. Murakami, H. R. Brown, C. J. Hawker, *J. Polym. Sci. Part A* **2016**, *54*, 1459–1467; d) A. A. Alzahrani, M. Saed, C. M. Yakacki, H. B. Song, N. Sowan, J. J. Walston, P. K. Shah, M. K. McBride, J. W. Stansbury, C. N. Bowman, *Polym. Chem.* **2018**, *9*, 121–130.

- [4] a) P. E. Farahani, S. M. Adelmund, J. A. Shadish, C. A. DeForest, *J. Mater. Chem.* **2017**, *5*, 4435–4442; b) C. P. Ramil, Q. Lin, *Curr. Opin. Chem. Biol.* **2014**, *21*, 89–95.
- [5] F. Trausel, C. Maity, J. M. Poolman, D. S. J. Kouwenberg, F. Versluis, J. H. van Esch, R. Eelkema, *Nat. Commun.* **2017**, *8*, 879.
- [6] a) A. Galán, G. Gil-Ramírez, P. Ballester, *Org. Lett.* **2013**, *15*, 4976–4979; b) Y.-C. Horng, P.-S. Huang, C.-C. Hsieh, C.-H. Kuo, T.-S. Kuo, *Chem. Commun.* **2012**, *48*, 8844–8846; c) N. Nishimura, K. Yoza, K. Kobayashi, *J. Am. Chem. Soc.* **2010**, *132*, 777–790; d) M. L. C. Quan, D. J. Cram, *J. Am. Chem. Soc.* **1991**, *113*, 2754–2755; e) R. Eelkema, K. Maeda, B. Odell, H. L. Anderson, *J. Am. Chem. Soc.* **2007**, *129*, 12384–12385.
- [7] Y. Jin Jeon, S.-Y. Kim, Y. Ho Ko, S. Sakamoto, K. Yamaguchi, K. Kim, *Org. Biomol. Chem.* **2005**, *3*, 2122–2125.
- [8] a) Y. Chen, Z. Huang, H. Zhao, J.-F. Xu, Z. Sun, X. Zhang, *ACS Appl. Mater. Interfaces* **2017**, *9*, 8602–8608; b) R. Oun, R. S. Floriano, L. Isaacs, E. G. Rowan, N. J. Wheate, *Toxicol. Res.* **2014**, *3*, 447–455; c) C. P. Carvalho, V. D. Uzunova, J. P. Da Silva, W. M. Nau, U. Pischel, *Chem. Commun.* **2011**, *47*, 8793–8795; d) S. Walker, R. Oun, F. J. McInnes, N. J. Wheate, *Isr. J. Chem.* **2011**, *51*, 616–624; e) L. Zou, A. S. Braegelman, M. J. Webber, *ACS Cent. Sci.* **2019**, *5*, 1035–1043.
- [9] a) X. Lu, L. Isaacs, *Angew. Chem. Int. Ed.* **2016**, *55*, 8076–8080; *Angew. Chem.* **2016**, *128*, 8208–8212; b) J. Mohanty, N. Thakur, S. Dutta Choudhury, N. Barooah, H. Pal, A. C. Bhasikuttan, *J. Phys. Chem. B* **2012**, *116*, 130–135; c) M. Shaikh, J. Mohanty, A. C. Bhasikuttan, V. D. Uzunova, W. M. Nau, H. Pal, *Chem. Commun.* **2008**, 3681–3683; d) F. Biedermann, D. Hathazi, W. M. Nau, *Chem. Commun.* **2015**, *51*, 4977–4980; e) G. Ghale, A. G. Lanctôt, H. T. Kreissl, M. H. Jacob, H. Weingart, M. Winterhalter, W. M. Nau, *Angew. Chem. Int. Ed.* **2014**, *53*, 2762–2765; *Angew. Chem.* **2014**, *126*, 2801–2805.
- [10] a) J. Brinkmann, D. Wasserberg, P. Jonkheijm, *Eur. Polym. J.* **2016**, *83*, 380–389; b) J. M. Chinai, A. B. Taylor, L. M. Ryno, N. D. Hargreaves, C. A. Morris, P. J. Hart, A. R. Urbach, *J. Am. Chem. Soc.* **2011**, *133*, 8810–8813; c) A. R. Urbach, V. Ramalingam, *Isr. J. Chem.* **2011**, *51*, 664–678.
- [11] a) W. L. Mock, T. A. Irra, J. P. Wepsiec, T. L. Manimaran, *J. Org. Chem.* **1983**, *48*, 3619–3620; b) T. C. Krasia, J. H. G. Steinke, *Chem. Commun.* **2002**, 22–23.
- [12] C. Klöck, R. N. Dsouza, W. M. Nau, *Org. Lett.* **2009**, *11*, 2595–2598.
- [13] G. Y. Tonga, Y. Jeong, B. Duncan, T. Mizuhara, R. Mout, R. Das, S. T. Kim, Y.-C. Yeh, B. Yan, S. Hou, V. M. Rotello, *Nat. Chem.* **2015**, *7*, 597.
- [14] a) V. Blanco, A. Carlone, K. D. Hänni, D. A. Leigh, B. Lewandowski, *Angew. Chem. Int. Ed.* **2012**, *51*, 5166–5169; *Angew. Chem.* **2012**, *124*, 5256–5259; b) V. Blanco, D. A. Leigh, U. Lewandowska, B. Lewandowski, V. Marcos, *J. Am. Chem. Soc.* **2014**, *136*, 15775–15780; c) V. Blanco, D. A. Leigh, V. Marcos, J. A. Morales-Serna, A. L. Nussbaumer, *J. Am. Chem. Soc.* **2014**, *136*, 4905–4908.
- [15] a) P. Michael, M. Biewend, W. H. Binder, *Macromol. Rapid Commun.* **2018**, *39*, 1800376; b) P. Michael, W. H. Binder, *Angew. Chem. Int. Ed.* **2015**, *54*, 13918–13922; *Angew. Chem.* **2015**, *127*, 14124–14128.
- [16] V. Hong, A. K. Udit, R. A. Evans, M. G. Finn, *ChemBioChem* **2008**, *9*, 1481–1486.
- [17] a) M. A. Tasdelen, Y. Yagci, *Angew. Chem. Int. Ed.* **2013**, *52*, 5930–5938; *Angew. Chem.* **2013**, *125*, 6044–6053; b) R. S. Stoll, S. Hecht, *Angew. Chem. Int. Ed.* **2010**, *49*, 5054–5075; *Angew. Chem.* **2010**, *122*, 5176–5200; c) C. Maity, F. Trausel, R. Eelkema, *Chem. Sci.* **2018**, *9*, 5999–6005.
- [18] S. De, S. Pramanik, M. Schmittel, *Angew. Chem. Int. Ed.* **2014**, *53*, 14255–14259; *Angew. Chem.* **2014**, *126*, 14480–14484.
- [19] C. Gaulier, A. Hospital, B. Legeret, A. F. Delmas, V. Aucagne, F. Cisnetti, A. Gautier, *Chem. Commun.* **2012**, *48*, 4005–4007.

- [20] S. I. Presolski, V. Hong, S.-H. Cho, M. G. Finn, *J. Am. Chem. Soc.* **2010**, *132*, 14570–14576.
- [21] C. Araman, L. Pieper-Pournara, T. van Leeuwen, A. S. B. Kampstra, T. Bakkum, M. H. S. Marqvorsen, C. R. Nascimento, G. J. Mirjam Groenewold, W. van der Wulp, M. G. M. Camps, H. S. Overkleeft, F. A. Ossendorp, R. E. M. Toes, S. I. van Kasteren, *bioRxiv* **2019**, 439323.
- [22] X. Peng, E. S. Nelson, J. L. Maiers, K. A. DeMali, *Int. Rev. Cell Mol. Biol.* **2011**, *287*, 191–231.
- [23] J. van Heemst, D. T. S. L. Jansen, S. Polydorides, A. K. Moustakas, M. Bax, A. L. Feitsma, D. G. Bontrop-Elferink, M. Baarse, D. van der Woude, G.-J. Wolbink, T. Rispens, F. Koning, R. R. P. de Vries, G. K. Papadopoulos, G. Archontis, T. W. Huizinga, R. E. Toes, *Nat. Commun.* **2015**, *6*, 6681.
- [24] H. Yang, Y. Liu, L. Yang, K. Liu, Z. Wang, X. Zhang, *Chem. Commun.* **2013**, *49*, 3905–3907.
- [25] a) A. E. Kaifer, *ChemPhysChem* **2013**, *14*, 1107–1108; b) L. Peng, A. Feng, M. Huo, J. Yuan, *Chem. Commun.* **2014**, *50*, 13005–13014.
- [26] W. S. Jeon, K. Moon, S. H. Park, H. Chun, Y. H. Ko, J. Y. Lee, E. S. Lee, S. Samal, N. Selvapalam, M. V. Rekharsky, V. Sindelar, D. Sobransingh, Y. Inoue, A. E. Kaifer, K. Kim, *J. Am. Chem. Soc.* **2005**, *127*, 12984–12989.
- [27] D. V. Berdnikova, T. M. Aliyeu, T. Paululat, Y. V. Fedorov, O. A. Fedorova, H. Ihmels, *Chem. Commun.* **2015**, *51*, 4906–4909.
- [28] P. Montes-Navajas, M. González-Béjar, J. C. Scaiano, H. García, *Photochem. Photobiol. Sci.* **2009**, *8*, 1743–1747.
- [29] D. Cherukaraveedu, P. T. Cowling, G. P. Birch, M. Bradley, A. Lilienkampf, *Org. Biomol. Chem.* **2019**, *17*, 5533–5537.
- [30] a) M. Süßner, H. Plenio, *Angew. Chem. Int. Ed.* **2005**, *44*, 6885–6888; *Angew. Chem.* **2005**, *117*, 7045–7048; b) J. Tomasek, J. Schatz, *Green Chem.* **2013**, *15*, 2317–2338.
- [31] E. Peris, *Chem. Rev.* **2018**, *118*, 9988–10031.

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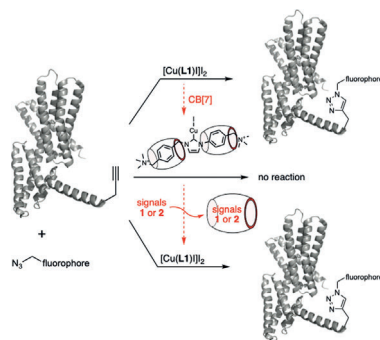
## Communications



## Host-Guest Systems

T. G. Brevé, M. Filius, C. Araman,  
M. P. van der Helm, P. L. Hagedoorn,  
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Conditional Copper-Catalyzed Azide–  
Alkyne Cycloaddition by Catalyst  
Encapsulation



**Labeling on demand:** Spatiotemporal control over click reactions, which are widely applied in (bio)chemical research, opens up new strategies for their application to small-molecule systems and (bio)macromolecules. In an effective supramolecular approach, a copper(I)–N-heterocyclic carbene click catalyst was inhibited through encapsulation by cucurbit[7]uril and activated by its release by chemical signals (see picture).