Communication

A Highly Efficient Single-Chain Metal-Organic Nanoparticle Catalyst for Alkyne-Azide "Click" Reactions in Water and in Cells

Yugang Bai, Xinxin Feng, Hang Xing, Yanhua Xu, Boo Kyung Kim, Noman Baig, Tianhui Zhou, Andrew A. Gewirth, Yi Lu, Eric Oldfield, and Steven C. Zimmerman

J. Am. Chem. Soc., **Just Accepted Manuscript** • DOI: 10.1021/jacs.6b04477 • Publication Date (Web): 16 Aug 2016

A Highly Efficient Single-Chain Metal-Organic Nanoparticle Catalyst for Alkyne-Azide "Click" Reactions in Water and in Cells

Yugang Bai,[1] Xinxin Feng,[1] Hang Xing,[1,2] Yanhua Xu,[1] Boo Kyung Kim,[1] Noman Baig,[1] Tianhui Zhou,[1] Andrew A. Gewirth,[1] Yi Lu,[1,2] Eric Oldfield[1] and Steven C. Zimmerman[1,*]

[1] Department of Chemistry, University of Illinois at Urbana-Champaign, Urbana, Illinois 61801, United States

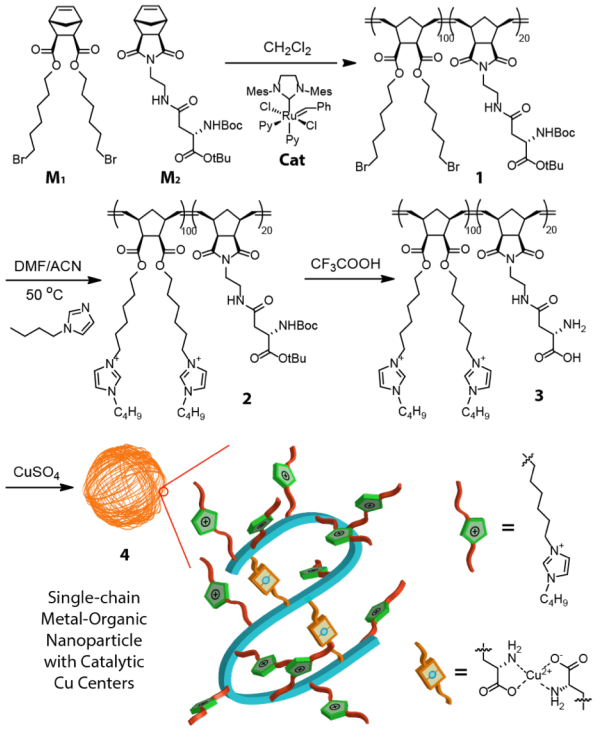
[2] Beckman Institute, University of Illinois at Urbana-Champaign, Urbana, Illinois 61801, United States

Supporting Information Placeholder

ABSTRACT: We show that copper-containing metal-organic nanoparticles (MONP) are readily synthesized via Cu(II)-mediated intramolecular crosslinking of aspartate-containing polyolefins in water. *In situ* reduction with sodium ascorbate yields Cu(I)-containing MONP that serve as highly efficient supramolecular catalysts for alkyne-azide "click chemistry" reactions, yielding desired 1,4-adducts at low parts-per-million catalyst levels. The nanoparticles have low toxicity and low metal loadings, making them convenient, green catalysts for alkyne-azide "click" reactions in water. The Cu-MONP enter cells and perform efficient, biocompatible click-chemistry, thus acting as intracellular nanoscale molecular synthesizers.

The key role played by transition metal catalysts in chemical reactions[1,2] has led to their integration into a wide array of more complex structures. Indeed, several types of macromolecular metal catalysts have been developed to increase catalyst selectivity, stability, and recyclability. For example, metal organic frameworks (MOFs) are heterogeneous lattices where the metal is integral to the 3D structure.[3] As catalysts, MOFs' highly organized structures represent one end of the spectrum[4] whereas at the other are homogenous or heterogeneous, polymer-supported catalysts that are much less defined structurally with one or more metal complexes attached to the side-chain or the end of a random coil polymer.[5] A new macromolecular motif was reported recently by Lemcoff and coworkers, which they named organometallic nanoparticles (OMNPs).[6-8] The OMNPs are single-chain linear polymers (SCNPs)[9] controllably "folded" by the formation of carbon-metal bonds into denser structures that contain metal centers and cavities, resembling metalloenzymes. Indeed, both Rh(I) and Ir(I) OMNPs showed catalytic activity in organic solvents. Pomposo and coworkers described copper containing SCNPs that catalyze coupling reactions and polymerizations, including in water.[10,11]

We recently reported several types of covalently cross-linked polymers termed organic nanoparticles (ONPs) that present an array of functional groups and enter cells in a controllable manner.[12-14] Here we describe water-soluble, copper-cross-linked single-chain, metal-organic nanoparticles (MONPs), as nanoscale catalysts. As with the Lemcoff and Pomposo systems, our Cu-containing MONPs were designed to share some structural similarity with metalloenzymes and in our case, to promote the copper-catalyzed alkyne-azide cycloaddition (CuAAC). The CuAAC "click" reaction was pioneered by Sharpless,[15] with significantly improved catalysts reported later.[16-19] We show herein that the easily synthesized Cu-MONPs have low toxicity and promote the regioselective ligation of terminal alkynes and azides with high efficiency at low parts-per-million (ppm) levels in water.



Scheme 1. Schematic illustration of the synthesis of the copper-containing MONP, and a cartoon showing the internal structure of the cross-linkage.

For maximum utility, MONPs should have three key properties: water solubility, catalytically active metal centers, and the ability to bind hydrophobic substrates. To install these properties, the ring-opening metathesis polymerization (ROMP)[20] of monomers **M₁** and **M₂** (Scheme 1) was performed using Grubbs 3rd generation catalyst[21] using a **M₁**:**M₂**:**Cat** = 100:20:1 ratio to give **1** (Scheme 1). Gel permeation chromatography (GPC) indicated a molecular weight close to the calculated value, and a low polydispersity index (1.04) for

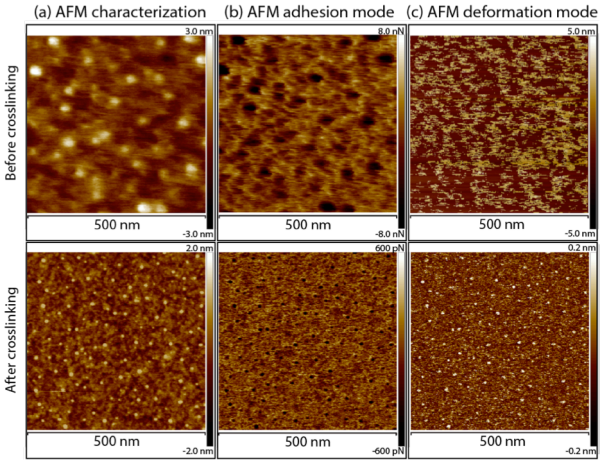


Figure 1. AFM characterization of the Cu-MONP (bottom) and its parent linear polymer (top) in (a) normal (height) mode, (b) adhesion mode and (c) deformation mode.

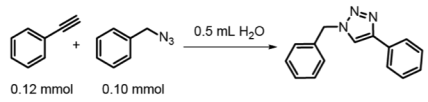
the parent linear polymer (see Figure S2). The polymer was quaternized with *N*-butylimidazole to give **2**, which was purified by precipitation. The imidazolium groups in **2** provide water solubility,[22] whereas the alkyl chains provide a hydrophobic interior to the MONP for substrate binding.

Amino acids form stable coordination complexes with Cu(II)[23] and Cu(I),[24,25] so **2** was prepared with a few aspartate-bearing co-monomer **M2**, allowing a limited number of copper ion mediated intra-chain crosslinks. Polymer **2** was deprotected with trifluoroacetic acid to give linear polymer **3**, which was intramolecularly crosslinked at high dilution in water using 0.5 equivalents of $CuSO_4$ per amino acid residue yielding Cu-MONP **4**. Dynamic light scattering showed the products to have slightly smaller radii of gyration (R_g, Figure S6) with no evidence of interchain crosslinking. The minimally reduced particle size is consistent with a small contraction from a few intrachain cross-links on a hydrophobically collapsed random-coil polynorbornene.

Atomic force microscopy (AFM) was used to visualize and probe the mechanical properties of polymer **3** and Cu-MONP **4**. The uncrosslinked polymers appear larger, less homogeneous than the Cu-MONPs (Figure 1a), similar to what was observed with ONPs.[12] The relatively narrow size distribution of the MONPs are consistent with a intramolecular crosslinking. Similar results were observed in adhesion mode (Figure 1b). Importantly, the adhesion force from the Cu-MONPs was much smaller than found with the linear polymers. For uncrosslinked linear polymers with random coil structures, the number of conformations available during macromolecular extension is high, leading to a greater adhesive force. In contrast, Cu-MONPs have more fixed conformations with a smaller entropic elasticity.[26] The difference in adhesion mode AFM further supports the formation of intramolecular crosslinks in the MONP synthesis. In deformation mode AFM, the Cu-MONPs show significantly higher rigidity and resistance to deformation (Figure 1c), consistent with the observation of previously reported dendritic ONPs,[27] providing additional evidence of crosslinking mediated by Cu(II). Likewise, there is a small increase in particle height (z) despite the significant diameter reduction observed in the xy plane after crosslinking (Figure S8).[27]

To compare the activity of the Cu-MONPs to polymeric copper catalysts reported previously for the Cu(I) catalyzed regioselective Huisgen ligation,[16-19,28-44] i.e., the "click" reaction, the cycloaddition between phenylacetylene and benzyl azide was investigated as a model system. Sodium ascorbate was used to reduce Cu(II) to Cu(I) in the OMNPs *in situ*,[45] and the reaction performed in pure water. Phenylacetylene was added in slight excess and the yield of the reaction

Table 1. "Click" reactions of benzyl azide and phenylacetylene with Cu-MONP or $CuSO_4$.



Cu-MONP level (ppm)	Cu Level (ppm)	NaAsc[a]	time (h)	T (°C)	Yield[b] (%)
1.0	10	+	24	50	58[c]
2.5	25	+	24	50	>99
5.0	50	+	24	50	>99
10	100	+	24	50	>99
2.5	25	+	72	25	>99
0	25[d]	+	336	25	9[e]
2.5	25	-	336	25	9[e]
0	0	-	24	50	6[e]
0	25[d]	+	24	50	6[e]

[a]Reactions marked "+" contained 2.5 mg of sodium *L*(+)-ascorbate (NaAsc). [b]By NMR integration (see procedure in Supporting Information). [c]Reaction was performed in 0.25 mL of water. [d]Because no Cu-MONP was used, the corresponding copper level was reached by adding $CuSO_4$ solution. [e]NMR yield of the 1,4-isomer. Crude product also contained approximately the same amount of 1,5-isomer for these entries.

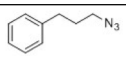
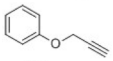
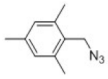
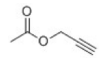
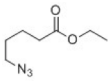
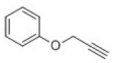
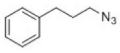
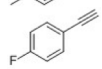
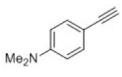
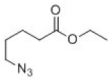
was calculated based on the integrated intensity of the ^{1}H NMR signal of the methylene group in benzyl azide with that in the product. In the presence of 2.5-10 ppm of Cu-OMNP with respect to benzyl azide at 50 °C, reactions went to completion in 24 h, with 1,4-triazoles as the only product. The 1,4-triazole was obtained in 58% yield with just 1 ppm Cu-MONP (Table 1). Without Cu(I), or with $CuSO_4$, only a low conversion of the starting material to product was observed, with a mixture of 1,4- and 1,5-triazoles, indicative of an uncatalyzed reaction.

To validate the generality of Cu-MONP catalysis, the cycloaddition reactions of additional substrate pairs was investigated. As can be seen in Table 2, seven different terminal alkynes and five different azides underwent Cu-MONP catalyzed click reaction, with isolated yields ranging from 83% to 98% at 5-30 ppm Cu-MONP after 24 h at 50 °C. For Entry 2 and 3, chloroform was added to improve the reaction kinetics (see Table S1 and discussion in Supporting Information). Preliminary data suggests that Cu-MONP is significantly less effective with highly hydrophilic substrates at preparative scale, such as propargyl alcohol and 6-azidohexanoic acid (see Supporting Information for additional detail). These results support the importance of hydrophobic binding, but further studies are needed to reach a firm conclusion.

For the examples shown, the catalyst loadings needed for Cu-MONP were significantly lower than those in most previous reports and comparable to the lowest catalyst loading reported in water, e.g., by Astruc,[17] and Yamada and Uozumi.[19] The fate of the Cu ion during and after the reaction was not determined. For example, it is beyond the scope of this study to determine if Cu is lost from the MONP. Even if metal loss occurs, it would represent a very low level of contamination. For the reaction catalyzed by the 2.5 ppm Cu-MONP, the 0.5 mL of water used as solvent contained only 2.5 nmol of Cu, equivalent to 0.32 ppm by weight, lower than the maximum contaminant level goal (MCLG) for Cu in drinking water (1.3 ppm), defined by the United States Environmental Protection Agency (EPA).

In addition to their potential utility in chemical synthesis, the low Cu concentrations needed for a highly reactive catalyst suggest the possibility of performing "click" reactions under biologically relevant

Table 2. "Click" reactions between various terminal alkynes and azides using Cu-MONP as catalyst[a]

	Alkyne	Azide	Cu-MONP (ppm)	NMR Yield[b] (%)	Isolated Yield (%)
1			5	93	91
2[c]			7.5	97	94
3[c]			5	95	94
4			10	>99	95
5			10	93	93
6			7.5	92	90
7			15	87	84
8			5	>99	96
9			5	>99	97
10			5	88	83
11			30	>99	98

[a]Unless otherwise noted, all reactions were carried out with 0.10 mmol of azide, 0.12 mmol of alkyne, 2.5 mg of sodium ascorbate in 0.5 mL of water at 50 °C for 24 h. Dosages of Cu-MONP were listed in the table for each reaction. [b]By NMR integration. (see Supporting Information). [c]50 μL of $CDCl_3$ was added to the reaction to delay the crystallization of the product and improve the kinetics.

conditions. Successful intracellular "click" was pioneered by Ting and coworkers using copper-chelating azides for biomolecular labeling,[46] and such intracellular reactions have also been found to be useful for the measurement of pH inside *E. coli*.[47] Both studies used >100 μM of ligands analogous to tris-[(1-benzyl-1*H*-1,2,3-triazol-4-yl)methyl]-amine (TBTA) developed by Sharpless and Fokin,[16] and copper concentrations of at least 40 μM.

To test the Cu-MONP for activity at low substrate and catalyst concentrations, 3-azido-7-coumarin (**5**) and 7-ethynyl-coumarin (**6**) were used as well-known fluorogenic coumarin derivatives that "light up" upon alkyne-azide cycloaddition. Results for two pairs of compounds are given in Figure S8 in the Supporting Information. The click efficiency for both pairs of compounds were similar with ≥90% conversion in 60 min at 1 μM Cu-MONP. The potential for intracellular Cu-MONP catalysis of the reaction between **5** and **7** was tested in live cells using confocal microscopy. Thus, NCI-H460 (human non-small cell lung carcinoma) and MDA-MB-231 (human breast cancer) cells were pre-incubated with PBS or Cu-MONP (0.5 μM in PBS) for 1 h. The PBS was removed and the cells washed three times. The substrates (100 μM) were added along with sodium ascorbate (2 mM) to effect Cu(II) reduction. As seen in Figure 2b, only weak fluorescence, likely from unreacted azidocoumarin, could be observed in the absence of Cu-MONP, whereas cells were intensely fluorescent in the presence of the catalyst. Cytotoxicity, hemolytic, and Amplex-Red/horseradish peroxidase[48] assays were performed and

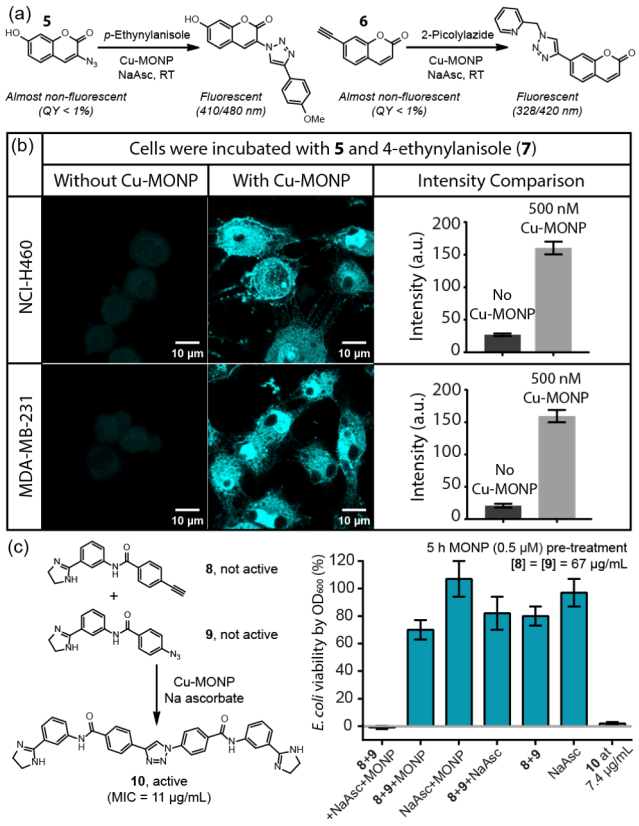


Figure 2. (a) Fluorogenic click compounds **5** and **6**. (b) Confocal microscopy study of intracellular catalysis of **4** (0 or 500 nM) on the "click" reaction between **5** (100 μM) and **7** (100 μM). (c) Model study of intracellular synthesis of antimicrobial bisamidine **10** from **8** and **9** in *E. coli*, catalyzed by **4**. NaAsc = sodium L-ascorbate.

revealed an acceptable biocompatibility for the Cu-MONP (Figures S10, S11 and S12 in the Supporting Information).

The apparent intracellular click reaction described above suggests the potential utility of MONP in the intracellular synthesis of more complex molecules. As a proof of concept, the modular assembly of an antimicrobial agent inside *E. coli* was tested. This experiment used amidine-containing alkyne **8** and azide **9**, which each showed minimal antimicrobial activity against *E. coli* (MIC > 300 μg/mL). The click product of the two, bisamidine **10,** showed much higher activity (MIC = 11 μg/mL).[49] Thus, *E. coli* with or without MONP **4** pretreatment and washing were treated with various combinations of **8**, **9** and sodium ascorbate. Strong inhibition of bacterial cell growth was observed in the experiment containing MONP, **8**, **9** and sodium ascorbate, whereas all other treatment combinations showed little inhibitory effect (Figure 2c). These data suggest the intracellular formation of **10** mediated by the Cu-MONP, and subsequent growth inhibition.

In summary, we developed a Cu-containing organic nanoparticle that catalyzes the CuAAC click reaction with high efficiency in water. Beyond their utility in chemical synthesis in water, our results demonstrate that Cu-MONPs can effect intracellular click reactions inside both bacteria as well as mammalian cells. The intracellular examples featured simple azides and alkynes, but more complicated modular assembly of complex molecules may be possible using appropriate MONP and bio-orthogonal reaction partners. Indeed, it may be possible to selectively synthesize inside cells, compounds that would otherwise be difficult to deliver.

ASSOCIATED CONTENT

Supporting Information

Detailed synthetic procedures and characterization data of the OMNP, "click" substrates and products were provided in the Supporting Information. This material is available free of charge via the Internet at http://pubs.acs.org.

AUTHOR INFORMATION

Corresponding Author

*sczimmer@illinois.edu

Notes

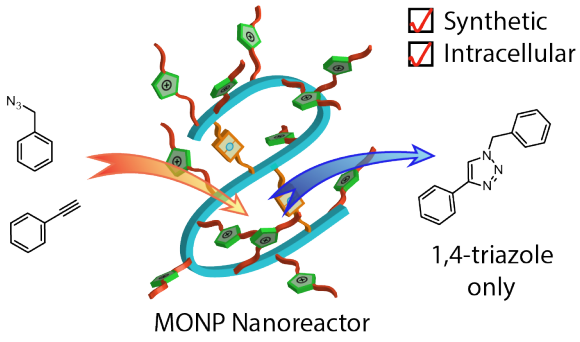
The authors declare no competing financial interests.

ACKNOWLEDGMENT

This work was supported by the National Science Foundation (CHE-1307404), the Binational Science Foundation (2014116), the U.S. National Institutes of Health (ES016865, GM65307 and CA158191), the Office of Science (BER), the U.S. Department of Energy (DE-FG02-08ER64568). Dr. Yang Liu is acknowledged for assistance with the confocal microscope studies.

REFERENCES

(1) Hartwig, J. F. *Organotransition Metal Chemistry: From Bonding to Catalysis*; Univ. Science Books: Herndon, VA, 2010.
(2) Hagen, J. *Industrial Catalysis: A Practical Approach*; Blackwell Science Publishing: England, 2006.
(3) Furukawa, H.; Cordova, K. E.; O'Keeffe, M.; Yaghi, O. M. *Science* **2013**, *341*, 1230444.
(4) Lee, J.; Farha, O. K.; Roberts, J.; Scheidt, K. A.; Nguyen, S. T.; Hupp, J. T. *Chem. Soc. Rev.* **2009**, *38*, 1450-1459.
(5) Hübner, S.; de Vries, J. G.; Farina, V. *Adv. Synth. Catal.* **2016**, *358*, 3-25.
(6) Berkovich, I.; Mavila, S.; Iliashevsky, O.; Kozuch, S.; Lemcoff, N. G. *Chem. Sci.* **2016**, *7*, 1773-1778.
(7) Mavila, S.; Diesendruck, C. E.; Linde, S.; Amir, L.; Shikler, R.; Lemcoff, N. G. *Angew. Chem. Int. Ed.* **2013**, *52*, 5767-5770.
(8) Mavila, S.; Rozenberg, I.; Lemcoff, N. G. *Chem. Sci.* **2014**, *5*, 4196-4203.
(9) Mavila, S.; Eivgi, O.; Berkovich, I.; Lemcoff, N. G. *Chem. Rev.* **2016**, *116*, 878-961.
(10) Sanchez-Sanchez, A.; Arbe, A.; Colmenero, J.; Pomposo, J. A. *ACS Macro Letters* **2014**, *3*, 439-443.
(11) Sanchez-Sanchez, A.; Arbe, A.; Kohlbrecher, J.; Colmenero, J.; Pomposo, J. A. *Macromol. Rapid. Comm.* **2015**, *36*, 1592-1597.
(12) Bai, Y.; Xing, H.; Vincil, G. A.; Lee, J.; Henderson, E. J.; Lu, Y.; Lemcoff, N. G.; Zimmerman, S. C. *Chem. Sci.* **2014**, *5*, 2862-2868.
(13) Bai, Y.; Xing, H.; Wu, P.; Feng, X.; Hwang, K.; Lee, J. M.; Phang, X. Y.; Lu, Y.; Zimmerman, S. C. *ACS Nano* **2015**, *9*, 10227-10236.
(14) Li, Y.; Bai, Y.; Zheng, N.; Liu, Y.; Vincil, G. A.; Pedretti, B. J.; Cheng, J.; Zimmerman, S. C. *Chem. Commun.* **2016**, *52*, 3781-3784.
(15) Kolb, H. C.; Finn, M. G.; Sharpless, K. B. *Angew. Chem. Int. Ed.* **2001**, *40*, 2004-2021.
(16) Chan, T. R.; Hilgraf, R.; Sharpless, K. B.; Fokin, V. V. *Org. Lett.* **2004**, *6*, 2853-2855.
(17) Deraedt, C.; Pinaud, N.; Astruc, D. *J. Am. Chem. Soc.* **2014**, *136*, 12092-12098.
(18) Kamata, K.; Nakagawa, Y.; Yamaguchi, K.; Mizuno, N. *J. Am. Chem. Soc.* **2008**, *130*, 15304-15310.
(19) Yamada, Y. M. A.; Sarkar, S. M.; Uozumi, Y. *J. Am. Chem. Soc.* **2012**, *134*, 9285-9290.
(20) Choi, T.-L.; Grubbs, R. H. *Angew. Chem. Int. Ed.* **2003**, *42*, 1743-1746.
(21) Hilf, S.; Kilbinger, A. F. M. *Nat. Chem.* **2009**, *1*, 537-546.
(22) Ng, V. W. L.; Tan, J. P. K.; Leong, J.; Voo, Z. X.; Hedrick, J. L.; Yang, Y. Y. *Macromolecules* **2014**, *47*, 1285-1291.
(23) van Heeswijk, W. A. R.; Eenink, M. J. D.; Feijen, J. *Synthesis* **1982**, *1982*, 744-747.
(24) Cerda, B. A.; Wesdemiotis, C. *J. Am. Chem. Soc.* **1995**, *117*, 9734-9739.
(25) Thomas, G.; Zacharias, P. S. *Polyhedron* **1985**, *4*, 811-816.
(26) Janshoff, A.; Neitzert, M.; Oberdörfer, Y.; Fuchs, H. *Angew. Chem. Int. Ed.* **2000**, *39*, 3212-3237.
(27) Lemcoff, N. G.; Spurlin, T. A.; Gewirth, A. A.; Zimmerman, S. C.; Beil, J. B.; Elmer, S. L.; Vandeveer, H. G. *J. Am. Chem. Soc.* **2004**, *126*, 11420-11421.
(28) Baig, R. B. N.; Varma, R. S. *Green Chem.* **2012**, *14*, 625-632.
(29) Berg, R.; Straub, J.; Schreiner, E.; Mader, S.; Rominger, F.; Straub, B. F. *Adv. Synth. Catal.* **2012**, *354*, 3445-3450.
(30) Collinson, J.-M.; Wilton-Ely, J. D. E. T.; Diez-Gonzalez, S. *Chem. Commun.* **2013**, *49*, 11358-11360.
(31) Diez-Gonzalez, S.; Nolan, S. P. *Angew. Chem. Int. Ed.* **2008**, *47*, 8881-8884.
(32) Kovacs, S.; Zih-Perenyi, K.; Revesz, A.; Novak, Z. *Synthesis* **2012**, *44*, 3722-3730.
(33) Lee, B.-H.; Wu, C.-C.; Fang, X.; Liu, C. W.; Zhu, J.-L. *Catal. Lett.* **2013**, *143*, 572-577.
(34) Liang, L.; Ruiz, J.; Astruc, D. *Adv. Synth. Catal.* **2011**, *353*, 3434-3450.
(35) Ozkal, E.; Llanes, P.; Bravo, F.; Ferrali, A.; Pericas, M. A. *Adv. Synth. Catal.* **2014**, *356*, 857-869.
(36) Park, I. S.; Kwon, M. S.; Kim, Y.; Lee, J. S.; Park, J. *Org. Lett.* **2008**, *10*, 497-500.
(37) Pathigoolla, A.; Pola, R. P.; Sureshan, K. M. *Appl. Catal. A: Gen.* **2013**, *453*, 151-158.
(38) Roy, S.; Chatterjee, T.; Pramanik, M.; Roy, A. S.; Bhaumik, A.; Islam, S. M. *J. Mol. Catal. A: Chem.* **2014**, *386*, 78-85.
(39) Shao, C.; Zhu, R.; Luo, S.; Zhang, Q.; Wang, X.; Hu, Y. *Tetrahedron Lett.* **2011**, *52*, 3782-3785.
(40) Shin, J.-A.; Lim, Y.-G.; Lee, K.-H. *J. Org. Chem.* **2012**, *77*, 4117-4122.
(41) Teyssot, M.-L.; Chevry, A.; Traikia, M.; El-Ghozzi, M.; Avignant, D.; Gautier, A. *Chem. Eur. J.* **2009**, *15*, 6322-6326.
(42) Wang, D.; Etienne, L.; Echeverria, M.; Moya, S.; Astruc, D. *Chem. Eur. J.* **2014**, *20*, 4047-4054.
(43) Wang, D.; Li, N.; Zhao, M.; Shi, W.; Ma, C.; Chen, B. *Green Chem.* **2010**, *12*, 2120-2123.
(44) Wang, W.; Wu, J.; Xia, C.; Li, F. *Green Chem.* **2011**, *13*, 3440-3445.
(45) Rostovtsev, V. V.; Green, L. G.; Fokin, V. V.; Sharpless, K. B. *Angew. Chem. Int. Ed.* **2002**, *41*, 2596-2599.
(46) Uttamapinant, C.; Tangpeerachaikul, A.; Grecian, S.; Clarke, S.; Singh, U.; Slade, P.; Gee, K. R.; Ting, A. Y. *Angew. Chem. Int. Ed.* **2012**, *51*, 5852-5856.
(47) Yang, M.; Jalloh, A. S.; Wei, W.; Zhao, J.; Wu, P.; Chen, P. R. *Nat. Commun.* **2014**, *5*.
(48) Hong, V.; Presolski, S. I.; Ma, C.; Finn, M. G. *Angew. Chem. Int. Ed.* **2009**, *121*, 10063-10067.
(49) For lead references on this class of antibacterial agents see: (a) Zhu, W.; Wang, Y.; Li, K.; Gao, J.; Huang, C. H.; Chen, C. C.; Ko, T. P.; Zhang, Y.; Guo, R. T.; Oldfield, E. *J. Med. Chem.* **2015**, *58*, 1215-1227. (b) Panchal, R. G.; Ulrich, R. L.; Lane, D.; Butler, M. M.; Houseweart, C.; Opperman, T.; Williams, J. D.; Peet, N. P.; Moir, D. T.; Nguyen, T.; Gussio, R. *Antimicrob. Agents Chemother.* **2009**, *53*, 4283-4291.


Synthetic
Intracellular
1,4-triazole
only
MONP Nanoreactor