Isolation of bis(copper) key intermediates in Cu-catalyzed azide-alkyne “click reaction”

Liqun Jin, Daniel R. Tolentino, Mohand Melaimi, Guy Bertrand*

The copper-catalyzed 1,3-dipolar cycloaddition of an azide to a terminal alkyne (CuAAC) is one of the most popular chemical transformations, with applications ranging from material to life sciences. However, despite many mechanistic studies, direct observation of key components of the catalytic cycle is still missing. Initially, mononuclear species were thought to be the active catalysts, but later on, dinuclear complexes came to the front. We report the isolation of the dinuclear species are involved in the kinetically favored pathway. The copper-catalyzed azide-alkyne cycloaddition (CuAAC) (1, 2) is the flagship of the “click chemistry” arsenal (3, 4). Because of the straightforward procedures, the high functional group tolerance, and the usually high yields, CuAAC has affected all fields of chemistry from material (5–8) to life sciences (9–13); however, the mechanism of the CuAAC is still under debate (14, 15). The first postulated mechanism involved mononuclear copper complexes, although more recently, the participation of bis(copper) complexes has been suggested on the basis of kinetic experiments (16) and computational investigations (17–19).

In 2013, Fokin and colleagues published a very elegant mechanistic study were thought to be the active catalysts, but later on, dinuclear complexes came to the front. We report the isolation of the dinuclear species are involved in the kinetically favored pathway.

The copper-catalyzed azide-alkyne cycloaddition (CuAAC) (1, 2) is the flagship of the “click chemistry” arsenal (3, 4). Because of the straightforward procedures, the high functional group tolerance, and the usually high yields, CuAAC has affected all fields of chemistry from material (5–8) to life sciences (9–13); however, the mechanism of the CuAAC is still under debate (14, 15). The first postulated mechanism involved mononuclear copper complexes, although more recently, the participation of bis(copper) complexes has been suggested on the basis of kinetic experiments (16) and computational investigations (17–19).

In 2013, Fokin and colleagues published a very elegant mechanistic study were thought to be the active catalysts, but later on, dinuclear complexes came to the front. We report the isolation of the dinuclear species are involved in the kinetically favored pathway.

Despite its thermodynamic stability, 1Cua reacts cleanly with benzyl azide in methylene chloride, affording 2Cua (figs. S11 and S12), which was isolated in 87% yield as an air- and moisture-stable, pale yellow solid. 1H and 13C NMR spectra show the presence of one benzyl and one phenyl group, as well as two magnetically nonequivalent CAAC moieties. X-ray diffraction studies of the corresponding tetrafluoroborate salt confirmed the 3,5-dimetallated-1,2,3-triazole structure 2Cua (Fig. 1C, center). Because dinuclear complexes of type 2Cua had never been postulated in any CuAAC mechanism, the next question was whether it was involved in the catalytic cycle. Addition of a stoichiometric amount of phenyl acetylene to 2Cua led, after 5 hours, to the quantitative release of the 1-benzyl-4-phenyl-1,2,3-triazole 3 accompanied by the regeneration of the bis-copper complex 1Cuaa. As an additional confirmation, the same reaction was repeated using 2Cua and a different alkyne (Ph₃C≡C=CH), which resulted in the isolation of 3 along with the corresponding σ,σ-bis-copper complex 1Cua b (Fig. 1C, right; figs. S13 and S14) in quantitative yields.

After having shown that the bis-copper complex 1Cuaa could be part of the catalytic cycle, we turned our attention to the corresponding mono-copper complex 1Cua. We found that a stoichiometric amount of benzyl azide slowly reacted with complex 1Cua, leading, after 16 hours, to the C-metallated heterocycle 2Cua (figs. S15 and S16), which was isolated as a pale yellow solid in 61% yield (29). Addition of one equivalent of phenyl acetylene to 2Cua led, after 5 hours, to the quantitative isolation of triazole 3 and regeneration of the mono-copper complex 1Cua.

These results as a whole suggest that both the mono- and bis-copper complexes could lead to the triazole 3, at least under stoichiometric conditions. However, the kinetic profile of the reactions of 1Cua and 1Cuaa with benzyl azide revealed critical rate acceleration when the bimetalllic complex is used [kobs (1Cuaa)/kobs (1Cua) > 94] (Fig. 1B and fig. S17) (30). In contrast, the protodemetallation of 2Cua and 2Cua a affording triazole 3 and regenerating 1Cuaa and 1Cua, respectively, proceeds at a similar rate (fig. S18). It should be noted that in both cases, the protodemetallation arises from the reaction with the terminal alkyne and, thus, does not necessitate an external Brønsted acid. These reactions lead back to the acetylide complexes 1Cuaa and 1Cua, which exclude the copper precatalyst (CAAC)CuOTf from the catalytic cycle.

The experiments discussed above reproduce parts of both postulated catalytic cycles under stoichiometric conditions. Each step proved to
be extremely clean and high-yielding, and could be achieved either under strictly dry and anaerobic conditions or using nondried solvent under air. To confirm that the isolated complexes are active catalytic species, phenyl acetylene was reacted with benzyl azide at room temperature in methylene chloride using 10 mol % of \(1\text{Cu}_2\alpha\) and \(2\text{Cu}_2\alpha\), and only 5 mol % of \(1\text{Cu}_2\alpha\) and \(2\text{Cu}_2\alpha\). The kinetic profiles of the reactions reveal that the catalytic activity of the dinuclear complexes \(1\text{Cu}_2\alpha\) and \(2\text{Cu}_2\alpha\) is drastically higher (94 and 99% yields of 3 after 10 hours) than that of mononuclear complexes \(1\text{Cu}_\alpha\) and \(2\text{Cu}_\alpha\) (2 and 12% yields of 3 after 10 hours) (Fig. 2A and fig. S19). These results are in agreement with those obtained for the stoichiometric reactions. Note that although the reactions promoted by \(1\text{Cu}_2\alpha\) and \(2\text{Cu}_2\alpha\) proceed at the same rate, a short initiation period is observed with \(1\text{Cu}_2\alpha\), which is concomitant with the formation of \(2\text{Cu}_2\alpha\); after the initiation period, the concentration of \(2\text{Cu}_2\alpha\) remains constant over the course of the catalytic reaction (Fig. 2B). Similar observations can be made from the reactions catalyzed by complexes \(1\text{Cu}_\alpha\) and \(2\text{Cu}_\alpha\) (Fig. 2C), demonstrating that metallated triazoles \(2\text{Cu}_\alpha\) and \(2\text{Cu}_2\alpha\) are the resting states of their respective catalytic cycles.

The results described above demonstrate that the catalytic reaction operates efficiently once \(1\text{Cu}_2\alpha\) is formed. When phenyl acetylene was reacted with benzyl azide at room temperature, in the presence of 10 mol % (CAAC)\text{Cu}OTf, we observed the near quantitative formation of triazole 3, but only after 35 hours. The kinetic profile (Fig. 2A) shows a long initiation period, followed by drastic rate acceleration. Because the formation of the bis(nuclear) acetylide \(1\text{Cu}_2\alpha\) from \(1\text{Cu}_\alpha\) is a fast reaction (vide supra), we conclude that the formation of \(1\text{Cu}_\alpha\) is the limiting step of CuAAC reaction when (CAAC)\text{Cu}OTf is the precatalyst. Indeed, when two equivalents of (CAAC)\text{Cu}OTf were added to phenyl acetylene, in the absence of benzyl azide, no apparent reaction occurred after 8 hours at ambient temperature (Fig. 2D). The trifluoromethanesulfonate anion is too weakly basic to efficiently promote the metallation of the alkyne (31). Addition of one equivalent of triethylamine to the previous solution cleanly afforded \(1\text{Cu}_2\alpha\) along with \(\text{Et}_3\text{NH}^+\text{OTf}^-\) within a few minutes. The effect of the presence of an additional base is also apparent on the kinetic profile of the catalytic reaction using 10 mol % (CAAC)\text{Cu}OTf and 5 mol % triethylamine (Fig. 2E). As expected, a significant shortening of the initiation period is observed.

In conclusion, the isolation of the mono- and bis-copper acetylide complexes \(1\text{Cu}_\alpha\) and \(1\text{Cu}_2\alpha\) demonstrates that although both species are active in the catalytic cycle, the dinuclear complex is involved in the kinetically favored pathway (Fig. 3). The stability of \(1\text{Cu}_2\alpha\) seems to be in contradiction with previous reports (21). The peculiar electronic properties of the CAAC ligand possibly play a role (32), but the use of a weakly coordinating trifluoromethanesulfonate ligand is crucial. Indeed, when (CAAC)\text{Cu}X complexes bear the more popular chloride or acetate X ligand, no bis(copper) complexes could be observed.
although kinetic data demonstrate their involvement. Moreover, when the trifluoromethanesulfonate is used, investigations of the dinuclear pathway permitted the isolation of a never-postulated bis(copper) triazole complex of type $2\text{Cu}_2^\text{a}$, which is the resting state of the catalytic cycle. The alkyne serves as the proton source for the demetallation of $2\text{Cu}_2^\text{a}$, which regenerates the $\pi,\sigma$-bis(copper) acetylide of type $1\text{Cu}_2^\text{a}$, leaving out complexes of type $1\text{Cu}_2^\text{a}$ from the catalytic cycle. These results led to the broader question of whether bis(metallic) complexes of type $1\text{Cu}_2^\text{a}$ are also key species in other copper-catalyzed organic reactions (33).

**MATERIALS AND METHODS**

**General**

$^1$H and $^{13}$C NMR spectra were recorded on Bruker Avance 300, Jeol ECA 500, and Varian Inova 500 spectrometers. NMR multiplicities are abbreviated as follows: $s$ = singlet, $d$ = doublet, $t$ = triplet, $q$ = quartet, $sept$ = septet, $m$ = multiplet, $br$ = broad signal. Chemical shifts are given in parts per million (ppm) and are referenced to SiMe$_4$ ($^1$H, $^{13}$C) and CFCl$_3$ ($^{19}$F). All spectra were obtained at 25°C in the solvent indicated. Coupling constants $J$ are given in hertz (Hz). Mass spectra were performed at the University of California San Diego Mass Spectrometry Laboratory. Melting points were measured with an electrothermal MEL-TEMP apparatus. Single crystal x-ray diffraction data were collected on a Bruker Apex II CCD (charge-coupled device) detector using Mo-Kα radiation ($\lambda = 0.71073\ \text{Å}$). Crystals were selected under...
oil, mounted on nylon loops, and then immediately placed in a cold stream of nitrogen. Structures were solved and refined using Olex2 and SHELXTL. The CAAC and 3,3-diphenyl-1-propyne were prepared following literature procedures, whereas all other starting materials were purchased from commercial sources.

**Synthesis of (CAAC)CuCCPh 1CuB**

n-BuLi (2.4 mmol, 2.5 M in hexane) was slowly added to a solution of phenyl acetylene (234 mg, 2.3 mmol) in THF (15 ml) at −78°C. After 30 min, a solution of (CAAC)CuOAc (1.0 g, 2.3 mmol) in THF (10 ml) was added, and the mixture was stirred at ambient temperature for 1 hour. Volatiles were removed under vacuum, and the remaining solid was extracted with benzene (3 × 10 ml). After removal of benzene under vacuum, the solid was washed with hexane (20 ml). 1CuB was obtained as a light yellow solid. Yield 88% (790 mg). mp 152°C [decomposition (dec.)].1H NMR (CD2Cl2, 500 MHz): δ = 7.48 (t, J = 7.8 Hz, 1 H, p-H), 7.34 (d, J = 7.8 Hz, 2 H, m-H), 7.23 (br, J = 7.1 Hz, 2 H, o-H), 7.14 (br, J = 7.1 Hz, 2 H, m-H)13CH(Ph)), 7.08 (br, J = 7.1 Hz, 1 H, p-H)13CH(Ph)). 2.88 (sept, J = 6.9 Hz, 2 H, CHMe2), 1.98 (s, 2 H, CH2), 1.96 to 1.87 (m, 2 H, CH2), 1.86 to 1.74 (m, 2 H, CH2), 1.39 to 1.30 (m, 18 H, CH3, CHCH3)13C NMR (CD2Cl2) 125 MHz): δ = 253.5 (C carbene), 145.6 (Cq), 135.1 (Cq), 131.8 (CHq), 129.9 (CHq), 128.1 (CHq), 127.7 (PhCO-Cu), 125.7 (CHq), 125.0 (CHq), 121.9 (CHq), 106.3 (PhCCu), 81.3 (Cq), 63.3 (Cq), 42.8 (CH2), 31.4 (CH2), 29.5, 29.4, 27.3, 22.4, 9.9; high-resolution mass spectrometry (HRMS) [electrospray ionization–time-of-flight mass spectrometry (ESI-TOFMS)]: mass/charge ratio (m/z) calculated for C39H32CuNNa+ 500.2349, found 500.2355.

**Synthesis of bis(copper) acetylide complexes 1Cu2a**

(CAAC)CuOTf (330 mg, 1 mmol) was added to a solution of 1CuA (480 mg, 1 mmol) in methane chloride (20 ml). The reaction was stirred for 5 min at ambient temperature. After removing the solvent under vacuum, 1Cu2a was obtained as a white solid. Yield 95% (952 mg). mp 174°C (dec.).1H NMR (CD2Cl2, 500 MHz): δ = 7.44 (t, J = 7.8 Hz, 2 H, p-H), 7.26 (d, J = 7.8 Hz, 4 H, m-H), 7.22 (t, J = 7.6 Hz, 1 H, p-H), 7.04 (t, J = 7.6 Hz, 2 H, m-H), 6.41 (d, J = 7.6 Hz, 2 H, o-H), 2.82 (sept, J = 6.9 Hz, 4 H, CHMe2), 2.05 (s, 4 H, CH2), 1.96 to 1.83 (m, 4 H, CH2), 1.80 to 1.68 (m, 4 H, CH2), 1.38 (s, 12H, CH3), 1.29 (d, J = 6.9 Hz, 12 H, CH3CH2), 1.18 to 1.02 (m, 24 H).13C NMR (CD2Cl2, 125 MHz): δ = 249.4 (C carbene), 145.3 (Cq-Aro), 134.7 (Cq-Aro), 132.0 (CH-Aro), 130.4 (CH-Aro), 129.4 (CH-Aro), 128.7 (CH-Aro), 125.4 (CH-Aro), 122.2 (PhCCu-Cu), 118.9 (Cq-Aro), 110.8 (PhCCu-Cu), 82.5 (Cq), 63.1 (Cq), 42.1 (CH2), 31.5 (CH3), 29.4, 27.2, 22.4, 9.9; HRMS (ESI-TOFMS): m/z calculated for C59H82Cu2N5+ 853.4517, found 853.4507.

**Synthesis of bis(copper) triazole complexes 2Cu2a**

Benzylic azide (40 mg, 0.30 mmol) was added to a solution of 1CuA (250 mg, 0.25 mmol) in methylene chloride (0.5 ml). After 1 hour, diethyl ether (15 ml) was added to induce the precipitation of the product. After filtration, 2Cu2a was isolated as a pale yellow solid. Yield 87% (246 mg). mp 175°C (dec.).1H NMR (CD2Cl2, 500 MHz): δ = 7.48 to 7.38 (m, 2 H, p-H), 7.34 to 7.14 (m, 12 H), 6.82 (br, 2 H), 5.11 (s, 2 H, CH2Ph), 2.77 (sept, J = 6.9 Hz, 4 H, CHMe2), 2.01 (s, 2 H, CH2), 1.96 (s, 2 H, CH2), 1.85 to 1.73 (m, 2 H, CH2), 1.73 to 1.61 (m, 4 H, CH2), 1.61 to 1.51 (m, 2 H, CH2), 1.34 (br, 12 H, CH3), 1.29 to 1.20 (m, 12 H, CH2CH3), 1.04 to 0.86 (m, 24 H).13C NMR (CD2Cl2, 125 MHz): δ = 252.7 (C carbene), 249.3 (C carbene), 156.7 (C triazole), 155.1 (C triazole), 145.4 (Cq), 145.3 (Cq), 136.7 (CqA), 135.2 (Cq), 134.8 (Cq), 133.3 (CqA), 130.3 (CHq), 130.2 (CHq), 129.1 (CHq), 128.8 (CHq), 128.2 (CHq), 128.1 (CHq), 128.0 (CHq), 127.1 (CHq), 125.2 (CHq), 125.1 (CHq), 125.4 (CHq), 82.3 (Cq), 81.9 (Cq), 63.3 (Cq), 63.0 (Cq), 57.1 (CH2Ph), 42.4 (CH2), 31.3 (CH2), 29.4, 27.0, 22.3, 9.7; HRMS (ESI-TOFMS): m/z calculated for C59H32Cu2N5+ 853.4517, found 853.4507.

**Protopodemetalation of 2Cu2a leading to the regeneration of 1CuA**

Phenyl acetylene (15 mg, 0.15 mmol) was added to the solution of 2Cu2a (136 mg, 0.12 mmol) in methylene chloride (5 ml). After stirring for 16 hours at room temperature, the solvent was removed under vacuum. The remaining solid was washed with diethyl ether (3 × 10 ml) to remove triazole 3, and after drying, 1Cu2a was obtained as a yellow solid. Yield 91% (108 mg). NMR data are identical to those of a sample prepared as described above.

**SUPPLEMENTARY MATERIALS**

Supplementary material for this article is available at http://advances.sciencemag.org/cgi/content/ full/1/5/e1500304/DC1

**REFERENCES AND NOTES**

2. C. W. Tornøe, C. Christensen, M. Meldal, Peptidotriazoles on solid phase: [1,2,3]-Triazoles by alkynie 1,3-dipolar cycloaddition and its application to click chemistry. J. Org. Chem. 67, 3057–3064 (2002).
27. Most copper acetylides are polymeric in nature (29) with the exception of a (NH)CuC ≡ CPh complex (16).
30. One of the reviewers suggested that traces of copper(I) dirt could be responsible for the monocopper cycloaddition, a hypothesis that cannot be totally ruled out. Consequently, the rate acceleration can only be greater than 94.

Acknowledgments: We are grateful to the U.S. Department of Energy (DE-FG02-13ER16370) for financial support of this work. Metrical data for the solid-state structures of 1Cu2, 1CuPh, and 2CuPh are available free of charge from the Cambridge Crystallographic Data Centre under reference numbers CCDC-1042482, CCDC-1042483, and CCDC-1042484, respectively. Competing interests: The authors declare that they have no competing interests.

Submitted 12 March 2015
Accepted 5 April 2015
Published 12 June 2015
10.1126/sciadv.1500304

Isolation of bis(copper) key intermediates in Cu-catalyzed azide-alkyne "click reaction"
Liqun Jin, Daniel R. Tolentino, Mohand Melaimi and Guy Bertrand

Sci Adv 1 (5), e1500304.
DOI: 10.1126/sciadv.1500304

ARTICLE TOOLS http://advances.sciencemag.org/content/1/5/e1500304

SUPPLEMENTARY MATERIALS http://advances.sciencemag.org/content/suppl/2015/06/09/1.5.e1500304.DC1

REFERENCES This article cites 31 articles, 1 of which you can access for free
http://advances.sciencemag.org/content/1/5/e1500304#BIBL

PERMISSIONS http://www.sciencemag.org/help/reprints-and-permissions

Use of this article is subject to the Terms of Service

Science Advances (ISSN 2375-2548) is published by the American Association for the Advancement of Science, 1200 New York Avenue NW, Washington, DC 20005. The title Science Advances is a registered trademark of AAAS.

Copyright © 2015, The Authors