Organic LETTERS

Letter

pubs.acs.org/OrgLett

Metal-Free Direct Amidation of Naphthoquinones Using Hydroxamic Acids as an Amide Source: Application in the Synthesis of an HDAC6 Inhibitor

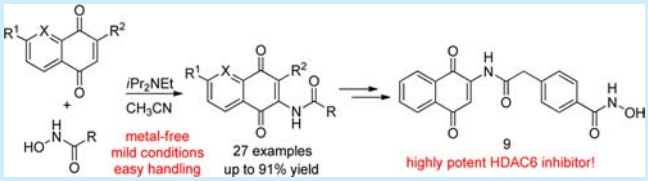
Cheng Zhang*,† and C. James Chou*,‡

†Department of Applied Chemistry, China Agricultural University, 2 Yuanmingyuan West Road, Beijing 100193, China

‡Department of Drug Discovery and Biomedical Sciences, Medical University of South Carolina, 280 Calhoun Street, MSC140 QF307, Charleston, South Carolina 29425, United States

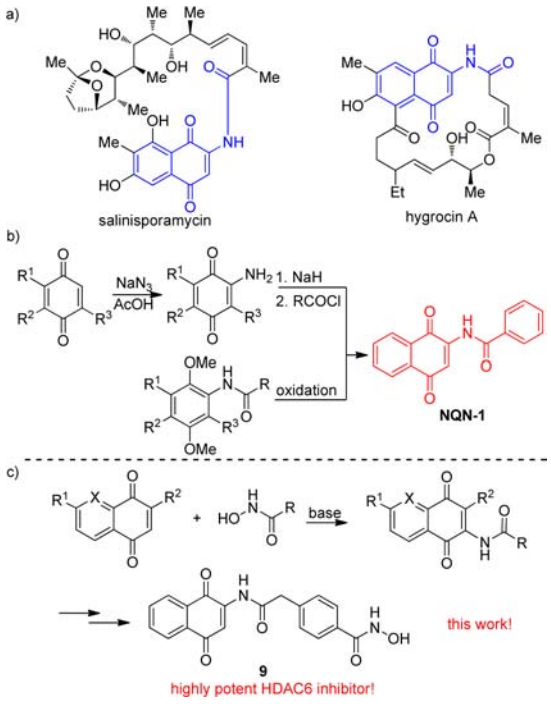
Supporting Information

ABSTRACT: A novel synthetic approach to amidoquinones by the reaction of naphthoquinones with hydroxamic acids under basic conditions was developed. The reaction is mild and operationally simple, and it affords high yields of amidoquinones. With this new method, a novel, very strong HDAC6 inhibitor, which showed high toxicity to AML cells, was successfully synthesized.



Amidoquinones are core structures of many biologically active natural products and synthetic molecules. Some of these compounds, such as hygrocin A and salinisporamycin (Scheme 1a), exhibit very important anticancer and antibiotic activities.[1a−e] Besides their biological applications, amidoquinones have also been used as metal chelators in catalysis.[1f] Recently, a novel amidoquinone-based HDAC6-selective inhibitor, **NQN-1**, that showed selective toxicity toward acute myeloid leukemia (AML) cells at micromolar concentration was developed.[2] To date, mainly two methods are available to access such amidoquinone structures (Scheme 1b): (1) An aminoquinone is obtained by directly warming a mixture of a quinone and NaN_3 in AcOH, and the aminoquinone is then reacted with an acyl chloride in the presence of strong base to form the final amidoquinone product. The strong base, such as NaH, is indispensable in the acylation reaction because the amino group is highly deactivated by the quinone ring, and the generation of a nitrogen anion with the strong base is necessary to make the nucleophilic substitution reaction proceed. As a result, the need to work with this corrosive and explosive NaN_3/AcOH reaction system and to handle the susceptible acylation reaction make this method considerably labor-intensive and time-consuming.[3] (2) The amidoquinone is directly formed by oxidation of the corresponding dimethoxy-*N*-phenylamide with a strong oxidant such as a hypervalent iodine compound. This method can afford various amidoquinones with different substituents.[4] However, substrates that are susceptible to strong oxidants cannot tolerate these reaction conditions. In this context, the development of more concise and environmentally benign synthetic methods to access these amidoquinone structures is highly desirable and of great significance.

Scheme 1. Amidoquinone Structures and Synthetic Methods



Many methods to access aryl C−N bonds have been reported, and the most reliable ones are Buchwald−Hartwig amination, the Ullman coupling reaction, and the Chan−Lam coupling reaction.[5] The advent of direct amination using hydroxylamine or its derivatives as amination reagents by N−O bond cleavage provides chemists with new powerful tools to create such C−N bonds, but these N−O bond cleavages usually need the

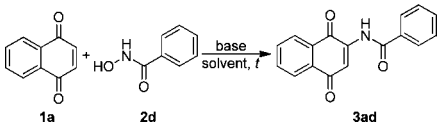
Received: September 12, 2016
Published: October 19, 2016

DOI: 10.1021/acs.orglett.6b02740
Org. Lett. 2016, 18, 5512−5515

assistance of transition metals.[6] Recently, metal-free synthetic methods to access arylamines have attracted more and more attention.[7] To our knowledge, March and Engenito[8] reported the first noncatalyzed direct amidation of aromatic compounds via N−O bond cleavage with hydroxamic acid using poly(phosphoric acid) (PPA) as a solvent, but this method cannot be practically useful because of its extremely limited substrate scope, low yields, and harsh conditions. In 2015, Chen and Wang[9] reported a direct amidation method to access aminophenol compounds with *N*-hydroxyindolinones, which involved a regioselective [1,3] arrangement. Very recently, Cheng, Li, and co-workers reported direct aminations of benzofuran-2(3*H*)-ones with hydroxylamine derivatives via a single electron transfer event in which the two types of reactants act as electron donors and electron receptors, repectively.[10] Despite these achievements, direct amidation of quinones has never been reported. We envisioned that the amidoquinone structure could be achieved by direct amidation of quinones with hydroxamic acid or its derivatives under specific reaction conditions. With our continuing interest in quinone-based organic molecules,[2,3c,11] we report here a metal-free direct amidation of naphthoquinones with hydroxamic acids (Scheme 1c) and its application in the synthesis of a highly potent HDAC6 inhibitor.

Initially, we employed 1,4-naphthoquinone (**1a**) and *N*-hydroxybenzamide (**2d**) as starting materials to interrogate different reaction conditions (Table 1). To our great delight, the

Table 1. Optimization of the Reaction Conditions[a]



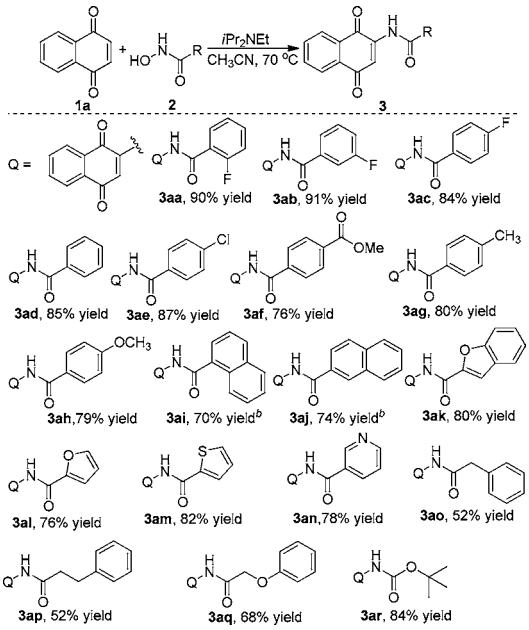
entry	solvent	base (equiv)	*t* (°C)	yield (%)[b]
1	1,4-dioxane	K_2CO_3 (2.0)	70	68
2	1,4-dioxane	−	70	none
3	THF	K_2CO_3 (2.0)	70	60
4	CH_2Cl_2	K_2CO_3 (2.0)	70	21
5	MeOH	K_2CO_3 (2.0)	70	none
6	DMF	K_2CO_3 (2.0)	70	72
7	CH_3CN	K_2CO_3 (2.0)	70	75
8	CH_3CN	*i*Pr_2NEt (2.0)	70	87
9	CH_3CN	*i*Pr_2NEt (1.0)	70	53
10	CH_3CN	Et_3N (2.0)	70	76
11	CH_3CN	pyridine (2.0)	70	69
12	CH_3CN	DBU (2.0)	70	messy
13[c]	CH_3CN	*i*Pr_2NEt (2.0)	25	80
14[d]	CH_3CN	*i*Pr_2NEt (2.0)	70	86

[a]Reactions were performed by warming a mixture of **1a** (0.6 mmol), **2d** (0.5 mmol), and the base (1.0 mmol) in the indicated solvent (4 mL) for 12 h. [b]Isolated yields based on **2d**. [c]The reaction was performed at room temperature. [d]The reaction was performed under argon.

desired product (**3ad**) was obtained in 68% yield when the starting materials were treated with K_2CO_3 at 70 °C in 1,4-dioxane (entry 1). However, no desired product was found when **1a** and **2d** were stirred under the same reaction conditions but in the absence of the base (entry 2). This indicated that the base is indispensable for the coupling reaction between **1a** and **2d**. Following the initial attempt, several other solvents were then screened for the reaction. CH_3CN was found to be the most suitable solvent, affording **3ad** in 75% yield (entry 7), while THF, CH_2Cl_2, MeOH, and DMF offered the desired product **3ad** in 60%, 21%, 0%, and 72% yield, respectively (entries 3−6). Besides K_2CO_3, other types of bases, including both organic and inorganic bases, were carefully tested for the reaction. Finally, *i*Pr_2NEt was proved to be the most efficient base for the coupling reaction, affording **3ad** in 87% yield (entry 8), while others, such as Et_3N, pyridine, etc., were also effective bases but gave lower yields (entries 10 and 11). However, the reaction was messy when the stronger organic base DBU was employed (entry 12). It was also found that 2 equiv of *i*Pr_2NEt (based on **2d**) is required for the best performance of the reaction, and the yield of **3ad** decreased when the loading of *i*Pr_2NEt was lowered (entry 9). The reaction was slightly less productive when performed at room temperature, giving an 80% yield of the final product (entry 13). There was no difference when the reaction was performed under argon atmosphere (entry 14), which indicated that oxygen and moisture do not affect the reaction.

Subsequently, the substrate scope of hydroxamic acids was explored with the most effective protocol. As shown in Scheme 2,

Scheme 2. Substrate Scope of Hydroxamic Acids[a]



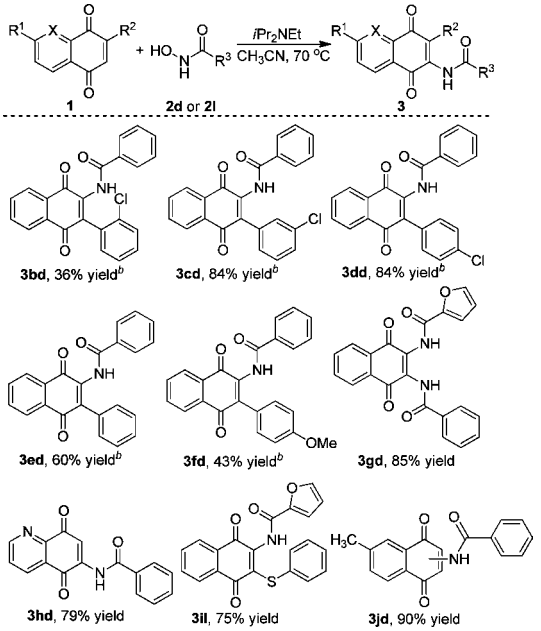
[a]Reactions were performed by warming a mixture of **1a** (0.6 mmol), **2** (0.5 mmol), and *i*-Pr_2NEt (1.0 mmol) in CH_3CN (4 mL) for 12 h. [b]The reaction was performed in DMF.

substitution of the hydroxamic acid at different positions of the phenyl ring did not affect the yield of the final product (**3aa−ac**). Electron-withdrawing or electron-donating substituents at the *para* position of the phenyl ring of the hydroxamic acid seemed to have little influence on the reaction, and the desired products were obtained in good yields (**3ae−ah**). *N*-hydroxynaphthamides have very poor solubility in CH_3CN, but reactions went smoothly in DMF and gave slightly lower isolated yields of the final products (**3ai** and **3aj**). Hydroxamic acids with heteroaromatic rings are also suitable substrates, giving the corresponding products in good yields (**3ak−an**). Alkyl hydroxamic acids are more challenging substrates, affording the desired substrates in moderate yields (**3ao−aq**). Notably, Boc-protected hydroxylamine can also be applied under the optimal reaction conditions, furnishing the product in high yield (**3ar**).

DOI: 10.1021/acs.orglett.6b02740
Org. Lett. 2016, 18, 5512−5515

To further test the limitations of this reaction, the scope of various quinones was also investigated (Scheme 3). The results

Scheme 3. Substrate Scope of Quinones[a]

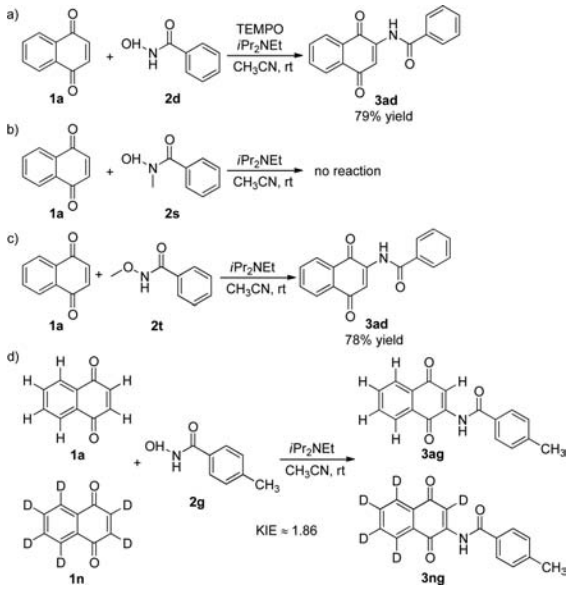


[a]Reactions were performed by warming a mixture of **1** (0.6 mmol), **2d** or **2l** (0.5 mmol), and *i*-Pr_2NEt (1.0 mmol) in CH_3CN (4 mL) for 12 h. [b]Pyridine was used as the base.

indicated that naphthoquinones with an electron-deficient group at the 2-position afford higher yields of the desired products (**3cd** and **3dd**), while naphthoquinones with an electron-donating group at the 2-position afford lower yields of the desired products (**3ed**–**fd** and **3il**). However, a 1,4-naphthoquinone with a 2-chlorophenyl substituent at the 2-position gave an extremely low yield of the corresponding product (**3bd**), probably because the steric hindrance hindered the formation of **3bd**. A 1,4-naphthoquinone with an amido substituent at the 2-position gave a high yield of **3gd**. Quinoline-5,8-dione is also a favorable substrate for this reaction that afforded a 79% yield of the desired product **3hd**. 6-Methyl-1,4-naphthoquinone gave a high yield of amido-substituted products, but it was a 1:1 mixture of 2- and 3-substituted products, which is difficult to purify (**3jd**). However, when 2-methylnaphthoquinone (**1k**), 1,4-benzoquinone (**1l**), or 2,5-dimethyl-1,4-benzoquinone (**1m**) was subjected to the standard conditions, no desired amidoquinone product was detected.[13]

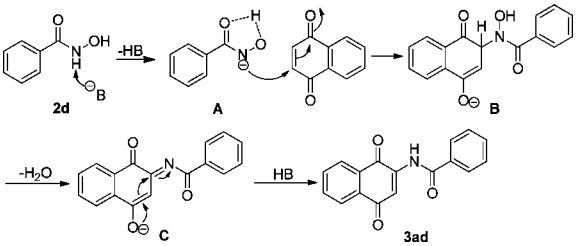
In order to probe the mechanism of the reaction, we carried out several control experiments as shown in Scheme 4. The model reaction proceeded well in the presence of excess 2,2,6,6-tetramethylpiperidin-1-oxyl (TEMPO), which indicated that the reaction does not go through a radical pathway (Scheme 4a). There was no reaction when 1,4- naphthoquinone was treated with *N*-methylhydroxamic acid (Scheme 4b). However, the desired product was obtained in 78% yield when 1,4-naphthoquinone was treated with *O*-methylhydroxamic acid (Scheme 4c). The results imply that the generation of amide anion is essential for the reaction. We also evaluated the kinetic isotope effect (KIE) for the reaction, and a small KIE value was observed (Scheme 4d). This indicated that the reaction does not involve direct C–H activation. We then proposed a plausible

Scheme 4. Control Experiments



mechanism based on the results obtained (Scheme 5). The reaction takes place via initial abstraction of H^+ from compound

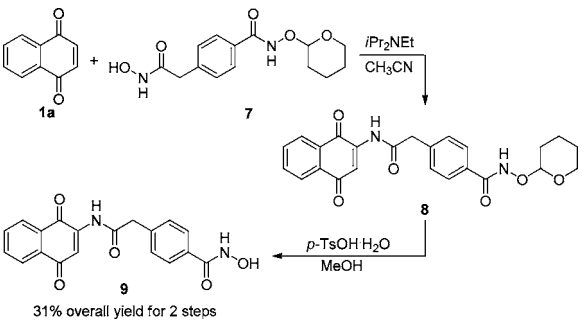
Scheme 5. Proposed Mechanism



2d by the base to form intermediate **A**,[12] which then attacks **1a** via nucleophilic addition, affording intermediate **B**. A molecule of water is abstracted from **B** to form intermediate **C**, and the final product **3ad** is then generated after electron rearrangement.

With great interest in amidoquinone-based HDAC6 inhibitors, we had been endeavoring to synthesize compound **9** but were unsuccessful.[13] With our newly developed method of direct amidation of 1,4-naphthoquinones, however, compound **9** was obtained in 31% overall yield (Scheme 6). Compounds **1a** and **7** were subjected to the optimal reaction conditions to yield compound **8**, and deprotection of **8** in MeOH with a catalytic

Scheme 6. Synthesis of Novel HDAC6 Inhibitor 9 by the Newly Developed Direct Amidation Reaction of Quinones



DOI: 10.1021/acs.orglett.6b02740
Org. Lett. 2016, 18, 5512–5515

amount of *p*-TsOH·H_2O afforded compound **9**. Biological studies of **9** showed that it has extremely high inhibition potency against human recombinant HDAC6 enzyme with an IC_{50} of 6 nM; it is stronger than SAHA (vorinostat), the first FDA-approved HDAC inhibitor in the clinic, which has an IC_{50} of 484 nM against HDAC6. Besides, Western immunoblot showed that **9** induces hyperacetylated tubulin because of HDAC6 inhibition. Upregulation of Hsp70 suggests HDAC6-associated Hsp90 inhibition followed by degradation of the oncogenic protein FLT-3 and STAT5 in AML cells. In the end, compound **9** exhibited high toxicity toward AML cells with an EC_{50} of 367 nM, compared with approximately 1 mM for SAHA, which indicates promising biological prospects.[13]

In conclusion, we have developed a new metal-free method to construct biologically valuable amidoquinone structures by the direct reaction of quinones with hydroxamic acids under mild basic conditions, and it unveiled a new N−O bond cleavage reaction of hydroxamic acids. The newly developed C−N bond formation reaction is convenient and operationally simple, has a broad substrate scope, and affords the final products in high yield (up to 91%). In particular, with this newly developed method, we successfully synthesized a highly potent HDAC6 inhibitor (IC_{50} = 6 nM) that is hard to access by usual methods, and the inhibitor exhibits promising biological activities, such as high toxicity toward AML cells (EC_{50} = 367 nM). Further biological studies are underway.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.orglett.6b02740.

Synthesis of quinones **1** and hydroxamic acids **2**, experimental procedures for products **3**, and ^1H and ^{13}C NMR spectra for **3** (PDF)

■ AUTHOR INFORMATION

Corresponding Authors

*E-mail: zhangc9711@cau.edu.cn, cheunggilbert@gmail.com.
*E-mail: chouc@musc.edu.

Author Contributions

Both authors have given approval to the final version of the manuscript.

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

This work was supported by the National Institutes of Health/National Cancer Institute (CA163452 to C.J.C.) and the Department of Drug Discovery and Biomedical Science, SCCP, Medical University of South Carolina. Support from the College of Science of China Agricultural University is also gratefully acknowledged.

■ REFERENCES

(1) (a) Hadden, M. K.; Lubbers, D. J.; Blagg, B. S. J. *Curr. Top. Med. Chem.* **2006**, *6*, 1173. (b) Le Brazidec, J.-Y.; Kamal, A.; Busch, D.; Thao, L.; Zhang, L.; Timony, G.; Grecko, R.; Trent, K.; Lough, R.; Salazar, T.; Khan, S.; Burrows, F.; Boehm, M. F. *J. Med. Chem.* **2004**, *47*, 3865. (c) Cai, P.; Kong, F.; Ruppen, M. E.; Glasier, G.; Carter, G. T. *J. Nat. Prod.* **2005**, *68*, 1736. (d) Matsuda, S.; Adachi, K.; Matsuo, Y.; Nukina, M.; Shizuri, Y. *J. Antibiot.* **2009**, *62*, 519. (e) Zhang, C.; Wang, M.; Fan, Z.; Sun, L.-P.; Zhang, A. *J. Org. Chem.* **2014**, *79*, 7626. (f) Wang, M.; Yang, Y.; Fan, Z.; Cheng, Z.; Zhu, W.; Zhang, A. *Chem. Commun.* **2015**, *51*, 3219.

(2) Inks, E. S.; Josey, B. J.; Jesinkey, S. R.; Chou, C. J. *ACS Chem. Biol.* **2012**, *7*, 331.

(3) (a) Fieser, L. F.; Hartwell, J. L. *J. Am. Chem. Soc.* **1935**, *57*, 1482. (b) Lien, J.-C.; Huang, L.-J.; Wang, J.-P.; Teng, C.-M.; Lee, K.-H.; Kuo, S.-C. *Chem. Pharm. Bull.* **1996**, *44*, 1181. (c) Josey, B. J.; Inks, E. S.; Wen, X.; Chou, C. J. *J. Med. Chem.* **2013**, *56*, 1007.

(4) (a) Nicolaou, K. C.; Sugita, K.; Baran, P. S.; Zhong, Y.-L. *Angew. Chem., Int. Ed.* **2001**, *40*, 207. (b) Nicolaou, K. C.; Sugita, K.; Baran, P. S.; Zhong, Y. L. *J. Am. Chem. Soc.* **2002**, *124*, 2221.

(5) (a) Guram, A. S.; Buchwald, S. L. *J. Am. Chem. Soc.* **1994**, *116*, 7901. (b) Paul, F.; Patt, J.; Hartwig, J. F. *J. Am. Chem. Soc.* **1994**, *116*, 5969. (c) Ma, D.; Cai, Q.; Zhang, H. *Org. Lett.* **2003**, *5*, 2453. (d) Jiao, J.; Zhang, X.-R.; Chang, N.-H.; Wang, J.; Wei, J.-F.; Shi, X.-Y.; Chen, Z.-G. *J. Org. Chem.* **2011**, *76*, 1180. (e) Chan, D. M. T.; Monaco, K. L.; Wang, R.-P.; Winters, M. P. *Tetrahedron Lett.* **1998**, *39*, 2933. (f) Lam, P. Y. S.; Clark, C. G.; Saubern, S.; Adams, J.; Winters, M. P.; Chan, D. M. T.; Combs, A. *Tetrahedron Lett.* **1998**, *39*, 2941. For recent reviews, please see: (g) Surry, D. S.; Buchwald, S. L. *Angew. Chem., Int. Ed.* **2008**, *47*, 6338. (h) Hartwig, J. F. *Acc. Chem. Res.* **2008**, *41*, 1534. (i) Monnier, F.; Taillefer, M. *Angew. Chem., Int. Ed.* **2009**, *48*, 6954. (j) Surry, D. S.; Buchwald, S. L. *Chem. Sci.* **2011**, *2*, 27. (k) Rauws, T. R. M.; Maes, B. U. W. *Chem. Soc. Rev.* **2012**, *41*, 2463. (l) Beletskaya, I. P.; Cheprakov, A. V. *Organometallics* **2012**, *31*, 7753. (m) Jiao, J.; Murakami, K.; Itami, K. *ACS Catal.* **2016**, *6*, 610.

(6) For selected examples, please see: (a) Matsuda, N.; Hirano, K.; Satoh, T.; Miura, M. *Org. Lett.* **2011**, *13*, 2860. (b) Yoo, E. J.; Ma, S.; Mei, T.-S.; Chan, K. S. L.; Yu, J.-Q. *J. Am. Chem. Soc.* **2011**, *133*, 7652. (c) Rucker, R. P.; Whittaker, A. M.; Dang, H.; Lalic, G. *J. Am. Chem. Soc.* **2012**, *134*, 6571. (d) Chen, Z.-Y.; Ye, C.-Q.; Zhu, H.; Zeng, X.-P.; Yuan, J.-J. *Chem. - Eur. J.* **2014**, *20*, 4237. (e) Ye, C.; Zhu, H.; Chen, Z. *J. Org. Chem.* **2014**, *79*, 8900. (f) Zhu, H.; Chen, P.; Liu, G. *J. Am. Chem. Soc.* **2014**, *136*, 1766. (g) Ye, Z.; Dai, M. *Org. Lett.* **2015**, *17*, 2190. (h) Yoon, H.; Lee, Y. *J. Org. Chem.* **2015**, *80*, 10244. (i) Zhou, S.; Yang, Z.; Chen, X.; Li, Y.; Zhang, L.; Fang, H.; Wang, W.; Zhu, X.; Wang, S. *J. Org. Chem.* **2015**, *80*, 6323. (j) Zhang, Z.; Yu, Y.; Liebeskind, L. S. *Org. Lett.* **2008**, *10*, 3005.

(7) (a) Kim, H. J.; Kim, J.; Cho, S. H.; Chang, S. *J. Am. Chem. Soc.* **2011**, *133*, 16382. (b) Kantak, A. A.; Potavathri, S.; Barham, R. A.; Romano, K. M.; DeBoef, B. *J. Am. Chem. Soc.* **2011**, *133*, 19960. (c) Antonchick, A. P.; Samanta, R.; Kulikov, K.; Lategahn, J. *Angew. Chem., Int. Ed.* **2011**, *50*, 8605. (d) Samanta, R.; Bauer, J. O.; Strohmann, C.; Antonchick, A. P. *Org. Lett.* **2012**, *14*, 5518. (e) Manna, S.; Serebrennikova, P. O.; Utepova, I. A.; Antonchick, A. P.; Chupakhin, O. N. *Org. Lett.* **2015**, *17*, 4588. (f) Xiao, Q.; Tian, L.; Tan, R.; Xia, Y.; Qiu, D.; Zhang, Y.; Wang, J. *Org. Lett.* **2012**, *14*, 4230. (g) Mlynarski, S. N.; Karns, A. S.; Morken, J. P. *J. Am. Chem. Soc.* **2012**, *134*, 16449. (h) Zhu, C.; Li, G.; Ess, D. H.; Falck, J. R.; Kürti, L. *J. Am. Chem. Soc.* **2012**, *134*, 18253. For a review, please see: (i) Coeffard, V.; Moreau, X.; Thomassigny, C.; Greck, C. *Angew. Chem., Int. Ed.* **2013**, *52*, 5684.

(8) March, J.; Engenito, J. S. *J. Org. Chem.* **1981**, *46*, 4304.

(9) Chen, Z.; Wang, Q. *Org. Lett.* **2015**, *17*, 6130.

(10) Yang, C.; Liu, Y.; Yang, J.-D.; Li, Y.-H.; Li, X.; Cheng, J.-P. *Org. Lett.* **2016**, *18*, 1036.

(11) Zhang, C.; McClure, J.; Chou, C. J. *J. Org. Chem.* **2015**, *80*, 4919.

(12) Ventura, O. N.; Rama, J. B.; Turi, L.; Dannenberg, J. J. *J. Am. Chem. Soc.* **1993**, *115*, 5754.

(13) For more information, please see the Supporting Information.

DOI: 10.1021/acs.orglett.6b02740
Org. Lett. 2016, 18, 5512−5515