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Effect of Resonance on the Clickability of Alkenyl Azides in the Strain-promoted Cycloaddition with Dibenzo-fused Cyclooctynes

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The clickabilities of various alkenyl and alkyl azides in the strain-promoted cycloaddition with dibenzo-fused cyclooctynes were investigated. Although alkenyl azides generally exhibited lower clickabilities than those of alkyl azides, a sterically-hindered alkenyl azide showed high reactivity comparable with those of alkyl azides. Theoretical analyses indicated that these unique reactivities are derived from the frontier molecular orbital interactions and distortability of the azido groups.

Keywords: Click chemistry, Azide, Resonance effect

The rise of click reactions such as the copper-catalyzed azide–alkyne cycloaddition (CuAAC) and the strain-promoted azide–alkyne cycloaddition (SPAAC) have improved the utility of organic azides in a broad range of disciplines, including materials chemistry, medicinal chemistry, and chemical biology.[1–6] Particularly, the clickabilities of various azides have gained increasing attention as a means to improve the efficiency of click conjugation and to achieve sequential click reactions.[7,8]

Previously, we discovered a unique enhancement of the clickability of aromatic azides, i.e., that the double SPAAC reaction of 2,6-diisopropylphenyl azide (**1c**) with Sondheimer diyne (**2**) is significantly more accelerated than that of unsubstituted phenyl azide (**1b**) with **2** (Figure 1A).[8d] We surmised that the enhanced reactivity of doubly sterically-hindered aromatic azide **1c** is due to the increased distortability of the azido group induced by the steric inhibition of resonance. Several analyses, including absorption spectroscopy and theoretical studies based on a density functional theory (DFT) (B3LYP/6-31G(d)) method, indicated that azide **1c** is predisposed to adopt a twisted structure, whereas azide **1b** rather prefers the planar state due to the resonance between its phenyl and azido groups (Figure 1B). Interestingly, 2,6-diisopropylphenyl azide (**1c**) showed almost ten-times higher reactivity than benzyl azide (**1a**) (Figure 1A). We speculated that the lower clickability of **1a** than that of **1c** is caused by stabilization of the azido group by hyper-conjugation with carbon–hydrogen bonds, which decreases the distortability of the azido group. However, the effect of the resonance remains unclear. Herein, we report the significant effects of resonance and its inhibition by steric hindrance in the clickability of various alkyl and alkenyl azides from experimental and theoretical aspects (Figure 1C).

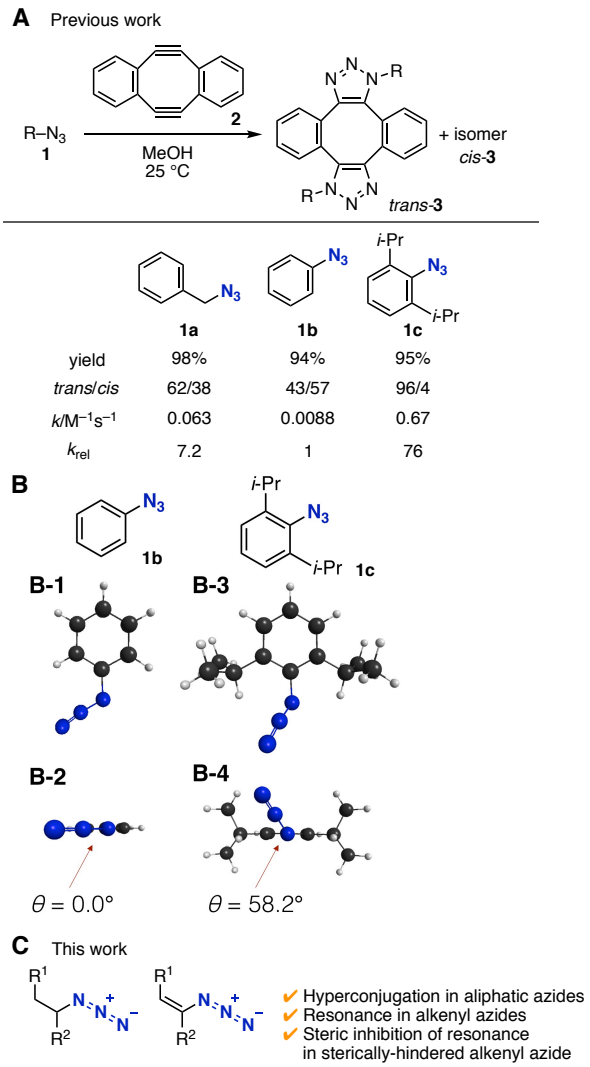
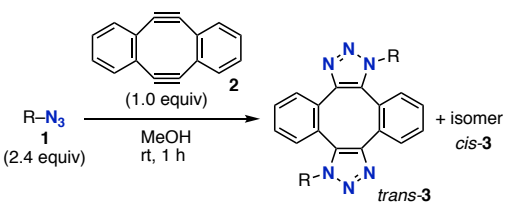


Figure 1. Clickability of various azides. (A) Double-click reactions of benzyl azide (**1a**), phenyl azide (**1b**), and 2,6-diisopropylphenyl azide (**1c**) with diyne **2**. (B) Overhead and side views of the ground state structures of **1b** and **1c**.[8d] (C) Alkyl and alkenyl azides.

We at first examined the clickability of alkyl and alkenyl azides **1** in the double-click reactions with Sondheimer diyne (**2**) (Table 1).[9,10] All reactions proceeded smoothly to afford a regioisomeric mixture of biscycloadducts **3** in excellent yields. The second-order rate

constants (k) for the first cycloadditions between azides **1** and diyne **2** clearly showed that the clickabilities of alkenyl azides **1e** and **1g** are lower than those of the corresponding saturated alkyl azides **1d** and **1f**. The reaction of 2-phenylethyl azide (**1d**) with diyne **2** was slightly faster than that of benzyl azide (**1a**) (entry 1 vs entry 2). Unsaturation from **1d** to *trans*-styryl azide (**1e**) decreased the clickability, indicating that the resonance between the azido and alkenyl groups retarded the cycloaddition (entry 2 vs entry 3). The reaction of secondary alkyl azide **1f** proceeded more slowly than those of primary alkyl azides **1a** and **1d** (entry 4 vs entries 1 and 2). Unsaturated azides such as 2-styryl azide (**1g**) and 4-phenyl-1-buten-2-yl azide (**1h**) also exhibited lower reactivities than that of **1f**, indicating that the alkenyl moiety decreased the clickability of azides (entries 5 and 6 vs entry 4). In contrast to the low reactivity of alkenyl azide **1g**, *trans*-1,2-diphenylvinyl azide (**1i**) showed similar reactivity to that of alkyl azide **1d**, despite the presence of an alkenyl and two bulky phenyl groups (entry 7). Thus, the introduction of a phenyl group to alkenyl azide **1g** significantly accelerates its click reaction with diyne **2**.

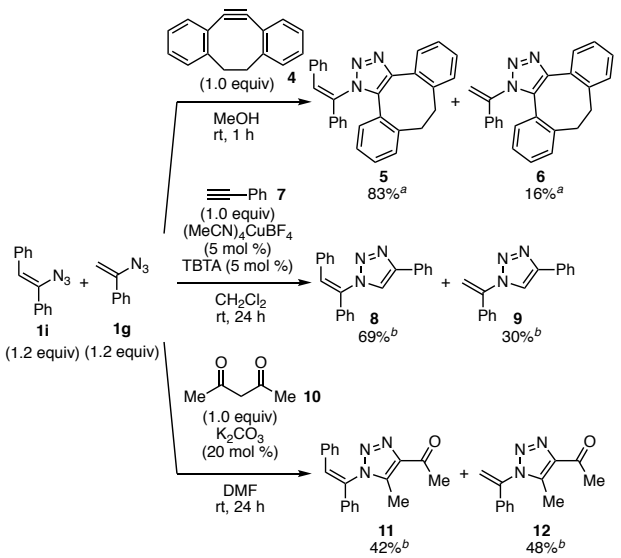
Table 1. Double-click reactions of diyne **2** with various alkyl and alkenyl azides **1**



Entry	R–N_3	1	3	Yield/%	*trans*:*cis*	k/$M^{-1}s^{-1}$ [c]	k_{rel}
1[a]	Ph, N_3	**1a**	**3a**	98	62/38	6.3×10^{-2}	16
2	Ph, N_3	**1d**	**3d**	99	57/43	9.6×10^{-2}	25
3	Ph, N_3	**1e**	**3e**	98	53/47	4.1×10^{-2}	11
4	Me, N_3, Ph	**1f**	**3f**	98	n.d.[b]	2.1×10^{-2}	5.4
5	N_3, Ph	**1g**	**3g**	98	49/51	3.9×10^{-3}	1
6	N_3, Ph	**1h**	**3h**	97	54/46	6.1×10^{-3}	1.6
7	Ph, N_3, Ph	**1i**	**3i**	quant.	75/25	8.9×10^{-2}	23

[a]From ref. 10. [b]Not determined. [c]See the Supporting Information for details.

We also performed competition experiments using an equimolar mixture of alkenyl azides **1i** and **1g** in three types of triazole formation reactions (Scheme 1). The SPAAC reaction with cyclooctyne **4**[4e] resulted in a preferential consumption of the bulkier alkenyl azide **1i** to afford cycloadduct **5** with good selectivity along with a small amount of cycloadduct **6** obtained from **1g** (Scheme 1, top). The CuAAC reaction[2] with terminal alkyne **7** afforded triazoles **8** and **9** without significant selectivity (Scheme 1, middle). The base-catalyzed triazole formation[11] with 1,3-diketone **10** provided an almost 1:1 mixture of triazoles **11** and **12** (Scheme 1, bottom). These results indicate that concerted triazole formation reactions between alkenyl azides and dibenzo-fused cyclooctynes are accelerated by increasing the steric hindrance of the azido group. This is similar to the enhanced clickability observed for aromatic azides upon introduction of two bulky *ortho*-substituents.



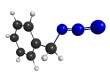
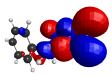
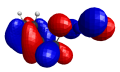
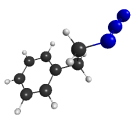
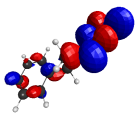
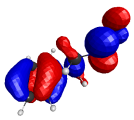
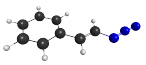
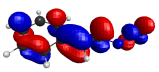
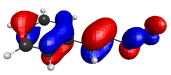
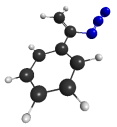
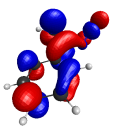
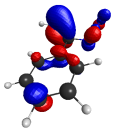
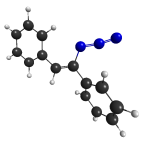
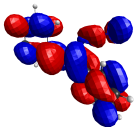
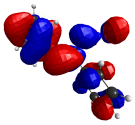
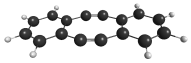
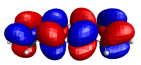
Scheme 1. Competitive triazole formation reactions. TBTA = tris[(1-benzyl-1*H*-1,2,3-triazol-4-yl)methyl]amine. [a]Isolated yields. [b] ^{1}H NMR yields.

To gain more insight into the clickability between alkyl and alkenyl azides in the SPAAC reaction with dibenzo-fused cyclooctynes, a DFT study was conducted for several azides **1** (Table 2). Although only slight differences were observed in the natural bond orbital analyses,[12] the frontier molecular orbital calculations clearly indicated the hyper-conjugation characteristics of azides **1**. For example, hyper-conjugation between the azido group and the neighboring C–H bonds was observed in the highest occupied molecular orbitals (HOMOs) of alkyl azides **1a** and **1d**. Comparisons of energy gaps between the lowest unoccupied molecular orbital (LUMO) and HOMO levels of azides **1** and diyne **2** suggest more favorable interactions between the frontier orbitals of alkenyl azides (**1e**, **1g**, and **1i**) and diyne **2** than those between alkyl azides (**1a** and **1d**) and **2**. Notably, the energy gap between the HOMO of the highly reactive azide **1i** and the LUMO of diyne **2** was the lowest (2.47 eV) among the azides examined.

The distortability of the azido groups in **1d**, **1e**, **1g**, and **1i** was theoretically evaluated by calculation of the rotation energy of the azido group (Figure 2). The low rotation energy of the azido group in alkyl azide **1d** indicates its higher distortability than those of the azido groups in alkenyl azides **1e**, **1g**, and **1i** (Figure 2A vs Figures 2B–2D). Azides **1e** and **1g** are most stable when the azido group lies in the same plane with the alkenyl group (Figures 2B and 2C). This causes the low distortability of the azido groups in these azides,

resulting in their low reactivities in the concerted click reactions. In contrast, alkenyl azide **1i** is stable as a partially-twisted structure, which renders the azido group slightly more distortable than those in alkenyl azides **1e** and **1g** (Figure 2D vs Figures 2B and 2C).

Table 2. Selected molecular orbitals contributing to the SPAAC reactions between azides **1** and diyne **2**.

1	Optimized structure[a]	Frontier orbitals[a]	
1a		 LUMO (−1.63 eV)	 HOMO (−6.33 eV)
1d		 LUMO (−1.61 eV)	 HOMO (−6.35 eV)
1e		 LUMO (−2.16 eV)	 HOMO (−5.48 eV)
1g		 LUMO (−2.15 eV)	 HOMO (−5.83 eV)
1i		 LUMO (−2.27 eV)	 HOMO (−5.33 eV)
2		 LUMO (−2.86 eV)	 HOMO (−5.24 eV)

[a]Optimized structures, molecular orbitals, and charge distributions were obtained by the theoretical analyses at the M11-L/6-31G(d) level. See the Supporting Information for details.

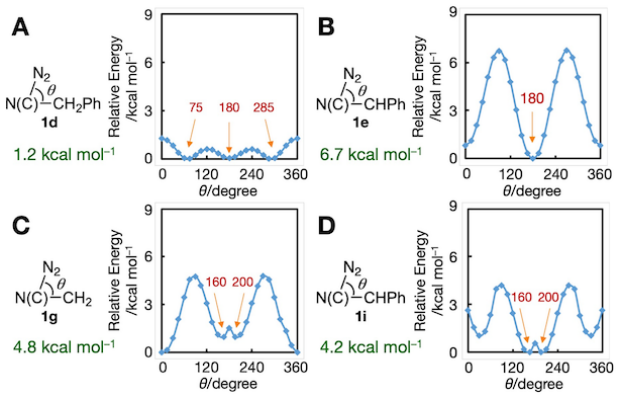
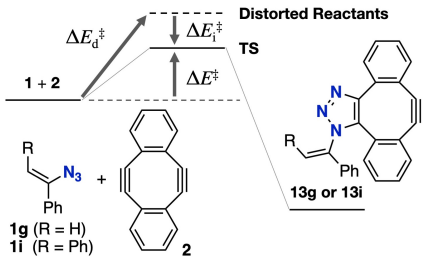


Figure 2. Rotation energy of the azido groups in **1d** (A), **1e** (B), **1g** (C), and **1i** (D) calculated at the M11-L/6-31G(d) level, where zero energies are set at the local minima. Rotation barrier (green) and dihedral angles in local minima (red) are shown.

Detailed theoretical analysis of the cycloaddition of alkenyl azides **1g** and **1i** with diyne **2** provided further insight into the enhancement of the clickability of **1i**. The potential energy profiles for the reactions of **1g** and **1i** with **2** are shown in Table 3. The order of calculated activation energies was in good agreement with that of the second-order rate constants shown in Table 1 (entries 5 and 7). Further analysis based on the distortion/interaction model[13] showed that the difference in the activation energies consists largely of the difference between the distortion energies of azides **1g** and **1i**. The lower distortion energy of **1i** than that of **1g** would be derived by the steric inhibition of resonance between the azido and alkenyl groups. This result and the analysis based on the frontier molecular orbitals show that the higher clickability of **1i** than that of **1g** is attributable to the enhanced distortability of the azido group induced by the inhibition of resonance with the additional bulky phenyl group, which surpassed the deceleration caused by the steric hindrance.

Table 3. DFT analysis of the cycloaddition of **1g** and **1i** with **2**[a]



Reactants	**1g** + **2**		**1i** + **2**	
Distorted reactants[b]	+18.8	+3.3	+16.3	+3.2
Distortion energy[b] ($\Delta E_d^{\ddagger}$)	+22.1		+19.5	
Interaction energy[c] ($\Delta E_i^{\ddagger}$)	−11.6		−12.7	
Activation energy[d] ($\Delta E^{\ddagger}$)	+10.5		+6.8	

[a]Distortion, interaction, and activation energies for the first cycloaddition from the most stable conformations obtained at the M11-L/6-31G(d) level are shown in kcal mol^{-1}. [b]Energy required to distort the geometry of each reactant to the transition state (TS). [c]Interaction energy between the distorted fragments at the TS. [d]Energy differences for each fragment between the optimized and the TS geometries.

In summary, we found that alkenyl azides generally exhibit lower clickabilities than those of alkyl azides in the SPAAC reactions with dibenzo-fused cyclooctynes, though a sterically-hindered alkenyl azide shows high reactivity comparable with those of alkyl azides. DFT analysis revealed that the frontier molecular orbital interactions between azides and cyclooctynes and the distortion energy of the azido group determine the clickabilities of alkenyl and alkyl azides. The development of a new convergent synthetic method for multifunctional molecular probes and construction of a chemical library by sequential click conjugations based on the diverse clickabilities of various azides are now underway in our group.

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References and Notes

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1 H. C. Kolb, M. G. Finn, K. B. Sharpless, *Angew. Chem., Int. Ed.* **2001**, *40*, 2004.

2 a) C. W. Tornøe, C. Christensen, M. Meldal, *J. Org. Chem.* **2002**, *67*, 3057. b) V. V. Rostovtsev, L. G. Green, V. V. Fokin, K. B. Sharpless, *Angew. Chem., Int. Ed.* **2002**, *41*, 2596. c) M. Meldal, C. W. Tornøe, *Chem. Rev.* **2008**, *108*, 2952. d) J. E. Hein, V. V. Fokin, *Chem. Soc. Rev.* **2010**, *39*, 1302. e) A. Mandoli, *Molecules* **2016**, *21*, 1174.

3 a) E. M. Sletten, C. R. Bertozzi, *Angew. Chem., Int. Ed.* **2009**, *48*, 6974. b) M. F. Debets, C. W. J. van der Doelen, F. P. J. T. Rutjes, F. L. van Delft, *ChemBioChem* **2010**, *11*, 1168. c) J. C. Jewett, C. R. Bertozzi, *Chem. Soc. Rev.* **2010**, *39*, 1272. d) J. Dommerholt, F. P. J. T. Rutjes, F. L. van Delft, *Top. Curr. Chem.* **2016**, *374*, 16. e) S. Yoshida, *Bull. Chem. Soc. Jpn.* **2018**, *91*, 1293.

4 a) G. Wittig, A. Krebs, *Chem. Ber.* **1961**, *94*, 3260. b) N. J. Agard, J. A. Prescher, C. R. Bertozzi, *J. Am. Chem. Soc.* **2004**, *126*, 15046. c) S. T. Laughlin, J. M. Baskin, S. L. Amacher, C. R. Bertozzi, *Science* **2008**, *320*, 664. d) J. A. Codelli, J. M. Baskin, N. J. Agard, C. R. Bertozzi, *J. Am. Chem. Soc.* **2008**, *130*, 11486. e) X. Ning, J. Guo, M. A. Wolfert, G.-J. Boons, *Angew. Chem., Int. Ed.* **2008**, *47*, 2253. f) A. A. Poloukhtine, N. E. Mbua, M. A. Wolfert, G.-J. Boons, V. V. Popik, *J. Am. Chem. Soc.* **2009**, *131*, 15769. g) M. F. Debets, S. S. van Berkel, S. Schoffelen, F. P. J. T. Rutjes, J. C. M. van Hest, F. L. van Delft, *Chem. Commun.* **2010**, *46*, 97. h) J. C. Jewett, E. M. Sletten, C. R. Bertozzi, *J. Am. Chem. Soc.* **2010**, *132*, 3688. i) J. Dommerholt, S. Schmidt, R. Temming, L. J. A. Hendriks, F. P. J. T. Rutjes, J. C. M. van Hest, D. J. Lefeber, P. Friedl, F. L. van Delft, *Angew. Chem., Int. Ed.* **2010**, *49*, 9422. j) E. H. P. Leunissen, M. H. L. Meuleners, J. M. M. Verkade, J. Dommerholt, J. G. J. Hoenderop, F. L. van Delft, *ChemBioChem* **2014**, *15*, 1446. k) S. Yoshida. Y. Hatakeyama, K. Johmoto, H. Uekusa, T. Hosoya, *J. Am. Chem. Soc.* **2014**, *136*, 13590. l) R. Ni, N. Mitsuda, T. Kashiwagi, K. Igawa, K. Tomooka, *Angew. Chem., Int. Ed.* **2015**, *54*, 1190. m) K. Kaneda, R. Naruse, S. Yamamoto, *Org. Lett.* **2017**, *19*, 1096. n) K. Igawa, S. Aoyama, Y. Kawasaki, T. Kashiwagi, Y. Seto, R. Ni, N. Mitsuda, K. Tomooka, *Synlett* **2017**, *28*, 2110. o) E. G. Burke, B. Gold, T. T. Hoang, R. T. Raines, J. M. Schomaker, *J. Am. Chem. Soc.* **2017**, *139*, 8029. p) S. Yoshida, T. Kuribara, H. Ito, T. Meguro, Y. Nishiyama, F. Karaki, Y. Hatakeyama, Y. Koike, I. Kii, T. Hosoya, *Chem. Commun.* **2019**, *55*, 3556. q) C. Lis, T. Berg, *Synlett*, **2019**, *30*, 939.

5 a) J. Lahann, *Click Chemistry for Biotechnology and Materials Science*, John Wiley & Sons, West Sussex, 2009. b) W. Xi, T. F. Scott, C. J. Kloxin, C. N. Bowman, *Adv. Funct. Mater.* **2014**, *24*, 2572.

6 a) P. Thirumurugan, D. Matosiuk, K. Jozwiak, *Chem. Rev.* **2013**, *113*, 4905. (b) C. S. McKay, M. G. Finn, *Chem. Biol.* **2014**, *21*, 1075.

7 Little attention has been paid to increase the reactivity of the azide. See: a) S. Bräse, C. Gil, K. Knepper, V. Zimmermann, *Angew. Chem., Int. Ed.* **2005**, *44*, 5188. b) S. Bräse, K. Banert, *Organic Azides: Syntheses and Applications*, John Wiley & Sons, West Sussex, 2010. c) Z. Yuan, G.-C. Kuang, R. J. Clark, L. Zhu, *Org. Lett.* **2012**, *14*, 2590. d) J. Dommerholt, O. van Rooijen, A. Borrmann, C. F. Guerra, F. M. Bickelhaupt, F. L. van Delft, *Nat. Commun.* **2014**, *5*, 5378. e) S. Xie, S. A. Lopez, O. Ramström, M. Yan, K. N. Houk, *J. Am. Chem. Soc.* **2015**, *137*, 2958. f) N. Münster, P. Nikodemiak, U. Koert, *Org. Lett.* **2016**, *18*, 4296. g) K. Banert, *Synthesis* **2016**, *48*, 2361. h) D. Huang, G. Yan, *Adv. Synth. Catal.* **2017**, *359*, 1600. i) T. Yokoi, H. Tanimoto, T. Ueda, T. Morimoto, K. Kakiuchi, *J. Org. Chem.* **2018**, *83*, 12103. j) D. Svatunek, N. Houszka, T. A. Hamlin, F. M. Bickelhaupt, H. Mikula, *Chem.–Eur. J.* **2019**, *25*, 754. k) T. Yokoi, T. Ueda, H. Tanimoto, T. Morimoto, K. Kakiuchi, *Chem. Commun.* **2019**, *55*, 1891.

8 a) T. Hosoya, T. Hiramatsu, T. Ikemoto, M. Nakanishi, H. Aoyama, A. Hosoya, T. Iwata, K. Maruyama, M. Endo, M. Suzuki, *Org. Biomol. Chem.* **2004**, *2*, 637. b) T. Hosoya, T. Hiramatsu, T. Ikemoto, H. Aoyama, T. Ohmae, M. Endo, M. Suzuki, *Bioorg. Med. Chem. Lett.* **2005**, *15*, 1289. c) T. Hosoya, A. Inoue, T. Hiramatsu, H. Aoyama, T. Ikemoto, M. Suzuki, *Bioorg. Med. Chem.* **2009**, *17*, 2490. d) S. Yoshida, A. Shiraishi, K. Kanno, T. Matsushita, K. Johmoto, H. Uekusa, T. Hosoya, *Sci. Rep.* **2011**, *1*, 82. e) S. Yoshida, Y. Misawa, T. Hosoya, *Eur. J. Org. Chem.* **2014**, 3991. f) T. Meguro, S. Yoshida, T. Hosoya, *Chem. Lett.* **2017**, *46*, 1137. g) S. Yoshida, K. Kanno, I. Kii, Y. Misawa, M. Hagiwara, T. Hosoya, *Chem. Commun.* **2018**, *54*, 3705. h) T. Meguro, N. Terashima, H. Ito, Y. Koike, I. Kii, S. Yoshida, T. Hosoya, *Chem. Commun.* **2018**, *54*, 7904. i) T. Meguro, S. Yoshida. K. Igawa, K. Tomooka, T. Hosoya, *Org. Lett.* **2018**, *20*, 4126. j) S. Yoshida, J. Tanaka, Y. Nishiyama, Y. Hazama, T. Matsushita, T. Hosoya, *Chem. Commun.* **2018**, *54*, 13499.

9 a) H. N. C. Wong, P. J. Garratt, F. Sondheimer, *J. Am. Chem. Soc.* **1974**, *96*, 5604. b) A. Orita, D. Hasegawa, T. Nakano, J. Otera, *Chem.–Eur. J.* **2002**, *8*, 2000. c) F. Xu, L. Peng, K. Shinohara, T. Morita, S. Yoshida, T. Hosoya, A. Orita, J. Otera, *J. Org. Chem.* **2014**, *79*, 11592.

10 I. Kii, A. Shiraishi, T. Hiramatsu, T. Matsushita, H. Uekusa, S. Yoshida, M. Yamamoto, A. Kudo, M. Hagiwara, T. Hosoya, *Org. Biomol. Chem.* **2010**, *8*, 4051.

11 E. P. J. Ng, Y.-F. Wang, B. W.-Q. Hui, G. Lapointe, S. Chiba, *Tetrahedron* **2011**, *67*, 7728.

12 See the Supporting Information for details.

13 a) D. H. Ess, K. N. Houk, *J. Am. Chem. Soc.* **2007**, *129*, 10646. b) D. H. Ess, G. O. Jones, K. N. Houk, *Org. Lett.* **2008**, *10*, 1633.