Letter

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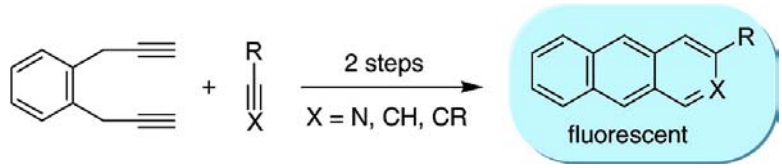
Org. Lett., **2008**, 10 (20), 4661-4664• DOI: 10.1021/ol8019549 • Publication Date (Web): 25 September 2008



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ORGANIC LETTERS

2008
Vol. 10, No. 20
4661−4664

Synthesis of Anthracene and Azaanthracene Fluorophores via [2+2+2] Cyclotrimerization Reactions

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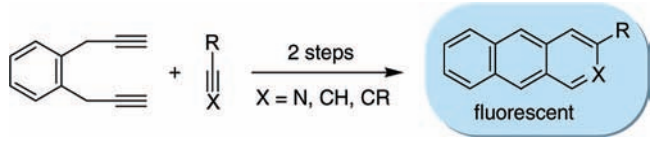
Received August 20, 2008

ABSTRACT



A highly convergent [2+2+2] cyclotrimerization approach to anthracenes and 2-azaanthracenes has been developed. It allows for the facile introduction of the anthracene moiety on alkyne and nitrile bearing molecules and the rapid construction of compound arrays. This is showcased in the assembly of a collection of fluorophores and their photochemical evaluation.

Anthracenes are core structures employed in a variety of practical applications, including potential therapeutics,[1] optical devices,[2] and polymeric materials.[3] Most importantly, anthracenes have interesting fluorescent properties and have received attention as imaging agents for cellular processes.[4]

Recently, we[5,6] and others[7] discovered significant microwave effects on transition-metal catalyzed [2+2+2] cyclotrimerization reactions, enabling their application as effective tools for the synthesis of arrays of carbo- and heterocyclic structures. Here, we describe the application of this methodology to the rapid assembly of fluorophores based on an anthracene and an azaanthracene scaffold. Although several synthetic routes to anthracenes have been reported,[8,9] the synthesis of 2-azaanthracenes is an undeveloped field. Moreover, we discovered that the synthesized 2-azaanthracenes have very unique fluorescent properties in contrast to regular anthracenes.

A classical [2+2+2] cyclotrimerization reaction involves the transformation of three or two alkynes and/or a nitrile to benzenes and pyridines (Scheme 1). The reactions are

[†] Both authors contributed equally to this work.

(1) (a) Alfred, L. J.; DiPaolo, J. A. *Cancer Res.* **1968**, *28*, 60. (b) Srinivasan, R.; Tan, L. P.; Wu, H.; Yao, S. Q. *Org. Lett.* **2008**, *10*, 2295. (c) Tan, W. B.; Bhambhani, A.; Duff, M. R.; Rodger, A.; Kumar, C. V. *Photochem. Photobiol.* **2006**, *82*, 20. (d) Piao, W. H.; Wong, R.; Bai, X. F.; Huang, J.; Campagnolo, D. I.; Dorr, R. T.; Vollmer, T. L.; Shi, F. D. *J. Immunol.* **2007**, *179*, 7415.

(2) (a) Hirose, K.; Shiba, Y.; Ishibashi, K.; Doi, Y.; Tobe, Y. *Chem.−Eur. J.* **2008**, *14*, 981. (b) Gimenez, R.; Pinol, M.; Serrano, J. L. *Chem. Mater.* **2004**, *16*, 1377.

(3) (a) Hargreaves, J. S.; Webber, S. E. *Macromolecules* **1984**, *17*, 235. (b) Rameshbabu, K.; Kim, Y.; Kwon, T.; Yoo, J.; Kim, E. *Tetrahedron Lett.* **2007**, *48*, 4755.

(4) (a) Wilson, A. J.; Hong, J.; Fletcher, S.; Hamilton, A. D. *Org. Biomol. Chem.* **2007**, *5*, 276. (b) Gassensmith, J. J.; Arunkumar, E.; Barr, L.; Baumes, J. M.; DiVittorio, K. M.; Johnson, J. R.; Noll, B. C.; Smith, B. D. *J. Am. Chem. Soc.* **2007**, *129*, 15054.

(5) Young, D. D.; Deiters, A. *Angew. Chem., Int. Ed.* **2007**, *46*, 5187.

(6) (a) Young, D. D.; Sripada, L.; Deiters, A. *J. Comb. Chem.* **2007**, *9*, 735. (b) Teske, J. A.; Deiters, A. *J. Org. Chem.* **2008**, *73*, 342.

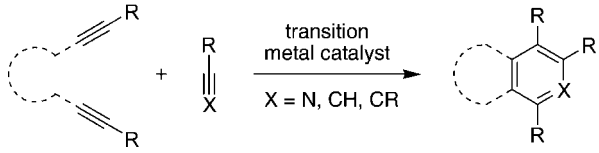
(7) (a) Zhou, Y.; Porco, J. A.; Snyder, J. K. *Org. Lett.* **2007**, *9*, 393. (b) Hrdina, R.; Kadlcikova, A.; Valterova, I.; Hodacova, J.; Kotora, M. *Tetrahedron: Asymmetry* **2006**, *17*, 3185. (c) Saaby, S.; Baxendale, I. R.; Ley, S. V. *Org. Biomol. Chem.* **2005**, *3*, 3365. (d) Efskind, J.; Undheim, K. *Tetrahedron Lett.* **2003**, *44*, 2837. (e) Schore, N. E. *Chem. Rev.* **1988**, *88*, 1081. (f) Schore, N. E. [2+2+2] Cycloadditions. In *Comprehensive Organic Synthesis*; Trost, B. M., Fleming, I., Paquette, L. A., Eds.; Pergamon Press: Oxford, 1991; Vol. 5, pp 1129. (g) Varela, J. A.; Saa, C. *Chem. Rev.* **2003**, *103*, 3787. (h) Yamamoto, Y. *Curr. Org. Chem.* **2005**, *9*, 503. (i) Gandon, V.; Aubert, C.; Malacria, M. *Chem. Commun.* **2006**, 2209. (j) Vollhardt, K. P. C. *Angew. Chem., Int. Ed.* **1984**, *23*, 539. (k) Bonnemann, H. *Angew. Chem., Int. Ed.* **1985**, *24*, 248.

(8) (a) House, H. O.; Campbell, W. J.; Koepsell, D. G. *J. Org. Chem.* **1972**, *37*, 1003. (b) Sangaiah, R.; Gold, A.; Toney, G. E. *J. Org. Chem.* **1983**, *48*, 1632. (c) Li, G.; Zhou, S.; Su, G.; Liu, Y.; Wang, P. G. *J. Org. Chem.* **2007**, *72*, 9830.

(9) Takahashi, T.; Li, S.; Huang, W. Y.; Kong, F. Z.; Nakajima, K.; Shen, B. J.; Ohe, T.; Kanno, K. *J. Org. Chem.* **2006**, *71*, 7967.

10.1021/ol8019549 CCC: $40.75
Published on Web 09/25/2008

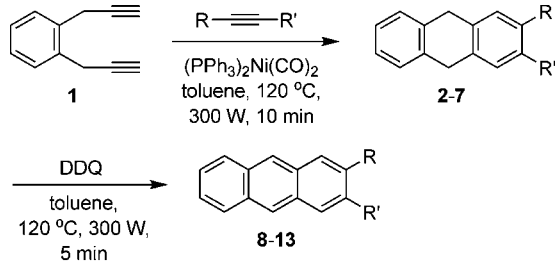
Scheme 1. General [2+2+2] Cyclotrimerization Reaction to (Fused) Benzenes and Pyridines



typically conducted under Co, Ni, Ru, or Rh catalysis, although other transition metals have been used as well.[10] To avoid chemoselectivity issues in the cyclotrimerization step, one of the alkynes can be immobilized on a solid-support,[5,11] or two alkynes can be tethered together. The latter leads to the synthesis of fused benzene and pyridine rings and has been applied in the assembly of a wide range of aromatic structures.

We envisioned the application of this strategy to the synthesis of anthracenes, and azanthracenes and thus assembled the known diyne **1**[9] in three steps from commercially available o-xylene dibromide with an overall yield of 72%. The precursor **1** was cyclotrimerized with a set of six alkynes containing alkyl chains, benzenes, hydroxy groups, nitriles, and imides. These reactions were conducted with 10 mol% $(PPh_3)_2Ni(CO)_2$ in toluene at 120 °C under microwave irradiation (300 W) in 10 min, delivering the tricyclic compounds **2**−**7** in 66−86% yield (Scheme 2).

Scheme 2. Anthracene Synthesis via [2+2+2] Cyclotrimerization

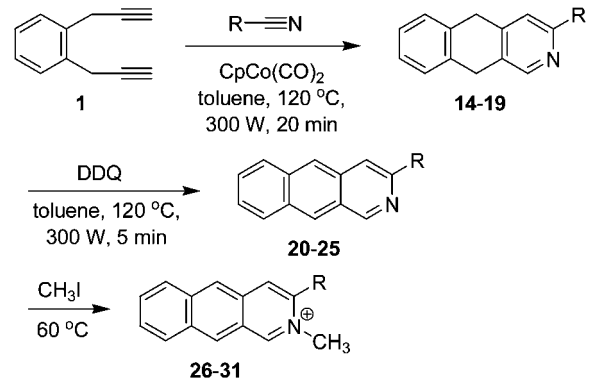


entry	R	R'	compd	yield	compd	yield
1	Bu	H	**2**	75%	**8**	70%
2	Ph	Ph	**3**	86%	**9**	78%
3	CH_2OH	H	**4**	66%	**10**	79%[a]
4	$(CH_2)_3CN$	H	**5**	74%	**11**	74%
5	$(CH_2)_3OH$	H	**6**	78%	**12**	70%
6	$(CH_2)_2$Pth	H	**7**	86%	**13**	76%[b]

[a]Oxidized to aldehyde. [b]Pth = phthalimide.

Reactions conducted thermally in the absence of microwave irradiation displayed diminished yields (e.g., 35% in the case of compound **4** under otherwise identical conditions). Byproducts observed in some reactions resulted from the dimerization of **1**, as well as cyclotrimerization of **1** with two monoalkyne molecules.

Scheme 3. Azanthracene Synthesis via [2+2+2] Cyclotrimerization



entry	R	compd	yield	compd	yield	compd	yield
1	Me	**14**	94%	**20**	75%	**26**	99%
2	Pr	**15**	87%	**21**	85%	**27**	99%
3	$(CH_2)_2OH$	**16**	83%	**22**	53%	**28**	99%
4	$CH{=}CH_2$	**17**	90%	**23**	80%	**29**	99%
5	Ph	**18**	87%	**24**	84%	**30**	99%
6	Pyr	**19**	80%	**25**	81%	**31**	99%[a]

[a]Pyr = pyridine.

The anthracences **8**−**13** were generated in yields of 70−79% through a rapid microwave-assisted oxidation with 2,3-dichloro-5,6-dicyanobenzoquinone (DDQ) (Scheme 2). This two-step procedure provides a flexible and facile approach to the introduction of an anthracene moiety into a wide range of alkynes.

We subsequently investigated the feasability of this route toward the synthesis of 2-azaanthracenes, a mostly unexplored class of compounds. Here, the diyne **1** was reacted with nitriles bearing a variety of functional groups, including alkyl and alkene chains, hydroxy groups, benzene, and pyridine rings. The reactions were conducted under $CpCo(CO)_2$ catalysis in toluene using microwave irradiation (300 W), delivering the cyclotrimerization products **14**−**19** in 80−94% yields. The change in catalyst system was necessary to achieve cyclotrimerization reactions toward

(10) (a) Das, S. K.; Roy, R. *Tetrahedron Lett.* **1999**, *40*, 4015. (b) Yamamoto, Y.; Arakawa, T.; Ogawa, R.; Itoh, K. *J. Am. Chem. Soc.* **2003**, *125*, 12143. (c) Torrent, A.; Gonzalez, I.; Pla-Quintana, A.; Roglans, A.; Moreno-Manas, M.; Parella, T.; Benet-Buchholz, J. *J. Org. Chem.* **2005**, *70*, 2033. (d) Tanaka, K.; Toyoda, K.; Wada, A.; Shirasaka, K.; Hirano, M. *Chemistry* **2005**, *11*, 1145. (e) Bonaga, L. V. R.; Zhang, H. C.; Moretto, A. F.; Ye, H.; Gauthier, D. A.; Li, J.; Leo, G. C.; Maryanoff, B. E. *J. Am. Chem. Soc.* **2005**, *127*, 3473. (f) Evans, P. A.; Sawyer, J. R.; Lai, K. W.; Huffman, J. C. *Chem. Commun.* **2005**, 3971. (g) Duong, H. A.; Cross, M. J.; Louie, J. *J. Am. Chem. Soc.* **2004**, *126*, 11438. (h) Duong, H. A.; Louie, J. *J. Organomet. Chem.* **2005**, *690*, 5098. (i) McCormick, M. M.; Duong, H. A.; Zuo, G.; Louie, J. *J. Am. Chem. Soc.* **2005**, *127*, 5030. (j) Yamamoto, Y.; Kinpara, K.; Saigoku, F.; Takagishi, H.; Okuda, S.; Nishiyama, H.; Itoh, K. *J. Am. Chem. Soc.* **2005**, *127*, 605. (k) Fatland, A. W.; Eaton, B. E. *Org. Lett.* **2000**, *2*, 3131.

(11) (a) Senaiar, R. S.; Young, D. D.; Deiters, A. *Chem. Commun.* **2006**, 1313. (b) Young, D. D.; Senaiar, R. S.; Deiters, A. *Chem.−Eur. J.* **2006**, *12*, 5563.

(12) (a) Rettig, W. *Applied fluorescence in chemistry, biology and medicine*' Springer-Verlag: Berlin; New York, 1999; p xix. (b) Valeur, B. *Molecular fluorescence: principles and applications*; Wiley-VCH: Weinheim, 2002; p xiv.

pyridines.[7e−k] In the case of thermal heating, but in the absence of microwave irradiation, very low product yields were obtained under otherwise identical conditions (e.g., 8% in case of compound **18**). The subsequent DDQ oxidation step proceeded smoothly and yielded the azaanthracences **20**−**25** in 53−85% yield. To investigate the effects of a permanently positively charged nitrogen center on the fluorescent properties of azaanthracenenes, and to increase their solubility in an aqueous environment, we methylated the azaanthracenes **20**−**25** in neat methyl iodide at 60 °C to obtain the salts **26**−**31** in quantitative yields.

A prominent feature of anthracenes is their intrinsic fluorescence,[12] a property that is largely unexplored in case of the corresponding 2-azaanthracenes. The developed array approach to both compound classes prompted us to investigate the fluorescent properties of the synthesized molecules, since combinatorial approaches to fluorophores have been successfully applied to biological imaging problems.[13] We were especially interested in their application as environmentally sensitive probes, and therefore investigated the dependence of their fluorescence spectra on (a) the polarity of the solvent, (b) the pH of the solvent, and (c) the presence of different metal cations. These experiments were conducted in a 96-well format (Figure 1).

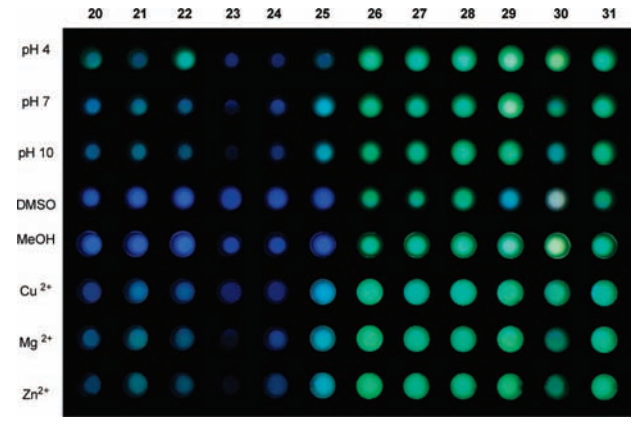


Figure 1. Synthesized azaanthracenes arrayed in a 96-well plate, exposed to a variety of conditions (see text) and excited with 365 nm UV light.

While the anthracenes **9**−**12** did not show any significant changes in fluorescence under different conditions, many of the azaanthracenes **20**−**25** exhibited environmental sensitivity (Figure 1). This can be explained by the ability of the nitrogen center to undergo coordination to the solvent, to protons, and to metal ions. Moreover, the azaanthracenes **20**−**25** exhibited generally higher fluorescence levels than the anthracenes **8**−**13** (see Supporting Information, Figures

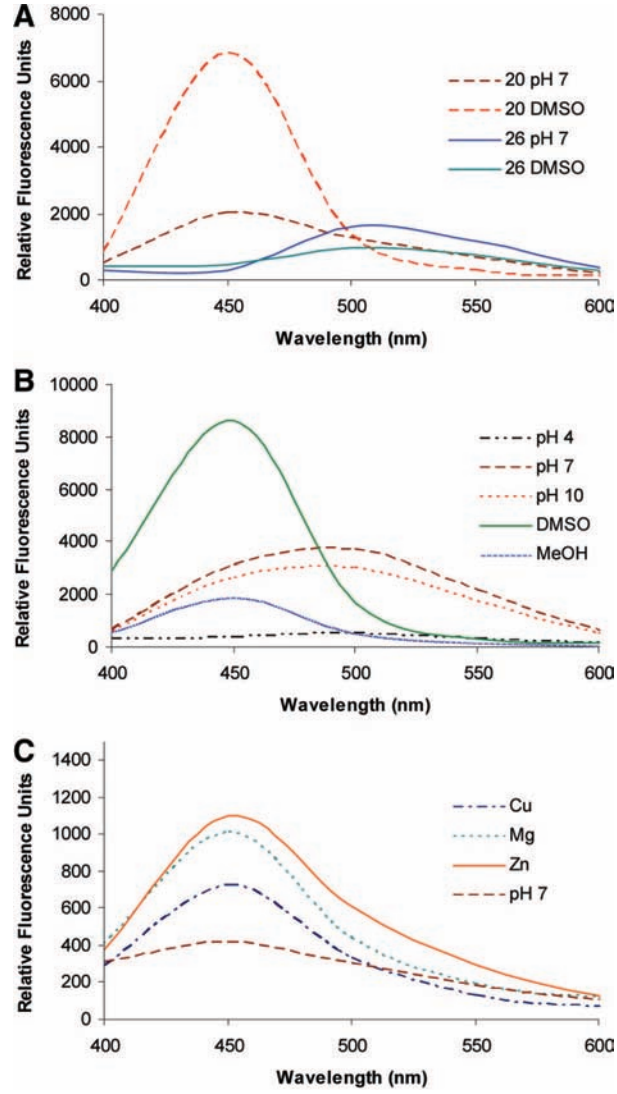


Figure 2. Fluorescence spectra (350 nm excitation wavelength) of (A) representative azaanthracenes **20** and **26** demonstrating the effect of quaterinization on the fluorescence properties in DMSO and H_2O; (B) the representative azaanthracene **22** in the presence of different solvents and different pH; and (C) the pyridylazaanthracene **25** in the presence of different divalent metal cations.

S1−S3). A significant change in fluorescence intensity was observed for most of the azaanthracenes **20**−**25** between protic (H_2O at pH 4−10) and aprotic (DMSO) solvents (Figure 1). More drastic changes in fluorescence emission were visually observed in other nonpolar solvents such as CH_2Cl_2 and toluene (see Supporting Information, Figures S2 and S3); however, these solvents were incompatible with the 96-well microtiter plates employed in the fluorescence measurements. This phenomenon has previously been investigated for 2-azaanthracene (**20**), finding that a polar solvent causes a broadening of the $\pi \rightarrow \pi_1^*$ energy levels resulting in a bathochromic shift.[14] A loss of solvent sensitivity was observed upon quaternization of the nitrogen center in **26**−**31**, and a moderate broadening of the emission spectrum occurred when compared to the non-quaternized

(13) (a) Rosania, G. R.; Lee, J. W.; Ding, L.; Yoon, H. S.; Chang, Y. T. *J. Am. Chem. Soc.* **2003**, *125*, 1130. (b) Schiedel, M. S.; Briehn, C. A.; Bauerle, P. *Angew. Chem., Int. Ed.* **2001**, *40*, 4677. (c) Yee, D. J.; Balsanek, V.; Sames, D. *J. Am. Chem. Soc.* **2004**, *126*, 2282.
(14) Zvolinskii, V. P. *Russ. Phys. J.* **1977**, *20*, 701.
(15) Piantadosi, C. A.; Suliman, H. B. *J. Biol. Chem.* **2006**, *281*, 324.

analogs (Figure 2A). Additionally, a visible bathochromic shift in emission could be detected upon quaternization from blue to green (see compounds **20**−**25** and **26**−**31** in Figure 1). At low pH (<4), protonation of the azaanthracenes **20**−**25** led to a general bathochromic shift (∼480 to 520 nm) of their fluorescence emission spectra, similar to their quaternized analogs **26**−**31** (Figure 2B). No changes in fluorescence were observed between pH 7−10. As expected, the azaanthracene **25** demonstrated a fluorescence sensitivity toward metal ions, as a result of the chelating ability of the bipyridyl motif. An increase in the fluorescence intensity of **25** was observed in the presence of different divalent metal ions, suggesting a trend in binding affinity $Cu^{2+} < Mg^{2+} \approx Zn^{2+}$ (Figure 2C).

We also examined the application of the synthesized compounds for the fluorescent labeling of mammalian cells. Compounds **20**−**31** (20 μM) were incubated with human embryonic kidney (HEK-293T) cells overnight in standard DMEM growth media. To our surprise, compound **24** selectively forms ∼200 nm sized particles within the cells but not outside the cells, as confirmed by confocal microscopy using a Leica TCS SP1 laser scanning confocal microscope (Figure 3). By imaging the cells on varying planes of focus, the upper and lower membranes are apparent with the aggregates inside of the cell, not on the surface (see supporting video, Supporting Information). No observable effect on the cell phenotype and the cell viability was observed (Promega Cell-Titer Blue assay).[15]

In summary, we have developed a rapid route to anthracenes and azaanthracenes via microwave assisted [2+2+2] cyclotrimerization reactions. These compounds have unique photochemical and biological properties and can act as environmentally sensitive dyes, metal sensors, pH sensors, and cellular stains. Future work will reveal further interesting phenomena and lead to the application of these fluorophores in a biological context.

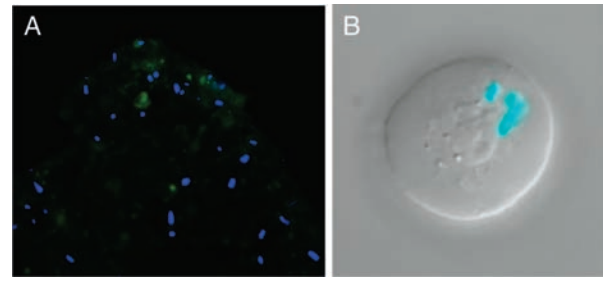


Figure 3. (A) HEK-293T cells incubated with azaanthracene **24** (20 μM) for 12 h were imaged at 365 nm UV irradiation (100 × magnification). The slight green color is a result of cell autofluorescence. (B) Confocal microscopy image of azaanthracene **18** crystallized inside a single human embryonic kidney cell (400 × magnification, false color image).

Acknowledgment. This research was supported by the Donors of the American Chemical Society Petroleum Research Fund and the Department of Chemistry at North Carolina State University. D.D.Y. acknowledges a graduate research fellowship from the ACS Medicinal Chemistry Division. A.C.M. was supported by the NCSU REU program. We also acknowledge T. J. Styslinger (NCSU) for his contributions to the work.

Supporting Information Available: General reaction protocols, analytical data, as well as ^{1}H NMR spectra and fluorescent properties for compounds **2**−**31**. This information is available free of charge via the Internet at http://pubs.acs.org.

OL8019549