Available online at www.sciencedirect.com
SCIENCE DIRECT®
Tetrahedron Letters 45 (2004) 4489–4493
Tetrahedron Letters

Concise synthesis of the core bicyclo[2.2.2]diazaoctane ring common to asperparaline, paraherquamide, and stephacidin alkaloids

Luke A. Adams, Chandele R. Gray and Robert M. Williams*

Department of Chemistry, Colorado State University, Fort Collins, CO 80523, USA

Received 16 March 2004; accepted 9 April 2004

Abstract—A versatile synthesis of the bicyclo[2.2.2]diazaoctane core structure of asperparaline, brevianamide, paraherquamide, and stephacidin natural products is demonstrated. This convergent synthesis relies on an intramolecular hetero Diels–Alder reaction to construct the key tetracycle from a diketopiperazine derived azadiene; which in turn was formed from prolinamide and a pyruvic acid derivative. The stereochemical outcome of the Diels–Alder reaction was found to favor the brevianamide stereochemistry.

Asperparaline A (**1**, aspergillimide) was isolated in 1997 by Hayashi from the fungus *Aspergillus japonicus* JV-23,[1] which had been obtained from soil samples collected in Sakai, Japan. Subsequently asperparalines B (**2**) and C (**3**) were also isolated by the same group and all three compounds were shown to have paralytic activity against silkworms.[2] Asperparaline A was also isolated independently by Everett's group,[3] from *Aspergillus* sp. IMI 337664. Like the structurally related paraherquamides,[4] the asperparalines display anthelmintic properties. Drugs developed with these characteristics have found global use, as highlighted by a World Health Organization study on the impact of deworming on childhood anemia in Tanzania.[5]

Significantly, recent reports concerning the isolation and structural elucidation of avrainvillamide **4**[6] and stephacidins A (**5**) and B (**6**),[7] has exemplified alkaloids that are structurally similar to the asperparalines and paraherquamides (Fig. 1), but have also shown promising anti-cancer activity. Of particular interest was the report that stephacidin B that exhibited selective, in vitro, antitumor activity against testosterone-sensitive prostrate LNCaP cell line with an IC_{50} value of 0.06 μM.[8]

The asperparalines, brevianamides, paraherquamides, and stephacidins are all structurally related by being

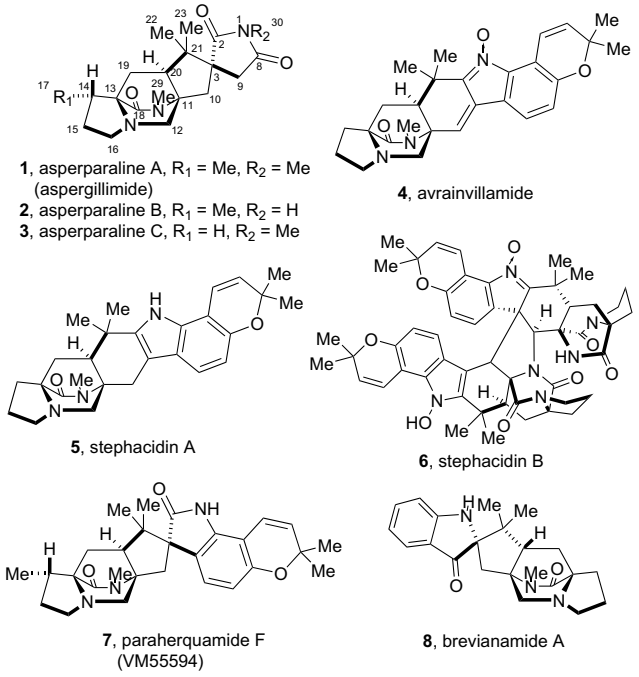


Figure 1. Structures of the bicyclo[2.2.2] indole alkaloids.

constituted with an unusual bicyclo[2.2.2]diazaoctane core ring system. It has been proposed that this key feature may have been formed biosynthetically by an intramolecular Diels–Alder reaction.[9] Our continued interest in the biomimetic total synthesis and biosynthesis of this entire family of prenylated indole alkaloids

Keywords: Biosynthetic Diels–Alder reaction; Bicyclo[2.2.2]diazaoctane.

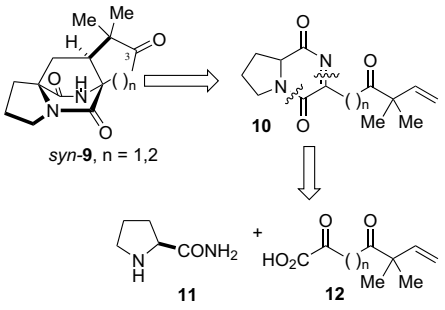
* Corresponding author. Tel.: +1-970-491-6747; fax: +1-970-491-3944; e-mail: rmw@chem.colostate.edu

0040-4039/$ - see front matter © 2004 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tetlet.2004.04.054

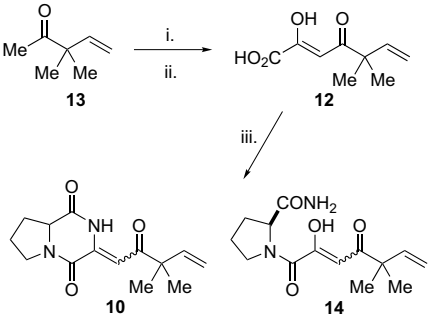
has led us to develop an extremely convergent synthesis of the bicyclo-[2.2.2] core, which we would like to report here.[10]

We envisioned that the unique *spiro*-succinimide structure of asperparaline could be derived from the functionalized tetracyclic compound **9**. It seemed plausible that **9** could be constructed by an intramolecular Diels–Alder reaction from the suitably oxidized diketopiperazine **10** (DKP), which in turn could be derived from prolinamide (**11**) and pyruvate derivatives **12** (Scheme 1).

Since prolinamide (**11**) was commercially available[11] we began our synthesis by constructing pyruvic acid **12**.



Scheme 1.



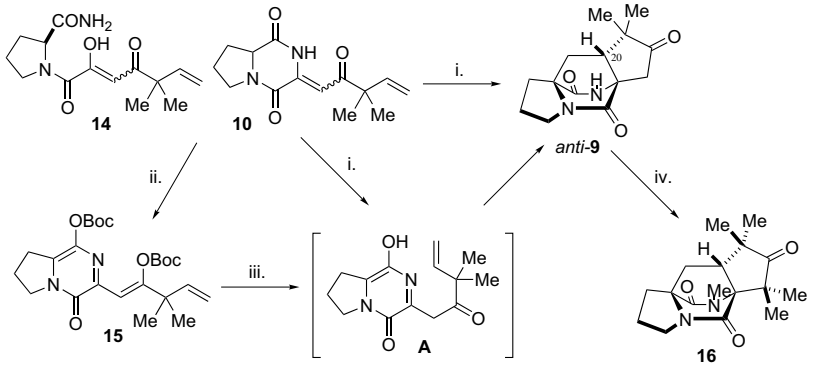
Scheme 2. Reagents and conditions: (i) $(EtO_2C)_2$, NaOH, EtOH, reflux 3 h, 65%; (ii) LiOH, THF, EtOH, H_2O, 87%; (iii) **11**, DCC, CH_2Cl_2, reflux 12 h, 61% based on **10**.

Ketone **13**, formed by the Barbier coupling of acetonitrile with prenyl bromide in the presence of zinc–silver couple,[12,13] was treated with sodium ethoxide and diethyl oxalate, yielding the desired pyruvate ester in 65% yield after silica gel purification. Lithium hydroxide hydrolysis of the ester proceeded smoothly to give the diketo-acid **12** in 87% yield (Scheme 2).

Several conditions were then screened for the synthesis of diketopiperazine **10** by coupling prolinamide **11** with pyruvic acid derivative **12**. The conditions for effecting the desired condensation are shown in Scheme 2.[14] In all cases, the coupling reactions yielded an inseparable mixture of compounds, which were believed to be the uncyclized amide **14** (281 [MH^+, 75%]) and the desired piperazinedione **10** (263 [MH^+, 100%]).

Treatment of the mixture of **10** and **14** with an excess of Boc_2O and 1.1 equiv of DMAP in CH_2Cl_2 at room temperature gave the bis-*O*-Boc-protected enol **15** (Scheme 3). Similar azadienes have been reported to undergo intermolecular Diels–Alder reactions under Brønsted or Lewis acidic conditions.[15] Thus, we treated **15** with 5 equiv of $AlCl_3$ in refluxing EtOAc, which gave the cycloadduct **9** as a single diastereomer in excellent yield.

Given the success of the Lewis acid-catalyzed intramolecular Diels–Alder reaction with the preformed azadiene **15**, we were then interested to see if similar results could be achieved directly with the diketopiperazine **10**. Reaction of the mixture of **10** and **14** with a solution of 4 M HCl in dioxane gave the intramolecular Diels–Alder product **9** in 45% yield (Scheme 3).[16] The remaining products identified were pyruvic acid derivative **12** and prolinamide **11**, therefore indicating that the starting mixture had contained uncyclized enol–amide **14** whose peptide bond had apparently been cleaved under these conditions. Once again the tetracyclic compound **9** had been formed as a single diastereomer and was identified by ^1H NMR to be the same as the $AlCl_3$ reaction product. Presumably both intramolecular Diels–Alder reactions go via the same intermediate (**A**; Scheme 3).



Scheme 3. Reagents and conditions: (i) 4 M HCl/dioxane, 45%; (ii) Boc_2O (2.75 equiv), DMAP (1.1 equiv), CH_2Cl_2, 28%; (iii) $AlCl_3$ (5 equiv), EtOAc, reflux 5 days, 81%; (iv) excess NaH, MeI, THF, reflux 1 h, 62%.

In order to determine, which diastereomer had been formed, that is, the relative configuration at C20, it was necessary to functionalize **9** by methylation of the nitrogen of the secondary amide. The pentamethylated compound **16** was smoothly synthesized from **9**, using excess sodium hydride and methyl iodide. ^{1}H NMR NOE experiments identified the diastereomer from the Diels–Alder reaction as that depicted in Scheme 3, where the C20 proton is *syn* to the proline ring and the cyclopentanone ring is *anti*. This is the opposite configuration at C20 in the asperparaline, paraherquamide, and stephacidin class of alkaloids, but is the same relative configuration as that found in the brevianamides (Fig. 1).

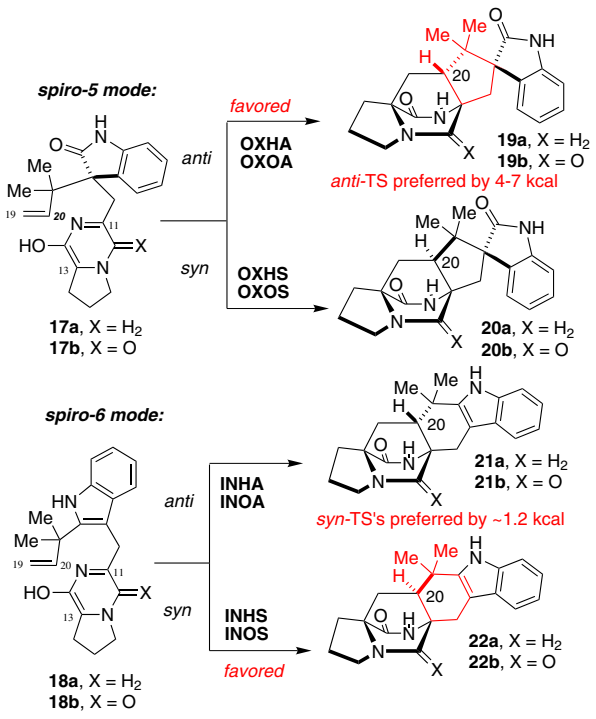
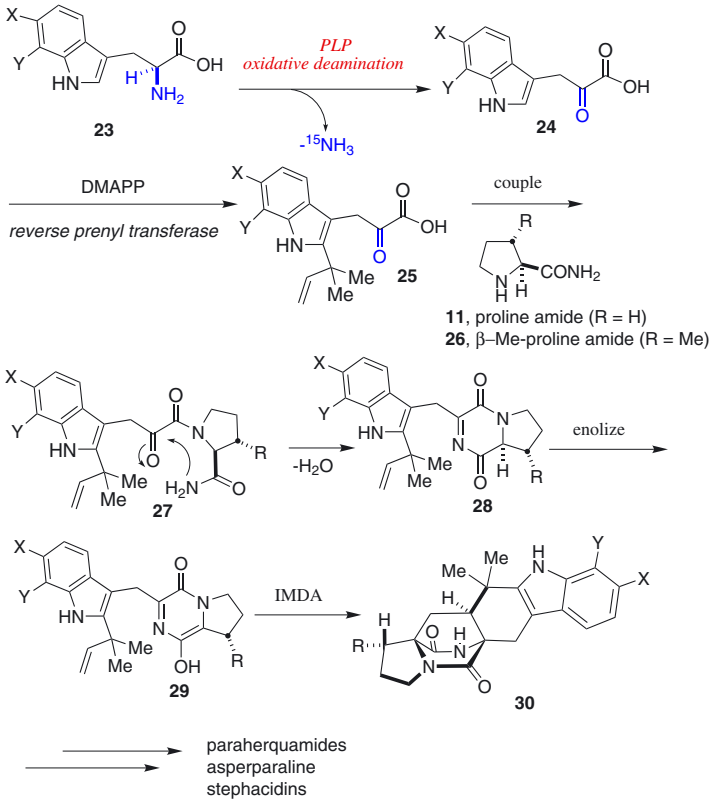


Figure 2. Ab initio calculation on the *spiro*-5 and *spiro*-6 modes of IMDA cycloaddition.[18]

These experimental results support the findings of our previously reported theoretical studies on the intramolecular Diels–Alder cycloaddition as related to the biosynthesis of the brevianamides,[17] paraherquamide A and VM99955 (Fig. 2).[18] Formation of the *anti-spiro*-5 systems (**19**, Fig. 2) was calculated to be favored by ~4–7 kcal/mol whereas the *spiro*-6 systems (**22**) were found to favor formation of the *syn-spiro* ring systems by ~1–2 kcal/mol.[19] Therefore, these constraints applied to the intramolecular cycloaddition reaction directed toward five-membered rings were found to be responsible for the observed large *anti* selectivity in the biosynthesis of brevianamides and hence can explain the observed *anti* selectivity at C20 of the Diels–Alder product **9**, from our results. It is interesting to note that we have experimentally observed a 2 ~ 2.5:1 *syn:anti* ratio of products (**22b**:**21b**) in the intramolecular Diels–Alder reactions of **18b** (and the corresponding β-methylproline-containing substrate) where a *spiro*-six-membered ring was constructed as directed toward the total synthesis of VM55599 and brevianamide B.[19–21]



Scheme 4. A new biogenetic hypothesis.

These results suggest both a reasonable synthetic approach to the asperparalines and stephacidins as well as a possible biogenetic construction as shown in Scheme 4. Thus, oxidative deamination of tryptophan or a reverse-prenylated tryptophan derivative by (for example) a PLP-dependent deaminase yields α-ketoacid **25**. Condensation with the appropriate proline amide derivative would result in a potentially spontaneous cascade of cyclodehydration, tautomerization and IMDA to afford the key hexacyclic substances **30**. Downstream oxidations, prenylations and final fashioning of each natural product's functional group configurations would lead to the paraherquamides, asperparalines, and stephacidins (through avrainvillamide).

We have demonstrated a very concise and convergent synthesis of a functionalized bicyclo[2.2.2]diazaoctane ring system. Current efforts in our group are concentrated on applying this synthetic strategy to the *spiro*-six system **10**, where $n = 2$. Both theoretical as well as experimental data support the notion that these IMDA cycloadditions should be *syn*-selective. Efforts to deploy this strategy to the total synthesis of the asperparalines and stephacidins, as well as related members of the paraherquamide family are under study in these laboratories.

Acknowledgements

This work was supported by the National Institutes of Health (Grant CA70375).

References and notes

1. Hayashi, H.; Nishimoto, Y.; Nozaki, H. *Tetrahedron Lett.* **1997**, *38*, 5655–5658.
2. Hayashi, H.; Nishimoto, Y.; Akiyama, K.; Nozaki, H. *Biosci. Biotechnol. Biochem.* **2000**, *64*, 111–115.
3. Banks, R. M.; Blanchflower, S. E.; Everett, J. R.; Manger, B. R.; Reading, C. *J. Antibiot.* **1997**, *50*, 840–846.
4. (a) Yamazaki, M.; Okuyama, E.; Kobayashi, M.; Inoue, H. *Tetrahedron Lett.* **1981**, *22*, 135–136; (b) Ondeyka, J. G.; Goegelman, R. T.; Schaeffer, J. M.; Kelemen, L.; Zitano, L. *J. Antibiot.* **1990**, *43*, 1375–1379; (c) Liesch, J. M.; Wichmann, C. F. *J. Antibiot.* **1990**, *43*, 1380–1386; (d) Blanchflower, S. E.; Banks, R. M.; Everett, J. R.; Manger, B. R.; Reading, C. *J. Antibiot.* **1991**, *44*, 492–497; (e) Shoop, W. L.; Haines, H. W.; Eary, C. H.; Michael, B. F. *Am. J. Vet. Res.* **1992**, *53*, 2032; (f) Schaeffer, J. M.; Blizzard, T. A.; Ondeyka, J. G.; Goegelman, R. T.; Sinclair, P. J.; Mrozik, H. *Biochem. Pharmacol.* **1992**, *43*, 679–684.
5. Guyatt, H. L.; Brooker, S.; Kihamia, C. M.; Hall, A.; Bundy, D. A. P. *Bull. World Health Organisation* **2001**, *79*, 695–703.
6. (a) Fenical, W.; Jensen, P.; Cheng, X. C. U.S. Patent 6,066,635, 2000; (b) Sugie, Y.; Hirai, H.; Inagaki, T.; Ishiguro, M.; Kim, Y.-J.; Kojima, Y.; Sakakibara, T.; Sakemi, S.; Sugiura, A.; Suzuki, Y.; Brennan, L.; Duignan, J.; Huang, L.; Sutcliffe, J.; Kojima, N. *J. Antibiot.* **2001**, *54*, 911–916.
7. Qian-Cutrone, J.; Krampitz, K.; Shu, Y.-Z.; Chang, L. P. U.S. Patent 6,291,461, 2001.
8. Qian-Cutrone, J.; Huang, S.; Shu, Y.-Z.; Vyas, D.; Fairchild, C.; Menendez, A.; Krampitz, K.; Dalterio, R.; Klohr, S. E.; Gao, Q. *J. Am. Chem. Soc.* **2002**, *124*, 14556–14557.
9. (a) Porter, A. E. A.; Sammes, P. G. *J. Chem. Soc., Chem. Commun.* **1970**, 1103; (b) Williams, R. M.; Kwast, E.; Coffman, H.; Glinka, T. *J. Am. Chem. Soc.* **1989**, *111*, 3064–3065; (c) Williams, R. M.; Glinka, T.; Kwast, E.; Coffman, H.; Stille, J. K. *J. Am. Chem. Soc.* **1990**, *112*, 808–821; (d) Stocking, E. M.; Williams, R. M. *Angew. Chem., Int. Ed.* **2003**, *42*, 3078–3115.
10. (a) Gray, C. R.; Sanz-Cervera, J. F.; Silks, L. A.; Williams, R. M. *J. Am. Chem. Soc.* **2003**, *125*, 14692–14693; for a review, see (b) Williams, R. M.; Cox, R. J. *Acc. Chem. Res.* **2003**, *36*, 127–139.
11. Alternatively prolinamide **11** was prepared according to: Vendeville, S.; Goossens, F.; Debreu-Fontaine, M.-A.; Landry, V.; Davioud-Charvet, E.; Grellier, P.; Scharpe, S.; Sergheraert, C. *Bioorg. Med. Chem.* **2002**, *10*, 1719–1729.
12. Rousseau, G.; Conia, J. M. *Tetrahedron Lett.* **1981**, *22*, 649–652.
13. Sanz-Cervera, J. F.; Glinka, T.; Williams, R. M. *Tetrahedron* **1993**, *49*, 8471–8482.
14. 3-(3,3-Dimethyl-2-oxopent-4-enylidene)-hexahydropyrrolo-[1,2-*a*]pyrazine-1,4-dione (**10**) and (*S*)-1-(2-hydroxy-5,5-dimethyl-4-oxohepta-2,6-dienoyl)pyrrolidine-2-carboxamide (**14**). Under argon; to a solution of prolinamide **11** (1.12 g, 9.81 mmol) in anhydrous CH_2Cl_2 (20 mL) was added acid **12** (1.99 g, 10.8 mmol) followed by 1,3-dicyclohexylcarbodiimide (3.04 g, 14.7 mmol). The cloudy yellow solution was heated at reflux for 12 h, before it was allowed to cool to room temperature; TLC (5:95 MeOH/CH_2Cl_2) showed 1,3-dicyclohexylcarbodiimide (R_f 0.26) and the mixture of diketopiperazine **10** and amide **14** (R_f 0.17 and 0.14); all three compounds were potassium permanganate active. Solvent was evaporated under reduced pressure and this crude mixture was purified by column chromatography on silica using a gradient of MeOH/CH_2Cl_2 (5:95 MeOH/CH_2Cl_2 to 20:80 MeOH/CH_2Cl_2) as eluent to give the mixture of diketopiperazine **10** and amide **14** (1.57 g, 61%) as an orange solid. Major peaks identified. ^{1}H NMR (300 MHz, $CDCl_3$) 1.26 (6H, s), 1.82–2.28 (4H, m), 3.38–3.90 (2H, m), 4.54 (1H, dd, $J = 7.4$, 3.3 Hz), 5.12 (1H, d, $J = 17.4$ Hz), 5.12 (1H, d, $J = 11.1$ Hz), 5.90 (1H, dd, $J = 17.4$, 10.6 Hz), 6.75 (1H, br s). IR (NaCl, neat): 2976, 2934, 1681, 1646, 1414 cm^{-1}. MS: m/z (FAB) 281 (**14**, MH$^+$, 75%), 263 (**10**, MH$^+$, 100%).
15. (a) Sanz-Cervera, J. F.; Williams, R. M.; Marco, J. A.; Lopez-Sanchez, J. M.; Gonzalez, F.; Martinez, M. E.; Sancenon, F. *Tetrahedron* **2000**, *56*, 6345–6358; (b) Jin, S.; Wessig, P.; Liebscher, J. *Eur. J. Org. Chem.* **2000**, 1993–1999; (c) Jin, S.; Wessig, P.; Liebscher, J. *J. Org. Chem.* **2001**, *66*, 3984–3997.
16. 21,21-Dimethyl-31,32-diazatetracyclo[5.5.2.0.0]tetradec-3,12,18-trione (**9**). The diketopiperazine **10**/amide **14** mixture (412 mg, 1.57 mmol) was dissolved in a solution of 4 M HCl in dioxane (35 mL). The bright yellow solution was stirred at room temperature for 20 h; TLC (EtOAc) showed the tetracycle **9** (R_f 0.22, potassium permanganate active). Solvent was evaporated under reduced pressure and this crude mixture was purified by column chromatography on silica using 5:95 MeOH/CH_2Cl_2 as eluent to give the desired tetracycle **9** (187 mg, 45%) as a colorless oil. ^{1}H NMR (300 MHz, $CDCl_3$) 1.02 (3H, s), 1.10 (3H, s),

1.81–2.16 (5H, m), 2.38 (1H, dd, $J = 10.3$, 6.6 Hz), 2.62 (1H, d, $J = 19.2$ Hz), 2.74 (1H, dt, $J = 13.0$, 6.5 Hz), 3.15 (1H, d, $J = 19.2$ Hz), 3.48 (2H, ddd, $J = 6.7$, 6.7, 1.8 Hz), 7.44 (1H, br s). ^{13}C NMR: 21.0, 24.7, 25.8, 28.9, 29.5, 40.1, 44.0, 47.2, 50.8, 63.8, 68.5, 168.4, 173.2, 218.1. IR (NaCl, neat): 1745, 1693 cm^{-1}. HRMS (FAB+): Calcd for $C_{14}H_{19}N_2O_3$: 263.1396. Found: 263.1405 (MH^+).

17. Domingo, L. R.; Sanz-Cervera, J. F.; Williams, R. M.; Picher, M. T.; Marco, J. A. *J. Org. Chem.* **1997**, *62*, 1662–1667.
18. Domingo, L. R.; Zaragoza, R. J.; Williams, R. M. *J. Org. Chem.* **2003**, *68*, 2895–2902.
19. Williams, R. M.; Sanz-Cervera, J. F.; Sancenon, F.; Marco, J. A.; Halligan, K. M. *Bioorg. Med. Chem.* **1998**, *6*, 1233–1241.
20. Stocking, E. M.; Sanz-Cervera, J. F.; Williams, R. M. *J. Am. Chem. Soc.* **2000**, *122*, 1675–1683.
21. Sanz-Cervera, J. F.; Williams, R. M. *J. Am. Chem. Soc.* **2002**, *124*, 2556–2559.