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Total synthesis of 3,4-dihydrobenzo[*h*]quinazolin-4-one and structure elucidation of perlolidine and samoquasine A

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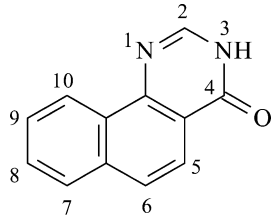
Abstract—The total synthesis of 3,4-dihydrobenzo[*h*]quinazolin-4-one is described. Based on the spectral data and chemical evidence, the structures of 3,4-dihydrobenzo[*h*]quinazolin-4-one, perlolidine, and samoquasine A are not the same. A structure for samoquasine A is suggested.

Benzo[*h*]quinazolines are poorly studied systems because of the absence of accessible and reliable methods of synthesis of such compounds. There are some published routes for the synthesis of 4-substituted 5,6-dihydrobenzo[*h*]quinazolines,[1] 2,3-disubstituted benzo[*h*]quinazolin-4(3*H*)-one,[2] and 1,2-dihydrobenzo[*h*]-quinazolines.[3] The diversity of pharmacological activities [1,4–7] (antidepressive, antitumor, anti-platelet aggregation, etc.) of benzo[*h*]quinazolines show that they are important heterocyclic systems. In 2000, a new cytotoxic alkaloid, samoquasine A, was isolated from the seeds of *Annona squamosa*.[8] This was the first example of a benzo[*h*]quinazoline alkaloid from a plant belonging to the Annonaceae. In previous work, including our studies,[9,10] Annonaceous plants have yielded many alkaloids but no compound's structure was related to the proposed structure for samoquasine A. The cytotoxic activity of samoquasine A prompted us to synthesize it and some analogues.



The modified methods[11,12] to synthesize 1-amino-2-naphthoic acid **5** were used (Scheme 1). 1-Nitronaphthalene **1** (0.03 mol) was added to a solution of ethyl cyanoacetate (0.09 mol), KOH (0.06 mol), and KCN (0.03 mol) in DMF (90 mL), then the mixture was stirred for 30 h at 55°C. After the solvent had been removed under vacuum, the residue was dissolved in ethanol (75 mL), and 20% aqueous NaOH solution (25 mL) was added. The mixture was heated to reflux for 10 h. After the ethanol was removed under reduced pressure, the aqueous solution was brought to pH 6.5 with aqueous concentrated acetic acid. The solution was partitioned with $CHCl_3$. The $CHCl_3$ extract obtained was purified by silica gel chromatography (*n*-hexane/$CHCl_3$ = 1:3) to provide 1-amino-2-naphthoic acid **5** (46%).[13] A microwave-assisted Niementowski reaction was used to prepare the final compound **6** from **5**. A mixture of 1-amino-2-naphthoic acid **5** (0.2 mmol) and formamide (1 mmol) was irradiated in a domestic microwave oven for 10 min (580 W). After cooling, the crude reaction product was purified by SPE (LiChrolut, RP-18, 40–63 μm, 500 mg, 6 mL, Merck) to afford **6** (74%). Acetamide and propionamide were used in analogous reactions to afford compounds **7** and **8**.

Compound **6** was obtained as a yellow powder, EIMS *m*/*z* 196 $[M]^+$, mp 244–245°C. The ^1H NMR (400 MHz) and ^{13}C NMR (100 MHz) data are given in Table 1. Using 2D NMR (^1H–^1H COSY, HMBC, HMQC, ROESY) analysis, the structure was unambiguously assigned as 3,4-dihydrobenzo[*h*]quinazolin-4-one. The structures of **7** (2-methyl-3*H*-benzo[*h*]-quinazolin-4-one) and **8** (2-ethyl-3*H*-benzo[*h*]-quinazolin-4-one) were similarly confirmed.[2]

However, the physical and spectra data of **6** were incompatible with those reported for samoquasine A.[8] There were some differences in the NMR data (Table

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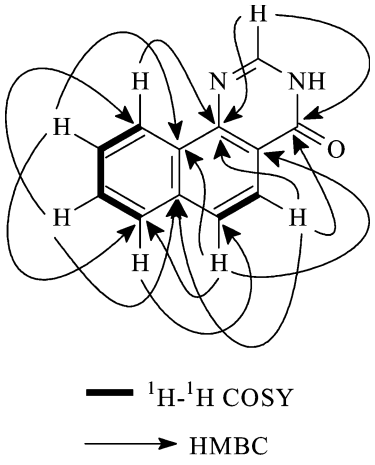
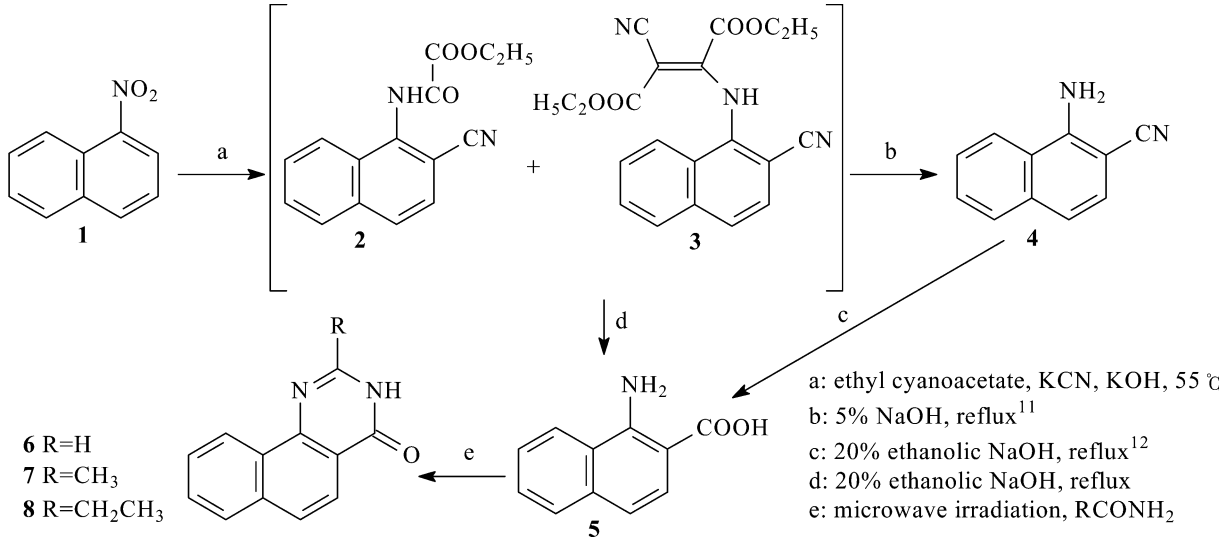
1). The downfield singlet for H-2 is significantly different.

In the present study on the stems of *A. squamosa*, we also isolated a diazaphenanthrene derivative.[14] It's physical and spectral data were similar to those of samoquasine A. The diazaphenanthrene had a molecular formula of $C_{12}H_8N_2O$ as revealed by HREIMS (m/z 196.0634, $[M]^+$, calcd 196.0636). ^1H and ^{13}C NMR data indicated that the molecule has one conjugated carbonyl and 11 olefinic carbons. On analysis by 2D NMR (COSY, ROESY, HMQC, HMBC), this compound was shown to be 2,9-diazaphenanthren-1(2*H*)-one. The EIMS peaks (m/z 196 $[M]^+$, 168 $[M-CO]^+$, 140 $[M-H_2CN]^+$) also confirmed the structure. To our knowledge, 2,9-diazaphenanthren-1(2*H*)-one (perlolidine) has not been isolated from any plants other than those of the Poaceae family, and no detailed NMR data were found.[15,16] The NMR data of perlolidine cannot be



2D NMR correlations of **6**

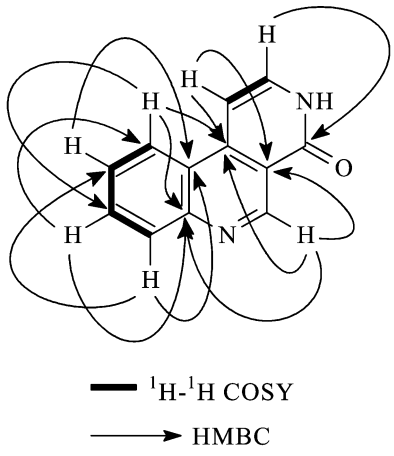


Scheme 1.

Table 1. NMR data of samoquasine A and **6**

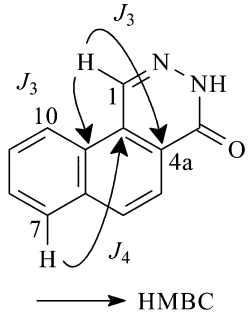
Position	Samoquasine A in $CDCl_3$		**6** in $CDCl_3$	**6** in d_6-DMSO	
	δ_H (J, Hz)[8]	δ_C[8]	δ_H (J, Hz)	δ_H (J, Hz)	δ_C
2	9.60 s	150.5	8.29 s	8.36 s	146.3
4	–	164.0	–	–	160.9
4a	–	118.7	–	–	119.2
5	7.55 d (7.3)	136.0	8.23 d (8.8)	8.06 d (8.8)	121.4
6	7.30 d (7.3)	101.6	7.91 d (8.8)	7.94 d (8.8)	126.9
6a	–	123.6	–	–	135.8
7	8.40 dd (1.0, 8.3)	125.0	7.95 dd (1.8, 7.5)	8.04 dd (1.8, 7.5)	128.2
8	7.71 dt (1.0, 8.3)	128.7	7.72 td (1.8, 7.5)	7.72 td (1.8, 7.5)	129.4
9	7.87 dt (1.0, 8.3)	132.8	7.75 td (1.8, 7.5)	7.77 td (1.8, 7.5)	127.3
10	8.12 dd (1.0, 8.3)	130.4	9.01 dd (1.8, 7.5)	8.90 dd (1.8, 7.5)	124.8
10a	–	148.5	–	–	129.6
10b	–	144.3	–	–	147.3
NH	–	–	10.54 br s	13.20 br s	–

measured in $CDCl_3$ because of its poor solubility, so that we cannot be sure whether perlolidine and samoquasine A are the same or not.



2D NMR correlations of perlolidine

There are currently 229 substances listed in the Beilstein Commander database with the molecular formula $C_{12}H_8N_2O$. But none of those can be considered as a possible structure for samoquasine A. Based on the NMR data and chemical evidence,[8] we suggest moving the nitrogen from position 1 to 2. Thus, samoquasine A may contain a naphthalene ring system and a pyridazinone ring. This structure could explain the downfield singlet signal at δ 9.60 (deshielded by the aromatic ring). The J_3 long-range correlations between H-1/C-4a and H-1/C-10a and the absence of a long-range correlation between H-1/C-4 are also consistent with the suggested structure for samoquasine A.



HMBC correlations of proposed structure for samoquasine A

In summary, a convenient method to synthesize 3,4-dihydrobenzo[*h*]quinazolin-4-one **6** and its analogues **7** and **8** was developed. This method could be used to prepare other 2,3-disubstituted benzo[*h*]quinazolin-4-ones to investigate their biological activities. Perlolidine has been isolated from the stems of *A. squamosa*. It is the first time that perlolidine has been isolated from a plant of a non-Poaceae family. Perlolidine exhibited mild cytotoxicity against NUGC and HONE-1 cancer cell lines in vitro (inhibitory ratio 63% and 62% at 50 μg/mL, respectively). The structures of **6** and perlolidine were confirmed by NMR data clearly and unambiguously. Compound **6**, perlolidine, and samoquasine A are definitely not the same. The possible structure of samoquasine A is 3,4-dihydrobenzo[*f*]phthalazin-4-one.

Acknowledgements

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References

1. Hirota, T.; Sasaki, K.; Ohtomo, H.; Uehara, A.; Nakayama, T. *Heterocycles* **1990**, *31*, 153–161.
2. Padmaja, J.; Satyanarayana Reddy, M.; Ratnam, C. V. *Indian. J. Chem. Sec. B* **1986**, *25*, 712–715.
3. Okada, E.; Masuda, R.; Hojo, M.; Tone, H.; Gotoh, N.; Huang, T. K. *Heterocycles* **1995**, *40*, 905–911.
4. *Fused Pyrimidines*, *Part 1: Quinazolines*, Armarego, W. L. F., Ed.; Wiley-Interscience: New York, 1967
5. Armarego, W. L. F. *Adv. Heterocycl. Chem.* **1979**, *24*, 1–62.
6. Parrot-Lopez, H.; Delacotte, J. M.; Renault, J. *J. Heterocycl. Chem.* **1986**, *23*, 1039–1043.
7. Askew, B. M. *Life Sci.* **1963**, *2*, 725–730.
8. Morita, H.; Sato, Y.; Chan, K. L.; Choo, C. Y.; Itokawa, H.; Takeya, K.; Kobayashi, J. *J. Nat. Prod.* **2000**, *63*, 1707–1708.
9. Leboeuf, M.; Cave, A.; Bhaumik, p. K.; Mukherjee, B.; Mukherjee, R. *Phytochemistry* **1982**, *21*, 2783–2813.
10. Kuo, R. Y.; Chang, F. R.; Wu, Y. C. *The Chinese Pharma. J.* **2002**, *54*, 155–173.
11. Tomioka, Y.; Ohkubo, K.; Yamazaki, M. *Chem. Pharm. Bull.* **1985**, *33*, 1360–1366.
12. Zhang, W.; Koehler, K. F.; Harris, B.; Skolnick, P.; Cook, J. M. *J. Med. Chem.* **1994**, *37*, 745–757.
13. 1-Amino-2-naphthoic acid **5**: mp 195–196°C; EIMS *m*/*z* 187 $[M]^+$; ^{1}H NMR (400 MHz, CD_3OD) δ: 7.04 (d, J=9.0 Hz, 1H), 7.44 (td, J=1.6, 8.4 Hz, 1H), 7.50 (td, J=1.6, 8.4 Hz, 1H), 7.56 (d, J=9.0 Hz, 1H), 7.70 (dd, J=1.6, 8.4 Hz, 1H), 8.12 (dd, J=1.6, 8.4 Hz, 1H). ^{13}C NMR (100 MHz, CD_3OD) δ: 107.4 (s), 116.4 (d), 123.6 (d), 125.5 (s), 126.1 (d), 126.2 (d), 128.9 (d), 129.1 (d), 137.3 (s), 149.5 (s), 175.2 (s).
14. Extraction and isolation of perlolidine: Fresh stems of *A. squamosa* (15 kg), collected from Pingtung, Taiwan, in May 2000, were extracted repeatedly with MeOH at room temperature. The combined MeOH extract was evaporated under reduced pressure to yield a dark-brown syrup (550 g). The syrup was partitioned between $CHCl_3$ and H_2O to give two layers. The $CHCl_3$ layers was extracted with 3% HCl to get a $CHCl_3$ solution and an acidic aq. layer. The aq. layer was basified with NH_4OH and extracted with $CHCl_3$ to give the crude alkaloid (2.1

g), which was subjected to silica gel chromatography, eluting with increasingly polar mixtures of $CHCl_3$–MeOH to afford 5 fractions. Fraction 2 was separated by silica gel chromatography and further purified by recrystallization to give perlolidine (5 mg): mp 280–282°C; EI MS m/z 196 $[M]^+$, 168 $[M-CO]^+$, 140 $[M-H_2CN]^+$; IR (KBr) cm^{-1} 3420, 1650; ^1H NMR (400 MHz, d_5-pyridine) δ: 7.32 (d, $J=7.2$ Hz, H-4), 7.65 (dd, $J=1.6$, 7.2 Hz, H-6), 7.84 (d, $J=7.2$ Hz, H-3), 7.85 (dd, $J=1.6$, 7.2 Hz, H-7), 8.40 (dd, $J=1.6$, 7.2 Hz, H-8), 8.48 (dd, $J=1.6$, 7.2 Hz, H-5), 10.21 (s, H-10), 13.05 (br s, NH). ^{13}C NMR (100 MHz, d_5-pyridine) δ: 118.6 (C-10a), 123.0 (C-4b), 123.7 (C-4), 124.6 (C-5), 127.3 (C-6), 130.6 (C-8), 131.2 (C-7), 135.9 (C-3), 142.8 (C-4a), 148.8 (C-8a), 150.4 (C-10), 162.8 (C-1).

15. Reifer, I.; White, E. P. *New Zealand J. Sci. Technol.* **1945**, *27B*, 242 and references therein.
16. Rocca, P.; Cochennec, C.; Marsais, F.; Thomas-dit-Dumont, L.; Mallet, M.; Godard, A.; Quéguiner, G. *J. Org. Chem.* **1993**, *58*, 7832–7838.