# SIMPLE SYNTHESIS OF A TERPENOPHENOL—CHLORIN CONJUGATE WITH AN AMIDE BOND

**E. V. Buravlev*, I. Yu. Chukicheva, D. V. Belykh, and A. V. Kuchin**

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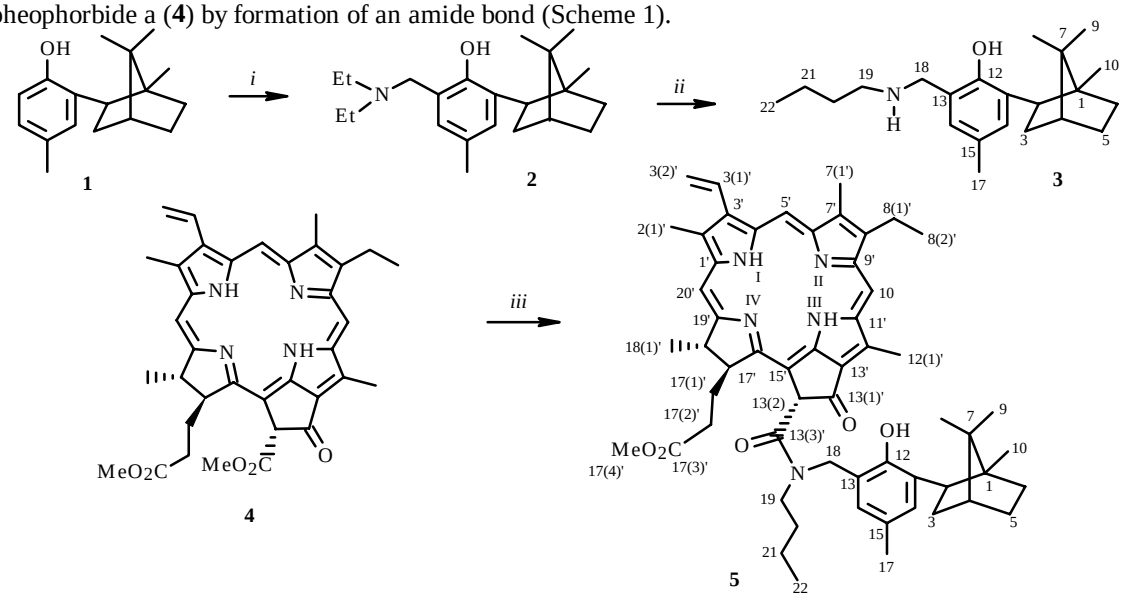
*A terpenophenol with a butylaminomethyl group was synthesized and conjugated to a chlorin macrocycle through formation of an amide bond without using activating reagents.*

**Key words:** terpenophenols, amines, methylpheophorbide a, aminomethylation, transamination, catalyst-free amidation, conjugate.

The synthesis of compounds with several pharmacophores is of great interest because the combination of several pharmacophores in a single molecule can not only enhance the already known physiological activity but also cause the appearance of new activity (for example, incorporation of a carborane into protohemin IX leads to a compound that is not only active as an agent for boron-neutron-capture therapy but also phototoxic, although the starting protohemin IX does not have this property) [1]. The affinity of chlorophyll derivatives for malignant neoplasms (i.e., the increase of photosensitizer concentration in a tumor above that in the surrounding healthy tissue) makes them effective as antitumor preparations with a different mechanism of action. Incorporation into chlorin of a terpenophenol may lead to more effective antitumor preparations by increasing the capability to interact with cell membranes [2, 3].

We synthesized a terpenophenol with a secondary aminomethyl group (**3**) and conjugated it to the *exo*-ring of methylpheophorbide a (**4**) by formation of an amide bond (Scheme 1).




*i*: $Et_2NH$, HCHO, benzene, reflux, 6 h, yield 92%; *ii*: $BuNH_2$ (excess), reflux, 58 h, yield 47%; *iii*: **3**, toluene, reflux, 6 h, yield 50%

Scheme 1.

Institute of Chemistry, Komi Scientific Center, Urals Division, Russian Academy of Sciences, 167982, Russia, Republic of Komi, Syktyvkar, ul. Pervomaiskaya, 48, fax: +7-(8212) 21 84 77, e-mail: buravlev-ev@chemi.komisc.ru. Translated from Khimiya Prirodnykh Soedinenii, No. 6, pp. 561-563, November-December, 2007. Original article submitted September 6, 2007.

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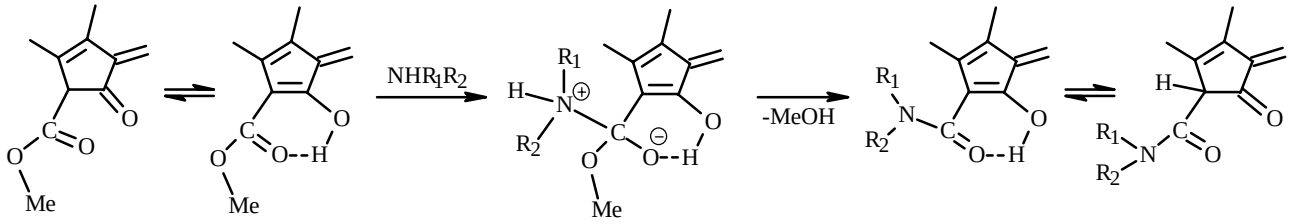
Using primary amines directly in a Mannich reaction with phenols can lead to the formation of dihydroxybenzoxazines and *bis*-[2-hydroxybenzyl]amines [3]. Therefore, **3** was synthesized using aminomethylation [4, 5] of 4-methyl-2-isobornylphenol (**1**) [6] followed by transamination of tertiary amine **2**. Compounds **2** and **3** were identified by NMR spectroscopy. Comparison of the spectra with those reported previously for *o*-isobornylphenol established that the terpenophenol moiety did not change [7]. Resonances in the NMR spectra were assigned based on a comparison of chemical shifts of analogous resonances in the spectra of compounds with similar structural features. The NMR spectra of **2** and **3** contained resonances characteristic of aminomethyl groups [8].

The terpenophenol moiety was introduced onto the periphery of methylpheophorbide a (**4**) via catalyst-free amidation of the 13(2)′-$CO_2$Me group by secondary amine **3**. During the reaction, just the 13(2)′-$CO_2$Me group of **4** was amidated and not the sterically more accessible ester of the 17′-propionate. This was confirmed by PMR spectroscopy. The spectrum of the reaction product **5** lacked a resonance for the protons of the 13(2)′-$CO_2$Me group. The shift to weak field of the resonance of the 13(2)′ proton argues in favor of a reaction involving the 13(2)′ ester. The greater reactivity of the 13(2)′-$CO_2CH_3$ group can be explained by its activation due to formation of a H-bond to the hydroxyl in the enol form of **4** (Scheme 2).




Scheme 2.

Because starting terpenophenol **1** and its aminomethyl derivatives **2** and **3** are mixtures of enantiomers, conjugate **5** is a mixture of diastereomers, the presence of which was apparent as a doubling of the resonances for the phenol protons of the terpenophenol OH group of **5**. Resonances of the other protons of the diastereomers of this compound were the same.

## EXPERIMENTAL

PMR and $^{13}$C NMR spectra in $CDCl_3$ were recorded on a Bruker DRX-400 spectrometer (operating frequency 400 MHz); IR spectra in thin layers and KBr disks, on a Specord M 80 instrument. MALDI mass spectra were obtained in a Vision-2000 spectrometer. Melting points were determined on a Kofler stage. The course of reactions was monitored by TLC on Sorbfil and Silufol plates. Plates were developed for **1**-**3** by treatment with $KMnO_4$ solution (15 g $KMnO_4$, 300 mL $H_2O$, 0.5 mL conc. $H_2SO_4$). Amines were developed using ninhydrin solution (0.5%) in ethanol with added glacial AcOH (3 vol %) followed by heating to 100-120°C.

Benzene and toluene were dried over $CaCl_2$ and distilled over metallic sodium. We used petroleum ether (fraction with bp 65-70°C). Diethylamine, *n*-butylamine, acetone, *i*-PrOH, and $CCl_4$ were freshly distilled. Column chromatography used silica gel (Alfa Aesar, 70/230 μ, wet packed) and chemically pure paraformaldehyde. Compound **1** was synthesized from *p*-cresol and camphene [7]. Methylpheophorbide a (**4**) was prepared from spirulin. The spectral properties of **4** agreed with those reported in the literature [9].

**2-((Diethylamino)methyl)-4-methyl-6-{exo-(1,7,7-trimethylbicyclo[2.2.1]hept-2-yl)}phenol (2).** 4-Methyl-2-isobornylphenol (**1**, 1.50 g, 6.1 mmol) and paraformaldehyde (0.22 g, 7.3 mmol) were dissolved in dry benzene (20 mL) at room temperature, treated with diethylamine (0.8 mL, 7.3 mmol), and refluxed for 6 h. After the reaction was finished, the excess of solvent was removed at reduced pressure. The reaction mixture was separated over a column (silica gel, 70/230 μ, eluent petroleum ether:$Et_2O$ with increasing fraction of the latter) to afford **2** (1.85 g, 92%) as a colorless oil. $C_{22}H_{35}NO$. IR spectrum (thin layer, $cm^{-1}$): 2960, 2884, 1470, 1390 (Me, $CH_2$); 1614 (C=C); 1248 (C–O); 1234 (C–N); 868, 780 (=C–H).

PMR spectrum (400 MHz, $CDCl_3$, δ, ppm, J/Hz): 0.78 (s, 3H, Me-10), 0.83 (s, 3H, Me-9), 0.91 (s, 3H, Me-8), 1.09 (t, 6H, J = 7.2, Me-20, Me-20′), 1.31-1.38 (m, 1H, H-3), 1.50-1.63 (m, 3H, H-4, $CH_2$-5), 1.81-1.88 (m, 2H, $CH_2$-6), 2.13-2.20

(m, 1H, H-3), 2.24 (s, 3H, Me-17), 2.50-2.70 (m, 4H, $CH_2$-19, $CH_2$-19′), 3.28 (t, 1H, J = 9.2, H-2), 3.60 and 3.78 (both d, 1H each, J = 14.0, 13.6, $CH_2$-18), 6.61, 7.01 (both br.d, both 1H, H-14, H-16), 11.40 (br.s, 1H, OH).

$^{13}$C NMR spectrum (100 MHz, $CDCl_3$, δ): 154.96 (C-12), 130.23 (C-11), 127.57 (C-14), 126.39 (C-15), 126.22 (C-16), 121.03 (C-13), 56.96 (C-18), 49.78 (C-1), 47.89 (C-7), 45.95 (C-19, C-19′), 45.76 (C-4), 44.67 (C-2), 39.64 (C-6), 33.81 (C-3), 27.57 (C-5), 21.51 (C-17), 20.97 (C-8), 20.36 (C-9), 12.10 (C-10), 11.04 (C-20, C-20′).

**2-((Butylamino)methyl)-4-methyl-6-{exo-(1,7,7-trimethylbicyclo[2.2.1]hept-2-yl)}phenol (3).** Amine **2** (1.0 g, 3.0 mmol) was refluxed in an excess of *n*-butylamine (1.5 mL, 15.0 mmol) for 48 h. After the reaction was finished, *n*-butylamine and diethylamine were removed at reduced pressure at 75°C. The reaction mixture was separated over a column (silica gel 70/230 μ, eluent petroleum ether:$Et_2O$:*i*-PrOH with increasing fraction of $Et_2O$) to afford **3** (0.47 g, 47%) as a yellow oil. Cald. for $C_{22}H_{35}NO$, $[M]^+$ 329.272; found, 329.277. IR spectrum (thin layer, $cm^{-1}$): 3328 (N–H); 2964, 2936, 2884, 1472, 1376 (Me, $CH_2$); 1614 (C=C); 1282 (C–N); 1232 (C–O), 866, 786 (=C–H).

PMR spectrum (400 MHz, $CDCl_3$, δ, ppm, J/Hz): 0.79 (s, 3H, Me-10), 0.84 (s, 3H, Me-9), 0.91 (s, 3H, Me-8), 0.92 (t, 3H, J = 7.2, Me-22), 1.31-1.40 (m, 3H, $CH_2$-21, H-3), 1.48-1.64 (m, 5H, H-4, $CH_2$-5, $CH_2$-20), 1.79-1.89 (m, 2H, $CH_2$-6), 2.11-2.20 (m, 1H, H-3), 2.24 (s, 3H, Me-17), 2.65 (t, 2H, J = 7.0, $CH_2$-19), 3.30 (t, 1H, J = 9.0, H-2), 3.93 (s, 2H, $CH_2$-18), 6.64, 7.02 (both d, 1H each, J = 1.2, 1.6, H-14, H-16), 12.02 (br.s, 2H, NH+OH).

$^{13}$C NMR spectrum (100 MHz, $CDCl_3$, δ): 12.18 (C-10), 13.92 (C-22), 20.35 (C-9), 20.90 (C-8), 21.48 (C-17), 27.55 (C-5), 29.67 (C-21), 31.74 (C-20), 33.94 (C-3), 39.66 (C-6), 44.69 (C-2), 45.77 (C-4), 47.88 (C-7), 48.28 (C-19), 49.76 (C-1), 52.88 (C-18), 121.50 (C-16), 126.09 (C-13), 126.45 (C-14), 127.73 (C-15), 130.65 (C-11), 154.05 (C-12).

**Methylpheophorbide a 13(2)-*N*-Butyl-*N*-(2-hydroxy-3-(exo-1,7,7-trimethylbicyclo[2.2.1]hept-2-yl)-5-methylbenzyl)amide (5).** Methylpheophorbide a (**4**, 0.10 g, 0.16 mmol) was dissolved in dry toluene (5 mL), treated with amine **3** (0.11 g, 0.33 mmol), and refluxed for 6 h. After the reaction was finished, the excess of solvent was removed at reduced pressure. The reaction mixture was separated over a column (silica gel 70/230 μ, eluent $CCl_4$:acetone with increasing fraction of the latter) to afford **5** (0.75 g, 50%) as dark green crystals, mp 107-109°C. Cald. for $C_{57}H_{69}N_5O_5$, $[MH]^+$ 904.53; found, 905.576. IR spectrum (KBr, $cm^{-1}$): 1624 (chlorin band), 1742 (ester C=O), 1706 (ketone C=O in the exocycle).

PMR spectrum (400 MHz, $CDCl_3$, δ, ppm, J/Hz): Aminomethylterpenophenol fragment: 0.65 (s, 3H, Me-10), 0.76 (s, 3H, Me-9), 0.87-0.89 (m, 3H, Me-22), 0.89 (s, 3H, Me-8), 1.44-2.46 (m, 10H, $CH_2$-3, $CH_2$-5, $CH_2$-20, $CH_2$-21, and $CH_2$-6), 2.36 (s, 3H, Me-17), 2.50-2.60 (m, 2H, $CH_2$-19), 3.30 (t, 1H, J = 8, H-2), 3.60 (m, 1H, H-4), 3.57-3.64 (m, 2H, $CH_2$-18), 5.15-5.18 (br.s, 1H, 12-C<u>OH</u>) (two diasteromers, 1:1), 6.95 and 7.16 (both s, 1H each, H-14, H-16); chlorin fragment: -1.65 (br.s, 1H, NH-III), 0.51 (br.s, 1H, NH-I), 1.64 [d, 3H, J = 7.2, Me-18(1)′], 1.69 [t, 3H, J = 8.0, Me-8(2)′], 2.04-2.47 [m, 4H, $CH_2$-17(1)′ and $CH_2$-17(2)′], 3.20 [s, 3H, Me-7(1)′], 3.37 [s, 3H, Me-2(1)′], 3.45 [s, 3H, Me-17(4)′], 3.62-3.71 [m, 2H, $CH_2$-8(1)′], 3.65 [s, 3H, Me-12(1)′], 4.96-4.46 (m, 2H, H-17′ and H-18′), 6.15 [dd, 1H, H(*cis*)-3(2)′, J = 11.6 and 1.2], 6.26 [dd, 1H, H(*trans*)-3(2)′, J = 18.0 and 1.2], 6.66 [s, 1H, H-13(2)′], 7.96 [dd, 1H, H-3(1)′, J = 17.6 and 11.6], 8.50 (s, 1H, H-20′), 9.33 (s, 1H, H-5′), 9.47 (s, 1H, H-10′).

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## REFERENCES

1. V. A. Ol′shevskaya, R. G. Nikitina, A. V. Zaitsev, M. A. Gyul′malieva, V. N. Luzgina, E. G. Kanonova, T. G. Morozova, V. V. Drozhzhina, M. L. Kaplan, V. N. Kalinin, and A. A. Shtil′, *Dokl. Akad. Nauk*, **399**, No. 6, 783 (2004).
2. A. F. Mironov, *Ross. Khim. Zh.*, **5**, 23 (1998).
3. A. F. Mironov, *SPIE Proceedings CIS Selected Papers "Laser use in oncology,"* Vol. **2728**, 150 (1996).
4. M. Tramontini, *Synthesis*, **12**, 703 (1973).
5. K.-W. Chi, Y. S. Ahn, K. T. Shim, T. H. Park, and J. S. Ahn, *Bull. Korean Chem. Soc.*, **20**, 973 (1990).

6. I. Yu. Chukicheva and A. V. Kuchin, *Ross. Khim. Zh.*, **3**, 21 (2004).
7. I. Yu. Chukicheva, A. V. Kuchin, L. V. Spirikhin, O. Ya. Borbulevich, A. V. Churakov, and A. I. Belokon′, *Khimiya i Komp′uternoe Modelirovanie, Butlerovskie Soobshcheniya*, No. 1, 9 (2003).
8. I. Yu. Chukicheva, E. V. Buravlev, L. V. Spirikhin, A. V. Churakov, and A. V. Kuchin, *Izv. Akad. Nauk, Ser. Khim.*, No. 10, 1754 (2006).
9. L. A. Tulaeva, D. V. Belykh, N. M. Yakovleva, I. A. Sel′kova, A. V. Rocheva, and A. V. Kuchin, *Izv. Vyssh. Uchebn. Zaved., Khim. Khim. Tekhnol.*, **49**, No. 4, 82 (2006).