Attempted Oxidative Deamination of *N*-Deacetylcolchicinoids with 3,5-Di(*tert*-butyl)-1,2-benzoquinone: Synthesis of 2*H*-1,4-Benzoxazine-Type Adducts

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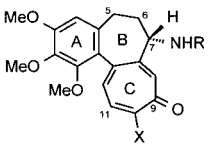
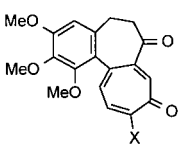
In an attempt to use 3,5-di(*tert*-butyl)-1,2-benzoquinone for the oxidative deamination of *N*-deacetylcolchicine (**4**) and *N*-deacetylthiocolchicine (= *N*-deacetyl-10-demethoxy-10-(methylthio)colchicine; **5**) to give the corresponding ketones **2** and **3**, the 2*H*-1,4-benzoxazine-type adducts **8**/**9** and **11**/**12**, respectively, were formed instead, showing a new and unexpected behavior of *Corey*'s reagent. The adducts were separated and spectroscopically characterized, and a plausible scheme of formation is reported.

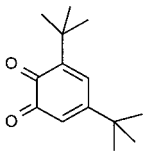
Introduction. – Colchicine (=(*S*)-*N*-(5,6,7,9-tetrahydro-1,2,3,10-tetramethoxy-9-oxobenzo[*a*]heptalen-7-yl)acetamide; **1**), one of the most important non-indole alkaloids, possesses a notable antimitotic activity mainly applied for the treatment of acute gout, amyloidosis of familial Mediterranean fever, and liver cirrhosis [1]. Moreover, colchicine shows a potent antileukemic activity *in vitro*, but its extreme toxicity is a major obstacle in the effective clinical use for the treatment of neoplastic diseases [2]. Therefore, the modification of the molecular structure of colchicine is an active area of research, spurred by the possibility to improve the pharmaceutical profile of the natural product by synthesizing less toxic and therapeutically more favorable analogues and to unravel the complex structure-activity relationships.



1 R = COMe , X = MeO
4 R = H , X = MeO
5 R = H , X = MeS
7 R = CHO , X = MeO
10 R = COMe , X = MeS



2 X=MeO
3 X=MeS



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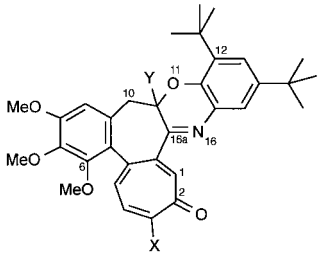

8 X = MeO, Y = H
9 X = MeO, Y = OH
11 X = MeS, Y = H
12 X = MeS, Y = OH

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In the frame of a systematic study on the pharmacological properties of new and known colchicinoids, we intended to investigate extensively the biological properties of the non-N-containing compounds 7-deacetamido-7-oxocolchicine (**2**), a metabolite of colchicine [3], and 7-deacetamido-7-oxothiocolchicine (**3**), the so-called colchicone and thiocolchicone, respectively (thiocolchicine = 10-demethoxy-10-(methylthio)colchicine). Both compounds are readily prepared in excellent yield by oxidative deamination of *N*-deacetylcolchicine (**4**) [4] and *N*-deacetylthiocolchicine (**5**) [5], respectively, with the *Rapoport* reagent (4-formyl-1-methylpyridinium benzenesulfonate) [6], following the protocol developed by *Banwell et al.* [4]. During our study, a literature search revealed a limited number of synthetic reagents for the conversion of primary amines to carbonyl compounds (for a leading reference, see [7]). Among these, the readily available 3,5-di(*tert*-butyl)-1,2-benzoquinone (DTBQ; **6**) developed by *Corey* and *Achiva* [8] seemed to be very promising: it is reported to be an extremely efficient reagent for the conversion of cyclohexylamine to cyclohexanone under mild conditions.

In an attempt to use this reagent for the synthesis of **2** and **3** from **4** and **5**, respectively, we observed a different reaction outcome: unexpectedly the adducts **8**/**9** and **11**/**12**, respectively, containing a 2*H*-1,4-benzoxazine moiety were formed. The structure determination of **8**, **9**, **11**, and **12** and a plausible mechanism of formation are the object of the present report.

Results and Discussion. – *N*-Deacetylcolchicine (**4**), obtained in 80% overall yield from *N*-deacetyl-*N*-formylcolchicine (**7**) [9] by the methodology reported by *Grieco* and co-workers for the hydrolysis of amides [10][2]), was reacted with 1 equiv. of DTBQ (**6**) in MeOH for 24 h. After hydrolysis with oxalic acid, the product mixture was separated by chromatography, affording the expected colchicone (**2**) in only 6% isolated yield. In addition two novel optically inactive compounds, **8** and **9**, were isolated in 22 and 6% yield, respectively, together with 50% of unreacted **4**. The 2*H*-1,4-benzoxazine-like structures of **8** and **9**, derived by condensation of **4** with **6** followed by functionalization at C(6), were established by spectroscopic means.

The MS of **8** exhibits a molecular ion at *m*/*z* 557, revealing immediately that a molecule of DTBQ is incorporated into the colchicine moiety. In fact, the ^{1}H-NMR spectrum shows the presence of 2 *s* (each 9 H) at δ 1.28 and 1.47 for two *t*-Bu groups, and of two aromatic protons (*meta*-oriented to each other) at δ 7.19 and 7.25 (J = 3 Hz), in addition to the signals of the colchicine moiety (see *Exper. Part*). These NMR data, compared to that of **1**, suggest that a profound modification has occurred in the ring-B region of **8**: the characteristic splitting pattern of the *ABCDX* system due to CH_2(5), CH_2(6), and H–C(7) of colchicinoids is missing and replaced by an *ABX* system with signals at δ 3.06, 3.29, and 4.88 (J_{AB} = 14 Hz, J_{AX} = J_{BX} = 3 Hz, apparent values). In the ^{13}C-NMR spectrum of **8**, the presence of a benzylic methylene C-atom (*t* at δ 36.0), an oxymethine C-atom (*d* at δ 75.8), and a non-protonated sp^2 C-atom directly linked to a heteroatom (*s* at δ 164.4) is suggested.

The second reaction product **9** is simply the 10a-hydroxy derivative of **8** ($M^{+\cdot}$ at *m*/*z* 573, 16 amu higher than that of **8**), as confirmed by the ^{1}H- and ^{13}C-NMR spectra, which show an *AB* system at δ(H) 3.22 and 3.46 (J = 13.5 Hz) and a *t* at δ(C) 35.0 for C(10), a *s* at δ(C) 95.6 for an oxygenated C-atom (C(10a)), and a *s* at δ(C) 164.6 for the substituted sp^2-C-atom (C(16a)).

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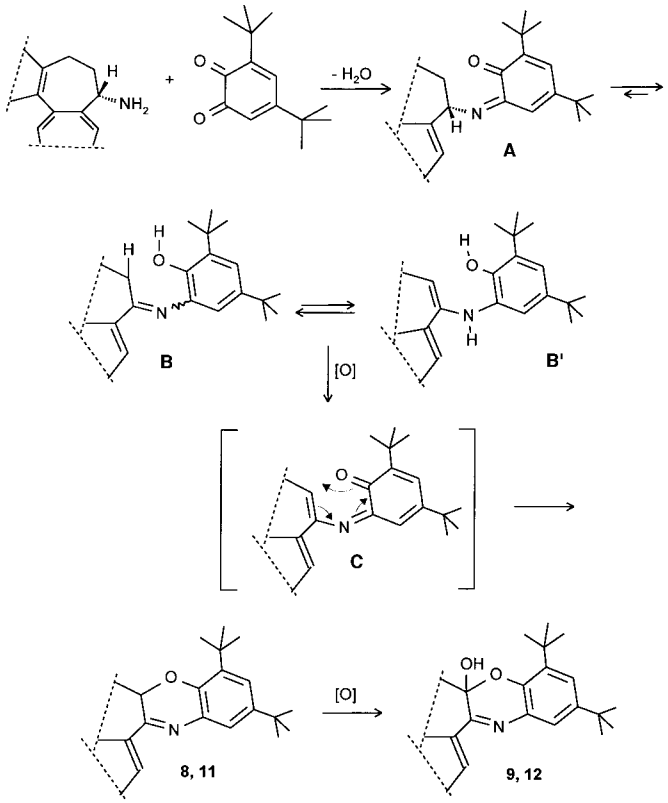
[2]) This methodology has been applied by *Lebeau* and co-workers to the deacetylation of cholchicine (**1**) [11]. However, in our hands, conversion of **1** to *N*-[(*tert*-butoxy)carbonyl]colchicine proceeded with unsatisfactory yield (15–20%), in spite of repeated experimentation.

The *N*-deacetylthiocolchicine (**5**), easily available from thiocolchicine (**10**) [12], behaved in a very similar way. On reaction with 1.1 equiv. of DTBQ (**6**), almost 48% of unaffected starting material **10** was recovered, thiocolchicone (**3**) was formed in 5% isolated yield, and **11** and **12**, the thio analogoues of **8** and **9**, were obtained in 24 and 5% yield, respectively.

Scheme



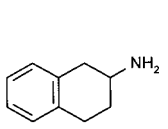
The formation of the new 2*H*-1,4-benzoxazine derivatives may be explained in terms of the pathway depicted in the *Scheme*. The condensation of **4** or **5** with DTBQ (**6**) proceeds uneventfully by initial condensation to the less hindered carbonyl group of **6** to form an intermediate *o*-quinone monoimine (**A**) [13]. This quinone monoimine undergoes a rapid spontaneous [1,5]-prototropic shift to give a *N*-substituted *o*-aminophenol represented as a mixture of the imine structure **B** and the enamine structure **B′** the rearrangement being thermodynamically favored by the aromatization of the *o*-quinone-monoimine system [13]. In the presence of the high potential of DTBQ (**6**)[3]), the intermediate *N*-substituted *o*-aminophenol **B**/**B′** is oxidized to another quinonoid state, generating the transient enamine-derived *o*-quinonemono-

[3]) For leading references concerning the use of high-potential quinones in the oxidation and dehydrogenation of N-compounds, see [14].

imine **C** which, by electrocyclization, leads to the 1,4-benzoxazines **8** or **11**[4]). The hydroxylated products **9** and **12** are formed by further oxidation of **8** and **11**, respectively, with DTBQ, presumably *via* an 1,4-benzoxazinium cation that is isolated in the hydrated form.

The reaction outcome is likely to result from a slow step of the initial condensation, compared with a fast step of dehydrogenation of the imine/enamine intermediates **B**/**B′** to form the enamine-derived *o*-quinone monoimine **C**. In fact, using 1.1 equiv. of DTBQ (**6**), the first formed aminophenol is readily further oxidized to the 1,4-benzoxazines, thus leaving most of the starting material unaffected. Acid hydrolysis of this reaction mixture allows only a small amount of the 7-ketones **2** and **3** to be isolated. With 2.1 equiv. of **6**, the ketones and starting amines were still obtained, although in small quantity, together with a substantial amount of the 2*H*-1,4-benzoxazine and 2*H*-2-hydroxy-1,4-benzoxazine derivatives. These benzoxazine derivatives are the only products isolated when 3.1 equiv. of **6** are used.

The behavior of **4** and **5** is different from that of cyclohexylamine and related amines, where an almost quantitative yield of the corresponding ketones is obtained after hydrolysis [9][13]. In an attempt to explain this difference, the tetrahydronaphthalin-2-amine **13** [16] was subjected to excess (3 equiv.) DTBQ (**6**) giving rise to a complex reaction mixture from which the only isolated product was the adduct **14**, albeit obtained in only 25% yield. No 3,4-dihydronaphthalin-2(1*H*)-one or other reaction products could be detected. The above results suggest that the formation of the benzoxazine products from **4**, **5**, and **13** is probably dependent on conjugation with the neighboring aromatic systems and the probable presence of the intermediate *N*-substituted *o*-aminophenol in solution in the enamine form **B′**, which is readily oxidized by DTBQ (**6**) to the enamine-derived *o*-quinone monoimine **C** [14]. At variance, in the case of cyclohexylamine, it has been shown [13] that the *N*-substituted *o*-aminophenol exists as the aromatic *Schiff* base **B**.



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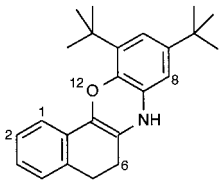

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In conclusion, the oxidative deamination of *N*-deacetylcolchicine (**4**) and *N*-deacetylthiocolchicine (**5**) with 3,5-di(*tert*-butyl)-1,2-benzoquinone (DTBQ; **6**) failed to give the corresponding ketones. Instead, the new adducts **8**, **9**, **11**, and **12** were obtained by the electrocyclic reaction of an intermediate enamine-derived *o*-quinone monoimine species, thus featuring an unprecedented reactivity of DTBQ. By

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[4]) The formation of a 1,4-benzoxazine has never been observed in the oxidative deamination of amines by DTBQ (**6**). The formation of a 2*H*-1,4-benzoxazine has been reported during the reaction of 2,4-di(*tert*-butyl)-6-(methoxyimino)cyclohexa-2,4-dien-1-one with *trans*-stilbene [15].

this reaction, derivatives of the colchicine nucleus functionalized at position 6 have been obtained for the first time.

Evaluation of **8**, **9**, **11**, and **12** as inhibitors of tubulin polymerization indicated that the compounds are devoid of any activity.

Experimental Part

General. The 3,5-di(*tert*-butyl)-1,2-benzoquinone was purchased from *Aldrich*. Colchicine (**1**), *N*-deacetyl-*N*-formyl colchicine (**7**), and thiocolchicine (**10**) were a gift of *Indena, S.p.A.*, Milano. All solvents were distilled and properly dried prior to use. During usual workup, all org. extracts were dried (Na_2SO_4 or Mg_2SO_4) and evaporated. Thin layer chromatography (TLC) for reaction monitoring: *Merck* silica gel *60* F_{254} plates; detection under UV light or spraying with *Pancaldi* reagent ($(NH_4)_6MoO_4$, $Ce(SO_4)_2$, H_2SO_4, H_2O) or alkaline $KMnO_4$ soln. Flash column chromatography (FC): *Merck* silica gel *60* (230–400 mesh). M.p.: *Kofler* hot-bench apparatus. ^{1}H- and ^{13}C-NMR Spectra: *Bruker AC300* (300 and 75.2 MHz, resp.); in $CDCl_3$; chemical shifts δ in ppm relative to $SiMe_4$ (= 0 ppm) as an internal standard, coupling constants *J* in Hz (apparent values). MS: *VG-7070EQ-HF* instrument (EI, 70 eV); *m*/*z* (rel. intensity in %).

N-*Deacetylcolchicine* (=*(*S*)-7-Amino-6,7-dihydro-1,2,3,10-tetramethoxybenzo[a]heptalen-9(5*H*)-one*; **4**). To a soln. of *N*-deacetyl-*N*-formylcolchicine (**7**; 3 g, 7.8 mmol) [8] and 4-(dimethylamino)pyridine (DMAP; 1 g, 8.2 mmol) in CH_2Cl_2 (30 ml), di(*tert*-butyl) pyrocarbonate (9.85 g, 45 mmol) was added at r.t. Stirring was continued for 24 h, then the temp. was increased to 40°, and di(*tert*-butyl) pyrocarbonate (9.85 g, 45 mmol) and DMAP (1 g, 8.2 mmol) in CH_2Cl_2 (20 ml) were added. After washing with H_2O and workup, the residue was purified by FC (AcOEt) to yield *N*-[(*tert*-butoxy)carbonyl]-*N*-deacetyl-*N*-formylcolchicine (3.21 g, 85%). Yellow solid. M.p. 95–99°. TLC (AcOEt/acetone 8:2): R_f 0.42. ^{1}H-NMR: 9.21 (*s*, 1 H); 7.20 (*s*, 1 H); 7.15 (*d*, *J* = 10.3, 1 H); 6.74 (*d*, *J* = 10.3, 1 H); 6.52 (*s*, 1 H); 5.00–5.15 (*m*, 1 H); 3.96 (*s*, 3 H); 3.93 (*s*, 3 H); 3.89 (*s*, 3 H); 3.63 (*s*, 3 H); 2.45–2.65 (*m*, 3 H); 1.9–2.0 (*m*, 1 H); 1.55 (*s*, 9 H). HR-MS: 485.2078 ($C_{26}H_{31}NO_8^+$; calc. 485.2050).

N-[(*tert*-Butoxy)carbonyl]-*N*-deacetyl-*N*-formylcolchicine (3.21 g, 6.62 mmol) in MeOH (20 ml) was added portionwise to a soln. of MeONa in MeOH (0.57 g of Na (24.78 mmol) in 60 ml of MeOH) at 0°. After 1 h, the soln. was neutralized by adding solid NH_4Cl. Evaporation and extraction with CH_2Cl_2 afforded pure *N*-[(*tert*-Butoxy)carbonyl]-*N*-deacetylcolchicine (2.06 g, 98%). Pale yellow solid. M.p. 80–84°. TLC (AcOEt/hexane 9:1): R_f 0.12. ^{1}H-NMR: 7.58 (*s*, 1 H); 7.27 (*d*, *J* = 10.3, 1 H); 6.83 (*d*, *J* = 10.3, 1 H); 6.53 (*s*, 1 H); 4.98 (*d*, *J* = 7.5, NH); 4.41 (*m*, 1 H); 4.01 (*s*, 3 H); 3.93 (*s*, 3 H); 3.90 (*s*, 3 H); 3.68 (*s*, 3 H); 2.2–2.6 (*m*, 4 H); 1.40 (*s*, 9 H). HR-MS: 429.2167 ($C_{24}H_{31}NO_6^+$; calc. 429.2151).

A soln. of *N*-[(*tert*-butoxy)carbonyl]-*N*-deacetylcolchicine (1.36 g, 2.98 mmol) in CH_2Cl_2 (30 ml) and CF_3COOH (3.3 ml) was stirred at r.t. under N_2 for 5 h. Evaporation, addition to toluene (50 ml), and evaporation gave a residue which was extracted wtih CH_2Cl_2 in the presence of aq. ammonia. FC purification (CH_2Cl_2/MeOH 19:1) gave **4** (1.04 g, 98%). M.p. 141–146°. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.36. ^{1}H-NMR: 7.87 (*s*, 1 H); 7.46 (*d*, *J* = 10.3, 1 H); 7.00 (*d*, *J* = 10.3, 1 H); 6.61 (*s*, 1 H); 4.20 (*m*, 1 H); 3.94 (*s*, 3 H); 3.93 (*s*, 3 H); 3.92 (*s*, 3 H); 2.6–2.8 (*m*, 1 H); 2.3–2.5 (*m*, 1 H). HR-MS: 357.1587 ($C_{20}H_{23}NO_5^+$; calc. 357.1576).

General Procedure for the Reaction of N-*Deacetylcolchicine* (**4**), N-*Deacetylthiocolchicine* (**5**), *and 1,2,3,4-Tetrahydronaphthalen-2-amine* (**13**) *with 3,5-Di(*tert*-butyl)-1,2-benzoquinone* (**6**). To a soln. of **4**, **5**, or **13** [13] (0.14 mmol) in MeOH (1 ml), 3,5-di(*tert*-butyl)-1,2-benzoquinone (**6**; 34 mg, 0.15 mmol) was added under N_2. Stirring was continued for 24 h, then THF (1 ml) and a soln. of oxalic acid (25 mg, 0.20 mmol) in H_2O (0.5 ml) was added. After 24 h, the hydrolysis was complete, the solvents were evaporated, and the residue taken up in H_2O and extracted with CH_2Cl_2. The residue of the reaction of **4** was chromatographed (CH_2Cl_2/MeOH 19:1) to give **2** (6%), **8** (22%), **9** (6%), and unreacted **4** (50%). The residue of the reaction of **5** was chromatographed (hexane/AcOEt 7:3) to give **3** (5%), **11** (24%), **12** (5%), and unreacted **5** (48%). The residue of the reaction of **13** was chromatographed ($CHCl_3$) to give **14** (30%).

With 2.1 equiv. of **6**, isolated yields were from **4**: **2** (4%), **8** (48%), **9** (15%), and recovered **4** (6%); from **5**: **3** (5%), **11** (51%), **12** (14%), and recovered **5** (5%). With 3.1 equiv. of **6**, isolated yields were from **4**: **8** (58%) and **9** (14%); from **5**: **11** (60%) and **12** (17%).

*5,6-Dihydro-1,2,3,10-tetramethoxybenzo[*a*]heptalene-7,9-dione* (**2**): Yellowish solid. M.p. 231°. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.62. ^{1}H- and ^{13}C-NMR: identical to those reported in [3][4]. HR-MS: 356.1283 ($C_{20}H_{20}O_6^+$; calc. 356.1260).

*12,14-Di(*tert-*butyl)-10,10a-dihydro-3,6,7,8-tetramethoxy-2*H-*benzo[4,5]heptaleno[2,1-*b*][1,4]benzoxazin-2-one* (**8**): Pale-yellow solid. M.p. 148–149°. TLC (CH_2Cl_2/MeOH 11:1): R_f 0.48. ^{1}H-NMR: 7.54 (*s*, 1 H); 7.31 (*d*, *J* = 11, 1 H); 7.25 (*d*, *J* = 3, 1 H); 7.19 (*d*, *J* = 3, 1 H); 6.85 (*d*, *J* = 11, 1 H); 6.65 (*s*, 1 H); 4.88 (*t*, *J* = 3, 1 H); 4.01 (*s*, 3 H); 3.87 (*s*, 3 H); 3.82 (*s*, 3 H); 3.52 (*s*, 3 H); 3.29 (*dd*, *J* = 14, 3, 1 H); 3.06 (*dd*, *J* = 14, 3, 1 H); 1.40 (*s*, 9 H); 1.28 (*s*, 9 H). ^{13}C-NMR: 178.8 (CO); 164.4 (C(16a)); 164.2 (C(3)); 153.0, 151.5 (C(6), C(8)); 147.0 (C(11a)); 144.4 (C(7)); 142.6 (C(15a)); 141.7 (C(5a)); 137.6 (C(1)); 136.4 (C(9a)); 136.1 (C(5)); 133.1 (C(12)); 133.0 (C(16b)); 131.6 (C(14)); 125.1 (C(5b)); 124.0, 123.0 (C(13), C(15)); 112.1 (C(4)); 109.1 (C(9)); 75.8 (C(10a)); 61.2, 61.1 (*Me*O–C(6), *Me*O–C(7)); 56.1, 55.8 (*Me*O–C(3), *Me*O–C(8)); 36.0 (C(10)); 34.7, 34.4 (Me_3C–C(12), Me_3C–C(14)); 31.4, 29.7 (2 Me_3C). HR-MS: 557.2789 ($C_{34}H_{39}NO_6^+$; calc. 557.2777).

*12,14-Di(*tert-*butyl)-10,10a-dihydro-10a-hydroxy-3,6,7,8-tetramethoxy-2*H-*benzo[4,5]heptaleno[2,1-*b*][1,4]-benzoxazin-2-one* (**9**): Pale-yellow solid. M.p. 178°. TLC (CH_2Cl_2/MeOH 11:1): R_f 0.28. ^{1}H-NMR: 7.69 (*s*, 1 H); 7.60 (*d*, *J* = 1.4, 1 H); 7.40 (*d*, *J* = 1.4, 1 H); 7.31 (*d*, *J* = 11, 1 H); 6.86 (*d*, *J* = 11, 1 H); 6.65 (*s*, 1 H); 4.00 (*s*, 3 H); 3.86 (*s*, 3 H); 3.81 (*s*, 3 H); 3.50 (*s*, 3 H); 3.46 (*d*, *J* = 13.5, 1 H); 3.22 (*d*, *J* = 13.5, 1 H); 1.47 (*s*, 9 H); 1.28 (*s*, 9 H). ^{13}C-NMR: 178.8 (CO); 164.6 (C(16a)); 162.6 (C(3)); 153.4, 151.4 (C(6), C(8)); 146.4 (C(11a)); 144.6 (C(7)); 141.9 (C(15a)); 140.2 (C(5a)); 138.5 (C(1)); 137.4 (C(9a)); 136.0 (C(5)); 133.4 (C(12)); 131.2 (C(16b)); 130.1 (C(14)); 125.6 (C(5b)); 124.4, 123.3 (C(13), C(15)); 114.5 (C(4)); 109.0 (C(9)); 95.7 (C(10a)); 61.1, 60.6 (*Me*O–C(6), *Me*O–C(7)); 56.4, 55.8 (*Me*O–C(3), *Me*O–C(8)); 35.0 (C(10)); 34.5, 34.2 (Me_3C–C(12), Me_3C–C(14)); 31.4, 29.9 (2 Me_3C). HR-MS: 573.2726 ($C_{34}H_{39}NO_7^+$; calc. 573.2735).

*5,6-Dihydro-1,2,3-trimethoxy-10-(methylthio)benzo[*a*]heptalene-7,9-dione* (**3**): Yellow solid. M.p. 231–235°. TLC (AcOEt/hexane 4:1): R_f 0.54. TLC (CH_2Cl_2/MeOH 9:1): R_f 0.61. ^{1}H- and ^{13}C-NMR: identical to those reported in [5]. HR-MS: 388.0987 ($C_{20}H_{20}O_5S^+$; calc. 388.0980).

*12,14-Di(*tert-*butyl)-10,10a-dihydro-6,7,8-trimethoxy-3-(methylthio)-2*H-*benzo[4,5]heptaleno[2,1-*b*][1,3]-benzoxazin-2-one* (**11**): Yellow solid. M.p. 106–108°. TLC (hexane/AcOEt 1:1): R_f 0.47. ^{1}H-NMR: 7.35 (*s*, 1 H); 7.30 (*d*, *J* = 11, 1 H); 7.27 (*d*, *J* = 3, 1 H); 7.20 (*d*, *J* = 3, 1 H); 7.09 (*d*, *J* = 11, 1 H); 6.64 (*s*, 1 H); 4.87 (*t*, *J* = 4, 1 H); 3.87 (*s*, 3 H); 3.83 (*s*, 3 H); 3.53 (*s*, 3 H); 3.30 (*dd*, *J* = 14, 4, 1 H); 3.06 (*dd*, *J* = 14, 4, 1 H); 2.45 (*s*, 3 H); 1.41 (*s*, 9 H); 1.28 (*s*, 9 H). ^{13}C-NMR: 182.3 (CO); 164.7 (C(16a)); 159.8 (C(3)); 153.8 (C(8)); 152.1 (C(6)); 147.2 (C(16b)); 144.9 (C(11a)); 143.2 (C(15a)); 142.3 (C(7)); 137.0 (C(5a)); 136.1 (C(5)); 135.5 (C(9a)); 135.1 (C(1)); 133.6 (C(12)); 132.3 (C(14)); 126.8 (C(4)); 125.8 (C(5b)); 124.6, 123.6 (C(13), C(15)); 109.8 (C(9)); 76.5 (C(10a)); 61.8, 61.6 (*Me*O–C(6), *Me*O–C(7)); 56.4 (*Me*O–C(8)); 36.6 (C(10)); 35.2, 35.0 (Me_3C–C(12), Me_3C–C(14)); 31.9, 30.3 (2 Me_3C); 15.7 (MeS). HR-MS: 573.2568 ($C_{34}H_{39}NO_5S^+$; calc. 573.2549).

*12,14-Di(*tert-*butyl)-10,10a-dihydro-10a-hydroxy-6,7,8-trimethoxy-3-(methylthio)-2*H-*benzo[4,5]heptaleno[2,1-*b*][1,4]benzoxazin-2-one* (**12**): Yellow solid. M.p. 228°. TLC (hexane/AcOEt 7:3): R_f 0.40. ^{1}H-NMR: 7.47 (*s*, 1 H); 7.38 (*d*, *J* = 2.5, 1 H); 7.30 (*d*, *J* = 2.5, 1 H); 7.26 (*d*, *J* = 10, 1 H); 7.07 (*d*, *J* = 10); 6.66 (*s*, 1 H); 3.85 (*s*, 3 H); 3.82 (*s*, 3 H); 3.50 (*s*, 3 H); 3.38 (*d*, *J* = 13, 1 H); 3.24 (*d*, *J* = 13, 1 H); 2.46 (*s*, 3 H); 1.48 (*s*, 9 H); 1.26 (*s*, 9 H). ^{13}C-NMR: 182.0 (CO); 163.4 (C(16a)); 160.1 (C(5)); 154.1 (C(8)); 152.0 (C(6)); 147.3 (C(16a)); 144.9 (C(15a)); 142.5 (C(7)); 141.1 (C(9a)); 137.9 (C(5a)); 136.3 (C(5)); 136.3 (C(1)); 131.9 (C(12)); 131.2 (C(14)); 127.2 (C(4)); 126.3 (C(5b)); 124.9, 124.0 (C(13), C(15)); 109.8 (C(9)); 96.3 (C(10a)); 61.9, 61.7 (*Me*O–C(6), *Me*O–C(7)); 56.5 (*Me*O–C(8)); 43.7 (C(10)); 35.7, 35.1 (Me_3C–C(12), Me_3C–C(14)); 31.4, 30.6 (2 Me_3C); 15.8 (MeS). HR-MS: 589.2506 ($C_{34}H_{39}NO_6S^+$; calc. 589.2498).

*9,11-Di(*tert-*butyl)-5,10-dihydro-6*H-*benzo[c]phenoxazine* (**14**): Amorphous solid. TLC ($CHCl_3$): R_f 0.55. ^{1}H-NMR: 7.81 (*dd*, *J* = 2.5, 1, 1 H); 7.52 (*d*, *J* = 2, 1 H); 7.3–7.5 (*m*, 3 H); 7.23 (*d*, *J* = 2, 1 H); 3.53 (*t*, *J* = 7.5, 2 H); 3.27 (*t*, *J* = 7.5, 2 H); 1.44 (*s*, 9 H); 1.37 (*s*, 9 H). HR-MS: 347.2271 ($C_{24}H_{29}NO^+$; calc. 347.2249).

REFERENCES

[1] H. G. Capraro, A. Brossi, in 'The Alkaloids', Ed. A. Brossi, Academic Press Inc., New York, 1984, Vol. XXIII, pp. 1–70.

[2] A. Brossi, H. J. C. Yeh, M. Chrzanowska, J. Wulff, E. Hamel, C. M. Lin, F. Quinn, M. Suffness, J. Silverton, *Med. Res. Rev.* **1988**, *8*, 77.

[3] T. H. Al-Tel, M. H. Abu-Zarga, S. S. Sabri, A. J. Frayer, M. Shamma, *J. Nat. Prod.* **1990**, *53*, 623.

[4] M. G. Banwell, S. C. Peters, R. J. Greenwood, M. F. Makay, E. Hamel, C. M. Lin, *Aust. J. Chem.* **1992**, *45*, 1577.

[5] Q. Shi, P. Verdier-Pinard, A. Brossi, E. Hamel, A. T. McPhail, K.-H. Lee, *J. Med. Chem.* **1997**, *40*, 961; U. Berg, H. Bladh, M. Hoff, C. Svensson, *J. Chem. Soc., Perkin Trans. 2* **1997**, 1697.

[6] T. F. Buckley, H. Rapoport, *J. Am. Chem. Soc*. **1982**, *104*, 4446.
[7] R. J. Baumgarten, *J. Chem. Ed*. **1996**, *43*, 398.
[8] E. J. Corey, K. Achiva, *J. Am. Chem. Soc*. **1969**, *91*, 1429.
[9] L. Canonica, B. Danieli, P. Manitto, G. Russo, *Chim. Ind.* **1967**, *49*, 1304.
[10] D. L. Flynn, R. E. Zelle, P. A. Grieco, *J. Org. Chem.* **1983**, *48*, 2424.
[11] P. Ducray, L. Lebeau, C. Mioskowski, *Helv. Chim. Acta* **1996**, *79*, 2346.
[12] P. Kerekes, P. N. Sharma, A. Brossi, C. F. Cignell, F. R. Quinn, *J. Med. Chem*. **1985**, *28*, 1204.
[13] R. F. Klein, L. M. Bargas, V. Horak, *J. Org. Chem.* **1988**, *53*, 5994.
[14] H.-D. Becker, in 'The Chemistry of Quinonoid Compounds. Part 1', Ed. S. Patai, John Wiley & Sons, Ltd., London, 1974, 398.
[15] D. N. Nicolaides, R. W. Award, E. A. Varella, *J. Heterocycl. Chem.* **1996**, *33*, 633.
[16] G. J. Cannon, J. A. Perez, J. P. Pease, J. P. Long, J. R. Flynn, D. B. Rusterholz, S. E. Dryer *J. Med. Chem.* **1980**, *23*, 745.

Received May 20, 1999