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Synthesis of Vinca Alkaloids and Related Compounds. Part LXVIII.[1] Two Diastereoisomeric Aspidosperma-Eburnea Type Bis-indoles: Their Synthesis and Structure Revisited

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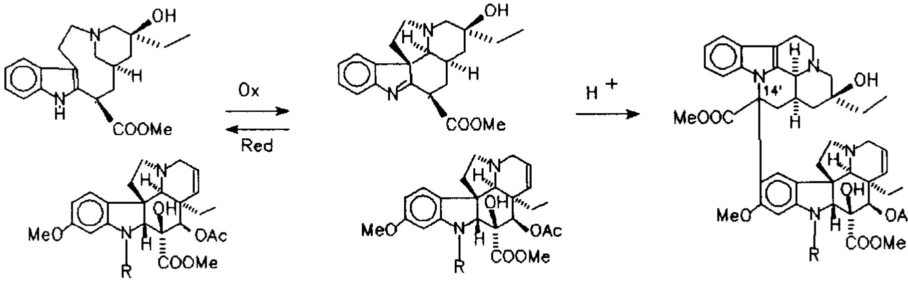
Abstract: With the aim of clarifying their previously incorrectly depicted structure, the indole-indoline type compounds 11 *and* 12 *were synthesized via different routes. The results presented here are a detailed account of the synthetic aspects of this work, and also redress some points of an earlier paper on this topic.*

Owing to their potential therapeutic benefit in cancer chemotherapy, bis-indole (indole-indoline) alkaloids have been a focus of much research for the past few decades. Many such compounds exhibit exceptional antitumour activity, and among these vinblastine (**1a**) and vincristine (**1b**)(Scheme 1) are widely used in clinical practice.[2]



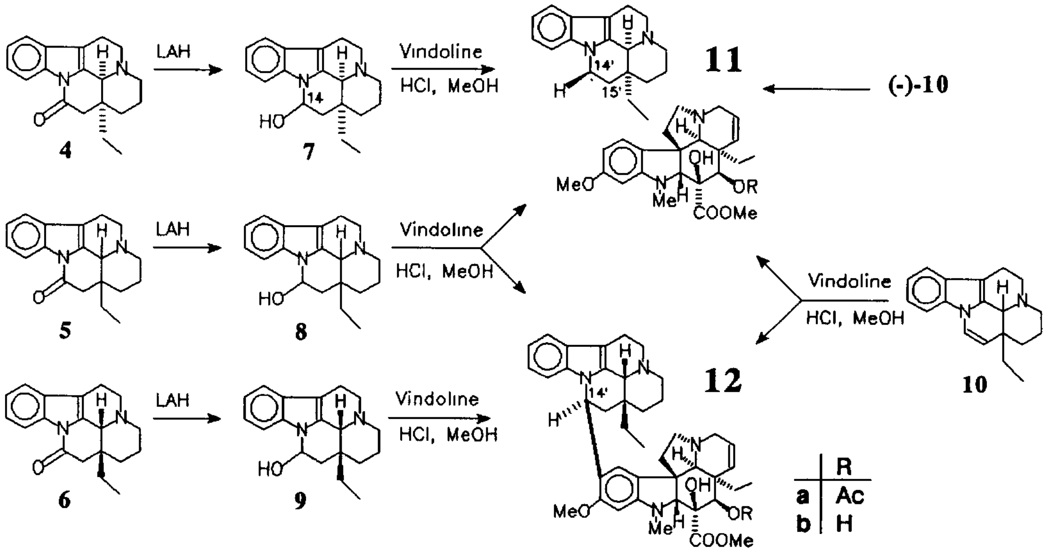
1 **2** **3**

a: R=Me; **b**: R=CHO

Scheme 1

One of our recent findings has shown that oxidation of **1** gives a Ψ-aspidosperma-aspidosperma type skeleton (**2**) via transannular cyclization.[3] Acid catalysis in turn triggers an aspidospermane → eburnane skeletal rearrangement of **2** into **3**.[4] Such a rearrangement is well known when starting from the "monomeric" vincadifformine, tabersonine[5,6] or vindoline[7], but not in the case of the more complex bis-indoles of type **2**. For the "monomeric" structures the rearrangement in all cases proceeds with retention of configuration at each point of ring anellation.[5-7] However, in exploring the synthetic scope and stereochemical details of these transformations involving various constitutional and stereoisomeric analogues of **1**, **2** and **3**, we have found that C-14' is particularly vulnerable to epimerization during the **2** → **3** rearrangement. Moreover, the stereostructural identification of compounds of type **3** has proved to be a notoriously challenging task (full details will be published in a forthcoming paper). This prompted us to look for suitable model compounds that would help provide a secure basis for resolving the relevant structure elucidation problems. Two such analogues are the aspidosperma-eburnea type diastereoisomers **11a** and **12a** (Scheme 2), first synthesized and structurally characterized by Takano and coworkers.[8] However, upon reproducing compounds **11a** and **12a** following Takano's work, we realized that the structure elucidation as well as the synthetic aspects of their paper requires reinvestigation.

First, as based on the vicinal $J_{\text{H-14',H-15'}}$ coupling constants, Takano *et al.* concluded that H-14' is "axial" in both **11a** and **12a**, and, by analogy with the similar coupling pattern of H-14 in eburnamine (**9**, OH=β)[9], they assigned the configuration of C-14' in **11a** and **12a**. Unfortunately, those authors depicted the structure of eburnamine incorrectly [the given formula is actually that of isoeburnamine (**9**, OH=α)[10] where H-14 is "equatorial" rather than "axial"], which led to erroneous graphical representations for the bis-indoles **11a** and **12a** as well.[8] As a result, the structure of these molecules has entered the general literature with the wrong C-14' configuration.[11] (Scheme 2 shows the correct configurations). We addressed this issue in a separate paper where we gave a detailed structural analysis of compounds **11a** and **12a** using NMR methods.[12] Our ^{1}H{^{1}H} NOE studies have also shown that these molecules exhibit a strongly biased two-site chemical exchange system due to hindered rotation about the bond connecting the two indole units. The kinetic characteristics of this exchange system are such that the minor conformer gives broad signals that are undetectable in a conventional ^{1}H NMR spectrum. However, in the NOE experiments this "hidden" exchange partner can lead to peculiar and easily misinterpretable effects, and this possibility has not been pointed out or demonstrated before. Moreover, such a "silent partner" might be expected to occur in a variety of related structures, and unawareness of its existence may result in false structural conclusions.[12]



Scheme 2

Secondly, on reproducing their work we noted that some of our synthetic observations were somewhat different from those reported by Takano *et al.* For this reason we took a more elaborate approach to producing compounds **11** and **12**. Here we report the results of this work, which also provides further insight into the chemical behaviour of this class of compounds.

Synthesis

We obtained compounds **11a** and **12a** via three different routes (Scheme 2). First, we followed Takano's procedure by coupling (±)-eburnamenine **10** with natural (-)-vindoline to give a mixture of **11a** and **12a**. In our hands the conversion of the dimers, especially that of **11a**, was extremely low. In all of our experiments therefore we applied reaction times that were significantly longer than specified by Takano *et al.*[8] However, longer reaction times promote the formation of the deacylated products **11b** and **12b**, which can then be reacylated to increase the yield of **11a** and **12a**.

Secondly, the reduction of racemic eburnamonine **5** gave an isomeric mixture of the alcohols **8** which, upon subsequent coupling with (-)-vindoline, afforded **11** and **12**.

Thirdly, in order to obtain **11** and **12** directly in isomerically pure forms, the enantiomeric vincamone **4** and eburnamonine **6** were both reduced into the respective C-14 epimeric mixtures of vincanol (**7**, OH=α) and epivincanol (**7**, OH=β), and isoeburnamine (**9**, OH=α) and eburnamine (**9**, OH=β). Alcohols **7** and **9** were then treated with (-)-vindoline to give **11a,b** and **12a,b**, respectively. The reaction time for coupling vindoline with **7** is ca. double that for **9**, which offers the possibility of synthesizing **11** and **12** diastereoselectively when starting from racemic eburnamonine **5** (see experimental section). (Takano *et al.* did not make mention of such a difference in reaction times.)[8]

EXPERIMENTAL

All reactions were carried out under inert atmosphere. As used below, the term "extractive workup" refers to the following procedure: extraction with the indicated solvents at pH 9, washing the organic layer with brine, drying over $MgSO_4$, concentration *in vacuo*, final drying (< 5 Torr) until constant weight. Most reactions were monitored by TLC using Merck $60F_{254}$ precoated silica gel on alumina sheets. Indole compounds were characterized with a 10% solution of ceric ammonium sulfate (CAS) in phosphoric acid as spray reagent or UV visualization. Flash chromatography was performed with the indicated solvents on Merck silica gel 60 (230-400 mesh); for gravity chromatography Merck silica gel 60 (70-230 mesh) was employed. Melting points are uncorrected. IR spectra were recorded on a Spectromom 2000 spectrometer. Optical rotation measurements were carried out with a Perkin-Elmer 241 polarimeter. Mass spectra were recorded on an AEI MS 902 double focusing spectrometer.

General procedure for the preparation of alcohols **7-9**

A solution of (-)-eburnamonine (**4**)[13,14] (0.80 g; 2.7 mmol) in dry THF (30 ml) was added to the suspension of $LiAlH_4$ (0.80 g) in THF (30 ml) and the resulting mixture was stirred at room temperature until **4** had been completely consumed (30 min). After quenching and the usual extractive workup the obtained mixture (**7**, 0.74 g, 92.5%) was used up directly for the coupling reaction.

The same process was applied for the preparation of the optically active **9** from (+)-eburnamonine (**6**)[13], and for racemic **8**, starting from (±)-eburnamonine (**5**)[15]. Compounds (-)-**10** and (±)-**10** were prepared from the corresponding alcohols by refluxing in pyridine for 15-20 h.

General method for the coupling of eburnamines with (-)-vindoline

A.) Starting from (±)-**8**. A mixture of (-)-vindoline (508 mg; 1.11 mmol) and (±)-eburnamine (330 mg; 1.11 mmol) in 40 ml of 2% HCl/MeOH solution was heated under reflux for 7 h [TLC monitoring, CH_2Cl_2-MeOH (20:1), R_F **12a** > **11a** > **12b** > **11b**]. After evaporation of the solvent *in vacuo* the residue was partitioned between dichloromethane (80 ml) and dilute ammonia solution (20 ml) at pH 9, and the aqueous phase was extracted with CH_2Cl_2 (3x20 ml). The combined organic solution was washed with dilute ammonia, then with brine, dried and evaporated under reduced pressure. The residue (760 mg in 5 ml of dry dichloromethane) was then treated with acetic anhydride (1.5 ml) and DMAP (50 mg), and was stirred at room temperature overnight. The solution was diluted with CH_2Cl_2 (50 ml), poured into saturated aq. $NaHCO_3$ solution and extracted with CH_2Cl_2 (3x25 ml). After the usual workup the products were separated by chromatography on silica gel with 0.5-2% MeOH in CH_2Cl_2. The first fraction contained 102 mg (20%) of nonreacted vindoline. The second fraction yielded 290 mg (35.5%) of **12a**, and from the third fraction 130 mg (16%) of **11a** was obtained.

11a: $[\alpha]_D^{20} = +24.2°$ (c = 1; $CHCl_3$) (lit.[8] $[\alpha]_D^{17} = -3.5°$), mp 190-192°C from ethyl acetate-hexane (lit.[8] 187-188°C).

IR (KBr): 2900-2850, 1740, 1620, 1260-1230, 1030 cm^{-1}.

MS (m/e, %): 734(M,75), 733(9), 675(13), 674(11), 587(25), 575(99), 574(74), 573(22), 495(32), 490(8.9), 467(55), 466(37), 367(14), 321(11), 287(31), 282(40), 265(11), 252(25), 208(12), 198(45), 135(100), 124(12), 122(57), 121(40), 107(33), 93(22).

12a: $[\alpha]_D^{20} = -199.9$°C (c = 1; $CHCl_3$) (lit.[8] $[\alpha]_D^{17} = -196°$), mp 234°C from methanol (lit.[8] 236-237 °C).

IR (KBr): 2950-2850, 1738, 1620, 1260-1220, 1040 cm^{-1}.

MS (m/e %): 734(M,96), 733(9.6), 675(15), 674(9.7), 587(19), 575(99), 574(79), 573(24), 495(33), 490(10), 467(58), 466(29), 453(10), 367(15), 321(15), 287(26), 282(56), 265(14), 252(31), 249(51), 208(57), 198(42), 135(100), 124(25), 122(47), 121(28), 107(33), 93(23).

Using the same quantities as under A.) the mixture was heated under reflux for 10 h. After extractive workup but without reacetylation 800 mg of crude product was obtained. Repeated chromatography on silica gel with 0.5-3% MeOH in CH_2Cl_2 afforded 82 mg (16%) of recovered vindoline, 220 mg (27%) of **12a**, 139 mg (17%) of **11a**, 62 mg (8%) of **12b**, and 77 mg (10%) of **11b**. (Total of **12**: 35%, total of **11**: 27%).

11b: $[\alpha]_D^{20} = +94.3°$ (c = 1; $CHCl_3$), mp 266-268°C from methanol (dec.).

IR (KBr): 2980-2880, 1740, 1610, 1230 cm^{-1}.

MS (m/e, %): 692 (M,100), 691(8.3), 577(33), 576(54), 575(87), 574(42), 573(14), 495(73), 494(22), 467(38), 466(23), 453(14), 346(9.8), 321(9.5), 287(13), 265(10), 252(32), 240(64), 208(11), 198(33), 135(64), 124(18), 122(47), 121(31), 107(28), 93(20).

12b: $[\alpha]_D^{20} = -168.3°$ (c = 1; $CHCl_3$), mp 181-183°C from methanol. IR (KBr): 2980-2880, 1740, 1610, 1230 cm^{-1}.

B.) Starting from **7**. A mixture of (-)-vindoline (708 mg; 1.55 mmol) and **7** (490 mg; 1.65 mmol) was heated in 2% HCl/MeOH (80 ml) under reflux for 10 h. After extractive workup the crude product was reacetylated with acetic anhydride (0.5 ml) and DMAP (20 mg) in dry CH_2Cl_2 (10 ml) by stirring at room temperature overnight. Repeated flash chromatography on silica gel, eluting first with 0.5-7% MeOH in CH_2Cl_2, then with a mixture of hexane-ethyl acetate-methanol-triethylamine (10:10:0.5:0.5), provided 106 mg (15%) of recovered vindoline and 638 mg (56%) of **11a**.

After heating under reflux for 18 h in 2% HCl/MeOH (25 ml) a mixture of (-)-vindoline (154 mg; 0.33 mmol) and **7** (100 mg; 0.33 mmol), extractive workup and separation with PLC as above afforded 111 mg (45%) of **11a** and 42 mg (18%) of **11b**, and the quantity of nonreacted vindoline decreased to 7% (11 mg).

C.) Starting from **9**. The reaction mixture of **9** (466 mg; 1.57 mmol) and (-)-vindoline (681 mg; 1.49 mmol) was refluxed for 2h in 2% HCl/MeOH (80 ml). The extractrive workup yielded 1.07 g of crude product. The repeated flash chromatography on silica gel, eluting first with 0.2-2.5% MeOH in CH_2Cl_2, then with a mixture of hexane-ethyl acetate-methanol-triethylamine (10:10:0.25:0.25), afforded 102 mg (15%) of recovered vindoline, 676 mg (62%) of **12a** and 52 mg (5%) of **12b**.

When the reacton mixture of **9** (100 mg; 0.33 mmol) and (-)- vindoline (154 mg; 0.33 mmol) was heated in 2 % HCl/MeOH (25 ml) under reflux for 9 h, extractive workup and preparative layer chromatography [Merck silica gel 60 $PF_{254+366}$ with CH_2Cl_2-MeOH (100:10)] yielded 148 mg (60%) of **12a** and 28 mg (12%) of **12b**, and the quantity of nonreacted vindoline decreased to 2% (3 mg).

D.) Starting from (-)-**10**. A solution of (-)-vindoline (328 mg; 0.72 mmol) and (-)-eburnamenine hydrocloride (224 mg; 0.72mmol) was heated under reflux for 10 h in 2% HCl/MeOH (50 ml). The usual extractive workup and purification by chromatography on silica gel with 0.5-3% MeOH in CH_2Cl_2 provided 260 mg (49%) of **11a**, 85 mg (17%) of **11b**, and 53 mg (16%) of vindoline was recovered.

General procedure for the base-catalyzed deacetylation

A solution of **11a** (300 mg; 0.41 mmol) and 0.5N methanolic sodium methoxide (3 ml) in dry methanol (30 ml) was stirred at room temperature until TLC indicated the complete consumption of the starting compound (10 days). Neutralization with glacial acetic acid was followed by evaporization *in vacuo* and an extractive workup with CH_2Cl_2 provided 240 mg (85%) of **11b**, which proved to be identical with the compound prepared as a side-product of the coupling reaction. Following the same procedure from **12a** (500 mg, 0.68 mmol), 420 mg (89%) of **12b** was obtained which could be matched spectroscopically with the sample prepared from the coupling reaction.

The above results are summarized in the following table:

Table. Attained Yields with Different Starting Compounds and Reaction Times

		YIELD (%)							
starting compound	reflux time(h)	regenerated vindoline	**11a**	**11b**	∑11	**12a**	**12b**	∑12	∑dimer
(±)-**10**	3	30	8	a	8	30	a	30	38
(±)-**8**	7	20	16		16[b]	35.5		35.5[b]	51.5
(±)-**8**	10	16	17	10	27	27	8	35	62
7	10	15	56		56[b]				56
7	18	7	45	18	63				63
(-)-**10**	10	16	49	17	66				66
9	2	15				62	5	67	67
9	9	2				60	12	72	72

[a] Not prepared. [b] After reacylation.

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REFERENCES AND NOTES

1. For part LXVII see: Nagy, T.; Szabó L.; Kovács A.; Tóth, G.; Kalaus, Gy.; Szántay, Cs. *Nat. Prod. Lett.,* in press.
2. Antitumor Bisindole Alkaloids from Catharanthus roseus (L.). In *The Alkaloids*; Brossi, A. Ed.; Academic Press: San Diego, 1990, Vol. 37, Chap. 1-6.
3. a) Honty, K.; Szabó, L.; Baitz-Gács, E.; Tamás, J.; Kajtár, M.; Szántay, Cs. IUPAC 14th Int. Symp. Chem. Nat. Prod., Poznan, Abstracts I, **1984** , p. 292. b) Belg. BE 901,446 **1985** , CA **1986** , *104* , 149228d
4. Szántay, Cs. *Pure and Appl. Chem.,* **1990** , *62*, 1299-1302.
5. For a summary see: a) Cordell, G. A.: The Aspidosperma Alkaloids. In *The Alkaloids*; Manske, R. H. F.; Rodrigo, R. G. A. Eds.; Academic Press: New York, 1979, Vol. 17, pp. 267-283. b) Saxton, J. E.: The Aspidosperma Group. In *The Monoterpenoid Indole Alkaloids*; Saxton, J. E. Ed.; Wiley-Interscience: New York, 1983; pp. 331-437.
6. For some recent studies on this rearrangement see: a) Hugel, G.; Lévy, J. *Tetrahedron* **1984** , *40,* 1067-1073. b) Magnus, P.; Pappalardo, P.; Southwell, I. *Tetrahedron* **1986** , *42*, 3215-3222. c) Palmisano, G.; Danieli, B.; Lesma, G.; Trupiano, F.; Pilati, T. *J. Org. Chem.* **1988** , *53*, 1056-1064. d) Lewin, G.; Poisson, J.; Schaeffer, C.; Volland, J. P. *Tetrahedron* **1990** , *46*, 7775-7786. e) Belattar, A.; Saxton, J. E. *J. Chem. Soc. Perkin Trans. I.* **1992** , 1583-1585.
7. Bölcskei, H.; Baitz-Gács, E.; Szántay, Cs. *Tetrahedron Lett.,* **1989** , *30*, 7245-7248.
8. Takano, S.; Hatakeyama, S.; Ogasawara, K. *Heterocycles* **1977** , *6*, 1311-1317.
9. Thomas, D. W.; Achenbach, H.; Biemann, K. *J. Am. Chem. Soc.* **1966**, *88*, 1537-1544.
10. Here we adopted the names of these compounds as used by Pfäffli and Hauth: Pfäffli, P.; Hauth, H; *Helv. Chim. Acta,* **1978**, *61*, 1682-1695. A different terminology has also been recently proposed: Lounasmaa, M.; Tolvanen, A.: Eburnamine-Vincamine Alkaloids. In *The Alkaloids*; Brossi, A. Ed.; Academic Press: New York, 1992, Vol. 42, Chap. 1. p. 1.
11. Cordell, G. A.; Saxton, J. E.: Bisindole Alkaloids. In *The Alkaloids;* Manske, R. H. F.; Rodrigo, R. G. A. Eds.; Academic Press: New York, 1981; Vol. 20, p. 190.
12. Szántay, Cs. Jr.; Demeter, Á.; Honty, K.; Kolonits, P.; Szántay, Cs. *Magn. Reson. Chem.,* **1993** , *31* , 773-785.
13. Novák, L.; Rohály, J.; Czibula, L.; Szántay, Cs. *Heterocycles,* **1977**, *6*, 1149-1156.
14. Szabó, L.; Sápi, J.; Kalaus, Gy.; Argay, Gy.; Kálmán A.; Baitz-Gács, E.; Tamás, J.; Szántay, Cs. *Tetrahedron*, **1983** , *39*, 3737-3747.
15. Kalaus, Gy.; Malkieh, N.; Katona I.; Kajtár-Peredi, M.; Koritsanszky, T.; Kálmán A.; Szabó L.; Szántay Cs. *J. Org. Chem.,* **1985** , *50*, 3760-3767.