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Bioactive Montanine Derivatives from Halide-induced Rearrangements of Haemanthamine-type Alkaloids. Absolute Configuration by VCD

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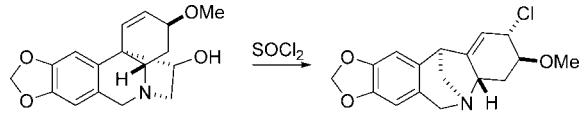
ABSTRACT



5 **7**

An unexpected rearrangement of haemanthamine-type alkaloids in the presence of halogenating agents has been found. Rearranged compounds present the 5,11-methanomorphantridine framework characteristic of montanine-type alkaloids. These compounds are difficult to obtain because of their scarcity in natural sources and because the synthetic approaches developed so far require numerous steps. Vibrational circular dichroism (VCD) spectroscopy was used to determine the absolute configuration of one of the rearranged compounds. Several rearranged alkaloids showed antimalarial activity.

Montanine-type alkaloids such as montanine (**1**), pancracine (**2**), coccinine (**3**) and manthine (**4**) belong to a subclass of *Amaryllidaceae* alkaloids.[1] These natural products have the core structure of the 5,11-methanomorphantridine ring system and differ only in the configurations of stereocenters at C-2 and C-3.[2] This type of alkaloid is isolated in very small amounts from some species of *Pancratium*[3] and *Narcissus*[4] genera. Because of their unique architecture and promising pharmacological potential,[5] these alkaloids have attracted much synthetic effort.[6] In this sense, most synthetic ap-

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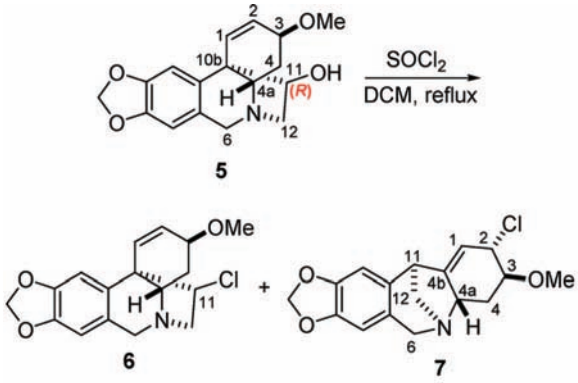
proaches involve many steps and low yield. For instance, recently Sha et al. have reported that the total synthesis of (−)-manthine has over 18 steps.[6e] We have found that montanine-type alkaloids can be easily obtained from alkaloids with a haemanthamine skeleton (which are more common and available compounds)[3] using several halogenating agents like thionyl chloride, thionyl bromide or DAST.

When haemanthamine (**5**) reacted with 1.5 equiv of $SOCl_2$ in DCM, two compounds were obtained (Scheme 1). One

Scheme 1. Reaction of Haemanthamine (**5**) with Thionyl Chloride



entry	equiv $SOCl_2$	6 (%)	7 (%)
1	1.5	14	31
2	1.5*	12	23
3	10	0	50
4	20	0	71

* = 1.5 equiv of pyridine was also added.

of them was identified as the expected 11-chlorohaemanthamine (**6**) (14%), where the hydroxyl group was substitued by chlorine.[7] The other one showed a molecular formula $C_{17}H_{18}NO_3Cl$ and NMR data that were not in accordance with the spectroscopic data of haemanthamine-type alkaloids.[8] Its ^1H NMR spectrum showed only one signal for an olefinic proton (δ 5.61, s, 1H). From the ^{13}C NMR spectrum, we observed the presence of a quaternary carbon at δ 151.6 instead of the quaternary carbon at δ 49.9 attributable to C-10b in **6**. 2D NMR studies pointed out a montanine-type skeleton for compound **7**. The HMBC spectrum showed the three-bond correlation of the aromatic proton H-10 to a doublet carbon at δ 44.8 assignable to C-11, which indicated us that a rearrangement occurred on the haemanthamine skeleton. Also, it was observed the followings three-bond correlations, H-11/C-1 and H-12/C-4b, which confirmed the proposed structure. From the ROESY spectrum, we determined the relative stereochemistry of the stereogenic centers in **7**. The chlorine group was established as α, because of the NOE effect between H-2 and the β-proton H-4a. On the other hand, the methoxy group was established as β, on the basis of the NOE effect observed between H-3 and H-12.

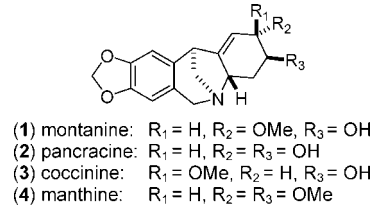
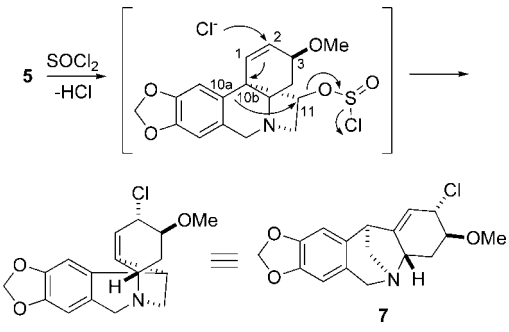


Figure 1. Structures of some montanine-type alkaloids.

Several attempts were performed to improve the yield. We used different ratio of thionyl chloride and the presence of base. For instance, from the reaction of haemanthamine with 1.5 equiv of $SOCl_2$ and 1.5 equiv of pyridine (entry 2), the products **6** (12%) and **7** (23%) were obtained in lower yield. The best results were achieved using a large excess of thionyl chloride. In fact, when we used 20 equiv of $SOCl_2$, the yield improved to 71% and only the rearranged product **7** was produced. A tentative mechanism for the formation of **7** is illustrated in Scheme 2. The chloride anion attacks at the

Scheme 2. Proposed Mechanism for the Rearrangement of Haemanthamine with Thionyl Chloride



carbon 2 with migration of the double bond and the σ-bond C10a−C10b. The antiperiplanar disposition of the C10a−C10b

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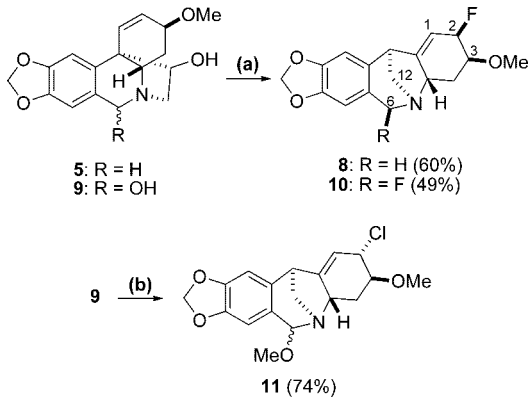
bond and the leaving group at C-11[9] favors the expulsion of the latter. However it is also possible to consider another mechanism through the formation of a nonclassical carbocation, since the orientation of the leaving group is *exo* in the azabicyclo [3.2.1] octane system. With the data in hand, we can not decide if the observed rearrangement is concerted or stepwise. Additional kinetic studies are required to discern between these two possibilities.

A related rearrangement was described by Wildman et al.[10] They observed that the treatment of the alkaloid crinamine with mesyl chloride followed by basic hydrolysis using strong conditions led to a rearranged product. This product could not be unequivocally characterized, because of the lack of NMR studies. The absence of published work on this type of rearrangement since Wildman's paper encouraged us to perform a deep study of the reaction.

To establish the versatility of this rearrangement, we treated haemanthamine (**5**) with 1.5 equiv of another halogenating agent like DAST [(diethylamino)sulfurtrifluoride], and the rearranged product **8** was obtained in 60% yield (Scheme 3). In this case, the rearrangement proceeded at low

Scheme 3. Rearrangement of Haemanthamine (**5**) and Haemanthidine (**9**)[a]



[a] Reagents and conditions: (a) DAST, DCM, −78 °C, 24 h; (b) 20 equiv $SOCl_2$, DCM, reflux, 5 h. Purification by preparative TLC with DCM:MeOH 9:1.

temperature and without the need of a large excess of the halogenating reagent to obtain a moderate yield. The main difference of **8** with respect to **7** was the β-disposition of the halogen group. The relative stereochemistry at C-2 was determined by the NOE effect between H-2 and H-3 and H-4α.

When haemanthamine (**9**), another alkaloid with a haemanthamine skeleton, was reacted with 3 equiv of DAST, the rearrangement was also observed (Scheme 3). The product **10** was obtained in 49% yield and it showed the same structure than **8**, with the difference that the hydroxyl group at C-6 was substituted by a fluorine atom in β-disposition. This stereochemistry was supported by the detected NOE effect between H-6 and H-12. The different stereochemistry at C-2 could be explained by the possible formation of a complex between the OMe group and DAST. This complex could favor the attack of the fluoride in the same face as the MeO group. We tried to obtain the corresponding brominated derivatives by using thionyl bromide ($SOBr_2$). Thus, when haemanthamine was treated with thionyl bromide, we obtained the corresponding rearranged brominated compound in 49% yield, which resulted to be very unstable. However, the ^{1}H NMR spectrum showed the same characteristics of the other rearranged derivatives, and the MS spectrum confirmed also the molecular formula of the product.

Haemanthidine (**9**) also reacted with thionyl chloride as predicted: the rearrangement was produced and the hydroxyl group at C-6 was also substituted by chlorine, obtaining **11** (74%). However, the chlorine at C-6 is easily displaced, and it was replaced by a methoxy group, probably from methanol, during the purification process. Thus, two signals for methoxy groups were observed in the NMR spectra, and the structure was elucidated as shown in Scheme 3.

With the purpose of setting unequivocally the absolute configuration of the rearranged product **7**, we analyzed this compound by vibrational circular dichroism (VCD). This technique is based on comparing the results of the measured IR and VCD spectra of the sample molecule with the corresponding calculated spectra.[11] VCD analysis has been used in recent years for determining the absolute configuration of several natural products,[12] which include sesquiterpenes, peptides, coumarins, and tropane alkaloids, among others.

Because the unambiguous conformational definition of the studied molecule is very relevant for the success of VCD spectroscopy, the theoretical spectrum of (2*S*,3*S*,4a*S*,11*S*)-**7** was obtained employing a molecular modeling protocol which initially involved the use of systematic and Monte Carlo conformational searching methodologies, followed by comparison of the resulting conformation to that obtained experimentally from detailed ^{1}H NMR measurements.[13]

The molecular model was constructed and subjected to a full minimization routine employing molecular mechanics (MMFF). During this process, the energy value was monitored as the convergence criterion to yield the global minimum energy structure at $E_{MMFF} = 91.870$ kcal/mol. This conformer was used as the starting point of a Monte Carlo search which afforded a total of four conformational structures in the initial 10 kcal/mol range. The second conformation showed $E_{MMFF} = 92.012$ kcal/mol and the remaining two, which are negligible, showed $E_{MMFF} = 97.852$ and 98.143 kcal/mol, accounting for a conformational ratio of 55.95 to 44.04% for the two lower energy structures. These two lower energy structures were then submitted to geometry optimization by DFT calculations at the B3LYP/6−31G(d) level of theory to provide an accurate model of the molecule. These two conformers are essentially superimposable, excepting for the conformation of the C-3 methoxy group, and account for a conformational ratio **7a**:

(9) For a 3D structure of **5**, see Supporting Information.

(10) Inubushi, Y.; Fales, H. M.; Warnhoff, E. W.; Wildman, W. C. *J. Org. Chem.* **1960**, *25*, 2153.

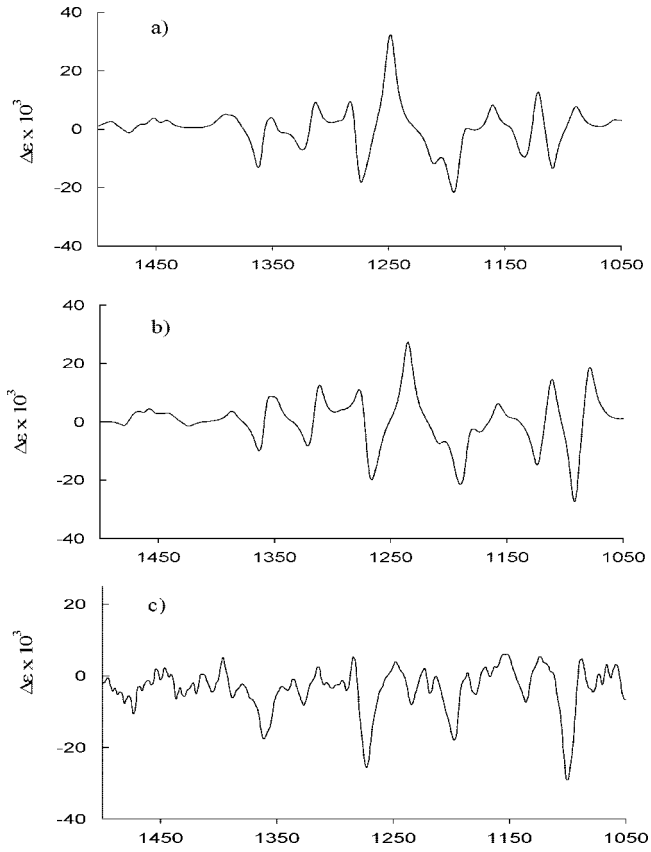


Figure 2. Comparison of calculated (a) DFT B3LYP/DGDZVP and (b) B3PW91/DGDZVP, and experimental (c) VCD spectra for **7**.

7b (for conformers, see Supporting Information) of 36.0:64.0, as deduced from application of the equation $\Delta G = -RT \ln K$. Conformers **7a** and **7b** were further optimized at the DFT B3LYP/DGDZVP level of theory to account for a 39.9:60.1 conformational distribution. After this structure optimization, the individual IR and VCD spectra for **7a** and **7b** were calculated at the B3LYP/DGDZVP and B3PW91/DGDZVP levels of theory. The calculated IR and VCD spectra of **7** were obtained by combining the individual spectra of **7a** and **7b** according to the Boltzmann conformational population. Detailed comparison of the calculated and experimental IR frequencies allowed calculation of an anharmonicity factor of 0.98 for the B3LYP/DGDZVP data, and of 0.97 for the B3PW91/DGDZVP data. The good agreement shown (Figure 2) between the experimental VCD spectrum of **7** and the final calculated DFT B3LYP/DGDZVP and B3PW91/DGDZVP VCD spectra directly allows the absolute configuration assignment to **7** as the 2*S*,3*S*,4a*S*,11*S* enantiomer.

Due to the antimalarial activity that presents some montanine-type alkaloids,[14] we decided to test our rearranged compounds for this activity. The results obtained for compounds **7**, **8** and **10** are summarized in Table 1. All derivatives showed significant activity against F-32 Tanzania strains of *Plasmodium falciparum*, compound **8** being the most active ($IC_{50} = 0.4$ μg/mL). These three compounds showed higher activity than the natural product pancracine (**2**), a montanine-type alkaloid isolated from several Amaryllidaceae species. From these results, we can conclude that the presence of one or more halogens is able to enhance the antimalarial activity.

Table 1. In Vitro Assays for Compounds **2**, **7**, **8** and **10** against *Plasmodium falciparum* F-32

compound	IC_{50} (μg/mL)
2	0.9 ± 0.04
7	0.4 ± 0.02
8	0.6 ± 0.04
10	0.7 ± 0.04
chloroquine	0.013

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Supporting Information Available: Experimental details, spectroscopic data for compounds **6**−**8**, **10** and **11**, and minimum energy conformers of **7**. This material is available free of charge via the Internet at http://pubs.acs.org.

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