A First Synthesis of Isofagaridine : Topoisomerase I Inhibitor

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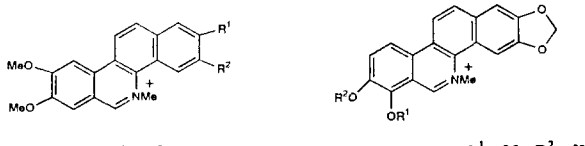
Naturally occurring benzo[*c*]phenanthridium alkaloids such as Nitidine (**1**) and Fagaronine (**2**) have been marked antitumor properties against leukemia even though these alkaloids exist toxicity problems as well as a narrow spectrum (Simeon *et al.*, 1989; Messmer *et al.*, 1972; Sufness *et al.*, 1979). More recently, much attention has been intensified as they were shown to inhibit HIV 1 and HIV 2 reverse transcriptases (Tan *et al.*, 1992).

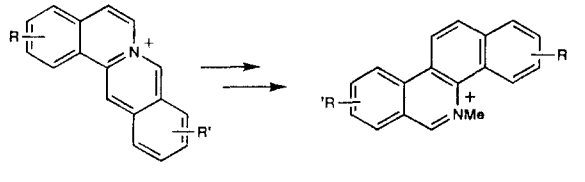
In 1993, Isofagaridine (**3**), was isolated and its structure was elucidated from the spectral data (Fang *et al.*, 1993). Through the bioassay-guided fractions of the roots of *Zanthoxylum nitidum*, this novel phenolic benzophenanthridine alkaloid showed to inhibit topoisomerase I-mediated DNA relaxation and stabilize the covalent complex between the enzyme and DNA. In connection with the bological activity of Fagaridine (**4**) which is a strong inhibitor of toposomerase II enzyme and being developed to the phase I clinical stage (Kobayashi *et al.*, 1993), the synthesis of Isofagaridine as well as the study of structure-activity relationship of substituents on aromatic ring attracted much attention to the researchers. Aiming at the convenient synthesis of Isofagaridine we tried to use the oxyfagarine (**5**) as a starting material because of its ready availability from a naturally abundant berberine according to our biogenetic transformation pathway.



Nitidine(1) : $R^1 + R^2 = OCH_2O$
Fagaronine(2) : R^1 = OH, R^2 = OMe

Isofagaridine(3) : R^1 = Me, R^2 = H
Fagaridine(4) : R^1=H, R^2 = Me

Scheme 1.



Protoberberine Benzo[c]phenanthridine

Scheme 2.

We have reported the convenient biomimetic methodology for the synthesis of all kinds of substituent pattern benzo[*c*]phenanthridine alkaloids (Hanaoka *et al.*, 1990; Hanaoka *et al.*, 1991). Regioselective demethylation of C-8 position on oxyfagaridine (**5**), an intermediate for the synthesis of Fagaridine (**4**), would afford the precursor for the synthesis of Isofagaridine because the strong hydrogen bonding between amide and hydroxyl group of C-7 position probably resists to be reacted with week base and electrophiles. Thus, a selective alkylation of dihydroxy compound supposed to be possible and be lead to the target compound, Isofagaridine.

RESULTS AND DISCUSSION

Attempts to demethylation of Oxyfagaridine (**5**) (under various conditions (ethanethiol/$AlCl_3$, BBr_3) (Node *et al.*, 1980; McOmie *et al.*, 1968) never gave the catechol (**8**) but rather the cleaveage of methylenedioxy group. Therefore, we investigated the oxidation of *o*-methoxy phenol to *o*-quinone. Such an oxidation of phenol to quinoid has been used in another instances (Reed *et al.*, 1988). Several oxidants such as Fremy's salt (Franck *et al.*, 1985), salcomine (Wakamatsu *et al.*, 1984) and cerium ammonium nitrate (CAN) (Orlemans *et al.*, 1988) are general reagents for preparing *o*- or / and *p*-quinone from the corresponding phenolic compounds. When the oxidation was performed in the potassium phosphate buffer solution with Fremy' salt or salcomine under the stream of oxygen resulted in recovering the starting material. However, oxidation of **5** with CAN in a mixed solution of CH_3CN and $CHCl_3$ at -15°C gave two products by monitoring on thin-layer chromatography, but we could isolate only the *p*-quinone (**7**) from the reaction mixture. As the *o*-quinone (**6**) produced might be too labile to be isolated due to a triketone structure, we tried to isolate it in a reduced form. So, the reaction mixture was immediately reduc-

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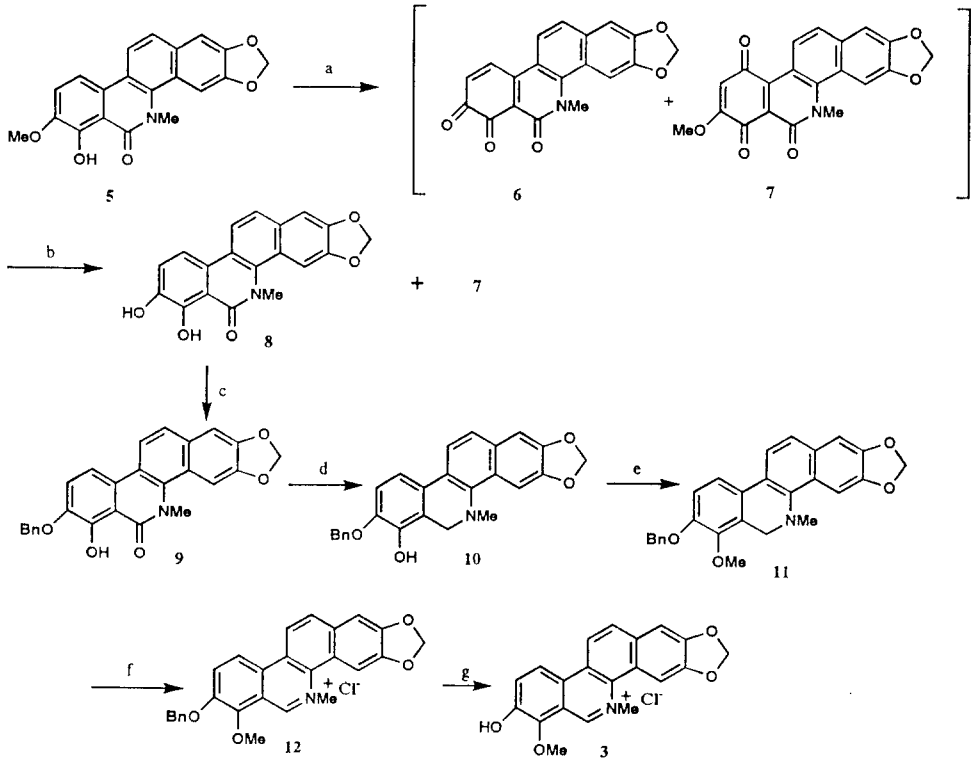

Scheme 3. *a.* $CAN/CHCl_3$-CH_3CN-H_2O; *b,* $Na_2S_2O_4$; *c,* $C_6H_5CH_2Cl$, K_2CO_3/CH_3COCH_3; *d,* LAH/THF then $NaBH_4$/MeOH; *e,* MeI, NaH/THF; *f,* DDQ, 5% NaOH/C_6H_6; *g,* *c*-HCl/EtOH

ed with acqueous sodium hydrosulfite to give the catechol (**8**) (38%) [mp 288-290°C($CHCl_3$). IR_{max} (KBr) cm^{-1} : 3450 (OH), 1640 (amide). ^{1}H-NMR δ : 8.03, 7. 58 (each 1H, AB-q, *J*=9.0 Hz, C_{11}-H and C_{12}-H), 7.66, 7.38 (each 1H, AB-q, *J*=9.0 Hz, C_9-H and C_{10}-H), 7. 58, 7.19 (each 1H, each s, C_1-H and C_4-H), 6.11 (2H, s, OCH_2O), 3.97 (3H, s, NCH_3)] and the *p*-quinone (**7**) (42%). The latter was probably produced by re-oxidation during column chromatography. The catechol (**8**) was regioselectively *o*-benzylated by treatment of benzyl chloride in the presence of potassium carbonate to give the monobenzyl ether (**9**) (91%) as we expected. The strong hydrogen bonding between amide ketone and 6-hydroxy group considered to be resist on above benzylation condition. Reduction of **9** with $LiAlH_4$ and $NaBH_4$, followed by methylation with methyl iodide and sodium hydride afforded the methyl ether (**11**) (85% from **9**). DDQ oxidation of **11** and subsequent *o*-debenzylation with HCl provided Isofagaridine (**3**) (83%) [mp 227-229°C (EtOH). IR_{max} (KBr) cm^{-1} (3450). ^{1}H-NMR (CF_3COOD) δ : 9.76 (1H, s, C_6-H), 8.62, 8.25 (each 1H, AB-q, *J*=9.0 Hz, C_{11}-H and C_{12}-H), 8.60, 8.11 (each 1H, AB-q, *J*=9.0 Hz, C_9-H and C_{10}-H), 8.10, 7.55 (each 1H, each s, C_1-H and C_4-H), 6.28 (2H, s, OCH_2O), 5.10 (3H, s, NCH_3). (Fang *et al.*, 1993, mp 226-228°C).

In conclusion, a first total synthesis of Isofagaridine was accomplished through the regioselective demethylation of *o*-methoxyphenol by way of *o*-quinone from oxyfagaridine which was transformed from a naturally abundant berberine.

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