Pergamon

Tetrahedron Letters, Vol. 35, No. 11, pp. 1789-1792, 1994
Elsevier Science Ltd
Printed in Great Britain
0040-4039/94 $6.00+0.00

0040-4039(94)E0142-K

The Squalestatins: Cleavage of the Bicyclic Core *via* the Novel 2,8,9-Trioxabicyclo[3.3.1]nonane Ring System.

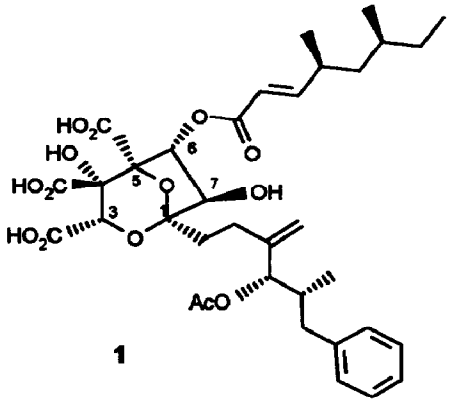
Daniele Andreotti, Panayiotis A. Procopiou* and Nigel S. Watson.

Medicinal Chemistry Department, Glaxo Group Research Limited, Greenford Road, Greenford, Middlesex, UB6 0HE, United Kingdom.

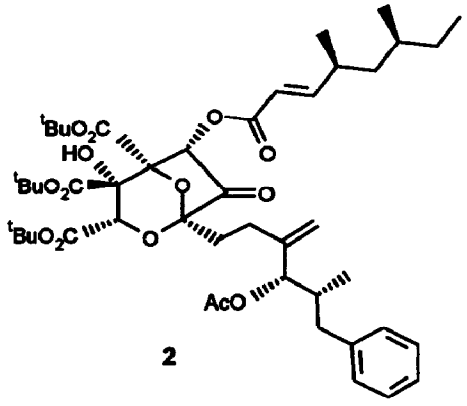
Abstract: Squalestatin 1 was converted into the γ-lactone analogue 5, *via* hydrolysis of the novel ortho ester anhydride 3, and thence into the acyclic derivative 8.

Recently we have described the isolation[1] and structure elucidation[2] of a family of novel and highly potent inhibitors of squalene synthase (SQS) termed the squalestatins. Subsequently the groups of Merck[3] and Tokyo Noko University[4] have reported the isolation of squalestatin 1 from different organisms. As part of our studies towards identification of the key features responsible for the biological activity of the squalestatins we have reported recently the preparation of the 6,7-dideoxy,[5] and 3-decarboxy[6] derivatives, and on the role of the tricarboxylic acid moiety.[7] Herein we disclose the preparation of analogues of 1 derived by formal cleavage of the 2,8-dioxabicyclo[3.2.1]octane ring system.

It was envisaged that oxidation of the 7-hydroxy group of the tris-*t*-butyl ester of 1 followed by Baeyer-Villiger oxidation of the derived ketone 2 should provide 3, incorporating the novel mixed anhydride-ortho ester grouping, and epoxide 4, hydrolysis of which would lead to cleavage of the squalestatin nucleus. Thus Baeyer-Villiger oxidation of ketone 2[5] using one equivalent of *m*-chloroperbenzoic acid (MCPBA) and buffered with sodium bicarbonate at 5°C gave cleanly the desired ortho ester 3 in 71% yield.[8] The use of larger amounts of MCPBA (2-3eq) and longer reaction time produced selectively the monoepoxide 4 which was isolated as a diastereoisomeric mixture (1:1).

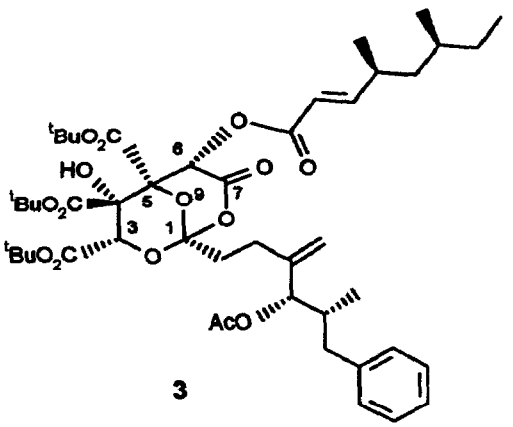


1

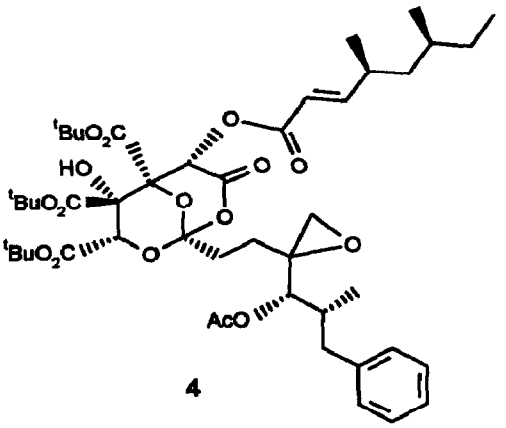


2

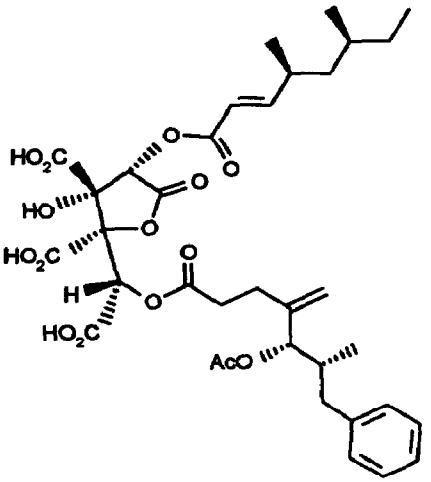
Deprotection of the *t*-butyl esters in 3 using formic acid gave only one product in quantitative yield. IR spectroscopy revealed the presence of a γ-lactone (1822 cm^{-1}), and the mass spectrum indicated a molecular



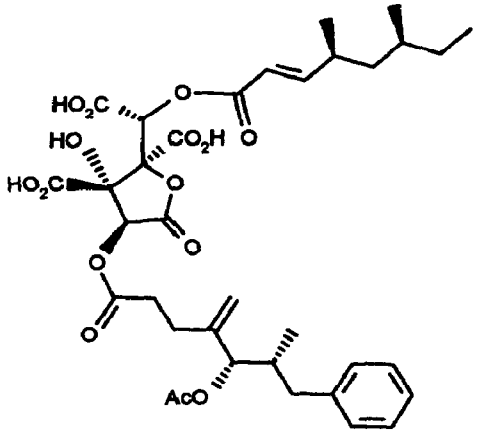
3



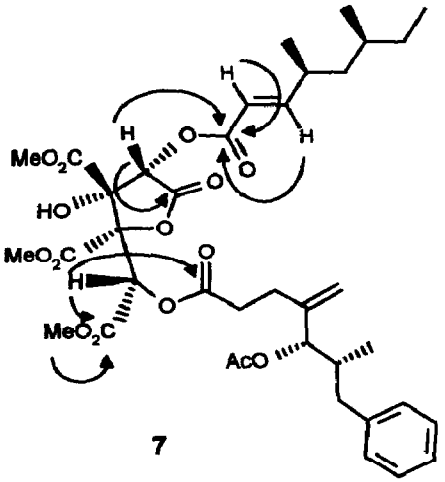
4



5



6



7

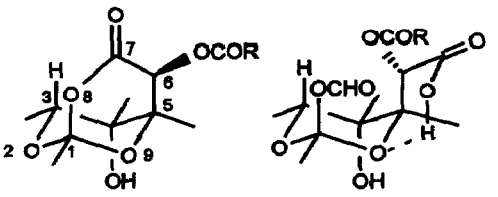
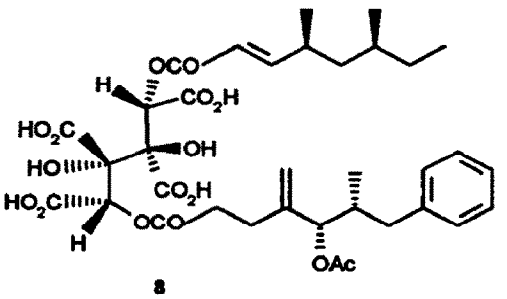


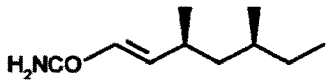
Figure 1

formula of $C_{35}H_{44}O_{15}$. The ^1H NMR spectrum indicated the presence of two 1H singlets, and signals for both lipophilic sidechains. In addition the ^{13}C and HMBC spectra were compatible with the structures of both γ-lactones **5** and **6**. In order to unambiguously identify the structure of this material it was treated with methyl iodide and sodium bicarbonate in DMF. NMR studies (HMBC) on the derived trimethyl ester established its structure as **7**.[9] The most significant long range $^1H \rightarrow ^{13}C$ correlations are shown diagrammatically which rule out the alternative structure **6** for the hydrolysis of **3**. Mechanistically, the acid catalysed cleavage of the bicyclic ring system present in **3** may be rationalised as follows: as O8 is a better leaving group than either O2 or O9, both of which have an electron pair oriented antiperiplanar to O8, initial cleavage of the C1-O8 bond would be expected to yield the lactonium ion, reaction of which with formic acid from the β-face would give an orthoester at C1 (Fig 1). O2 has a pair of electrons oriented antiperiplanar to the C1-O9 bond whose polarisation would be greatly enhanced by hydrogen bonding between O9 and the newly generated carboxylic acid at C7. This "push-pull" mechanism would facilitate the selective cleavage of the C1-O9 bond and thus generate the ester on O2. Lactonisation of the derived dihydroxy carboxylic acid would lead selectively to the γ-lactone **5**.

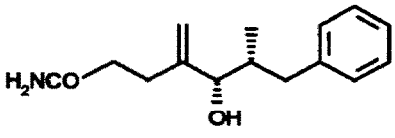
The lack of significant rat SQS inhibitory activity associated with lactone **5** (IC_{50} >1 μM) when compared with the natural product **1** (IC_{50} 12 nM) may be due to the different orientation of its tricarboxylic acid moiety. We have shown[10] that a monocyclic 1,3-dioxane analogue of **1** retains potent SQS inhibitory activity, and it was of particular interest to establish the inhibitory properties of an acyclic analogue. Selective γ-lactone hydrolysis of the tripotassium salt of **5** with aqueous ammonia gave after freeze-drying the tetracarboxylic acid **8** as its tripotassium salt.[11] **8** was stable in the solid state, but was unstable in solution for periods greater than 6h. Prolonged treatment of **5** with aqueous ammonia in methanol gave the amides **9**[12] and **10**[13], and δ-lactone **11**.[14] Compound **8** possesses reduced but significant enzyme inhibitory activity (IC_{50} 500 nM).



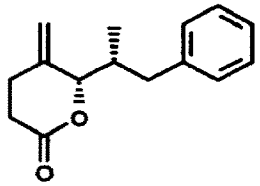
8



9



10



11

Acknowledgement: We are indebted to Dr Philip J. Sidebottom, Mr Sean Lynn and Mr Chris J. Seaman for conducting some detailed NMR experiments and for spectral interpretations.

References and Notes

1. Dawson, M. J.; Farthing, J. E.; Marshall, P. S.; Middleton, R. F.; O'Neill, M. J.; Shuttleworth, A.; Stylli, C.; Tait, R. M.; Taylor, P. M.; Wildman, H. G.; Buss, A. D.; Langley, D.; Hayes, M. V. *J. Antibiotics* **1992**, *45*, 639.
2. Sidebottom, P. J.; Highcock, R. M.; Lane, S. J.; Procopiou, P. A.; Watson, N. S. *J. Antibiotics.* **1992**, *45*, 648.
3. Hensens, O. D.; Dufresne, C.; Liesch, J. M.; Zink, D. L.; Reamer, R. A.; VanMiddlesworth, F. *Tetrahedron Letts.* **1993**, *34*, 399.
4. Hasumi, K.; Tachikawa, K.; Sakai, K.; Murakawa, S.; Yoshikawa, N.; Kumazawa, S.; Endo, A. *J. Antibiotics*, **1993**, *46*, 689.
5. Lester, M. G.; Giblin, G. M. P.; Inglis, G. G. A.; Procopiou, P. A.; Ross, B. C.; Watson, N. S. *Tetrahedron Letts.* **1993**, *34*, 4357.
6. Chan, C.; Inglis, G. G. A.; Procopiou, P. A.; Ross, B. C.; Srikantha, A. R. P.; Watson, N. S. *Tetrahedron Letts.* **1993**, *34*, 6143.
7. Watson, N. S.; Bell, R.; Chan, C.; Cox, B.; Hutson, J. L.; Keeling, S. E.; Kirk, B. E.; Procopiou, P. A.; Steeples, I. P.; Widdowson, J. *Bioorg. Med. Chem. Letts.* in press.
8. Data for **3**: ν_{max} ($CHBr_3$) 1768, 1732 cm^{-1}; δ_H ($CDCl_3$) 0.99 (d, J 7 Hz, 3H), 1.40, 1.46, 1.68 (3s, 27 H), 2.11 (s, 3H), 2.73 (dd, J 14 and 5 Hz, 1H), 4.19 (s, 1H), 4.72 (s, 1H), 5.01, 5.05 (2s, 2H), 5.14 (d, J 5 Hz, 1H), 5.81 (d, J 16 Hz, 1H), 6.67 (s,1H), 6.97 (dd, J 16 and 8 Hz, 1H), 7.1 - 7.3 (m, 5H); δ_C ($CDCl_3$) 169.9, 167.2, 163.8, 161.6, 158.5, 144.5, 140.0, 128.9, 128.1, 125.7, 117.4, 115.6, 112.2, 87.5, 79.3, 72.7, 71.7, 64.5, 43.0, 39.6, 36.6, 36.5, 34.4, 31.6, 29.5, 27.8, 27.7, 27.5, 24.2, 20.9, 19.9, 18.5, 13.7, 10.9; MS (DCI NH_3 + ve) *m/z* 890 $(M + NH_4)^+$, 873 $(M + H)^+$. Found: C, 65.01; H, 8.03. $C_{47}H_{68}O_{15}$ requires C, 64.70; H, 7.85%.
9. Data for **7**: ν_{max} ($CHBr_3$) 3469, 1822, 1749, 1737, 1648, 1244, 1023 cm^{-1}; δ_H ($CDCl_3$) 1.02 (d, J 7 Hz, 3H), 3.75, 3.88, 3.90 (3s, 9H), 4.20 (s, 1H), 5.05 (d, J 5 Hz, 1H), 5.00, 5.10 (2s, 2H), 5.80 (s, 1H), 5.84 (d, J 16 Hz, 1H), 5.91 (s, 1H), 6.94 (dd, J 16 and 8 Hz, 1H), 7.1-7.30 (m, 5H); δ_C ($CDCl_3$) 170.4, 170.1, 167.9, 167.0, 165.2, 165.1, 164.2, 159.0, 144.2, 140.1, 129.0, 128.3, 126.0, 117.0, 112.5, 89.6, 79.9, 79.3, 72.3, 71.6, 55.0, 53.4, 53.3, 43.1, 39.7, 37.0, 34.6, 31.9, 31.8, 29.6, 26.4, 21.0, 20.0, 18.9, 14.1, 11.0; MS (TSP NH_3 + ve) *m/z* 764 $(M + NH_4)^+$; HRMS (LSIMS + ve) found *m/z* 747.3216 $(M + H)^+$ $C_{38}H_{51}O_{15}$ requires 747. 3228. Found: C, 61.32; H, 6.79. $C_{38}H_{50}O_{15}$ requires C, 61.11; H, 6.75%.
10. Sharratt, P. J.; Hutson, J. L.; Inglis, G. G. A.; Lester, M. G.; Procopiou, P. A.; Watson, N. S. *Bioorg. Med. Chem. Letts.* submitted for publication.
11. Data for **8**: ν_{max} (KBr) 3363, 1726, 1619 cm^{-1}; δ_H (D_2O), 0.92 (d, J 7 Hz, 3H), 2.10 (s, 3H), 4.88 and 4.96 (2s, 2H), 4.96 (s, 1H), 5.58 (s, 1H), 5.84 (d, J 16 Hz, 1H), 6.82 (dd, J 16 and 8 Hz, 1H), 7.1-7.4 (m, 5H); δ_C (D_2O) 174.6, 174.2, 174.0, 173.9, 167.9, 158.1, 144.5, 140.7, 129.3, 128.5, 128.1, 118.2, 111.2, 80.0, 77.0, 42.6, 39.1, 39.0, 36.1, 36.0, 34.0, 31.5, 29.2, 25.8, 20.5, 19.3, 18.4, 13.4, 10.5; MS (LSIMS - ve) *m/z* 721 $(M - H)^-$, 703 $(M - H - H_2O)^-$, 551 $(M - H - C_{10}H_{18}O_2)^-$, 169 $(C_{10}H_{17}O_2)^-$, 155; HRMS (LSIMS - ve) found *m/z* 703.2608 $(M - H - H_2O)^-$ $C_{35}H_{43}O_{15}$ requires 703.2602.
12. Data for **9**: $[\alpha]_D$ + 56.8° (*c* 0.88 in MeOH); ν_{max} (KBr) 3335, 3183, 1678, 1636, 1601 cm^{-1}; δ_H ($CDCl_3$) 1.03 (d, J 7 Hz, 3H), 2.40 (m, 1H), 5.79 (d, J 16 Hz, 1H), 6.90 (dd, J 16 and 8 Hz, 1H); 6.90 (dd, J 16 and 8 Hz, 1H); MS (CI NH_3 + ve) *m/z* 187 $(M + NH_4)^+$, 170 $(M + H)^+$; HRMS (EI) found *m/z* 169.1468 (M^+) $C_{10}H_{19}NO$ requires 169.1467.
13. Data for **10**: $[\alpha]_D$ - 9.2° (*c* 1.2 in MeOH); ν_{max} ($CHBr_3$) 3521, 3399, 1678 cm^{-1}, δ_H ($CDCl_3$) 0.86 (d, J 7 Hz, 3H), 2.76 (dd, J 14 and 6 Hz, 1H), 3.92 (d, J 5 Hz, 1H), 4.95 and 5.12 (2s, 2H), 5.45 - 5.75 (br, 2H), 7.1 - 7.3 (m, 5H); MS (TSP NH_3 + ve) *m/z* 248 $(M + H)^+$, 230 $(MH - H_2O)^+$; HRMS (LSIMS +ve) found *m/z* 248.1650 $(M + H)^+$ $C_{15}H_{22}NO_2$ requires 248.1650.
14. Data for **11**: ν_{max} (KBr) 1747 cm^{-1}; δ_H ($CDC1_3$) 0.99 (d, J 7 Hz, 3H), 2.1 - 2.28 (m, 1H), 2.5 - 2.7 (m, 5H), 2.86 (dd, J 14 and 7 Hz, 1H), 4.70 (br s, 1H), 4.93 and 5.09 (2s, 2H), 7.13 - 7.33 (m, 5H); MS (TSP NH_3 + ve) *m/z* 248 $(M + NH_4)^+$, 231 $(M + H)^+$; HRMS (EI) found *m/z* 230.1318 (M^+) $C_{15}H_{18}O_2$ requires 230.1307.

(*Received in UK* 23 *December* 1993; *accepted* 14 *January* 1994)