Stereoselective synthesis and stereochemistry of seven isomeric spiroacetal structures based on the C17–C28 fragment (CD rings) of spongistatin 1

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Dedicated to Prof. Don Cameron on the occasion of his retirement
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Abstract

Brown allylation methodology has been employed to provide seven of the eight possible isomers of the (*ZE*)(*EZ*) spiroacetal system representing the CD ring fragment (C17–C28) of spongistatin 1. The stereochemistry of the isomers follows from high–field NMR examinations which furnish insights into the trends in the NMR data for these ketal structures.

Keywords: Stereoselective synthesis, spiroacetal, spongistatin, high-field nuclear magnetic resonance

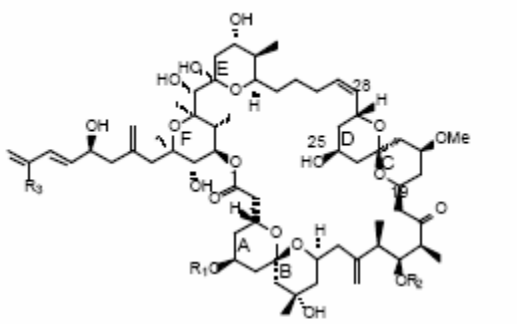
Introduction

In 1993–1994, the isolation of several series of marine–based macrolides was reported. These spongipyrans comprised the spongistatins, cinachyrolides and altohyrtins which exhibit extremely potent anti–tumour activity.[1-7] There has been enormous interest in the pharmacology, structures and stereochemistry of these systems which are available in only meagre amounts from their sponge sources. A number of syntheses of key sub– structures,[8] and several total syntheses[9,10,11] have been reported. Synthesis of altohyrtin C (identical with spongistatin 2) and altohyrtin A (spongistatin 1) have defined the relative and absolute stereochemistry of the natural compounds[5] which is shown below in Figure 1, and considered to represent this spongipyran group. The members of these families of metabolites differ with respect to the substituents at C5, C15 and C50.

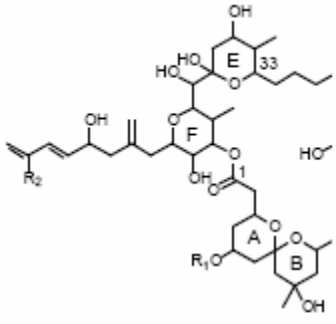
Sub–structure syntheses have focussed on the retrosynthetically attractive AB and CD spiroacetal assemblies and the EF tetrahydropyran moiety. These partial syntheses are

noteworthy as it may be that the most efficient total synthesis will select from the list of syntheses of the key, readily linked, sub–structures. Much of this synthetic endeavour has been summarised.[6,8]

In this report, we describe synthetic approaches to the CD spiroacetal system (C17–C28)[12] and at the time this work commenced (1995), there was stereochemical uncertainty regarding the relationship between the spongistatins, altohyrtins and cinachyrolide groups.[5] Our approach, at the outset, was to control the stereochemistry at C19 and C27 by asymmetric induction methods, and separate and examine the suite of spiroacetals that would result from lack of control at C21, C25 and the created spirocentre, C23. The structures of the resulting spiroacetals would provide insights into the thermodynamics[13] of these systems, whereas the fully assigned NMR spectra would clarify chemical shift trends and nOe's that would be valuable when additional stereocontrol was incorporated.



	R_1	R_2	R_3	
Spongistatin 1	Ac	Ac	Cl	(= altohyrtin A)
Spongistatin 2	Ac	Ac	H	(= altohyrtin C)
Spongistatin 3	H	Ac	Cl	(= 5-desacetylaltohyrtin A)
Spongistatin 4	Ac	H	Cl	(= cinachyrolide A)
Spongistatin 6	Ac	H	H	
altohyrtin B	Ac	Ac	Br	



	R_1	F
Spongistatin 5	H	Cl
Spongistatin 7	H	H
Spongistatin 8	Ac	H
Spongistatin 9	Ac	Cl

Figure 1. Structures of the spongistatins, altohyrtins, cinachyrolide A.

The CD system is stabilised by a single anomeric effect (in contrast with the AB spiroacetal system) and hence there is a question whether the required *ZE* or *EZ* system would be favoured, and which isomer(s) within each of these systems. This feature has been considered and regulated in a number of syntheses,[9,12] and equilibration–separation– equilibration cycles have been utilised. Given the free–energy similarities, choice of protection groups on the various oxy–functions could influence these equilibria. (In the assignment of the *ZE*, *EZ* descriptors,[14] the C ring has priority over the D ring, and the first descriptor always refers to the C ring, and the second to the D ring. In addition, if the ring substituents attached to the carbon atoms linked to the tetrahydropyranyl (ring) oxygen, are *cis*, then *Z* is used, and if *trans*, *E* is applied.) CD spiroacetal.

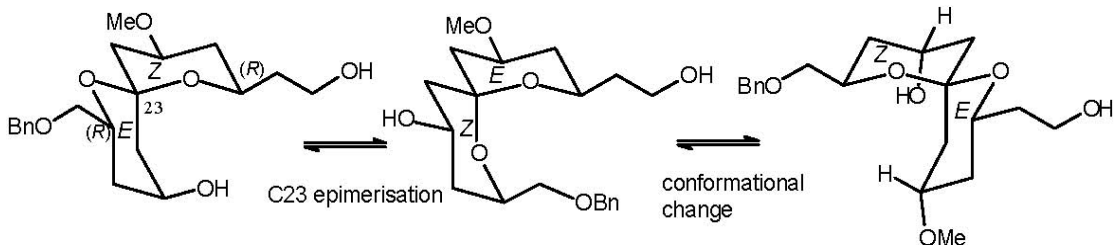


Figure 2. Illustrative spirocentre epimerisation and conformational reversal: *ZE* ≡ *EZ* interconversion in the CD system.

Results and Discussion

Excision of the CD spiroacetal unit from spongistatin leads to spiroacetal **1** which may be modified to **1a** and unravelled to ketohexol **2**. The latter was planned to result from dithiane coupling of two ∃ oxy epoxides **3** and **4**, as shown below.

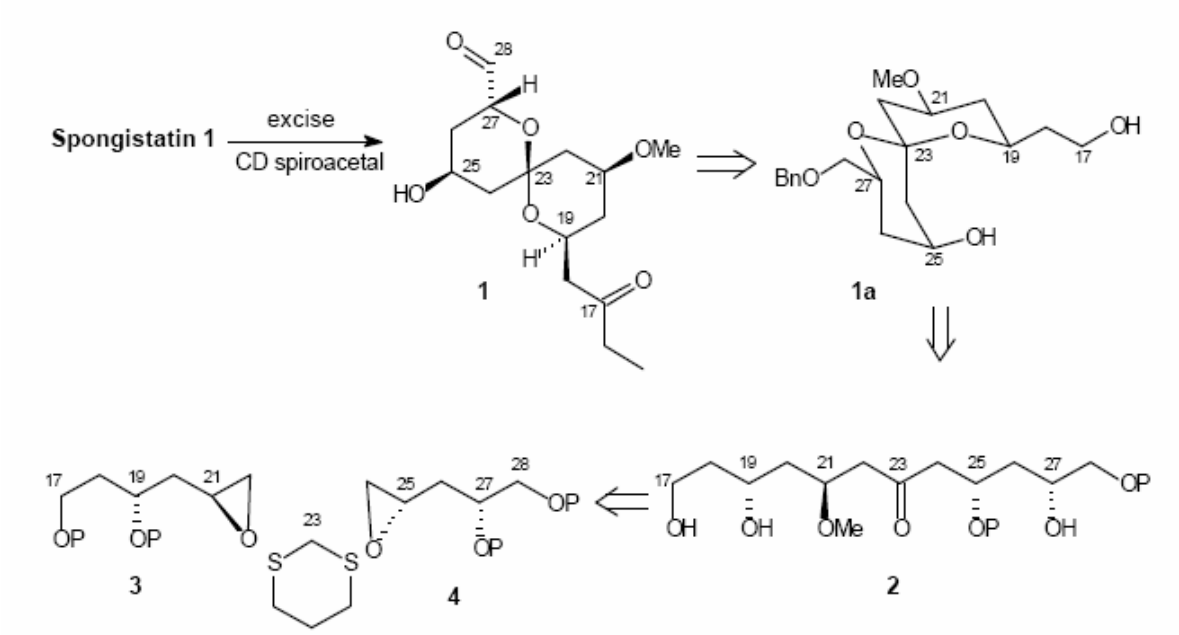
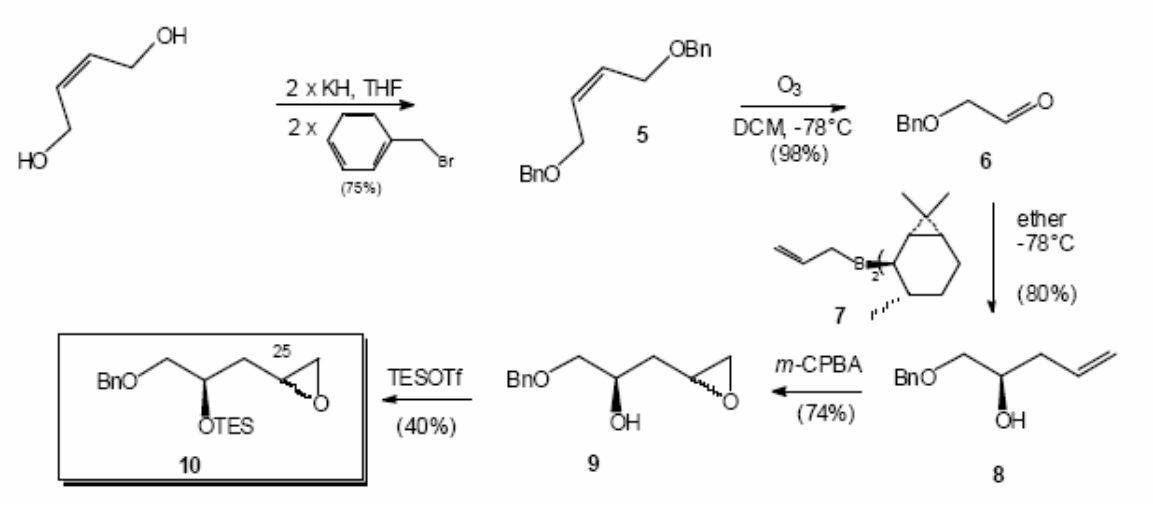


Figure 3. Retrosynthetic analysis of the CD spiroacetal.

Arrangement **2** incorporates four stereogenic centres which increases to five on spiroacetalisation (C23). Our approach to open–chain precursor **2** incorporated control at C27 and C19, so that in principle, the final spiroacetal could be a mixture of eight diastereomers, with centers at C25, C23 and C21 uncontrolled. Construction of the chiral epoxides **3** and **4** was achieved with moderate to high asymmetric induction at (C19, C21) and (C25, C27) as outlined below. The route to epoxide **4** utilised the *bis*–benzyl ether **5** of 2–buten–1,4–diol and ozonolysis provided the benzyloxy aldehyde **6**[15] Asymmetric allylation of this aldehyde, employing

Brown's *B*–allybis(isocaranyl)borane (4–^{d}Icr2Ball, **7**),[16] provided **8** with an *ee* exceeding 95%, based on NMR analysis of its Moshers ester. This was consistent with Brown's original reports and agrees with the results of Paterson.[12(b)]

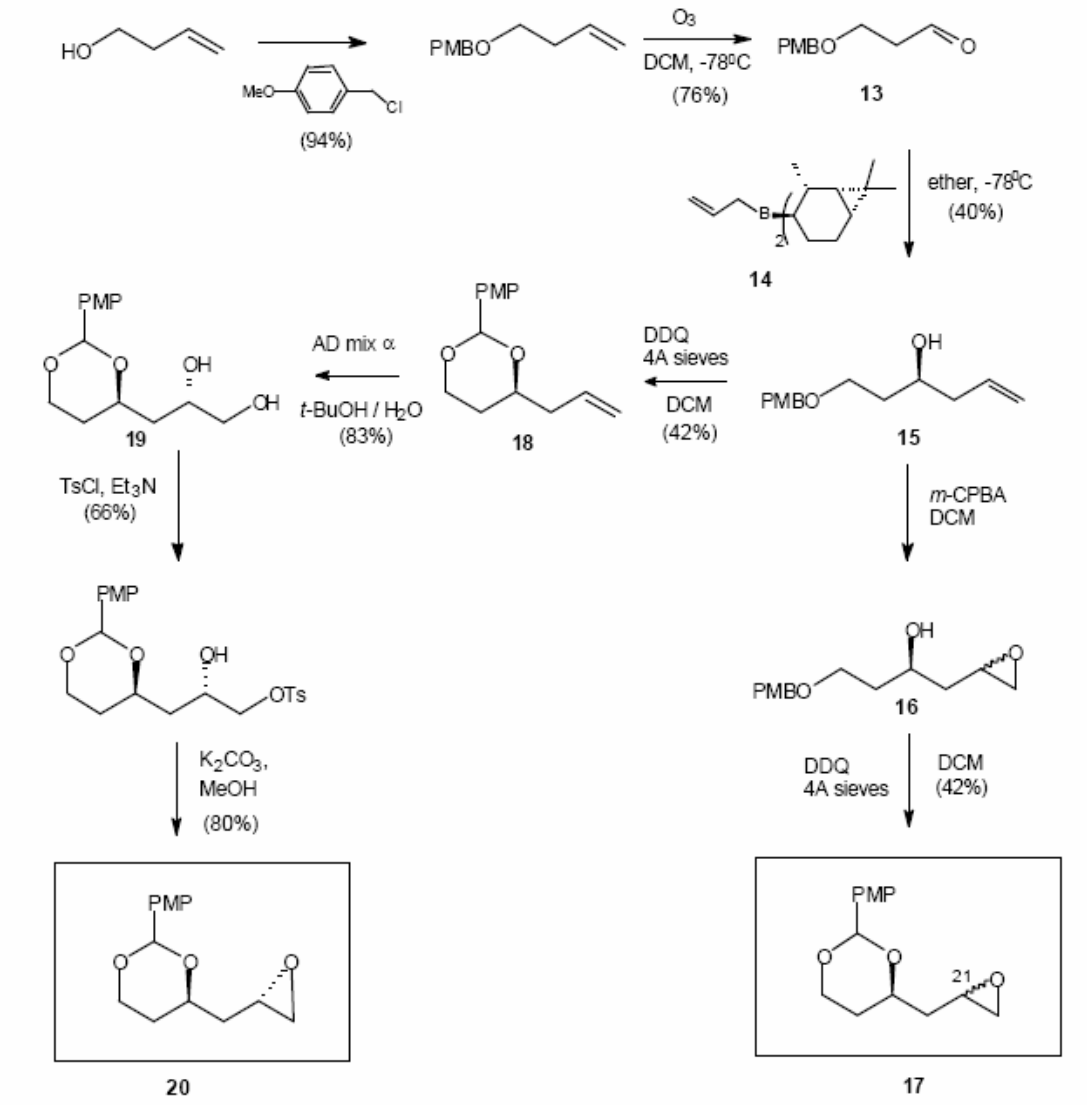
Epoxidation (*m*–CPBA) of **8** provided the desired epoxide **9** as a mixture of diastereomers, which were protected as the TES ethers, **10**. These were prone to decomposition and were stored at low temperature (–78° C). (Scheme 1)



Scheme 1. Synthesis of the chiral epoxide **10**.

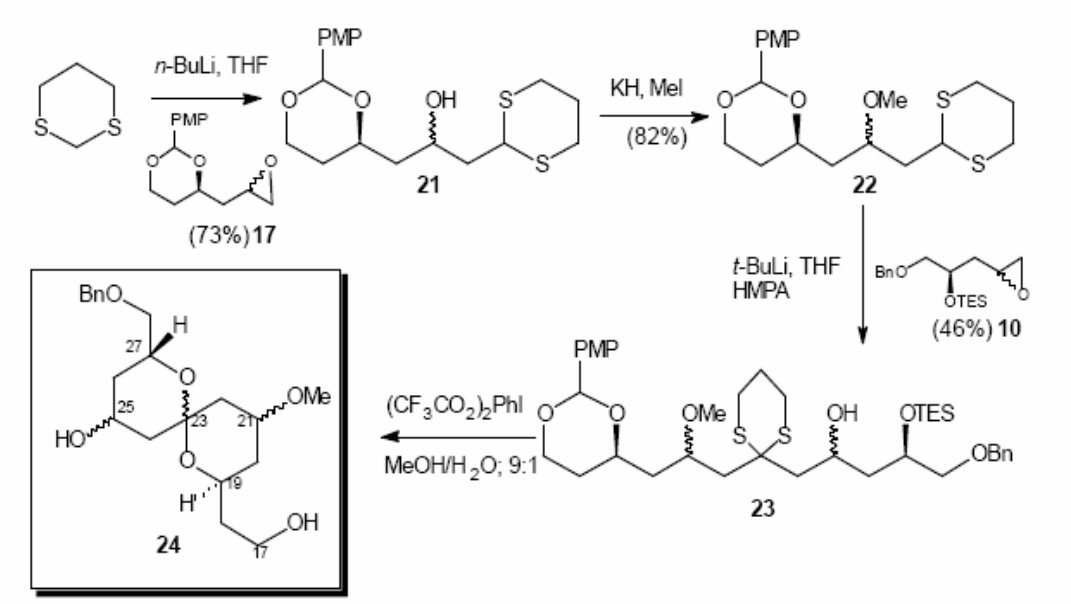
Epoxide **20** (cf.3) was synthesised, as for **10** but starting from the *p*– methoxybenzyl derivative of but–3–enol. Ozonolysis (careful TLC to monitor any over oxidation) provided aldehyde **13** which with Brown's carenyl–based allylating reagent **14** provided the desired allyl alcohol **15** (>95% *ee* by Mosher ester analysis) but in moderate yield. Elaboration to the epoxides **17** and **20** proceeded as shown in Scheme 2, with **17** being a mixture of epimers at C21.

Thus, epoxidation with *m*–CPBA of the hydroxy alkene **15** provided hydroxy epoxide **16**, and oxidative cyclisation of the *p*–methoxybenzyl group with DDQ in the presence of 4Δ sieves, afforded the required epoxide **17** in 42% yield. This epoxide was used in initial studies of the dithiane alkylations described below. Alternatively, DDQ-mediated cyclisation of the *p*–methoxybenzyl group of **15** provided the alkene **18** which served as a substrate for the Sharpless dihydroxylation protocol to deliver diol **19**. Transformation of the diol to the chiral epoxide **20** proceeded via the corresponding tosylate.



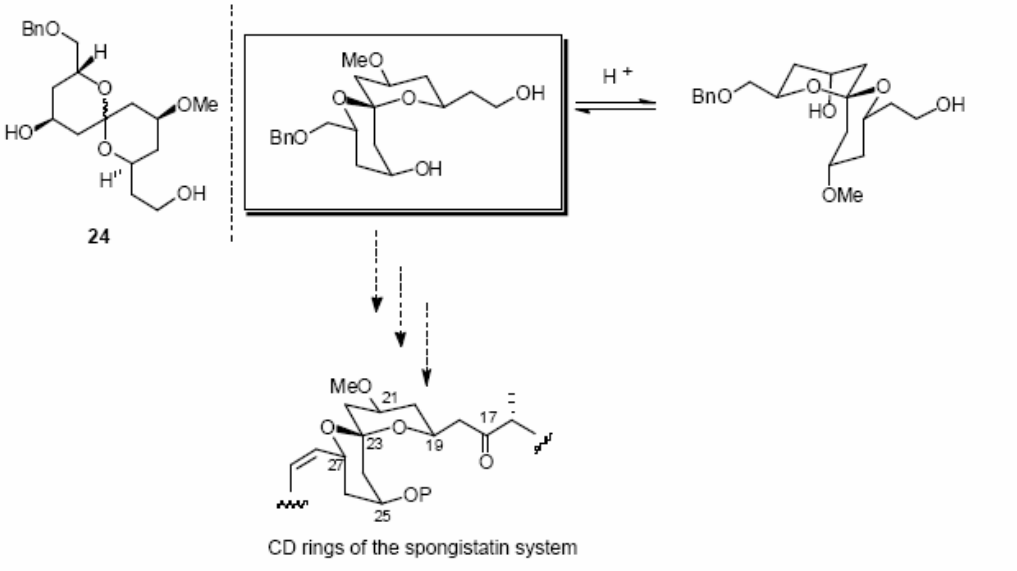
Scheme 2. Synthesis of the Chiral Epoxides **17** and **20**.

The coupling of 1,3–dithiane and the epoxide **17** at low temperature (–40 °C) (Scheme 3) proceeded efficiently (73%) to provide the monalkylated adduct **21** which was protected as the methyl ether. Suitable conditions for the opening of epoxide **10** by deprotonated **22** proved difficult to optimise. In our hands, ^{t}butyl lithium in THF containing 10% HMPA (freshly distilled) provided the best outcome and **23** was obtained as a diastereomeric mixture. Treatment of this with Stork's reagent, (bistrifluoroacetoxyiodobenzene) unmasked the carbonyl moiety, removed the *p*–methoxybenzyl acetal and the triethylsilyl group and effected cyclisation to a mixture of spiroacetals **24**. The high resolution mass spectrum exhibited a molecular ion (M=366) and fragmentation ions consistent with the spiroacetal structure **24**. For example, ions corresponding to the loss of OCH_3 (*m/z* 335), CH_3OH (*m/z*, 334), $–CH_2CH_2OH$ (*m/z*, 321), H_2O and CH_3OH (*m/z*, 316) and $–CH_2OBn$ (*m/z*, 245) are prominent, along with ions for oxygenated pyran fragments (eg. *m/z*, 237).[17,18]



Scheme 3. Synthesis of spiroacetal **24** via dithiane coupling reactions.

This spiroacetal system **24** contains five stereogenic centres, and as two of these (C19 and C27) were installed asymmetrically, there remains the possible formation of eight diastereoisomers. Reverse phase HPLC (acetonitrile/water) successfully separated the diastereomers and sufficient of each was obtained to enable high–field NMR studies, although in some cases, only very small amounts (sub–milligram) were available, and accurate optical rotations could not be determined. Benzene-d6 was employed as solvent to avoid possible acid catalysed equilibration (See Scheme 4) of the purified spiroacetals, a problem encountered previously with simpler systems in choroform.[19]



Scheme 4. Notional acid catalysed isomerization of spiroacetal **24**.

NMR Analysis of the Stereoisomers

Seven isomers of spiroacetal system **24** (see Figure 4) were of sufficient purity to allow analysis of the proton and carbon NMR data utilising a combination of high field 1–D and 2–D NMR and nOe analysis (at 400, 500 and 750 MHz). These isomers are referred to as **A**–**G** for convenience and ease of comparisons, and are shown in Figure 4, together with the remaining *ZE/EZ* isomer, **H**, which was not formed in sufficient amount for separation and NMR identification. It is of interest that the relative amounts of the seven isomers **A**-**G** formed under our spiroacetalisation conditions, are comparable, and no isomer predominates over the others by a factor exceeding 4 or 5. Given the difficulties in calculating equilibrium positions in acetal isomerisations, meaningful reconciliation of the data with computed relative free energies is extremely difficult.[20] Comparative analysis of the proton and carbon shifts (See Tables 1 and 2) reveals a number of characteristic resonances for certain centres in the *Z,E* spiroacetal system that mirror earlier findings on simpler systems.[19]

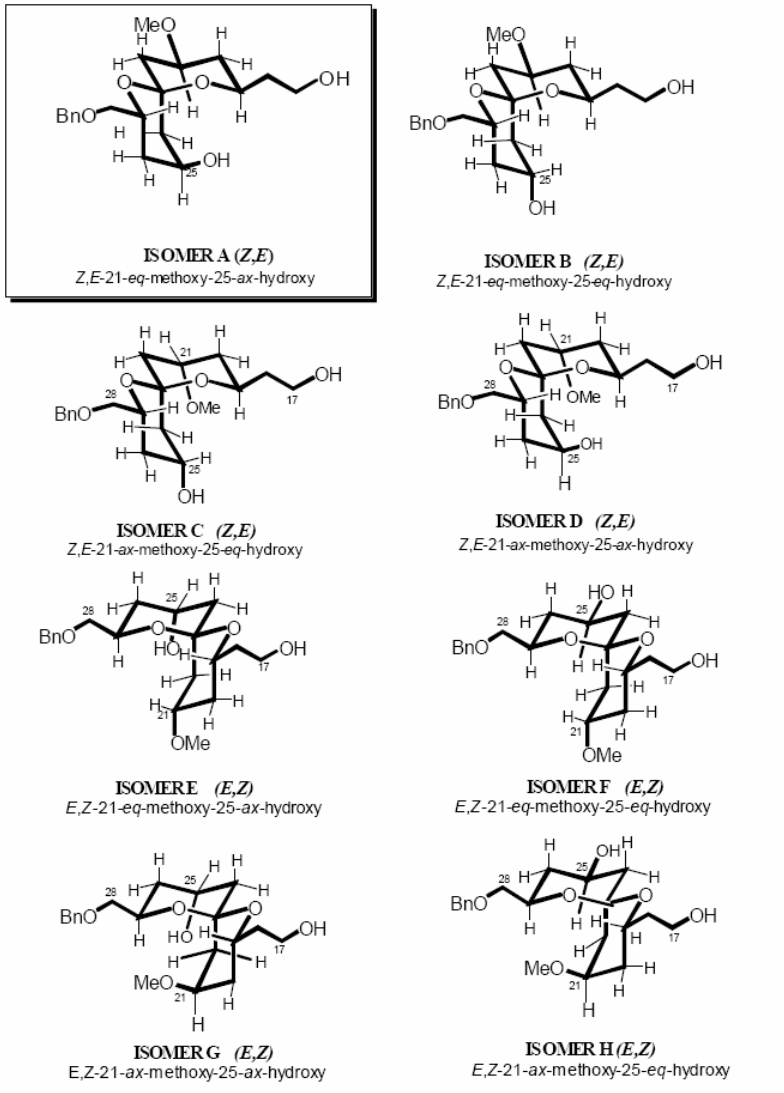


Figure 4. Isomers A–H of spiroacetal system 24

The ^{1}H and ^{13}C NMR chemical shifts for isomers **A**–**G** are presented in Tables 1 and 2, and a summary of ^{1}H shifts with coupling patterns and nOe data is given in Table 3.

Table 1 ^{1}H Chemical shift assignments

Assignment	Isomer **A**	Isomer **B**	Isomer **C**	Isomer **D**	Isomer **E**	Isomer **F**	Isomer **G**	Cinachyrolide A*
H17A	3.52	3.48	3.60	3.60	3.60	3.60	3.65	
H17B	3.81	3.62	3.64	3.73	3.68	3.68	3.69	
H18A	1.43-1.50	1.38	1.38-1.52	1.54	1.43-1.51	1.44	1.45-1.58	2.84
H18B	1.43-1.50	1.57	1.72-1.90	1.74	1.55-1.63	1.60	1.45-1.58	2.87
H19ax	3.56	3.35	4.01	4.12	4.13	4.26	4.58	4.11
H20ax	1.15	1.15	1.38-1.52	1.49	1.18	1.15	1.21	1.03
H20eq	1.68	1.64	1.38-1.52	1.49	1.78-1.88	1.81	1.59	2.01
H21	3.09	3.16	3.38	3.42	3.69	3.58	3.22	3.57
OMe21	3.04	3.04		2.96	3.13	3.11	3.12	
H22ax	1.70	1.70	1.72-1.90	1.69	1.26	1.17	1.19	1.20
H22eq	2.03	2.14	1.72-1.90	1.81	2.44	2.21	2.27	2.06
H24ax	0.97	1.04	1.19	1.25	1.35-1.40	1.55	1.33-1.41	1.65
H24eq	1.94	2.24	2.64	2.40	1.78-1.88	1.72	1.76	2.4
H25	3.76	3.85	3.98	3.92	3.88	3.58	3.90	4.03
HO25					2.13			
H26ax	1.43-1.50	1.32	1.26	1.40	1.35-1.40	1.36	1.33-1.41	1.65
H26eq	1.58	1.90	1.72-1.90	1.64	1.78-1.88	1.67	1.83	1.65
H27ax	4.69	4.28	4.23	4.67	4.20	3.50	4.24	5.1
H28A	3.41	3.38	3.33	3.36	3.38	3.30	3.55	5.4
H28B	3.47	3.46	3.45	3.45	3.60	3.45	3.64	

Note: The equatorial and axial protons on the carbon adjacent to the spiro centre in the E ring of the *ZE/EZ* system **24** are displaced markedly down- and up-field respectively**.**

Table 2. ^{13}C Chemical Shift Assignments

Assignment	Isomer **B**	Isomer **C**	Isomer **D**	Isomer **E**	Isomer **F**
C17	60.10	60.60	59.80	60.05	60.30
C18	39.00	38.90	38.80	39.02	38.80
C19	69.00	68.50	68.20	67.87	68.40
C20	38.10	35.40	35.00	37.99	37.90
C21	74.20	23.10	72.50	73.00*	64.40
C22	42.90	39.80	40.80	42.00	44.40
C23	100.20	99.80	99.10	99.92	99.60
C24	39.40	42.70	38.00	34.94**	38.50
C25	64.20	64.50	64.60	61.69	72.80
C26	38.50	38.00	35.10	44.69**	36.40
C27	69.60	69.20	64.70	70.68	70.50
C28	73.63	73.90	74.00	72.62	73.50
OMe	55.30	55.80	55.50	55.13	55.10
ArCH2O	73.61	73.50	73.30	73.04*	73.20
Ar	127.80		127.76	127.85	127.88
Ar	128.10		128.13	128.12	127.24(3C)
Ar	128.30		128.48	128.61	128.59
Ar	128.5(2C)		139.00	138.96	139.06
Ar	128.70				

Asterisked shifts may be interchangeable.

Table 3

Isomer **A**			
Proton shift	Assignment	Coupling pattern	nOe Correlations
3.52	H17A	m	
3.81	H17B	m	
1.43-1.50	H18A	m, overlap with H18B and H26ax	
1.43-1.50	H18B	m, overlap with H18A and H26ax	
3.56	H19ax	m	H24eq, H18A&B, H20eq
1.15	H20ax	ddd, *J*=11.6, 11.6, 11.6 Hz	
1.68	H20eq	m, overlap with H22ax	
3.09	H21ax	m	H24eq, H22eq, H20eq
3.04	OMe21	s, 3H	
1.70	H22ax	dd, *J*~12.2, 12.2 Hz overlap with H20eq	
2.03	H22eq	ddd, *J*=12.4, 4.5, 1.65 Hz	
0.97	H24ax	dd, *J*=14.6, 3.68 Hz	

Table 3. Continued

1.94	H24eq	dm, *J*~14.7, 4.3, 1.8 Hz	H19ax, H21ax, H25eq
3.76	H25eq	m	H24ax&eq, H26ax&eq
	HO25		
1.43-1.50	H26ax	m, overlap with H18A and H18B	
1.58	H26eq	dm, *J*~12 Hz	
4.69	H27ax	m	H26eq
3.41	H28A	dd, *J*=9.9, 4.95 Hz	
3.47	H28B	dd, *J*=9.8, 4.80 Hz	
Isomer **B**			
Proton shift	Assignment	Coupling Pattern	nOe effects
3.48	H17A	m	
3.62	H17B	m	
1.38	H18A	m	
1.57	H18B	m	
3.35	H19ax	m	H18A&B, H24eq, H25, H21
1.15	H20ax	ddd, *J*=11.7, 11.7, 11.7 Hz	
1.64	H20eq	dm, *J*~12.4, ~1.9 Hz	
3.16	H21ax	m	H19, H20eq, H22eq, H24eq
3.04	OMe21	s, 3H	
1.70	H22ax	dd, *J*~12.2, 11.7 Hz	
2.14	H22eq	ddd, *J*=12.4, 4.6, 1.8 Hz	
1.04	H24ax	dd, *J*=13, 11.4 Hz	
2.24	H24eq	ddd, *J*~12.9, 4.3, 1.8 Hz	H19, H21, H25
3.85	H25ax	m	H27, H19, H24 eq, H26eq
	HO25		
1.32	H26ax	ddd, *J*=11.9, 11.9, 11.9 Hz	
1.90	H26eq	dm, *J*~12.2, 2.1 Hz	
4.28	H27ax	m	H25, H26eq, H28A&B
3.38	H28A	dd, *J*=9.8, 5.1 Hz	
3.46	H28B	dd, *J*=9.8, 5.1 Hz	
Isomer **C**			
Proton shift	Assignment	Coupling pattern	nOe Effects
3.60	H17A	m (app. Quintet)	
3.64	H17B	m, br	
1.38-1.52	H18A	m, superimposed on H20ax & H20eq	
1.72-1.90	H18B	m, superimposed on H22ax & H22eq & H26eq	
4.01	H19ax	m,	H24eq, H18A&B,

Table 3. Continued

1.38-1.52	H20ax	m, superimposed on H18A & H20eq	
1.38-1.52	H20eq	m, superimposed on H18A & H20ax	
3.38	H21eq	m, app quintet *J*=4.2 Hz	H20eq&ax, H22eq&ax
	OMe21		
1.72-1.90	H22ax	m, superimposed on H18B & H22eq & H26eq	
1.72-1.90	H22eq	m, superimposed on H18B & H22ax & H26eq	
1.19	H24ax		
2.64	H24eq	dm, *J*~13.1 Hz	H19
3.98	H25ax	M	H24eq, H27, H19
	HO25		
1.26	H26ax		
1.72-1.90	H26eq	m, superimposed on H18B & H22ax & H22eq	
4.23	H27ax	M	H25, H28A&B
3.33	H28A	dd, *J*=9.9, 4.4 Hz	
3.45	H28B	dd, *J*=9.9, 5.7 Hz	
Isomer **D**			
Proton Shift	Assignment	Coupling Pattern	nOe effects
3.60	H17A	M	
3.73	H17B	M	
1.54	H18A	M	
1.74	H18B	M	
4.12	H19ax	M	H24eq, H18A&B, H20eq
1.49	H20ax	m, superimposed on H20eq	
1.49	H20eq	m, superimposed on H20ax	
3.42	H21eq	M	H20ax&eq, H22ax&eq
2.96	21OMe	S	
1.69	H22ax	dd, *J*=13.7, 5 Hz	
1.81	H22eq	dd, *J*=13.7, 4.4 Hz	
1.25	H24ax	dd, *J*=14.7, 3.6 Hz	
2.40	H24eq	dt, *J*=14.8, 2.3, 2.3 Hz	H19ax, H25eq, H24ax
3.92	H25eq	narrow m	H24eq&ax, H26ax&eq)
	25OH		
1.40	H26ax	td, *J*=12.7, 2.9 Hz	
1.64	H26eq	dm, *J*=13 Hz	
4.67	H27ax	M	H26eq&ax, H28A&B
3.36	H28A	AB of ABX, *J*=10, 4.2 Hz	

Table 3. Continued

3.45	H28B	AB of ABX, *J*=10, 5.8 Hz	
4.36	$ArCH_2O$	s, superimposed	
4.36	$ArCH_2O$	s, superimposed	
7.04-7.61	Aromatics x 5	M	
Isomer **E**			
Proton shift	Assignment	Coupling pattern	nOe effects
3.60	H17A	m, superimposed on H28B	
3.68	H17B	M	
1.43-1.51	H18A	M	
1.55-1.63	H18B	M	
4.13	H19ax	m, app. Tq	H21, H18, H20eq
1.18	H20ax	br q, *J*~11.55 Hz	
1.78-1.88	H20eq	m, overlap with H26eq, H24eq	
3.69	H21ax	tt, *J*=11.2, 4.4 Hz	H19, H27, H22eq
3.13	21OMe	S	
1.26	H22ax	dd, *J*=12.7, 11.35 Hz	H24eq, H20ax
2.44	H22eq	ddd, *J*=12.7, 4.4, 2.0 Hz	
1.35-1.40	H24ax	m, superimposed on H26ax	H25
1.78-1.88	H24eq	m, overlap with H20eq, H26eq	H25
3.88	H25eq	M	H24ax/H26ax, H24eq/H26eq
2.13	25OH		
1.35-1.40	H26ax	m, superimposed on H24ax	H25
1.78-1.88	H26eq	m, overlap with H20eq, H24eq	H25
4.20	H27ax	m, app. Quintet	
3.38	H28A	AB of ABX, *J*=9.9,6.2 Hz	
3.60	H28B	M	
4.38	$ArCH_2O$	AB JAB=12.3	
4.42	$ArCH_2O$	AB JAB=12.3	
7.11-7.31	Aromatics x 5	M	
Isomer **F**			
Proton shift	Assignment	Coupling Pattern	nOe effects
3.60	H17A	m	
3.68	H17B	m	
1.44	H18A	m	
1.60	H18B	m	
4.26	H19ax	m	H20eq, H18A, H21
1.15	H20ax	br q, *J*~11.9 Hz (overlap withH24ax)	

Table 3. Continued

1.81	H20eq	dm, *J*~12Hz + small coupling~2.3Hz	
3.58	H21ax	m, superimposed on H25	
3.11	21OMe	s	
1.17	H22ax	dd, *J*=12.8, 11.3 Hz (overlap with $H20_{ax}$)	
2.21	H22eq	ddd, *J*=12.8, 4.4 , 1.9 Hz H27, H21/H25	
1.55 H24ax dd, *J*=13.0, 8.2 Hz			
1.72	H24eq	ddd, *J*=12.9, 4.9, ~1 Hz	
3.58	H25ax	m, superimposed on H21	
25OH			
1.36	H26ax	ddd, *J*~12, 12, 12 Hz	
1.67	H26eq	dm, *J*~12 Hz	
3.50	H27ax	br m	H22eq, H26eq, H25
3.30	H28A	AB of ABX, *J*=9.9,4.4 Hz	
3.45	H28B	AB of ABX, *J*=9.8, 5.9 Hz	
4.4	$ArCH_2O$	AB JAB~12 Hz	
4.4	$ArCH_2O$	AB JAB~12 Hz	
7.11-7.41	Aromatics x 5	m	
Isomer **G**			
Proton shift	Assignment	Coupling pattern	nOe effects
3.65	H17A	m	
3.69	H17B	m (app. Sextet)	
1.45-1.58	H18A	m, superimposed on H18B	
1.45-1.58	H18B	m, superimposed on H18A	
4.58	H19ax	br m,	*J*~14 Hz + small coupling
1.21	H20ax	br t, *J*~14 Hz	
1.59	H20eq	br d, *J*~13.5 Hz	
3.22	H21eq	app. quintet *J*=3.4, 3.4, 3.4, 3.4 Hz	H22ax&eq, H20ax&eq
3.12	21OMe	s	
1.19	H22ax	dd, 15.5, 3.8 Hz	
2.27	H22eq	ddd, *J*=15.4, 2.2, 3.0 Hz	H27
1.33-1.41	H24ax	m, superimposed on H26ax	
1.76	H24eq	ddd, *J*=13.0, 4.5, 1.0 Hz	
3.90	H25eq	m, app. Octet, coupled to OH also	H24ax&eq and H26ax&eq
25OH			
1.33-1.41	H26ax	m, superimposed on H24ax	

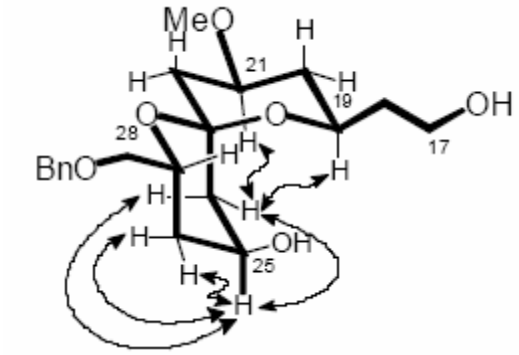
Table 3. Continued

1.83	H26eq	br dt, *J*=13.7, 5.3, 5.3 Hz	
4.24	H27ax	m, app. Quintet, *J*=6 Hz	H22eq
3.55	H28A	AB of ABX, *J*=9.8, 7.0 Hz	
3.64	H28B	AB of ABX, *J*=9.8, 5.35 Hz	

Stereochemical Assignments

High field nOe studies were used to assign the configurations of the isomers **A**–**G**, with emphasis on the relative configuration of the CD ring and the *axial* or *equatorial* orientations of the methoxy and hydroxy groups at C21 and C25 respectively. None of the isomers **A**–**G** appeared to favour a conformation in which either of the benzyloxymethyl or 2N –hydroxyethyl groups was predominantly *axially* orientated.

Isomer **A:** *Z,E*–(19R,21S, 23S,25S,27R)–**24**. Examination of the spectra of isomer **A** confirmed its stereochemical correlation with the natural CD spiroacetal fragment.[21] Specifically the CD ring configuration was assigned as *Z,E* with the C21 methoxy and C25 hydroxy groups concluded to be *equatorially* and *axially* disposed respectively, based on observed nOe's. (The Z $_{OH}$ or E nature of ring C (that bears the methoxy group) is always given first.) A strong nOe between the equatorial proton on C24 and the C19 proton together with the downfield shift of the C27 proton relative to the C19 proton (due to the deshielding 1,3–syn interaction with the axial C–O bond of ring C)[19] strongly supported the *Z,E* configuration for the CD ring system.



Isomer A

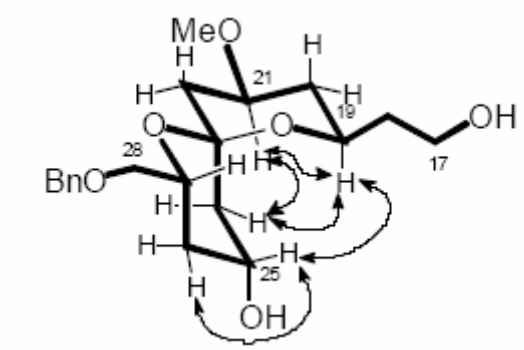
The C21 methoxy group was *equatorially* oriented on the basis of the cross ring nOe's of the C21 proton and the *equatorial* proton on C24 and lack of any nOe's to the adjacent ring protons at C20 and C22. In constrast, the hydroxyl substituent on C25 was disposed *axially* because nOe's are detected between the C25 proton and adjacent *axial* and *equatorial* protons on C24 and C26, but not detected to either C19 or C27 (ie. no *diaxial* interaction or cross ring proximity).

These stereochemical assignments for **A** were consistent with the patterns of proton–proton coupling, interpreted with the aid of 2D COSY and HSQC experiments. The analyses that follow for the isomers **B** through **G** provided cohesive and consistent sets of assignments and are based

on the types of interpretations outlined above.

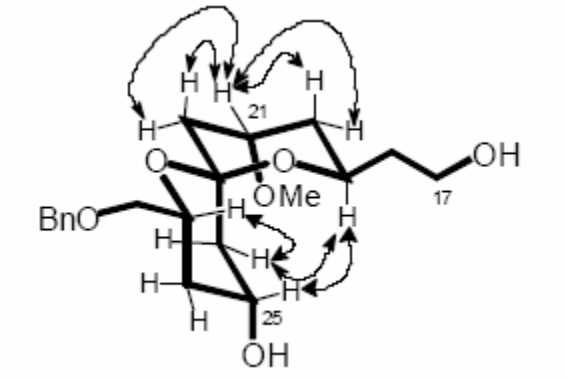
Isomer **B**: *Z,E*–(19R,21S,23S,25R,27R)–24. Analyses of spectra of isomer **B** identified the same strong nOe between a proton on C24 and C19 as for Isomer **A**. When considered with the downfield shift of the proton on C27 relative to the C19 proton, the assignment of the CD ring system as *Z,E* configured is strongly indicated.

The C21 methoxy was assigned as *equatorial*, from the cross ring nOe of its *geminal* proton to the *equatorial* C24 proton, and absence of nOe's to adjacent protons. Similarly, the C25 hydroxy substituent was also assigned as *equatorial*, based on observed *diaxial* and cross ring nOe's of H25 with protons on C27 (not indicated on accompanying structure) and C19, respectively.



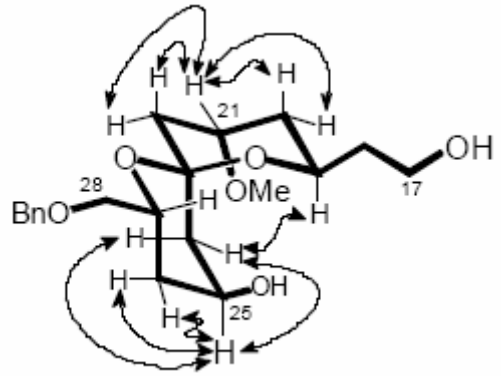
Isomer B

Isomer **C**: *Z,E*–(19R,21R,23S,25R,27R)–**24**. The spectra of isomer **C** exhibited the characteristic nOe between the protons on C24 and C19, and the downfield shift of the proton on C27 relative to the C19 proton, as in isomers **A** and **B**. Consequently the CD ring was designated as *Z,E*. The C21 methoxy group was deemed to be axially oriented from the nOe's of its geminal proton to adjacent axial and equatorial protons on C20 and C22 and absence of nOe's to either C19 or C24 (ie. no diaxial interaction or cross ring proximity). The observed diaxial and cross ring nOe's of the C25 proton to the C27 (not shown) and C19 protons respectively, supported assignment of the C25 hydroxyl substituent as *equatorial*.

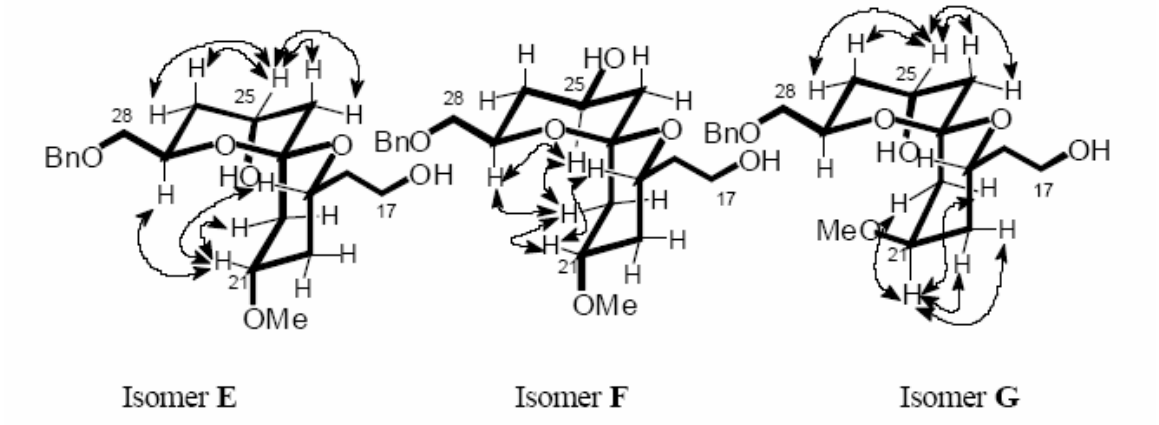


Isomer C

Isomer **D**: *Z,E*–(19R,21R,23S,25S,27R)–**24**. The *Z,E* nature of the CD ring system of isomer **D** was also based upon the presence of a strong nOe between the *equatorial* proton on C24 and the C19 proton, together with the downfield shift of the proton on C27 relative to the C19 proton, as described above for isomers **A**, **B** and **C**. As with Isomer **C**, the C21 methoxy was concluded to be *axial* because of the nOe's between its *geminal* proton (H21) and adjacent *axial* and *equatorial* protons on C20 and C22 rather than with either C19 or C24 (ie. no *diaxial* interaction or cross ring proximity). The C25 $_{\text{Isomer D}}$ hydroxyl was assigned as *axial* from the observed nOe's between the C25 proton and adjacent *axial* and *equatorial* protons on C24 and C26.



Isomers **E, F** and **G** are located in the alternative *E,Z* system (for the C and D rings respectively), and again considerations of nOe's, chemical shifts and coupling patterns lead to the stereochemical conclusions for these isomers. These are not discussed in detail, but the important nOe's are shown on the structures below, and full listings of NMR data are presented in the Tables.



Isomer **E** Isomer **F** Isomer **G**

Conclusions

Seven of the eight possible isomers of the (*Z,E) (E,Z*) spiroacetal system representing the CD fragment (C17–C28) of spongistatin 1 incuding the one whose stereochemistry correlates with

the natural fragment, have been characterised by high– field NMR measurements. These provide useful sets of comparison data for this system and similar oxygenated spiroacetal sub–structures. Stereocontrol at C19 and C27 was incorporated using Brown's allylation methodology. The Sharpless asymmetric dihydroxylation procedure has been explored as a vehicle for the introduction of further stereocontrol. For example, incorporation of epoxide **20** into the general scheme would control the configuration at C21, and similarly, control at C25 could be enforced.

Experimental Section

General Procedures. All operations involving air–sensitive reagents were performed under an inert atmosphere using syringe and cannula techniques. Glassware was assembled hot, evacuated and purged with nitrogen. THF and ether were distilled under nitrogen from sodium benzophenone ketyl. DCM was distilled from calcium hydride. ^{1}H and ^{13}C NMR spectra were recorded at the frequencies stated on a Bruker DMX750, AMX500, AMX400 or AC200F NMR spectrometer. Unless otherwise stated all ^{1}H NMR spectra were referenced to residual $CHCl_3$ (δ 7.24) and all ^{13}C NMR spectra were referenced to the central component of the $CDCl_3$ triplet at δ 77.0. 750 MHz ^{1}H and 187 MHz ^{13}C spectra were obtained on sub–milligram samples using NMR microtubes purchase from the Aldrich chemical company. Reverse phase HPLC was performed on a Dynamax 60A C18 column. GC–MS analyses were carried out on a Hewlett Packard HP5890 GC using a 30mx0.25mm BP5 column and a HP5970 mass selective detector. High resolution mass spectra were obtained from a Kratos MS25RFA instrument. Optical rotations were measured on a Perkin–Elmer 241 MC polarimeter using the sodium D line (589nm).

2–Benzyloxyethanal (6) was prepared from the bis–benzyl ether of 2–butene–1,4– diol 5 as described elsewhere.[15] The product was purified by flash chromatography (silica; 30% EtOAc in hexane) to provide the required aldehyde **6**, (9.5 g, 93%). ^{1}H NMR: ($CDCl_3$ 200 MHz) δ 4.09 (s, 2H), 4.62 (s, 2H), 7.35 (m, 5H), 9.35 (s, 1H). ^{13}C NMR: 73.60, 75.20, 128.00 (2C), 128.17, 128.55 (2C), 136.73, 200.43 (CHO).

Bis–(2–isocarenyl)–allylborane (7). To a cooled (–10° C) and stirred solution of borane–dimethylsulfide complex (10 mL, 100 mmol) in dry THF (200 mL) under nitrogen, 2–carene (30g, 200 mmol) was added dropwise with the solution maintained at –10°C throughout the addition. The reaction mixture was then stored at 0° C for 24 h, and the product appeared as white needles. The THF was decanted using a cannula and the solid was washed with cold ether (0° C, 3 x 100 mL) with the exclusion of air, and then dried under vacuum (1 mbar, 1 h). This bis–(2–isocarenyl)borane hydride (12.39 g, 43.3 mmol) was then transferred (glove–bag) into a dry, round bottomed flask and ether (17 mL) was added and the mixture stirred at 0° C. Dry methanol (spectrometric grade, 3 Δ sieves) (3.6 mL) was introduced dropwise over 10 minutes, after which the reaction mixture was stirred for a further 3 h at 0° C. The solvent and excess

methanol were then evaporated (1 mmbar, 3 hours). After diluting with dry ether (17 mL), the reaction mixture was cooled to –78° C and allyl magnesium bromide (41.5 mL, 1.0 M, 41.5 mmol) was added dropwise from a pressure equalising dropping funnel. The reaction mixture was stirred for a further 15 minutes at –78° C and then for 1 h at room temperature. The product was used immediately.

(*R*)–5–benzyloxy–4–hydroxypentene (8). Aldehyde **6** (1.62 g, 10.77 mmol) was dissolved in precooled (–78° C) dry ether, and added *via* cannula to freshly prepared allyl borane **7** cooled to –78° C. After 2 hours at –78° C, the reaction was quenched with 3 N NaOH (5 mL) followed by 10 mL of 30% aq. H_2O_2 and left in the freezer overnight. After a further 2–3 h of reflux, the organic layer was separated, and the aqueous layer extracted after treatment with brine. The organic layer was dried ($MgSO_4$) then purified by flash chromatography (silica; 20% EtOAc in hexane) to provide the desired product **8** (1.22 g, 61.5%). $[\alpha]_D^{23}$ –1.8 (c, 2.4, Et_2O). (95% *ee* by Mosher ester analysis). GC/MS: *m/z* 192 (M^+), 174 (2%). 151 (2), 139 (2), 107 (8), 105 (8), 92 (22), 91 (100). ^{1}H NMR: (CDCl3, 200 MHz) d 7.37–732 (m, 5H), 5.90–5.77 (m, 1H), 5.16–5.08 (m, 2H), 4.56 (s, 2H), 3.94–3.85 (m, 1H), 3.55–3.51 (dd, *J* = 10, 3.7 Hz, 1H), 3.41–3.36 (dd, *J* = 9.7, 7.4 Hz, 1H), 2.37 (d, *J* = 3.5 Hz, 1H), 2.28 (t, *J* = 6.6 Hz, 2H). ^{13}C NMR: ($CDCl_3$, 50 MHz) d 134.21, 132.24, 129.84, 128.21, 127.21, 123.21, 74.01, 73.82, 69.86. 38.72. HRMS: $C_{12}H_{16}O_2Na$ requires 215.1048. Measured, 215.1049.

1–Benzyloxy–4,5–epoxypentan–2–ol (9). To the alkene **8** (1.0 g, 5.2 mmol), stirred in DCM (10 mL) at room temperature, was added *m*CPBA (2.2 g of a 60% mixture, 1.5 equivs), in one portion. The reaction was monitored by TLC (silica, 50% EtOAc in hexane; anisaldehyde) and after 5 h a further 0.5 g of *m*CPBA was added. Stirring was continued for an hour after which time the reaction was quenched by the addition of saturated $NaHCO_3$ solution (10 mL). The aqueous phase was separated and extracted with DCM (3 x 20 mL). The combined organic layers were combined, dried ($MgSO_4$) and concentrated under reduced pressure. The residue was purified by flash column chromatography (silica, 50% EtOAc in hexane) to yield the desired epoxide **9**, (0.80 g, 74%). GC/MS: (M^+, O) 139 (6%), 107 (13), 105 (8), 92 (26), 91 (100), 87 (12), 69 (11). ^{1}H NMR: ($CDCl_3$, 200 MHz) d 1.42 – 1.93 (m, 2H), 2.51 (m, 1H), 2.67 (br s, 1H, OH), 2.78 (m, 1H), 3.11 (m, 1H) 3.35 – 3.52 (m, 2H), 4.05 (m, 1H), 4.56 (s, 2H), 7.25– 7.35 (m, 5H). ^{13}C NMR: ($CDCl_3$) 50 MHz) Two diastereomers d 35.92, 36.07, 46.62, 47.08, 49.59, 49.68, 68.28, 73.35, 73.91, 74.23, 127.70 (4C), 127.78 (2C), 128.42 (4C), 137.79 (2C), 150.35, 150.40. HRMS: $C_{12}H_{16}O_3$ requires 208.1099. Measured, 208.1103.

1–Benzyloxy–4,5–epoxy–2–triethylsiloxy–pentane (10). The alcohol **9** (1.1 g, 5.3 mmol) was dissolved in DCM (50 mL) under N2 and cooled to –78° C. 2,6–Lutidine (0.84 g, 0.923 mL) 7.9 mmol) was added *via* syringe and the resulting mixture stirred for 15 minutes at –78° C. Triethylsily triflate (1.68 g, 143 mL, 6.35 mmol) was then added *via* syringe and the reaction monitored by tlc (silica, 10% EtOAc in hexane) and quenched at –60° C with water (30 mL) when no starting material remained. The reaction was diluted with DCM (20 mL). The aqueous layer was separated and extracted with DCM (2 x 50 mL), dried ($MgSO_4$) and concentrated under reduced pressure. The residue was purified *via* flash column chromatography (silica, 7%

EtOAc in hexane) to yield protected epoxy diol, **10**, (0.5 g, 34%). GC/MS: *m/z* 293 (M^+ –29, 0.2%), 221 (2), 210 (3), 187 (3), 185 (3), 173 (3), 146 (4), 145 (31), 117 (20), 115 (12), 92 (10), 91 (100). ^{1}H NMR: ($CDCl_3$, 200 MHz) Two diastereomers (1:1) d 0.51 (1, *J* = 7.7 Hz, 3 x 2H, $SiCH_2CH_3$) 0.55 (q, *J* = 7.7 Hz, 3 x 2H, $SiCH_2CH_3$) 0.88 (t, *J* = 7.7 Hz, 3 x 3H, $SiCH_2CH_3$), 1.59 – 1.65 (m, 2 x 1H), 1.68 (t, *J* = 5.7 Hz, 2 x 1H), 2.37 (dd, *J* = 5.2, 2.7 Hz, 1H), 2.40 (dd, *J* = 5.2, 2.7 Hz, 1H), 2.64 (dd, *J* = 5, 4 Hz, 1H), 2.68 (dd, *J* = 5,4 Hz, 1H), 2.96 – 2.98 (m, 2 x 1H), 3.32 – 3.45 (m, 2 x 2H), 3.93 – 4.01 (m, 2 x 1H) 4.44 (s, 2H $PhCH_2$) 7.16 – 7.25 (m, 2 x 5H aromatic). ^{13}C NMR: ($CDCl_3$, 100 MHz) Two diastereomers d 4.86 (3C), 4.89 (3C), 6.74 (3C, 6.75 (3C), 37.97, 38.05, 46.70, 47.55, 49.26, 49.46, 69.24, 69.50, 73.27, 73.35, 74.28, 74.63, 127.49, 127.50, 127.58 (2C), 127.59 (2C), 128.25 (2 x 2C), 138.20, 138.27.

4–(*p*–Methoxybenzyloxy)–but–1–ene. But–3–enol was converted to the p– methoxybenzyl ether in the normal way with KH and then 4–methoxy–benzylchloride, in 89% yield. The crude product (12 g, 89%) was used without further purification. GC/MS: *m/z* 192 (M^+, 3.7%), 161 (5.2), 136 (5.8), 121 (100), 91 (6.8), 89 (3.8), 78 (14.6), 77 (14.8), 55 (6.4). ^{1}H NMR: ($CDCl_3$, 200 MHz) d 2.70 (qt, *J* = 6.7, 1.4 Hz, 2H), 3.84 (t, *J* = 6.7 Hz, 2H), 4.13 (s, 3H, OMe), 4.79 (s, 2H, H5), 5.34 – 5.49 (m, 2H), 6.07 – 6.20 (m, 1H), 7.22 (d, *J* = 8.7 Hz, 2H), 6.83 (d, *J* = 8.7 Hz, 2H). ^{13}C NMR: ($CDCl_3$, 50 MHz) 34.16, 55.15, 69.21, 72.46, 113.67 (2C), 116.23, 129.16 (2C), 130.46, 135.24, 159.06. HRMS: $C_{11}H_{17}O_2$ (M–CH_3) requires 177.0914. Measured, 177.0917.

3–(4–Methoxybenzyl)oxypropanal (13). The above alkene (5.4 g, 28 mmol) was treated with O_3 in the normal manner, but importantly, the reaction was monitored by TLC (silica; 30% EtOAc in hexane; product, R_f 0.27) and quenched immediately when no starting material remained. The product was purified by flash column chromatography (silica, 25% EtOAc in hexane) to give 3–(4–methoxybenzyl)oxypropanal (1.3 g, 24%). (This low yield could be improved by more rapid work–up). GC/MS: *m/z* 194 (M^+, 10.1%) 137 (63.6), 135 (6.5), 122 (9.7), 121 (100), 109 (15.0), 94 (8.4), 91 (10.9), 78 (19.5), 77 (27). ^{1}H NMR: ($CDCl_3$; 200 MHz) d 2.65 (td, *J* = 6.1, 1.9 Hz, 2H), 3.76 (t, *J* = 6.1 Hz, CH_2CH_2 overlapping with d 3.77), 3.77 (s, 3H, OMe), 4.44 (s, 2H, $PhCH_2$), 6.85 (d, *J* = 8.7 Hz, 2H), 7.22 (d, *J* = 8.7 Hz, 2H), 9.27 (t, *J* = 1.9 Hz, CHO). ^{13}C NMR: ($CDCl_3$ 50 MHz) d 43.82 (2C), 55.22 (OMe), 63.47, 72.86, 113.78 (2C), 129.30 (2C), 129.87, 159.25, 201.20.

***Bis*–(4–isocarenyl)–allylborane (14).** *Bis*–(4–isocarenyl)–allylborane was prepared as described for *bis*–(4–isocarenyl)–borane hydride. The product was not isolated but used immediately.

4–Methoxybenzyl protected enediol (15). 3–(4–Methoxybenzyloxy)propanal **13** was treated with borane **14** in identical fashion as for 2–benzyloxyethanol **6**, to provide homallyl alcohol **15** in 40% yield, after column chromatography (silica, 20% EtOAc in hexane). GC/MS: *m/z* 236 (M^+, 1%), 189 (1), 163 (1), 137 (33), 136 (10), 122 (10), 121 (100), 91 (6). ^{1}H NMR: ($CDCl_3$, 200 MHz) d 1.91 (m, 2H), 2.40 (dd, 2H), 3.31 (t, *J* = 5.7 Hz, 2H), 3.72 (s, 3H, OMe), 4.34 (s, 2H), 4.89 – 4.92 (m, 1H), 5.04 – 5.12, (m, 2H), 5.23 (m, 1H), 6.91, (d, *J* = 8.7 Hz, 2H), 7.32 (d, *J* = 8.7 Hz, 2H). ^{13}C NMR: ($CDCl_3$) d 38.73, 41.81, 55.13, 68.47, 70.24, 113.71, 117.36, 129.20, 129.91, 134.81, 159.14. HRMS: $C_{14}H_{20}O_3Na$ requires 259.1329. Measured, 259.1307.

4–Methoxybenzylidene acetals (18). Alcohol **15** (1.5 g, 6.36 mmol) was added to a suspension

of dry, powdered 3Δ sieves (8g) in DCM (20 mL) under N_2. After stirring for 30 minutes, DDQ (2.16 g) was added, and after stirring for a further 10 minutes, the reaction was complete as judged by TLC (silica, 30% EtOAc in hexane). The reaction mixture was filtered through celite and washed with 100 mL DCM. The filtrate was washed with sodium bicarbonate (3 x 150 mL), dried ($MgSO_4$) and concentrated under reduced pressure. The crude product was then purified by flash chromatography (15% EtOAc in hexane) to give the 4–methoxybenzylidene acetals **18** (0.50 g, 33%). GC/MS: *m/z* 234 (M^+, 21%) 233 (23), 203 (3), 193 (33), 137 (34), 136 (41), 135 (100), 121 (20), 109 (21), 108 (22), 107 (10) 94 (11), 92 (10). ^{1}H NMR: ($CDCl_3$, 200 MHz) (major + minor isomers). d 1.47 – 1.56 (m, 1H), 1.69 – 1.84 (m, 1H), 2.24 – 2.50 (m, 2H), 3.78 (s, 3H, OMe), 3.81 – 3.93 (m, 2H, OCH_2) 4.09 – 4.29 (m, 1H, OCH), 5.04 – 5.18 (m, 2H, $CH=CH_2$), 5.77 – 5.98 (m, 1H, $CH=CH_2$), 5.87 (dm, *J* = 8.8 Hz), 7.42 (dm, *J* = 8.8 Hz, 2H). ^{13}C NMR: d 30.68, 40.38, (C3 & C5), 55.19, (OMe), 66.89, 76.51, 101.00, 113.49, 117.26, 127.26, 131.28, 133.86, 159.77.

Asymmetric dihydroxylation of 18 to protected tetrol, (19). This was conducted in the usual way using *t*–butanol/water (1:1) under N_2, with AD–mix–∀. The reaction mixture was stirred at 0° C and monitored by TLC (silica, 30% EtOAc in hexane). After 3 days the reaction was quenched by the addition of 20% $Na_2S_2O_3$ aqueous solution and the reaction stirred for a further hour. After concentration to remove *t*–butanol, brine was added and the mixture extracted with ether (5 x 5 mL). The combined ether layers were dried ($MgSO_4$) concentrated and flash chromatographed (silica; 5% MeOH in DCM) to give diol (19) (0.75 g, 44% yield). ^{1}H NMR: ($CDCl_3$, 400 MHz) (Isomer Mixture). d 1.44 – 1.55 (m, 2 x 1H), 1.68 – 1.72 (m, 2 x 2H), 1.8 – 1.94 (m, 2 x 1H), 3.44 – 3.50 (m, 2 x 1H), 3.56 – 3.66 (m, 2 x 1H), 3.93 – 4.30 (m, 2 x 2H), 4.11 (m, 2 x 1H), 4.21 (m, 2 x 1H), 5.45 (s, 1H, major diastereomer), 5.48 (m, 1H, minor diastereomer), 6.85 (dm, *J* = 8.8 Hz, 2 x 2H), 7.35 (dm, *J* = 8.8 Hz, 2H minor isomer), 7.36 (dm, *J* = 8.8 Hz) major diastereomer. ^{13}C NMR: Major diastereomer: d 31.21, 38.84, 55.27, 66.89, 66.94, 68.68, 74.31, 101.17, 133.64 (2C), 127.24 (2C), 131.05, 159.96. Minor diastereomer: d 31.41, 38.98, 55.27, 66.50, 66.93, 71.00, 76.81, 101.20, 113.72 (2C), 127.25 (2C), 130.73, 160.07. HRMS: $C_{14}H_{20}O_5$ requires 268.13107. Measured, 268.1312.

Monotosylation of diol (19). This was conducted in the usual way with pyridine as solvent, tosyl chloride (0.34 g, 1.8 mmol) was added in one portion. The reaction was monitored and by TLC (silica; 50% EtOAc in hexane). After stirring at room temperature for 5 hours the reaction was stored overnight at 0° C. More tosyl chloride (100 mg) was added and the reaction was stirred for a further 6 h at room temperature. The reaction mixture was then diluted with ether (20 mL), washed with saturated aqueous $CuSO_4$ (3 x 20 mL), then with saturated aqueous NaHCO3 (2 x 20 mL), dried ($MgSO_4$) and concentrated under reduced pressure. The residue was purified by flash column chromatography (silica; 40% EtOAc in hexane) to provide the mono tosylate, (0.5 g, 66%). ^{1}H NMR: ($CDCl_3$, 400 MHz) (Two diastereomers). d 1.42 – 1.51 (m, 2 x 1H, CH_2), 1.69 (dd, *J* = 6.44, 5.56 Hz, 2 x 1H), 1.75 – 1.87 (m, 2 x 2H), 2.42 (s, 2 x 3H, PhMe), 3.78 (s, 2 x 3H, OMe), 3.89 – 4.19 (m, 2 x 5H), 4.21 (ddd, *J* = 11.44, 4.92, 1.04 Hz, 2 x 1H), 5.42 (s, 1H, major diastereomer), 5.44 (s, 1H, minor diastereomer), 6.84 (dm, *J* = 8.8 Hz, 2H,

minor diastereomer), 6.85 (dm, *J* = 8.5 Hz, 2H, major diastereomer), 7.29 – 7.35 (m, 2 x 4H, tosylate aromatics), 7.75 (dm, *J* = 8.8 Hz, 2 x 2H, aromatic). ^{13}C NMR: ($CDCl_3$, 100 MHz) Major diastereomers: d 21.60 (MePh), 31.19, 38.48, 55.29 (MeOPh), 66.17, 66.85, 73.65, 73.71, 101.12, 113.63 (2C), 127.24, 127.96 (2C), 129.88, 129.91, 130.96, 132.70, 145.03, 159.98. Minor diastereomer: d 20.99, 31.23, 38.74, 55.29 (superimposed on major isomer), 60.37, 68.42, 73.11, 76.15, 101.12 (superimposed on major isomer), 113.68 (2C), 127.13 (2C), 127.85 (2C), 129.91 (superimposed on major isomer), 130.70, 132.75, 144.95, 159.98 (superimposed on major isomer).

Protected epoxy diol (20). To the above tosylate (0.5 g, 1.2 mmol) in dry methanol (15 mL), anhydrous K_2CO_3 (0.33 g, 2.37 mmol) was added. The reaction mixture was stirred at room temperature and was complete after 1 h, as judged by TLC (silica; 40% EtOAc in hexane). The reaction was concentrated to remove the methanol, then diluted with water and extracted with ether (3 x 30 mL), dried (MgSO4) and concentrated to provide the desired epoxide (20) (0.24 g, 80%). GC/MS: *m/z* 250 (M^+, 18%), 219 (2), 193 (11), 176 (2), 152 (11), 135 (100), 108 (14), 94 (9), 77 (24), 41 (20). The crude product was purified by flash column chromatography. ^{1}H NMR: (2 diastereomers about benzylidene CH) 7.42, 7.45 (d ar, 2 x 2H, CH), 6.82, 6.88 (d ar, 2 x 2H, CH), 5.42, 5.46 (s, 2 x 1H, CHPMP), 3.84, 4.25 (m, 2 x 3H, CH and CH_2 in acetal ring), 3.80 (m, 2 x 2H, CH_2O), 3.78 (s, 2 x 3H, MeO), 2.86 – 2.94 (m, 2 x 4H, CH2S), 1.60 – 2.10 (m, 2 x 8H). ^{13}C NMR: ($CDCl_3$, 2 isomers) 164.48, 163.29 (MeOC), 131.55, 131.24 (ar 2CH), 129.89, 130.01 (ar quarternary C), 113.63, 114.18 (ar, 2CH), 101.00, 100.89 (benzylidene CH), 76.15, 76.12 (CH_2CHOCH), 74.67 (COH), 73.76, 72.67 (CH_2O), 55.15, 55.45 (OMC), 49.97 (CH_2S), 33.16, 32.79 ($2CH_2S$), 29.76, 28.52, 28.39, 27.78, 27.71, 27.03 (CH_2 signals). HRMS: $C_{14}H_{18}O_4$ requires 250.1204. Measured, 250.1208.

Epoxide (17). Enediol **15** was treated with m–CPBA in DCM in the normal way to provide protected epoxydiol **16** which was treated with DDQ in DCM (4 Δ sieves) to provide the protected epoxydiol **17**, which was purified as described for **20**. ^{1}H NMR: ($CDCl_3$, 200 MHz) (4 diastereomers, resolved into pairs about the benzylidene CH centre at this field strength). d 1.48 – 160 (m, 2H), 1.73 – 2.07 (m, 2H), 2.49 – 2.56 (m, 2 x 1H), 2.75 – 2.83 (m, 2 x 1H), 3.10 – 3.19 (m, 2 x 1H), 3.78 (s, 2 x 3H, OMe), 3.83 – 4.30 (M, 1H), 5.47 (s, 1H, minor isomer), 5.51 (s, 1H, major isomer), 6.87 (dm, *J* = 8.8 Hz, 2 x 2H), 7.40 (br d, *J* = 8.4 Hz, 2 x 2H). ^{13}C NMR: ($CDCl_3$, 50 MHz) Major isomer d 31.46, 38.36, 47.27, 49.16, 55.19 (superimposed on minor isomer, OMe), 66.69, 74.75, 100.93, 113.48 (2C), 127.19 (2C), 131.34, 159.77. Minor isomer d 31.46, 39.36. 46.27, 48.86, 55.19 (superimposed on minor isomer, OMe) 66.79, 74.75, 100.93, 113.48 (2C), 127.19 (2C), 131.34, 159.77. Minor isomer d 30.76, 39.37, 46.72, 48.86, 55.19 (superimposed on major isomer, OMe) 66.82, 74.34, 101.04, 113.52 (2C), 127.24 (2C), 131.06, 159.82.

Monoalkylation of 1,3–dithiane (21). ^{n}Butyl lithium (2.62 mL of a 2.2 M solution in hexane, 5.76 mmol) was added *via* syringe to 1,3–dithiane (0.48 g, 1.92 mmol) dissolved in dry THF (20 mL) under N_2 and cooled to –35° C. The reaction mixture was stirred at –35° C for 2 hours, after which the epoxide **17** in THF (5 mL) was added *via* cannula. The reaction with stirred for a

further 1 hour at –35° C and monitored *via* TLC (silica; 50% EtOAc in hexane). The sluggish reaction was allowed to warm to room temperature and quenched by the addition of EtOAc (20 mL) and then saturated aqueous $NaHCO_3$ (30 mL). The mixture was concentrated to remove THF, then extracted with EtOAc (3 x 40 mL), dried ($MgSO_4$) and concentrated. The crude product was purified by flash column chromatography (silica; 40% EtOAc in hexane) to provide dithiane 21 in 73% yield. GC/MS: *m/z* 278 (9%), 260 (23), 161 (22), 159 (25), 145 (36), 133 (30), 132 (28), 127 (30), 119 (77), 106 (23), 101 (35), 75 (24), 74 (25), 73 (28), 59 (42), 55 (32). ^{1}H NMR: ($CDCl_3$, 400 MHz, four diastereomers, two resolved) d 1.15 – 2.10 (m, 10H), 2.7 – 2.95 (m, 4H), 3.76 (s, OMe, minor isomer), 3.77 (s, 3H), 3.9 – 4.0 (td, 1H), 4.07 – 4.28 (m, 4H), 5.44 (s, 1H, major isomer), 5.46 (s, OCHO minor isomer), 6.82 – 6.87 (m, 2H), 7.33 – 7.37 (m, 2H). ^{13}C NMR: (CDCl3, 100 MHz) Major isomer: d 25.87, 30.03, 30.32, 31.01, 42.51, 42.93, 44.13, 55.25, 65.22, 66.93, 74.46, 101.15, 113.59 (2C), 127.25 (2C) 131.05, 159.90. Minor Isomer: d 25.97, 30.09, 30.48, 31.38, 42.72, 42.93, 43.80, 55.25, 66.88, 67.53, 77.5, 101.15, 113.63 (2C), 127.17 (2C) 130.70, 160.01. This alcohol **21** was carried on to the ether **22**.

Methylation of dithiane 21 to ether 22. The alcohol **21** (0.48 g, 1.3 mmol) dissolved in THF (10 mL) was added via cannula (5 mL wash) into a stirred suspension of KH (0.22 g of a 35% suspension in oil; washed with hexane and THF) at 0° C under N_2. After 5 minutes the reaction mixture was warmed to room temperature and stirred for 15 minutes. After recooling to 0° C, MeI (1.85 g, 0.81 mL, 13 mmol) was added neat, via syringe. After stirring for 10 minutes at 0° C the reaction mixture was stirred at room temperature for 20 minutes, then quenched by the careful addition of cold water. The mixture was concentrated to remove THF, then extracted with EtOAc (3 x 40 ml), dried ($MgSO_4$) and concentrated. The residue was purified by flash column chromatography (silica, 25% EtOAc in hexane) to provide the methyl ether 22 (0.27 g, 60%). GC/MS: *m/z* 384 (M^+ 30%), 383 (18), 263 (23), 231 (67), 216 (30), 193 (19), 175 (52), 159 (33), 145 (38), 141 (34), 137 (90), 136 (91), (lower m/z). ^{1}H NMR: ($CDCl_3$, 400 MHz) (Mixture of four diastereomers, 2 diastereomers resolved). d 1.44 – 1.53 (m, 2 x 1H), 1.64 – 2.09 (m, 2 x 7H), 2.74 – 2.91 (m, 2 x 4H, SCH_2), 3.33 (s, 3H, OMe), 3.39 (s, 3H, OMe), 3.68 – 3.79 (M, 2 x 1H), 3.77 (s, 2 x 3H, OMe), 3.90 – 4.02 (m, 2 x 2H), 4.12 – 4.25 (2 x 2H), 5.45 (s, 1H, OCHO), 5.46 (s, 1H, OCHO), 6.85 (dm, *J* = 8.8 Hz, 2H), 6.86, (dm, *J* = 8.8 Hz, 2H) 7.40 (dm, *J* = 8.8 Hz, 2 x 2H). ^{13}C NMR: ($CDCl_3$, 100 MHz) 25.90, 26.00 (CH_2), 30.10 (2C, CH_2), 30.34, 30.36 (CH_2), 31.58, 31.73 (CH2), 39.25, 39.83 (SCH2), 40.81, 41.50 (SCH_2), 43.34, 43.69 (SCHS) 55.26, 55.27 (OMe), 56.54, 57.75 (OMe), 66.93, 66.96 (OCH_2), 73.73, 73.92, 74.07, 74.11 (OCH), 100.82, 100.83 (OCHO), 113.50 (2C), 113.55 (2C), 127.22 (4C), 131.30, 131.38, 159.78, 159.82. HRMS: $C_{19}H_{28}O_4S_2$. Na requires 407.1326. Measured, 407.1342.

Alkylation of dithiane 22 to 23. To dithiane 22 (0.17 g, 0.44 mmol) dissolved in dry THF (2.5 mL; distilled off Na and then $LiAlH_4$) and cooled to –30°C (dry ice/CCl_4) under N2, was added ^{t}BuLi (288 µL, 0.48 mmol). After stirring for 10 minutes, HMPA (156 µL, 0.88 mmol) was added *via* syringe and the mixture stirred at –30°C for a further 1 hour. Epoxide 10 (0.17 g, 170 µL, 0.53 mmol) was added neat *via* syringe, the mixture stirred for a further 10 minutes at –30° C then warmed to 0° C and stirred for a further 1 hour. The reaction was quenched with water,

extracted with ether (3 x 20 mL), dried ($MgSO_4$) and concentrated. The residue was purified *via* flash chromatography (silica; 25% EtOAc in hexane) to provide the bis-alkylated dithiane **23**, 135 mg, (43%). ^{1}H NMR: ($CDCl_3$) 400 MHz) d 0.56 – 0.65 (m, 2 x 2H, $SiCH_2CH_3$), 0.91 – 0.97 (m, 3 x 3H, $SiCH_2CH_3$), 1.50 – 2.29 (m, 12H), 2.59 – 2.86 (m, 4H), 3.25 – 3.49 (m, 5H), 3.75 – 4.55 (m, 11H), 5.43 – 5.47 (m, 1H), 6.83 – 6.88 (m, 2H), 7.21 – 7.33 (m, 5H), 7.39 – 7.41 (m, 2H). HRMS: $C_{37}H_{58}O_7S_2Si$ requires 706.3393. Measured, 706.3395. Further characterisation of this diastereomeric mixture prior to spiroacetalisation was not attempted.

Spiroacetalisation of 23 to 24. Dithiane **23** (70 mg in MeOH/water (9:1, 5 mL) was treated with *bis*–trifluoroacetoxy iodobenzene (Stork's reagent: 140 mg) and the mixture was stirred and allowed to warm to room temperature. The reaction was monitored by TLC (silica; 5% MeOH in DCM) and quenched after 15 minutes by the addition of saturated aqueous $NaHCO_3$ (5 mL). The mixture was concentrated to remove MeOH and then extracted with ether (3 x 10 mL) and EtOAc (1 x 20 mL). The combined organic layers were dried ($MgSO_4$) and concentrated under reduced pressure. Initial purification of the residue by flash column chromatography (silica, 5% MeOH in DCM) was followed by gradient elution on semi–preparative reverse phase HPLC (75% acetonitrile/water through to 95% acetonitrile/water) to provide the diastereomers A – G whose spectra are tabulated and discussed in the text. Representative HPLC times for some of the spiroacetal isomers under these conditions were 16.6mins for isomer C and 11.6 mins for isomer F. Isomers B and E were obtained as a mixture which eluted at13.1mins. The peak area ratios of (B and E):C:F was 2.4:1.2:1. Isomers B and E were separated by further reverse phase HPLC elutions to afford pure B and E in a ratio of 1.6:1. Isomers A, D and G were separated under similar chromatographic conditions and were obtained in relatively similar quantities to B,E,C and F with the exception of isomer A of which only a small amount was obtained. HRMS: $C_{20}H_{30}O_6$ requires 366.2042. Measured, 366.2038.

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