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PSEUDOGUAIANOLIDES AND OTHER SESQUITERPENE LACTONES FROM *GAILLARDIA* SPECIES

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Key Word Index—*Gaillardia aristata*; *G. pulchella*; Compositae; sesquiterpene lactones; pseudoguaianolides; seco-neopulchellin derivatives; dugaldiolide derivatives.

Abstract—The aerial parts of two *Gaillardia* species afforded in addition to known sesquiterpene lactones 23 new pseudoguaianolides, two dugaldiolide derivatives and two seco-neopulchellin derivatives. The structures were elucidated by spectroscopic methods. Biogenetic relationships are discussed briefly.

INTRODUCTION

The North American genus *Gaillardia*, previously a member of the tribe Helenieae, is now placed in the tribe Heliantheae, subtribe Gaillardiinae [1, 2]. Several species of this genus have been studied chemically. In addition to some characteristic acetylenic compounds [3] and thymol derivatives [4], pseudoguaianolides seem to be typical for this genus and perhaps for the whole subtribe [5]. Also from the aerial parts of *G. aristata* Pursh. and *G. pulchella* Foug. several sesquiterpene lactones have been isolated [6–9] while the roots gave thymol derivatives [4] and acetylenic epoxysulfones [3]. We now have studied again the aerial parts of *G. aristata* and *G. pulchella* from North Carolina and cultivated material from the Botanical Garden in Berlin. The results will be discussed in this paper.

RESULTS AND DISCUSSION

The aerial parts of *G. pulchella* Foug. collected in North Carolina afforded pulchellin (**1a**) [10] and neopulchellin (**8a**) [11] and the seco derivative **23c**. The aerial parts of *G. aristata* Pursh. collected in Colorado, gave the eudesmanolides pulchellin C (**14a**) [12], pulchellin B (**14b**) [12] and pulchellin E (**14c**) [12], the pseudoguaianolides **10b**, **11c** and **12** as well as the seco derivative **23a**. A larger amount of *G. pulchella*, cultivated in the Botanical Garden at Berlin, afforded pulchellin (**1a**), the corresponding 6β-acetoxy derivative **1b**, spathulin (**1c**) [13], the 2-*O*-angelate **2a** [14], the 2-*O*-methyl butyrate **2b**, the 2-*O*-isovalerate **2c**, the known lactones **3a** [15], **3b** [7] and **3c** [15] as well as the corresponding isobutyrate **3d**. Furthermore the diacetates **4a**, **4b**, **5a–5c**, the diesters **6a–6d**, the angelates **7a** and **7b**, neopulchellin (**8a**) [11], the corresponding angelate **8b**, the triol **9a**, the angelates **10a** and **11a**, the methyl butyrate **11b**, florilenalin (**13**) [16], the angelate **18**, the corresponding methyl butyrate **19** and the seco derivative **23a** were isolated. The separation of the complex mixture of lactones was achieved by combination of repeated thin layer and high pressure liquid chromatography.

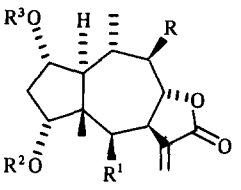
The structure of **1b** followed from the molecular formula and the ^{1}H NMR spectral data (Table 1) which were in part close to those of spathulin [13]. The missing 9-acetoxy group, however, caused the expected changes. Spin decoupling allowed a clear assignment of the signals of H-9α and H-9β which displayed threefold doublets at δ1.41 and 2.45. Furthermore the stereochemistry at all centres was established by NOE difference spectroscopy. Irradiation of the 5-methyl signal showed clear effects of H-2β, H-3β, H-4β, H-6, H-8 and H-10. Thus the configuration at C-2 differed from that of flexuosin A, where a 2β-hydroxyl group [17] was proposed.

The structures of **2b** and **2c** clearly followed from the ^{1}H NMR spectra (Table 1) as all signals, except those of the ester residue, were close to those of the angelate **2a** [14]. The nature of the ester group could be deduced from the typical ^{1}H NMR signals though these esters could not be separated. Spin decoupling allowed the assignment of all signals (Table 1). As in all other sesquiterpene lactones which have been isolated from the *Gaillardia* species the esterification of the 2-hydroxyl group caused a downfield shift of H-1.

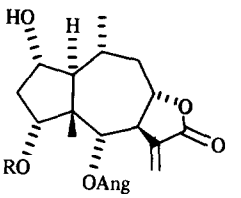
The molecular formula and the ^{1}H NMR spectrum of **3d** (Table 1) showed that this lactone was closely related to **3a–3c** [7, 15], only one of the ester groups being changed. The typical ^{1}H NMR signals indicated the presence of the corresponding isobutyrate. The relative position of the ester residues followed from the identical chemical shifts of H-6 and of the acetate methyl in the spectra of **3b** and **3d**. Especially the shift of the acetate methyl differs depending on its position in these lactones as followed from careful comparison of several fully or partial acetylated compounds.

Accordingly also the relative position of the ester groups in **4a** and **4b** could be deduced from the ^{1}H NMR spectra (Table 1). The presence of the free hydroxyl of course followed from the chemical shift of H-4 while that of H-6 and H-9 was the same as in spathulin (**1c**) indicating the same position of the acetate groups in **4a** and **4b**.

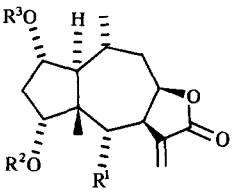
The ^{1}H NMR spectra of **5a** and of the inseparable mixture of **5b** and **5c** (Table 2) showed that in the lactones



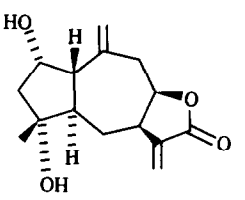
	R	R^1	R^2	R^3
1a	H	H	H	H
1b	H	OAc	H	H
1c	OAc	OAc	H	H
2a	OH	OH	H	Ang
2b	OH	OH	H	Mebu
2c	OH	OH	H	i Val
3a	OH	OAc	H	Ang
3b	OH	OAc	H	Mebu
3c	OH	OAc	H	i Val
3d	OH	OAc	H	i Bu
4a	OAc	OAc	H	Mebu
4b	OAc	OAc	H	i Val
5a	OH	OAc	Ac	Ang
5b	OH	OAc	Ac	Mebu
5c	OH	OAc	Ac	i Val
6a	OH	OH	Ac	Ang
6b	OH	OH	Ac	Mebu
6c	OH	OH	Ac	i Val
6d	OH	OH	Ac	i Bu



7a R = H **7b** R = iVal



	R^1	R^2	R^3
8a	H	H	H
8b	H	Ang	H
9a	OH	H	H
9b	OH	H	Ac
9c	OAc	Ac	Ac
10a	OAng	H	H
10b	OAng	H	Ac
11a	OH	Ang	H
11b	OH	Mebu	H
11c	OH	i Val	H
12	OAng	i Val	H



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a free hydroxyl at C-9 was present. Again the chemical shifts of H-2, H-4 and H-6 and the shifts of the acetate methyls indicated the relative position of the three ester functions. The nature of the ester groups also followed from the ^{1}H NMR spectra. All signals were assigned by spin decoupling. The same was true for the lactones **6a**–**6d** which again only differed in the nature of the ester group at C-2. While **6a** and **6d** could be separated by HPLC **6b** and **6c** were obtained as an inseparable mixture. However, as the concentration of both differed slightly all signals could be assigned from the spectrum of the mixture.

The ^{1}H NMR spectra (Table 3) of the lactones **7a** and **7b** differed characteristically from those in Tables 1 and 2 by a typical doublet around δ5.25 (J = 11 Hz) which turned out to be the signal of H-6 as proved by spin decoupling. The spectra also showed that **7b** was the isovalerate of **7a**, the H-4 doublet being shifted downfield in the spectrum of **7b** while the other signals were influenced only to a small extent. In the spectrum of **7a** some signals were overlapping multiplets. However, all signals could be assigned by spin decoupling. In the spectrum of **7b** the isovalerate residue showed unusual

Table 1. ^{1}H NMR spectral data of **1b**, **2b**, **2c**, **3d**, **4a** and **4b** (400 MHz, $CDCl_3$, TMS as internal standard)

	1b	$CDCl_3$–C_6D_6 2:1	**2b**		**2c**	**3d**	**4a**	**4b**
H-1	1.95 *m*	1.77 *dd*		2.34 *dd*		2.34 *dd*	2.42 *dd*	2.39 *dd*
H-2	4.15 *dddd*	3.86 *dddd*		5.00 *ddd*		4.99 *ddd*	4.99 *ddd*	4.99 *ddd*
H-3α	1.70 *dd*	1.48 *dd*		1.65 *dd*		1.56 *dd*	1.56 *dd*	1.59 *dd*
H-3β	2.43 *ddd*	2.18 *ddd*		2.64 *ddd*		2.58 *ddd*	2.62 *ddd*	2.61 *ddd*
H-4	3.82 *dd*	3.62 *dd*		3.88 *d*		3.83 *dd*	3.86 *dd*	3.85 *dd*
H-6	6.02 *d*	5.90 *d*		4.93 *d*		6.01 *d*	6.02 *d*	6.00 *d*
H-7	3.14 *dddd*	2.86 *dddd*		3.00 *dddd*		3.16 *dddd*	3.26 *dddd*	3.23 *dddd*
H-8	4.62 *ddd*	4.42 *ddd*	4.61 *ddd*		4.60 *ddd*	4.56 *t*	4.63 *t*	4.63 *t*
H-9α	1.41 *ddd*	1.17 *ddd*		} 3.30 *dt*		} 3.33 *dt*	} 4.85 *t*	} 4.84 *t*
H-9β	2.45 *ddd*	2.24 *ddd*						
H-10	1.95 *m*	1.71 *dddq*		1.95 *m*		1.90 *ddq*	2.05 *m*	2.05 *m*
H-13	6.26 *d*	6.19 *d*		6.43 *d*		6.32 *d*	6.31 *d*	6.31 *d*
H-13′	5.42 *d*	5.28 *d*		5.69 *d*		5.49 *d*	5.50 *d*	5.49 *d*
H-14	1.25 *d*	1.07 *d*		1.16 *d*		1.16 *d*	0.98 *d*	0.97 *d*
H-15	0.74 *s*	0.50 *d*	0.93 *s*		0.94 *s*	0.81 *s*	0.83 *s*	0.82 *s*
OCOR	2.03 *s*	1.84 *s*	2.28 *tq*		2.16 *d*	2.53 *qq*	2.35 *tq*	2.18 *d*
			1.65 *m*		2.08 *m*	1.16 *d*	1.68 *ddq*	2.07 *s*
			1.45 *m*		0.95 *d*		1.46 *ddq*	0.94 *d*
			0.93 *t*				0.91 *t*	0.94 *d*
							1.12 *d*	2.15 *s*
							2.15 *s*	2.03 *s*
							2.04 *s*	

J (Hz): 1, 2 = 6; 1, 10 = 10; 2, 3α = 1.5; 2, 3β = 8.5; 3α, 3β = 15; 3β, 4 = 4.5; 6, 7 = 4; 7, 8 = 9; 7, 13 = 3.5; 7, 13′ = 3; 10, 14 = 7; compound **1b**: 2, OH = 7; 4, OH = 4.5; 8, 9α = 12; 8, 9β = 3; 9α, 9β = 13; 9α, 10 = 11; 9β, 10 = 3; compounds **2b**, **2c**, **3d** and **4b**: 8, 9 = 9, 10 = 9.5; OCOR: see Table 2.

Table 2. ^{1}H NMR spectral data of **5a–5c** and **6a–6d** (400 MHz, $CDCl_3$, TMS as internal standard)

	5a	**5b**		**5c**	**6a**	**6b**	**6c**	**6d**
H-1	2.40 *dd*		2.37 *dd*		2.29 *dd*	2.26 *dd*	2.26 *dd*	2.25 *dd*
H-2	5.13 *ddd*		5.05 *ddd*		5.13 *ddd*	5.05 *ddd*	5.05 *ddd*	5.04 *ddd*
H-3α	1.66 *dd*		1.59 *dd*		1.65 *dd*	1.57 *dd*	1.57 *dd*	1.54 *dd*
H-3β	2.68 *ddd*		2.64 *ddd*		2 72 *ddd*	2.68 *ddd*	2.68 *ddd*	2.67 *ddd*
H-4	4.81 *d*		4.79 *d*		4.93 *d*	4.91 *d*	4.91 *d*	4.91 *d*
H-6	5.83 *d*	5.82 *d*		5.81 *d*	4.40 *dd*	4.41 *dd*	4.41 *dd*	4.41 *br d*
H-7	3 22 *dddd*		3.21 *dddd*		3.05 *dddd*	3.04 *dddd*	3.04 *dddd*	3.04 *dddd*
H-8	4.56 *t*	4.56 *t*		4.55 *t*	4.61 *t*	4.60 *t*	4.61 *t*	4.61 *t*
H-9	3.37 *dt*		3.37 *dt*		3.33 *dt*	3.33 *dt*	3.33 *dt*	3.33 *t*
H-10	1.92 *m*		1.91 *m*		1.92 *m*	1.88 *ddq*	1.88 *m*	1.88 *ddq*
H-13	6.33 *d*		6.33 *d*		6.45 *d*	6.43 *d*	6.43 *d*	6.44 *d*
H-13′	5.50 *d*		5.50 *d*		5.59 *d*	5.62 *d*	5.63 *d*	5.61 *d*
H-14	1.18 *d*		1.17 *d*		1.17 *d*	1.16 *d*	1.16 *d*	1.15 *d*
H-15	0.91 *s*		0.90 *s*		1.03 *s*	1.01 *s*	1.00 *s*	1.02 *s*
OCOR	6.09 *qq*	2.34 *tq*		2.15 *d*	6.09 *qq*	2.30 *tq*	2.16 (2H)	2.50 *qq*
	1.98 *dq*	1.65 *ddq*		2.08 *m*	1.99 *dq*	1.64 *ddq*	2.08 *m*	1.15 *d*
	1.87 *dq*	1.46 *ddq*		0.94 *d*	1.87 *dq*	1.47 *ddq*	0.95 *d* (6H)	
		6.88 *t*				0.90 *t*		
		1.11 *d*				1.11 *d*		
OAc	2.11 *s*	2.12 *s*		2.11 *s*	2.08 *s*	2.09 *s*	2 10 *s*	2.10 *s*
	2.02 *s*	2.02 *s*		2.02 *s*				
OH					5.53 *d*	1.83 *d*	2.53 *d*	
					3.14 *d*	1.83 *d*	3.14 *d*	

J (Hz): 1, 2 = 6; 2, 3α = 2; 2, 3β = 9; 6, 7 = 3; 7, 13 = 3.5; 7, 13′ = 3; 8, 9 = 9.5; 9, 10 = 10; 10, 14 = 7; compounds **5a–5c**: 1, 10 = 11; 3α, 3β = 15; 3β, 4 = 5; 7, 8 = 9; 9, OH = 1.5; compounds **6a–6d**: 1, 10 = 11.5; 3α, 3β = 16.5; 3β, 4 = 4.5; 6, OH = 3; 7, 8 = 9.5; OAng: 3′, 4′ = 7; 3′, 5′ = 4′, 5′ = 1.5; OMebu: 2′, 3′ = 3′, 4′ = 2′, 5′ = 7; $3_1'$,$3_2'$ = 14; OiVal: 3′, 4′ = 3′, 5′ = 7, OiBu: 2′, 3′ = 2′, 4′ = 7.

Table 3 ^{1}H NMR spectral data of **7a** and **7b** (400 MHz, $CDCl_3$, TMS as internal standard)

	7a	**7b**	
H-1	2.34 *dd*	2.39 *dd*	
H-2	4 15 *m*	4.27 *br dd*	
H-3α	1.57 *m*	1.57 *dd*	
H-3β	2.33 *m*	2.62 *ddd*	
H-4	3 84 *br d*	4.81 *d*	
H-6	5.28 *d*	5.22 *d*	
H-7	3.38 *dddd*	3 59 *dddd*	
H-8	4 18 *ddd*	4.18 *ddd*	
H-9α	1.53 *m*	1.60 *ddd*	
H-9β	2 39 *ddd*	2.35 *ddd*	
H-10	1.89 *m*	1.89 *m*	
H-13	6 23 *d*	6.21 *d*	
H-13′	5.55 *d*	5.57 *d*	
H-14	1 27 *d*	1 28 *d*	
H-15	0 99 *s*	1.02 *s*	
OCOR	6 32 *qq*	6 17 *qq*	2.14 *dd**
			2.22 *dd**
	2 05 *dq*	1 98 *dq*	2.05 *m*
	2 00 *dq*	1.87 *dq*	0 90 *d*
OH	2.90 *br s*		0.89 *d*

*J = 14 and 7 Hz

J (Hz): 1, 2 = 6, 1, 10 = 10.5; 2, 3α = 2 5; 2, 3β = 9; 3α, 3β = 15, 3β, 4 = 5; 6, 7 = 11; 7, 8 = 9; 7, 13 = 3; 7, 13′ = 2 5; 8, 9α = 12; 8, 9β = 3; 9α, 9β = 13; 9α, 10 = 10, 9β, 10 = 3; 10, 14 = 7; OCOR: see Table 2

pairs of double doublets for the α-protons, indicating steric hindrance to free rotation of the ester group. The stereochemistry of **7a** and **7b** followed from the couplings observed, especially if models were inspected, and from comparison with the data of **10a**, obviously an isomer of **7a**. While the Cotton effect of **10a** clearly showed the presence of a *cis*-8,12-lactone [18], **7a** showed no clear Cotton effect as is the case of some other pseudoguaianolides [19]. The presence of a *trans*-8,12-lactone in **7a** and **7b**, however, followed from the couplings $J_{6,7}$ and $J_{8,9\alpha}$, which obviously required a trans-diaxial orientation of H-6 and H-7 as well as of H-7 and H-8. Further support for a 6β-proton in **7a** was provided by the relative chemical shifts of H-6 in the spectrum of **7a** and in those of **3–5** as H-6α is deshielded by the 4α-oxygen function. Similarly the signals of the angelate residue were shifted downfield in the spectrum of **7a**. However, as in that of **7b** the angelate signals were at somewhat lower fields suggesting a hydrogen bond was present (**2a**, **3a**, **5a**, **8b**, **10a**, **10b**, **11a**).

The ^{1}H NMR spectrum of **8b** (Table 4) was in part very close to that of neopulchellin (**8a**) [11]. A drastic downfield shift of H-4 in the spectrum of **8b** compared with **8a** showed that an ester group was at C-4; its nature could again be deduced from the typical ^{1}H NMR signals.

From the spectrum of **9a** and its molecular formula $C_{15}H_{22}O_5$, the structure of 6α-hydroxyneopulchellin could be deduced, especially if the ^{1}H NMR spectra (Table 4) of the corresponding mono- and triacetate (**9b** and **9c**) were considered too. Characteristic differences of the H-8 couplings in the spectra of **9a** and **9b** compared with those of **9c** indicated small changes in the conformation. Spin decoupling in the usual way allowed the assignment of all signals indicating that the oxygen functions were at C-2, C-4 and C-6 while the couplings showed that they all were α-orientated. The presence of a 8,12-*cis*-lactone followed from the couplings which corresponded to those of neopulchellin. Furthermore it was characteristic that the H-13 signals always showed a smaller allylic coupling, if compared with those of the *trans*-isomers, and the chemical shift of H-7 was always at lower fields if compared with the corresponding 8,12-*trans*-lactones.

In a similar way the structures of **10a** and **10b** as well as those of **11a–11c** easily could be deduced from the ^{1}H NMR spectra (Table 4 and 5). The position of the angeloyloxy group in **10a** and **10b** followed directly from the chemical shift of H-6 if it is compared with the shift in the spectrum of **9c**. The presence of a 2α-acetoxy group in **10b** was deduced from the downfield shift of the H-2 signal in the ^{1}H NMR spectrum compared with that in the spectrum of **10a**.

The position of the ester groups in **11a–11c** followed from the chemical shift of H-4 (Table 5). In this case the methyl butyrate and the corresponding isovalerate could be separated by HPLC. The signals of H-2 always were broad multiplets. However, addition of deuteriobenzene changed this signal to a broadened double doublet (Table 5) indicating the same stereochemistry as in all the other pseudoguaianolides.

The ^{1}H NMR spectrum of **12** (Table 5) showed that it was most likely an angelate of **11c**. The H-6 doublet was shifted to δ5.31, while in the spectra of lactones with a saturated ester group at C-6 (**9c**) this doublet was at higher fields. Furthermore NOE difference spectroscopy established the stereochemistry as on irradiation of H-15 clear effects were visible for H-2, H-4, H-6 and a small one for H-10. Irradiation of H-7 caused an NOE of H-1 and also of the isovalerate methylene signal. Inspection of a model showed that only an ester group at C-4 could give this effect as the ester group at C-6 was orientated equatorially. These observations favoured an isovalerate group at C-4, most likely hydrogen-bonded with the hydroxyl at C-2. The methylene protons of the isovalerate displayed pairs of doublets which may support this assumption. Furthermore the chemical shifts of the angelate proton agreed with this structure.

The structures of **18** and **19** could be deduced easily from the ^{1}H NMR spectra (Table 6) which were close to that of the corresponding 2α-tigloyloxy-dugaldiolide isolated from a *Dugaldia* species, where the structure and the stereochemistry was established rigorously [20]. The ^{1}H NMR spectrum of **19** showed that a pair of enantiomers at C-2′ was present because several pairs of signals were visible (Table 6). A separation of these isomers by HPLC was not successful though a small enrichment of one isomer occurred. Compounds **18** and **19** are most likely formed via the bisepoxide **16** as shown in Scheme 1.

The structures of the lactones **23a** snd **23c** were deduced from the ^{1}H NMR spectra (Table 7) and the mass spectra. Irradiation of the broad singlet at δ5.67, obviously a signal of an olefinic proton, caused changes of the signal at δ1.87 (H-15), 2.42 (H-1) and 3.43 (H-7). The identity of the H-7 signal was established by its simplification on irradiation. Further spin decoupling allowed the assignment of all signals leading to the whole sequence

Table 4. ^{1}H NMR spectral data of **8b**, **9a**, **9b**, **9c**, **10a** and **10b** (400 MHz, $CDCl_3$, TMS as internal standard)

	8b	**9a**	C_6D_6	**9b**	**9c**	**10a**	**10b**
H-1	1.73 *dd*	2.01 *dd*	1.79 *dd*	2.38 *dd*	2.32 *dd*	2.08 *m*	2.39 *dd*
H-2	3.60 *m*	4.02 *ddd*	3.74 *ddd*	4.88 *ddd*	4.93 *ddd*	4.03 *m*	4.89 *ddd*
H-3α	1.64 *dd*	1.53 *dd*	1.35 *dd*	1.48 *dd*	1.45 *dd*	1.58 *dd*	1.53 *dd*
H-3β	2.78 *ddd*	2.62 *ddd*	2.37 *ddd*	2.72 *ddd*	2.77 *ddd*	2.58 *ddd*	2.70 *ddd*
H-4	4.81 *d*	4.04 *d*	3.83 *d*	4.09 *br d*	5.00 *d*	3.91 *br d*	3.93 *br d*
H-6	1.54 *m*	3.67 *br d*	3.32 *br d*	3.71 *br d*	5.13 *d*	5.31 *d*	5.34 *d*
H-7	3.43 *m*	3.50 *dddd*	3.27 *dddd*	3.54 *dddd*	3.72 *dddd*	3.74 *dddd*	3.76 *dddd*
H-8	4.81 *ddd*	4.81 *ddd*	4.52 *ddd*	4.81 *ddd*	4.83 *ddd*	4.81 *ddd*	4.80 *ddd*
H-9α*	1.85 *br d*	1.89 *m*	1.70 *m*	1.91 *m*	1.96 *br dd*	1.98 *m*	1.9–2.05 *m*
H-9β*	2.15 *m*				2.15 *m*	2.08 *m*	
H-10	2.10 *m*	1.96 *m*		1.94 *m*	2.00 *m*	1.98 *m*	
H-13	6.22 *d*	6.34 *br d*	6.23 *br d*	6.38 *br d*	6.28 *dd*	6.23 *br d*	6.23 *br d*
H-13′	5.44 *d*	5.83 *br d*	5.56 *br d*	5.82 *br d*	5.54 *br d*	5.53 *br d*	5.52 *br d*
H-14	1.27 *d*	1.25 *d*	1.10 *d*	1.09 *d*	1.13 *d*	1.24 *d*	1.12 *d*
H-15	0.94 *s*	0.89 *s*	0.63 *s*	0.93 *s*	1.01 *s*	0.91 *s*	0.94 *d*
OCOR	6.14 *qq*			2.05 *s*	2.13 *s*	6.18 *qq*	6.19 *qq*
	2.05 *dq*		2.91 *br s*	2.90 *br s*	2.05 *s*	1.97 *br d*	1.98 *dq*
	1.94 *dq*		(OH)	(OH)	2.01 *s*	1.96 *br s*	1.95 *dq*

*Not first order.

J (Hz): 1, 2 = 6; 1, 10 = 11; 2, 3α = 2.5; 2, 3β = 9; 3α, 3β = 15.5; 3β, 4 = 5; 6, 7 = 12; 7, 8 = 8; 7, 13 = 1.7; 7, 13′ = 1.5; 8, 9α = 4; 8, 9β = 12; 9α, 9β = 13; 10, 14 = 6.5; compounds **9a**/**9b**: 8, 9α = 6; 8, 9β = 9.

Table 5. ^{1}H NMR spectral data of **11a**–**11c** and **12** (400 MHz, $CDCl_3$, TMS as internal standard)

	11a	**11b**	**11c**	$CDCl_3$–C_6D_6 2:1	**12**	C_6D_6
H-1	1.90 *m*	1.95 *m*	1.90 *m*	1.70 *m*	2.05 *m*	1.96 *m*
H-2	4.14 *m*	4.12 *m*	4.12 *m*	3.68 *br dd*	4.13 *br t*	3.32 *m*
H-3α	1.59 *dd*	1.50 *dd*	1.53 *dd*	1.30 *dd*	1.57 *dd*	1.43 *dd*
H-3β	2.77 *ddd*	2.71 *ddd*	2.71 *ddd*	2.38 *ddd*	2.68 *ddd*	2.20 *ddd*
H-4	5.03 *br d*	4.97 *d*	5.00 *br d*	4.84 *br d*	4.82 *d*	4.86 *d*
H-6	3.69 *br d*	3.68 *br d*	3.67 *br d*	3.15 *m*	5.31 *d*	5.39 *d*
H-7	3.56 *dddd*	3.58 *dddd*	3.56 *dddd*	3.30 *dddd*	3.81 *br dd*	3.59 *m*
H-8	4.83 *ddd*	4.85 *ddd*	4.83 *ddd*	4.49 *ddd*	4.84 *ddd*	4.29 *ddd*
H-9α	2.00 *m*	1.95 *m*	2.00 *m*	1.65 *m*	1.96 *br dd*	1.56 *ddd*
H-9β					2.05 *m*	1.74 *ddd*
H-10					2.05 *m*	1.37 *m*
H-13	6.34 *br d*	6.35 *br d*	6.36 *br d*	6.19 *br d*	6.22 *br d*	6.30 *dd*
H-13′	5.83 *br d*	5.78 *br d*	5.75 *br d*	5.49 *br d*	5.49 *br d*	5.36 *dd*
H-14	1.27 *br d*	1.26 *br d*	1.27 *br d*	1.02 *d*	1.29 *d*	0.96 *d*
H-15	0.99 *br s*	0.97 *br s*	0.97 *br s*	0.62 *br s*	0.96 *s*	0.57 *s*
OCOR	6.10 *qq*	2.38 *tq*	2.20 *d*	2.03 *m*	6.08 *qq*	5.72 *qq*
	2.01 *dq*	1.74 *ddq*	2.15 *tq*	0.97 *d*	1.92 *dq*	1.97 *dq*
	1.93 *dq*	1.47 *ddq*	0.99 *d*		1.82 *dq*	1.86 *dq*
		0.94 *t*			2.22 *dd*	2.14 *d*
		1.18 *d*			2.12 *dd*	2.28 *m*
		2.84 *d* (OH)			2.06 *m*	0.94 *d*
					0.99 *d*	0.97 *d*

J (Hz): 1, 2 = 6; 2, 3α = 2.5; 2, 3β = 9; 3α, 3β = 15.5; 3β, 4 = 5.5; 6, 7 = 10; 7, 8 = 8; 7, 13 = 1.8; 7, 13′ = 1.5; 8, 9α = 3.5; 8, 9β = 12; 9α, 9β = 14; 9α, 10 ~ 1; 9β, 10 = 7.5; 10, 14 = 6.5; OCOR: see Table 2 (OiVal in **12**: 2′, 2′ = 16; 2′, 3′ = 3′, 4′ = 7).

Table 6. ^{1}H NMR spectral data of **18** and **19** (400 MHz, $CDCl_3$, TMS as internal standard)

	18	$CDCl_3$–C_6D_6 2:1	**19**	C_6D_6
H-1	2.29 *dd*	2.09 *dd*	2.22 *dd*	2.10 *dd*
H-2	4.83 *ddd*	4.70 *ddd*	4.79 *ddd**	4.83 *ddd**
H-3	2.50 *dd*	2.29 *dd*	2.47 *dd**	2.40 *dd**
H-3′	1.74 *dd*	1.52 *dd*	1.65 *dd*	1.60 *dd*
H-6	2.31 *dd*	2.01 *dd*	2.31 *dd*	1.69 *dd*
H-6′	2.10 *d*	1.77 *d*	2.08 *d*	1.53 *d*
H-7	3.29 *dd*	2.95 *dd*	3.28 *dd*	2.79 *dd*
H-8	4.92 *ddd*	4.54 *dd*	4.91 *ddd*	4.20 *ddd*
H-9	2.08 *br dd*	1.77 *ddd*	2.04 *br dd*	1.84 *m*
H-9′	1.65 *ddd*	1.31 *dd*	1.65 *m*	1.35 *m*
H-10	1.78 *m*	1.63 *m*	1.75 *m*	1.55 *m*
H-13	3.91 *dd*	3.68 *dd*	3.90 *dd*	3.79 *dd*
H-13′	3.53 *dd*	3.27 *dd*	3.53 *dd*	3.26 *dd*
H-14	1.00 *d*	0.82 *d*	0.98 *d*	0.78 *d*
H-15	1.22 *s*	0.98 *s*	1.21 *s*	0.85 *s**
OH	2.02 *t*	1.66 *t*	1.93 *t*	1.60 *br t*
OCOR	6.06 *qq*	6.07 *qq*	2.31 *ddq**	2.24 *ddq**
	1.98 *dq*	1.87 *dq*	1.65 *m*	1.55 *m*
	1.86 *dq*	1.75 *dq*	1.44 *m*	1.35 *m*
			0.89 *t**	0.86 *t**
			1.11 *d**	1.08 *d**

*Signals are split by *ca* 1–2 Hz.

J (Hz): 1, 2 = 7; 1, 10 = 11; 2, 3α = 2; 2, 3β = 9; 3α, 3β = 15.5; 6α, 6β = 14; 6β, 7 = 7, 7, 8 = 9.5; 8, 9α = 2; 8, 9β = 5; 9α, 9β = 15.5; 9α, 10 = 11.5; 10, 14 = 6.5; 13, 13′ = 11.5; 13, OH = 6; 13′, OH = 7; OCOR: see Table 2.

of all protons and thus to the structure **23a**. The spectrum of **23c** was very similar to that of **23a** and showed exactly the same splitting pattern of all signals. Only the H-4 signals were shifted downfield, indicating that the primary hydroxyl was esterified. The ^{1}H NMR spectrum showed in addition to an acetate methyl the typical signals of an angelate. To establish the relative position of the acetate group, **23a** was transformed to the diacetate **23b**. However, the chemical shifts of H-2 and H-4 both were close to those of **23c** though the H-4 signals now were collapsed to a double doublet. Inspection of the mass spectrum of **23c** showed a clear fragment at *m/z* 302 corresponding to $C_{18}H_{22}O_4$. This obviously required loss of ethyl acetate which is only possible if the acetoxy group is at C-4 by splitting the 2,3-bond combined with a hydrogen transfer. The corresponding fragment (*m/z* 262) was also present in the spectrum of **23b**. Remarkable is the fragment *m/z* 107 $[C_8H_{11}]^+$ which also was present in several of the pseudoguaianolides. Though the identity of this ion is not known, its formation most likely also requires a 4,5-seco-intermediate. The presence of an 8,12-*cis*-lactone was supported by a negative Cotton-effect [18] and by the chemical shift of H-7 which is always at higher fields in *trans*-8,12-xanthanolides [21]. Most likely **23a** was formed by fragmentation of **21** followed by reduction of the resulting aldehyde **22** as shown in Scheme 1, which further shows that perhaps all 8,12-*cis*-lactones isolated from the subtribe *Gaillardiinae* may be derived from the common precursor **15**, which itself could be formed from a germacranolide-1,10-epoxide [22].

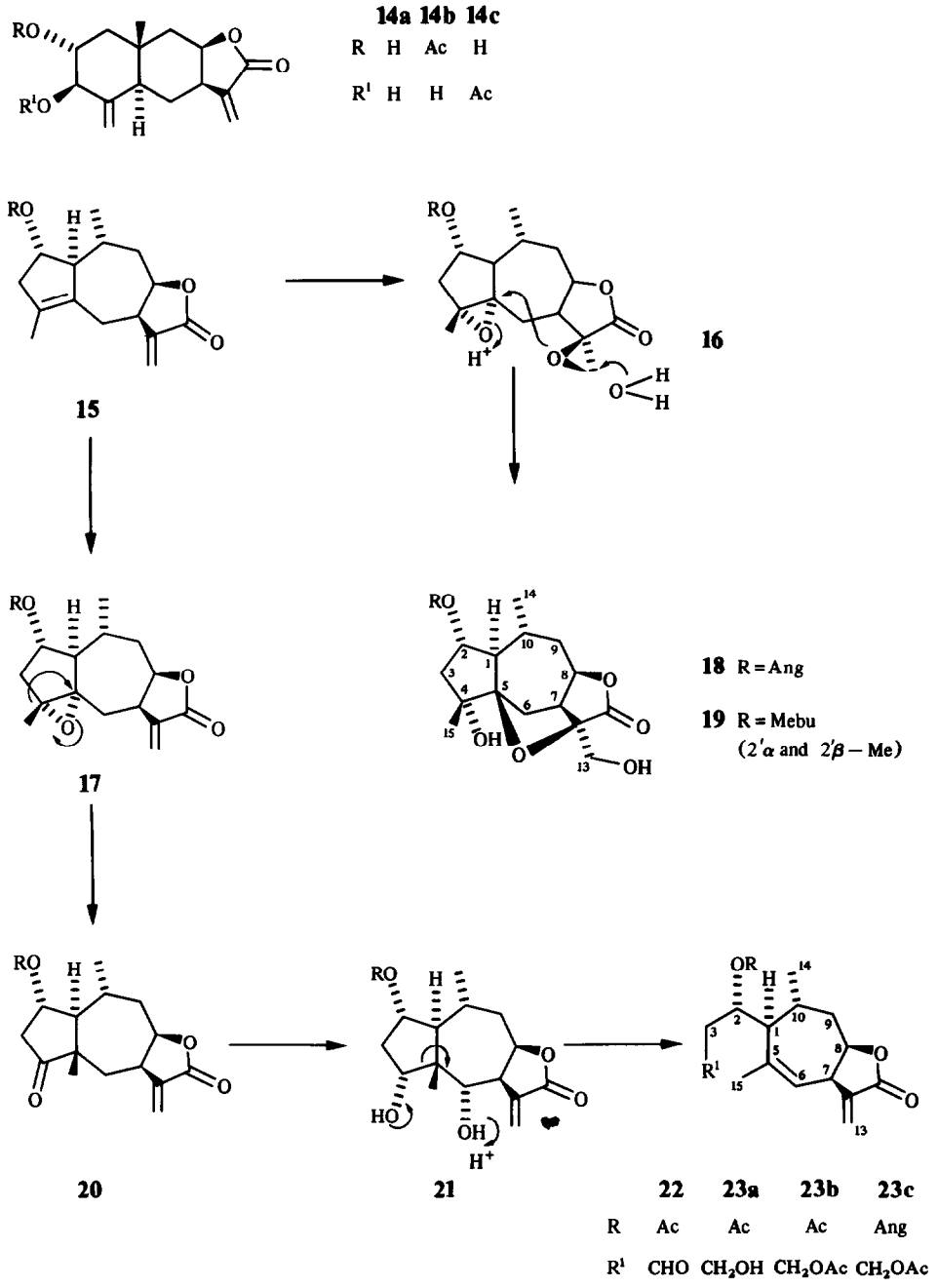
EXPERIMENTAL

General methods. Air dried plant material was cut into small pieces and extracted with Et_2O–petrol–MeOH, 1:1:1, at room temp. for 15 hr. After evaporation under red. pres. the residue was treated with MeOH to remove long chain saturated hydrocarbons. MeOH soln was evaporated under red. pres., C_6H_6 was added and again the soln was evaporated to remove traces of MeOH. The obtained crude material was first separated by column chromatography (CC, silica gel) into six fractions (petrol, Et_2O–petrol 1:10, 1:3, 1:1, Et_2O and finally Et_2O–MeOH, 10:1). From these fractions 400 MHz ^{1}H NMR spectra were measured and those fractions which showed interesting signals (not only those of saturated compounds) were further separated by TLC (SiO_2 PF 254, detection by UV light and by spraying with $KMnO_4$-soln). The extracts of the zones were again investigated by 400 MHz ^{1}H NMR. If still mixtures were present, which could not be separated by repeated TLC, for further separation HPLC (RP 8, MeOH–H_2O mixtures, analytical columns, repeated injection of *ca* 1–2 mg in MeOH each time) was used as well as TLC on aluminium sheets (0.1 mm, SiO_2 PF 254). Known compounds were identified by comparing the 400 MHz ^{1}H NMR spectra with those of authentic material or by rigorous structure elucidation using all spectroscopic methods and by comparing the data with those from the literature. Quantities were determined by weight.

Gaillardia pulchella (*voucher RMK* 9309, *collected in North Carolina*). The extract from 220 g aerial parts gave by CC polar fractions (Et_2O and Et_2O–MeOH, 10:1). TLC (SiO_2, Et_2O–petrol, 1:1) of the Et_2O-fraction gave crude **23c** (R_f 0.42) which was further purified by HPLC (MeOH–H_2O, 7:3, R_t 6.5 min) affording 6 mg **23c**. The more polar CC fraction gave by repeated TLC (Et_2O–petrol, 3:1, three developments) 35 mg pulchellin (**1a**), mp 165° and 20 mg neopulchellin (**8a**), mp 167°

Gaillardia aristata (*voucher RMK* 9087, *collected in Colorado*). The extract from 240 g aerial parts gave by CC a polar fraction (Et_2O–MeOH, 10:1) its ^{1}H NMR spectrum indicated a complex mixture of methylene lactones (H-13 signals). This mixture was first further separated by TLC (Et_2O–petrol, 3:1) The least polar zone gave 50 mg pure pulchellin B (**14b**), mp 216°. The next zone afforded 45 mg pulchellin E (**14c**), mp 180°. The mother liquor of this fraction afforded by HPLC (MeOH–H_2O, 13:7) 5 mg **14c** (R_t 1.0 min), 3 mg **11c** (R_t 2.6 min), 3 mg **23a** (R_t 3.5 min), 3 mg **10b** (R_t 4.8 min) and 3 mg **12** (R_t 6.9 min). The most polar TLC zone gave 95 mg pulchellin C (**14a**), mp 199°.

Second collection of G. pulchella (*Botanical Garden, Berlin, voucher* 22/83). The extract from 1 kg air dried material gave by CC a polar fraction with Et_2O–MeOH, 10:1 and 5:1 (5.5 g). This was further separated by medium pressure liquid chromatography (MPLC) using 200 g silica gel (30–60 μ) with $CHCl_3$ and raising amounts of MeOH affording fractions (25 ml each) which were combined as follows: A (1–4, $CHCl_3$), B (5–25, 2% MeOH), C (25–50, 2% MeOH), D (51–80, 5% MeOH), E (81–95, 5% MeOH), F (96–109, 10% MeOH), and G (110–150, 10% MeOH). Fraction A was separated again by MPLC ($CHCl_3$–C_6H_6–Et_2O, 1:1:1) into three fractions (A_1–A_3). HPLC (MeOH–H_2O, 3:2) of A_1 gave eight fractions (A_{11}–A_{18}). A_{11} (R_t 2.7 min) gave 15 mg **3a**, mp 231°, A_{12} (R_t 3.2 min) 15 mg **3c**, A_{13} (R_t 3.7 min) 10 mg **3b**, A_{14} (R_t 4.0 min) 5 mg **23a**, A_{15} (R_t 5.1) was a mixture of **5a**–**5c** and **7b**, which was further separated by HPLC (MeOH–H_2O, 11:9) affording 2 mg **7b** (R_t 6.8 min), 3 mg **5a** (R_t 8.6 min), 20 mg of a mixture of **5a**–**5c** (R_t 9.5 min) and 4 mg of **5b** and **5c** (*ca* 2:1) (R_t 9.8 min) (**5b** and **5c** could not be separated by HPLC or TLC).



Scheme 1.

A_{16} was separated by TLC ($CHCl_3$–C_6H_6–Et_2O, 1:1:1) affording 2 mg **8b** (R_f 0.40). A_{17} gave 5 mg **4a** (R_t 9.8 min.) and A_{18} 4 mg **4b** (R_t 10.6 min) which was purified by repeating HPLC (MeOH–H_2O, 3:2).

HPLC (MeOH–H_2O, 3:2) of A_2 gave a mixture of 30 mg **3a–3c** (*ca* 2:2:3) (R_t 2.8 min) and of 30 mg **5a–5c** (*ca* 3:3:1) (R_t 5.5 min) and HPLC (MeOH–H_2O, 3:2) of A_3 afforded 60 mg **3a–3c** (R_t 3.0 min).

HPLC (MeOH–H_2O, 3:2) of fraction B gave five fractions (B_1–B_5). Repeated HPLC (MeOH–H_2O, 1:1) of B_1 gave 2 mg **3d** (R_t 4.8 min), of B_2 5 mg **11a** (R_t 5.8 min), of B_3 4 mg **11b**, of B_4 (HPLC, MeOH–H_2O, 11:9) 5 mg **10a** (R_t 5.5 min) and of B_5 (HPLC, MeOH–H_2O, 11:9) 2 mg **7a** (R_t 6.9 min).

HPLC (MeOH–H_2O, 11:9) of fraction C combined with D gave 5 mg **1b** (R_t 3.5 min), 6 mg **6d** (R_t 5.0 min) and 40 mg of a mixture of **6a–6c** which was further separated by HPLC (MeOH–H_2O, 1:1) affording 3 mg **6a** (R_t 13.2 min) and a mixture of **6b** and **6c** (R_t 13.8 min).

HPLC (MeOH–H_2O, 11:9) of fraction E gave 20 mg **9a** (R_t 1.2 min), 3 mg **13** (R_t 2.0 min), 20 mg **1a** (R_t 3.1 min), 10 mg **8a** (R_t 3.5 min), 10 mg **1c** (R_t 3.8 min) and a mixture which gave by repeated HPLC (MeOH–H_2O, 11:9) 2 mg **18** (R_t 6.0 min) and 1.5 mg **19** (R_t 6.7 min).

From fraction F 550 mg **1c**, mp 260°, were obtained by

Table 7. ^{1}H NMR spectral data of **23a–23c** (400 MHz, $CDCl_3$, TMS as internal standard)

	23a	C_6D_6	**23b**	**23c**
H-1	2.42 *br dd*	2.04 *br dd*	2.44 *br dd*	2.44 *br dd*
H-2	5 39 *ddd*	5.43 *ddd*	5.36 *ddd*	5.42 *ddd*
H-3	1.82 *m*	1.47 *m*	2.00 *m*	2.02 *m*
H-4	3.67 *ddd*	3.45 *ddd*	4.08 *dd*	4.19 *ddd*
H-4′	3.50 *ddd*	3 30 *ddd*		4.10 *ddd*
H-6	5.67 *br s*	5.26 *br s*	5.67 *br s*	5.67 *br s*
H-7	3.43 *br d*	2.98 *br d*	3.43 *br d*	3.42 *br d*
H-8	4.34 *ddd*	4.22 *ddd*	4.29 *ddd*	4.29 *ddd*
H-9α	2 58 *ddd*	2 36 *ddd*	2.62 *ddd*	2.63 *ddd*
H-9β	1.67 *ddd*	1.37 *ddd*	1.68 *ddd*	1.68 *ddd*
H-10	2.25 *m*	1.74 *m*	2.00 *m*	2.22 *m*
H-13	6.22 *d*	6.19 *d*	6.22 *d*	6.22 *d*
H-13′	5.58 *d*	5.05 *d*	5.59 *d*	5 58 *d*
H-14	1.18 *d*	0.82 *d*	1.18 *d*	1.18 *d*
H-15	1.87 *ddd*	1.63 *ddd*	1 89 *ddd*	1.89 *ddd*
OCOR	2 07 *s*	1.55 *s*	2.05 *s*	6.08 *qq*
			2.03 *s*	1.99 *qq*
				1 89 *dq*
				2.02 *s*

J (Hz): 1, 2 = 5; 1, 6 = 1, 7 ~ 1; 1, 10 = 5; 2, 3 = 4; 2, 3′ = 8.5; 6, 7 = 2; 7, 8 = 9; 7, 13 = 7, 13′ = 3; 8, 9α = 6.5; 8, 9β = 11; 9α, 9β = 13; 9α, 10 = 7; 9β, 10 = 2.5; 10, 14 = 7; compound **23a**: 3, 4 = 3, 4′ = 3′, 4 = 5, 3′, 4′ = 9; 4, 4′ = 13; compound **23b**: 3, 4 = 6; 3′, 4 = 7; compound **23c**: 3, 4 = 5.5; 3′, 4 = 5.5; 3, 4′ = 6; 3′, 4′ = 8; 4, 4′ = 11.5, OAng: 3_1, 4_1 = 7, 3_1, 5_1 = 4_1, 5_1 = 1 5.

crystallization. The mother liquor was combined with fraction G. HPLC (MeOH–H_2O, 11:9) gave 5 mg **9a** (R_t 3 5 min.), 10 mg **2a** (purified by repeated HPLC, MeOH–H_2O, 1:1, R_t 3.9 min) and 10 mg **2b** and **2c** (*ca* 1:1) (R_t 4.3 min).

The purity of all compounds was tested by HPLC and TLC (Et_2O–petrol or $CHCl_3$–C_6H_6–Et_2O mixtures) and by their 400 MHz ^{1}H NMR spectra.

9-Desacetoxyspathulin (**1b**). Colourless crystals, mp 220°; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3610 (OH), 1770 (γ-lactone), 1735 (OAc); MS *m/z* (rel. int.): 264.135 [M – HOAc]$^+$ (3) (calc. for $C_{15}H_{20}O_4$: 264.135), 246 [264 – H_2O]$^+$ (11), 228 [246 – H_2O]$^+$ (4), 107 [C_8H_{11}]$^+$ (100).

$$[\alpha]_{24°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\ \text{nm}}{+14 \quad +17 \quad +20 \quad +33} \ (CHCl_3,\ c\ 0.1).$$

6β,9β-Dihydroxypulchellin-2-O-[2-methylbutyrate] and isovalerate (**2b** *and* **2c**). Not separated colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1770 (γ-lactone), 1730 (OCOR); MS *m/z* (rel. int.): 280.131 [M – HOCOR]$^+$ (8) (calc. for $C_{15}H_{20}O_5$: 280.131), 262 [280 – H_2O]$^+$ (16), 107 [C_8H_{11}]$^+$ (70), 85 [C_4H_9CO]$^+$ (40), 57 [85 – CO]$^+$ (100).

6β-Acetoxy-9β-hydroxypulchellin-2-O-isobutyrate (**3d**). Colourless crystals, mp 215°; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3590 (OH), 1770 (γ-lactone), 1760 (OAc), 1730 (OCOR); MS *m/z* (rel. int.): 322.142 [M – HOCOR]$^+$ (3) (calc. for $C_{17}H_{22}O_6$: 322.142), 262 [322 – HOAc]$^+$ (5), 244 [262 – H_2O]$^+$ (4), 218 [262 – CO_2]$^+$ (6), 107 [C_8H_{11}]$^+$ (48), 71 [C_3H_7CO]$^+$ (100), 55 [C_4H_7]$^+$ (62).

6β,9β-Diacetoxypulchellin-2-O-[2-methylbutyrate] (**4a**). Colourless crystals, mp 166°; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3590 (OH), 1780 (γ-lactone), 1750 (OAc), 1735 (OCOR); MS *m/z* (rel. int.): 406.199 [M – HOAc]$^+$ (0 2) (calc. for $C_{22}H_{30}O_7$. 406.199), 364 [M – HOCOR]$^+$ (10), 346 [364 – H_2O]$^+$ (1), 304 [364 – HOAc]$^+$ (3), 262 [304 – ketene]$^+$ (7), 244 [304 – HOAc]$^+$ (27), 226 [244 – H_2O]$^+$ (7), 85 [C_4H_9CO]$^+$ (64), 57 [85 – CO]$^+$ (100).

$$[\alpha]_{24°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\ \text{nm}}{+13.5 \quad +14.1 \quad +15.3 \quad +21\,8} \ (CHCl_3,\ c\ 0.17).$$

CD (MeCN): $\Delta\varepsilon_{273}$ +0.1, last reading $\Delta\varepsilon_{220}$ strongly negative.

6β,9β-Diacetoxypulchellin-2-O-isovalerate (**4b**). Colourless crystals, mp 164°; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3580 (OH), 1780 (γ-lactone), 1750 (OAc), 1735 (OCOR); MS *m/z* (rel. int.): 466.220 [M]$^+$ (0.8) (calc for $C_{24}H_{34}O_9$: 466.220), 407 [M – OAc]$^+$ (2.5), 406 [M – HOAc]$^+$ (1), 364 [M – HOCOR]$^+$ (8), 304 [364 – HOAc]$^+$ (5), 262 [304 – ketene]$^+$ (10), 244 [304 – HOAc]$^+$ (27), 85 [C_4H_9CO]$^+$ (71), 57 [85 – CO]$^+$ (100).

$$[\alpha]_{22°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\ \text{nm}}{+11\,8 \quad +12.9 \quad +14.6 \quad +18.3} \ (CHCl_3,\ c\ 0.10).$$

4-O-Acetyl-6β-acetoxy-9β-hydroxypulchellin-2-O-angelate (**5a**). Colourless crystals, mp 152°; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3590 (OH), 1780 (γ-lactone), 1750 (OAc), 1715 (OCOR); MS *m/z* (rel. int.): 464.205 [M]$^+$ (0.2) (calc. for $C_{24}H_{32}O_9$: 464.205), 405 [M – OAc]$^+$ (1), 404 [M – HOAc]$^+$ (0.8), 305 [405 – HOCOR]$^+$ (5), 245 [305 – HOAc]$^+$ (11), 83 [C_4H_7CO]$^+$ (100), 55 [83 – CO]$^+$ (67).

$$[\alpha]_{24°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\ \text{nm}}{-29 \quad -30 \quad -33 \quad -52} \ (CHCl_3,\ c\ 0.22).$$

4-O-Acetyl-6β-acetoxy-9β-hydroxypulchellin-2-O-[2-methylbutyrate] and 2-O-isovalerate (**5b** *and* **5c**). Not separated colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3600 (OH), 1780 (γ-lactone), 1745 (OAc), 1735 (OCOR); MS *m/z* (rel. int.): 466.220 [M]$^+$ (0.6) (calc. for $C_{24}H_{34}O_9$: 466.220), 407 [M – OAc]$^+$ (5), 406 [M – HOAc]$^+$ (2), 364 [M – HOCOR]$^+$ (1), 346 [406 – HOAc]$^+$ (1.5), 304 [364 – HOAc]$^+$ (3), 262 [304 – ketene]$^+$ (8), 244 [304 – HOAc]$^+$ (10), 107 [C_8H_{11}]$^+$ (24), 85 [C_4H_9CO]$^+$ (62), 57 [85 – CO]$^+$ (100).

4-O-Acetyl-6β,9β-dihydroxypulchellin-2-O-angelate (**6a**). Colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3580 (OH), 1765 (γ-lactone), 1740 (OAc), 1720 (OCOR); MS *m/z* (rel. int.): 362.173 [M – HOAc]$^+$ (1.2) (calc for $C_{20}H_{26}O_6$: 362.173), 322 [M – HOCOR]$^+$ (4), 304 [322 – H_2O]$^+$ (1.5), 262 [322 – HOAc]$^+$ (28), 107 [C_8H_{11}]$^+$ (88), 83 [C_4H_7CO]$^+$ (100), 55 [83 – CO]$^+$ (60).

$$[\alpha]_{22°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\ \text{nm}}{-10 \quad -11.5 \quad -12.3 \quad -21.5} \ (CHCl_3,\ c\ 0.26).$$

4-O-Acetyl-6β,9β-dihydroxypulchellin-2-O-[2-methylbutyrate] and isovalerate (**6b** *and* **6c**). Not separated colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3590 (OH), 1765 (γ-lactone), 1740 (OAc), 1720 (OCOR); MS *m/z* (rel. int.): 364.189 [M – HOAc]$^+$ (0.8) (calc. for $C_{20}H_{28}O_6$: 364.189), 322 [M – HOCOR]$^+$ (3.5), 304 [322 – H_2O]$^+$ (2), 262 [322 – HOAc]$^+$ (74), 107 [C_8H_{11}]$^+$ (100), 85 [C_4H_9CO]$^+$ (50), 57 [85 – CO]$^+$ (80).

4-O-Acetyl-6β,9β-dihydroxypulchellin-2-O-isobutyrate (**6d**). Colourless crystals, mp 147°; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3580 (OH), 1765 (γ-lactone), 1720 (OCOR); MS *m/z* (rel. int.): 350.173 [M – HOAc]$^+$ (0.7) (calc. for $C_{19}H_{26}O_6$: 350.173), 322 [M – HOCOR]$^+$ (2), 304 [322 – H_2O]$^+$ (1.5), 262 [322 – HOAc]$^+$ (50), 107 [C_8H_{11}]$^+$ (100), 71 [C_3H_7CO]$^+$ (54). CD (MeCN): $\Delta\varepsilon_{270}$ +0.1, $\Delta\varepsilon_{233}$ −0.44.

6α-Angeloyloxypulchellin (**7a**). Colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1770 (γ-lactone), 1730 (OCOR); MS *m/z* (rel. int.): 364.189 [M]$^+$ (0.4) (calc. for $C_{20}H_{28}O_6$: 364.189), 346 [M – H_2O]$^+$ (0.2), 265 [346 – OAc]$^+$ (1), 264 [M – HOCOR]$^+$ (1), 247 [265 – H_2O]$^+$ (2), 246 [264 – H_2O]$^+$ (2.5), 229 [247 – H_2O]$^+$ (2), 228 [246 – H_2O]$^+$ (1.2), 107 [C_8H_{11}]$^+$ (44), 83

$[C_4H_7CO]^+$ (100), 55 $[83-CO]^+$ (90). CD (MeCN): $\Delta\varepsilon_{275}$ +0.1; $\Delta\varepsilon_{238}$ −0.41.

6α-Angeloyloxypulchellin-4-O-isovalerate (**7b**). Colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3590 (OH), 1765 (γ-lactone), 1720 (OCOR); MS *m/z* (rel. int.): 430.236 $[M-H_2O]^+$ (0.5) (calc. for $C_{25}H_{34}O_6$: 430.236), 364 $[M-O{=}C{=}CHCHMe_2]^+$ (0.5), 349 $[M-OAng]^+$ (2.3), 348 $[M-HOAng]^+$ (1), 346 $[M-HOiVal]^+$ (2), 328 $[430-HOiVal]^+$ (3.5), 247 $[349-HOiVal]^+$ (22), 229 $[247-H_2O]^+$ (24), 85 $[C_4H_9CO]^+$ (32), 83 $[C_4H_7CO]^+$ (100), 87 $[85-CO]^+$ (50), 55 $[83-CO]^+$ (62).

Neopulchellin-4-O-angelate (**8b**). Colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3620 (OH), 1770 (γ-lactone), 1730, 1715 (OCOR); MS *m/z* (rel. int.): 348.194 $[M]^+$ (0.2) (calc. for $C_{20}H_{28}O_5$: 348.194), 330 $[M-H_2O]^+$ (0.2), 266 $[M-O{=}C{=}C(Me){=}CHMe]^+$ (0.4), 249 $[M-OCOR]^+$ (10), 248 $[M-HOCOR]^+$ (8), 230 $[248-H_2O]^+$ (10), 83 $[C_4H_7CO]^+$ (100), 55 $[83-CO]^+$ (42).

8α-Hydroxyneopulchellin (**9a**). Colourless oil, IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3570 (OH), 1760 (γ-lactone); MS *m/z* (rel. int.): 264.136 $[M-H_2O]^+$ (2) (calc. for $C_{15}H_{20}O_4$: 264.136), 246 $[264-H_2O]^+$ (2.8), 228 $[246-H_2O]^+$ (2.2), 218 $[246-CO]^+$ (7), 107 $[C_8H_{11}]^+$ (100).

$$[\alpha]_{22°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\,nm}{+21.4 \quad +22.6 \quad +26.1 \quad +48.4} (CHCl_3,\ c\ 1.31)$$

Compound **9a** (3 mg) was acetylated (0.5 ml Ac_2O, 80°, 90 min). After evaporation the residue was purified by TLC ($CHCl_3$–C_6H_6–Et_2O, 1:1:1, three developments) to give 2 mg **9b**, colourless crystals (Et_2O–petrol) mp 196°; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1760 (γ-lactone), 1730 (OAc); MS *m/z* (rel. int.): 324.157 $[M]^+$ (0.4) (calc. for $C_{17}H_{24}O_6$: 324.157), 306 $[M-H_2O]^+$ (1 2), 264 $[M-HOAc]^+$ (3.5), 246 $[264-H_2O]^+$ (20), 228 $[246-H_2O]^+$ (8), 218 $[246-CO]^+$ (20), 107 $[C_8H_{11}]^+$ (100).

$$[\alpha]_{22°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\,nm}{+42.5 \quad +47.5 \quad +57.5 \quad +100} (CHCl_3,\ c\ 0\,04).$$

Triacetate **9c**. Compound **9a** (3 mg) was acetylated in the presence of 5 mg 4-dimethylaminopyridine [22] in 0.5 ml Ac_2O at 80°. TLC (Et_2O–petrol, 3:1) gave 2 mg **9c**, colourless oil; MS *m/z* (rel. int.): 348.157 $[M-HOAc]^+$ (1) (calc. for $C_{19}H_{24}O_6$: 348.157), 288 $[348-HOAc]^+$ (5), 228 $[288-HOAc]^+$ (22), 107 $[C_8H_{11}]^+$ (100)

6α-Angeloyloxyneopulchellin (**10a**). Colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3610 (OH), 1770 (γ-lactone), 1730 (OCOR); MS *m/z* (rel. int.): 364.189 $[M]^+$ (1.5) (calc. for $C_{20}H_{28}O_6$· 364.189), 346 $[M-H_2O]^+$ (1), 265 $[M-OCOR]^+$ (4), 264 $[M-HOCOR]^+$ (1.5), 247 $[265-H_2O]^+$ (5), 246 $[264-H_2O]^+$ (2), 229 $[247-H_2O]^+$ (6), 107 $[C_8H_{11}]^+$ (18), 83 $[C_4H_7CO]^+$ (100), 55 $[83-CO]^+$ (76). CD (MeCN): $\Delta\varepsilon_{253} = -2.6$

6α-Angeloyloxyneopulchellin-2-O-acetate (**10b**). Colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}. 3600 (OH), 1775 (γ-lactone), 1755 (OCOR), MS *m/z* (rel. int.): 406.201 $[M]^+$ (0.8) (calc. for $C_{22}H_{30}O_7$. 406.201), 346 $[M-HOAc]^+$ (1), 247 $[346-OCOR]^+$ (3), 246 $[346-HOCOR]^+$ (6), 107 $[C_8H_{11}]^+$ (34), 83 $[C_4H_7CO]^+$ (100), 55 $[83-CO]^+$ (35).

6α-Hydroxyneopulchellin-4-O-angelate (**11a**). Colourless crystals, mp 137° (Et_2O–petrol); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3590 (OH), 1760 (γ-lactone), 1720 (OCOR); MS *m/z* (rel. int.): 364.189 $[M]^+$ (1) (calc. for $C_{20}H_{28}O_6$: 364.189), 346 $[M-H_2O]^+$ (0.8), 265 $[M-OCOR]^+$ (2.2), 264 $[M-HOCOR]^+$ (2), 247 $[265-H_2O]^+$ (8), 246 $[264-H_2O]^+$ (4), 229 $[247-H_2O]^+$ (6), 107 $[C_8H_{11}]^+$, (18), 83 $[C_4H_7CO]^+$ (100), 55 $[83-CO]^+$ (82).

$$[\alpha]_{22°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\,nm}{-51\,5 \quad -53.9 \quad -58.5 \quad -107.3} (CHCl_3,\ c\ 0.33).$$

6α-Hydroxyneopulchellin-4-O-[2-methyl butyrate] (**11b**). Colourless crystals, mp 160° (Et_2O–petrol); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3600 (OH), 1770 (γ-lactone), 1730 (OCOR); MS *m/z* (rel. int.): 366.204 $[M]^+$ (1.5) (calc. for $C_{20}H_{30}O_6$: 366.204), 348 $[M-H_2O]^+$ (4), 265 $[M-OCOR]^+$ (2), 264 $[M-HOCOR]^+$ (6), 247 $[265-H_2O]^+$ (3), 246 $[264-H_2O]^+$ (6), 218 $[246-CO]^+$ (10), 107 $[C_8H_{11}]^+$ (48), 85 $[C_4H_9CO]^+$ (30), 57 $[85-CO]^+$ (100).

$$[\alpha]_{22°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\,nm}{-9.3 \quad -9.3 \quad -10.5 \quad -16.3} (CHCl_3;\ c\ 0.40).$$

6α-Hydroxyneopulchellin-4-O-isovalerate (**11c**). Colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3600 (OH), 1770 (γ-lactone), 1735 (OCOR); MS *m/z* (rel. int.): 348.194 $[M-H_2O]^+$ (4) (calc. for $C_{20}H_{28}O_5$: 348.194), 264 $[M-HOCOR]^+$ (8), 246 $[264-H_2O]^+$ (20), 218 $[246-CO]^+$ (15), 107 $[C_8H_{11}]^+$ (60), 85 $[C_4H_9CO]^+$ (54), 57 $[85-CO]^+$ (100).

6α-Angeloyloxyneopulchellin-4-O-isovalerate (**12**). Colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3620 (OH), 1780 (γ-lactone), 1740 (OCOR); MS *m/z* (rel. int.): 448.246 $[M]^+$ (0.2) (calc. for $C_{25}H_{36}O_7$: 448.246), 349 $[M-OAng]^+$ (2), 348 $[M-HOAng]^+$ (6), 346 $[M-HOiVal]^+$ (1), 328 $[346-H_2O]^+$ (1), 247 $[348-OiVal]^+$ (8), 229 $[247-H_2O]^+$ (11), 85 $[C_4H_9CO]^+$ (14), 83 $[C_4H_7CO]^+$ (100), 57 $[85-CO]^+$ (32), 55 $[83-CO]^+$ (52). CD (MeCN): $\Delta\varepsilon_{255} = -0.21$

2α-Angeloyloxydugaldiolide (**18**). Colourless crystals, mp 94° (Et_2O–petrol); IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3605, 3540, 3400 (OH), 1780 (γ-lactone), 1730, 1717 (OCOR); MS *m/z* (rel. int.): 280.131 $[M-HOCOR]^+$ (3) (calc. for $C_{15}H_{20}O_5$: 280 131), 262 $[280-H_2O]^+$ (23), 83 $[C_4H_7CO]^+$ (100), 55 $[83-CO]^+$ (86).

$$[\alpha]_{22°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436 \quad 365\,nm}{+25.3 \quad +27.4 \quad +30.9 \quad +54.0 \quad +83.7} (CHCl_3;\ c\ 0.43).$$

2α-[2-Methylbutyryloxy]-dugaldiolide (**19**). Colourless oil; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3580, 3380 (OH), 1770 (γ-lactone), 1725 (OCOR); MS *m/z* (rel. int.): 280.131 $[M-HOCOR]^+$ (6) (calc for $C_{15}H_{20}O_5$: 280.131), 262 $[280-H_2O]^+$ (60), 244 $[262-H_2O]^+$ (3), 85 $[C_4H_9CO]^+$ (32), 57 $[85-CO]^+$ (100).

4,5-Seco-neopulchell-5-ene-2-O-acetate (**23a**). Colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 3620 (OH), 1780 (γ-lactone), 1740, 1240 (OAc); MS *m/z* (rel. int.): 248.141 $[M-HOAc]^+$ (10) (calc. for $C_{15}H_{20}O_3$: 248.141), 107 $[C_8H_{11}]^+$ (100), 91 (21). CD (MeCN): $\Delta\varepsilon_{282} = -2\,5$. Compound **23a** (3 mg) was acetylated (0.5 ml Ac_2O, 60 min, 80°). After evaporation and TLC (Et_2O–petrol, 3:1, R_f 0.4) 2.5 mg **23b** were obtained, colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1775 (γ-lactone), 1745, 1240 (OAc); MS *m/z* (rel. int.): 290.152 $[M-HOAc]^+$ (1) (calc. for $C_{17}H_{22}O_4$. 290.152), 248 $[M-ketene]^+$ (2), 230 $[M-HOAc]^+$ (11), 215 $[230-Me]^+$ (4), 107 $[C_8H_{11}]^+$ (100).

4-O-Acetyl-4,5-seco-neopulchell-5-ene-2-O-angelate (**23c**). Colourless oil; IR $\nu_{max}^{CCl_4}$ cm^{-1}: 1780 (γ-lactone), 1745, 1240 (OAc), 1720 (OCOR); MS *m/z* (rel. int.). 390.204 $[M]^+$ (0.3), (calc. for $C_{22}H_{30}O_6$. 390.204), 348 $[M-ketene]^+$ (0.6), 330 $[M-HOAc]^+$ (1.6), 302.151 $[M-EtOAc]^+$ (0.5), 230 $[330-HOAng]^+$ (22), 107 $[C_8H_{11}]^+$ (93), 83 $[C_4H_7CO]^+$ (100), 55 $[83-CO]^+$ (47).

$$[\alpha]_{22°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436\,nm}{-37.5 \quad -37.5 \quad -46.7 \quad -75.0} (CHCl_3;\ c\ 0.57).$$

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