New Mono- and Bisdesmosidic Triterpene Glycosides from *Pittosporum angustifolium* Lodd.

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This work is respectfully dedicated to the loving memory of Adelheid Bombor, a unique character and the most amiable and courageous person I have ever known (C. B.)

Fifteen new mono- and bisdesmosidic triterpene saponins, named pittangretosides J, K, M, Q–Z, A_1, and B_1, along with three known compounds were isolated from the leaves of *Pittosporum angustifolium*. By spectroscopic, mass spectrometric and chemical evidence, their structures were established as glycosides of A_1- and R_1-barrigenol, barringtogenol C and camelliagenin A backbones.

Key words: Pittosporum angustifolium, Pittosporaceae, Triterpene Saponins, Pittangretosides

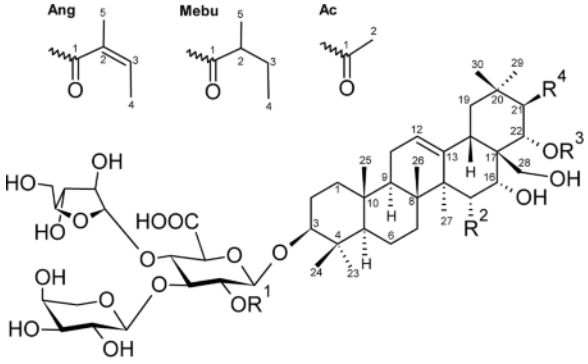
Introduction

Pittosporum angustifolium Lodd. (Pittosporaceae) is a small tree, growing endemically in Australia's inland areas. The plant is colloquially known as "gumby gumby", and various medical preparations of the leaves, seeds and fruits are used in the field of Aboriginal ethnomedicine and further for the complementary treatment of malignant diseases [1, 2].

In recent phytochemical studies we have reported the isolation and characterization of twelve new acylated triterpene saponins of the A_1- and R_1-barrigenol type, named pittangretosides A–I and N–P, from the leaves [2] and the seeds [3] of *P. angustifolium*. Depending on the acylation pattern of those compounds, cytotoxic activity against different cancer cell lines has been observed [2, 3]. Additionally, five known polyphenolic constituents have been isolated from the leaves [4]. Although the polyphenolic phytochemistry of other *Pittosporum* species remains almost completely uninvestigated, triterpene glycosides could be characterized as dominant secondary metabolites in a few *Pittosporum* species [5 – 9], as well. Our ongoing phytochemical screening of the leaves of *P. angustifolium* resulted in the identification of another eighteen triterpene saponins. Fifteen of these are described for the first time as natural products, and their isolation and structural elucidation is herewith reported.

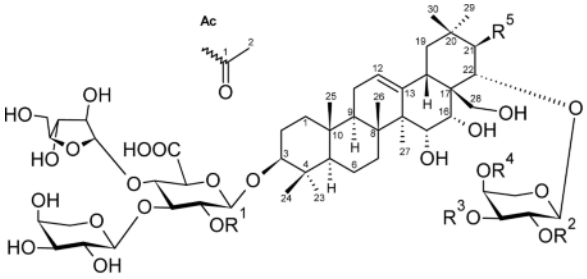
Results and Discussion

The defatted crude 80 % (v/v) ethanol extract of the leaves was purified and fractionated by chromatographic procedures, using Sephadex LH20, silica gel and RP18 solid phase extraction. For the isolation of compounds **1**–**18** (Figs. 1 and 2), obtained purified subfractions were subjected to semipreparative HPLC. New natural products **1**–**15** were named pittangretosides J, K, M, Q–Z, A_1, and B_1.



	R¹	R²	R³	R⁴
1	Gal	OH	H	H
2	Glc	OH	H	H
4	Gal	H	Ang	H
6	Gal	OH	Ang	OAng
7	Gal	H	Ang	OAng
13	Glc	OH	H	OAng
14	Gal	H	Ac	OAng
15	Glc	H	Ac	OAng
16	Glc	OH	Ac	OAng
17	Glc	OH	Mebu	H
18	Glc	OH	Ang	OAng

Fig. 1. Monodesmosidic triterpene saponins isolated from the leaves of *Pittosporum angustifolium*.



	R¹	R²	R³	R⁴	R⁵
3	Glc	Ac	Ac	Ac	OH
5	Glc	H	H	H	OH
8	Gal	Ac	Ac	Ac	OH
9	Glc	H	Ac	H	OH
10	Glc	H	H	Ac	OH
11	Gal	Ac	Ac	Ac	H
12	Glc	Ac	Ac	Ac	H

Fig. 2. Bisdesmosidic triterpene saponins isolated from the leaves of *Pittosporum angustifolium*.

According to their spectroscopic data, compounds **16**–**18** were identified as the known R_1-barrigenol glycosides 21β-angeloyloxy-22α-acetoxy-3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-15α,16α,28-triol (**16**) [5], 22α-(2-methylbutyroyloxy)-3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-15α,16α,28-triol (**17**) [6], and 21β-angeloyloxy-22α-angeloyloxy-3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-15α,16α,28-triol (**18**) [5].

Pittangretoside J (**1**) displayed in its high-resolution ESI mass spectrum a quasimolecular ion $[M–H]^-$ at $m/z = 1091.5256$ (neg. mode), predictive of a molecular formula of $C_{52}H_{84}O_{24}$. Seven tertiary methyl groups at $\delta_H = 0.92$, 0.98, 0.99, 1.00, 1.04, 1.10, and 1.39 ppm, an olefinic proton resonance at $\delta_H =$ 5.38 ppm in the ^{1}H NMR spectrum (Table 1) and further comparison of the NMR spectroscopic data of **1** with those of known structures [2] confirmed an A_1-barrigenol (olean-12-ene-3β, 15α, 16α, 22α, 28-pentol) aglycone backbone. The ^{1}H NMR spectrum showed four anomeric protons at $\delta_H = 4.49$ (d, $J = 7.6$), 4.87 (d, $J = 7.0$), 4.95 (d, $J = 8.0$), and 5.14 (br s) ppm, corresponding to $\delta_C = 105.7$, 103.8, 103.9, and 108.5 ppm in the HMQC spectrum (Table 2). As one of the anomeric carbon atoms ($\delta_C = 105.7$) showed a cross peak with H-3 of the aglycone part in the HMBC spectrum, and the chemical shift of C-3 at $\delta_C = 90.4$ ppm was indicative of a glycosylation, too, an attachment of the oligosaccharide chain to C-3 of the aglycone could be assigned. By using extensive H-H COSY, HMBC, and HMQC experiments, the glycoside moiety was determined as a 2,3,4-trisubstituted β-glucuronopyranosic acid, as recently identified for A_1-barrigenol glycosides from *P. angustifolium* [2]. Unequivocal assignments were made by HMBC cross peaks between H-1 of a β-galactopyranose and $\delta_C = 80.8$ ppm (GlcA-2), H-1 of an α-arabinopyranose and $\delta_C = 79.9$ ppm (GlcA-3), and H-1 of an α-arabinofuranose and $\delta_C = 74.7$ ppm (GlcA-4). Thiazolidine carboxylate analysis by GC-MS revealed a D configuration for β-glucuronic acid and β-galactose as well as an L configuration for α-arabinose. In contrast to the A_1-barrigenol gly-

Table 1. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the aglycone moieties of compounds **1**–**3** in CD_3OD (*J* in Hz)[a].

Position	**1**		**2**		**3**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
1	39.7	1.64, 1.01	39.9	0.98, 1.63	39.5	1.01, 1.65
2	26.0	1.73, 1.94	n. d.	1.73, 1.95	23.5	1.76, 1.65
3	90.4	3.20	91.5	3.17 dd (5.0;11.5)	91.2	3.23 dd (4.8;13.7)
4	39.5	–	40.5	–	39.4	–
5	55.5	0.79 d (11.7)	56.5	0.79 d (11.8)	55.6	0.80 d (12.0)
6	18.3	1.44, 1,58	n. d.	1.39, 1.57	18.6	1.44, 1.55
7	35.8	1.70, 1.78	36.9	1.72, 1.77	36.2	1.75, n. d.
8	41.4	–	42.4	–	41.2	–
9	47.9	1.57	48.3	1.55	47.2	1.58
10	36.9	–	38.0	–	36.8	–
11	23.7	1.88, 1.96	n. d.	1.91	23.5	1.89, 1.94
12	124.7	5.38 brs	125.2	5.36 brs	126.1	5.46 br s
13	143.8	–	144.8	–	143.0	–
14	47.9	–	48.9	–	47.5	–
15	67.3	3.94 d (4.4)	68.5	3.93 d (4.5)	67.6	3.66
16	72.5	4.15 d (4.4)	73.1	4.14 d (4.5)	73.0	3.65
17	44.3	–	n. d.	–	48.7	
18	42.0	2.16 dd (3.5;14.5)	43.3	2.15 dd (3.5;14.6)	40.4	2.56 brd (14.1)
19	46.8	1.01, 2.37 t (13.7)	47.5	1.00, 2.37 t (13.5)	46.6	2.46 t (13.4), 1.07
20	31.6	–	32.3	–	35.5	–
21	45.1	1.44, 2.07 t (12.5)	45.4	1.43, 2.06 t (12.5)	76.5	4.07
22	73.0	4.05 dd (5.5;12.0)	73.9	4.05 dd (5.5;12.0)	83.9	4.07
23	27.8	1.10 s	28.2	1.08 s	27.4	1.10 s
24	15.3	0.99 s	16.8	0.87 s	15.8	0.89 s
25	16.1	1.00 s	16.1	0.97 s	15.1	1.01 s
26	16.8	1.04 s	17.7	1.02 s	16.8	1.04 s
27	19.4	1.39 s	20.7	1.37 s	19.7	1.37 s
28	67.5	3.26 d (10.5), 3.55 d (10.5)	68.7	3.25 d (10.5), 3.53 d (10.5)	60.8	3.24 d (11.3), 3.65 d (11.3)
29	32.8	0.92 s	33.5	0.91 s	28.8	0.98 s
30	24.5	0.98 s	24.6	1.08 s	18.2	0.93 s
Acyl	–	–	–	–	Ac [Ara (p) II C-2]	
1	–	–	–	–	169.6	–
2	–	–	–	–	20.8	2.15 s
	–	–	–	–	Ac [Ara (p) II C-3]	
1	–	–	–	–	170.2	–
2	–	–	–	–	20.6	1.98 s
	–	–	–	–	Ac [Ara (p) II C-4]	
1	–	–	–	–	170.4	–
2	–	–	–	–	20.7	2.11 s

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; Ara (p) II, second arabinopyranose at C-22; Ac, acetic acid.

cosides recently isolated from *P. angustifolium* [2], no additional signals of acyl substituents were observed and H-15 ($\delta_H = 3.94$), H-16 ($\delta_H = 4.15$), H-22 ($\delta_H = 4.05$), and H-28 ($\delta_H = 3.26$, 3.55 ppm) showed normal shifts for unsubstituted hydroxymethine protons and one primary alcoholic function, respectively. Thus, the new natural product pittangretoside J (**1**) was elucidated as 3β-[β-D-galactopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-15α,16α,22α,28-tetrol.

The NMR spectroscopic data of pittangretoside K (**2**) displayed a strong similarity to those of compound **1** (Tables 1 and 2). Since the ESI mass spectrum revealed the same molecular formula of $C_{52}H_{84}O_{24}$, which was deduced from a quasimolecular ion [M–

Table 2. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the sugar moieties of compounds **1–3** in CD_3OD (J in Hz)[a].

Position	**1**		**2**		**3**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
C-3		GlcA		GlcA		GlcA
1	105.7	4.49 d (7.6)	105.3	4.45 d (7.6)	104.6	4.57 d (6.5)
2	80.8	3.93	79.8	3.92	78.9	3.94
3	79.9	3.88	80.0	3.87	79.5	3.89
4	74.7	3.88	74.6	3.89	73.7	3.82
5	79.5	3.75	79.9	3.87	78.9	3.82
6	n. d.	–	n. d.	–	n. d.	–
		Gal		Glc		Glc
1	103.8	4.87 d (7.0)	102.6	5.00 d (7 .0)	101.9	5.03 d (7.8)
2	73.5	3.58	75.9	3.20	74.9	3.22
3	75.3	3.49	77.7	3.38	77.2	3.40 t (9.0)
4	73.0	3.65	72.2	3.12 t (9.2)	71.4	3.12 t (9.6)
5	77.4	3.48	78.0	3.30	77.8	3.32
6	61.9	3.67, 3.75	63.2	3.58, 3.82	63.4	3.67 dd (5.1;11.3), 3.87
		Ara (p)		Ara (p)		Ara (p)
1	103.9	4.95 d (8.0)	103.7	4.94 d (7.9)	103.1	4.92 d (7.8)
2	73.0	3.65	72.9	3.58	72.3	3.60
3	75.1	3.52	74.1	3.51	73.1	3,52
4	70.2	3.75	70.2	3.75	69.5	3.79
5	67.2	3.57, 3.83	67.2	3.55, 3.83	66.6	3.52 dd (9.6;3.1), 3.84
		Ara (f)		Ara (f)		Ara (f)
1	108.5	5.14 br s	107.9	5.19 br s	107.4	5.10 brs
2	81.5	3.95 br s	81.5	3.96 br s	81.0	3.97 brs
3	79.5	3.75	79.3	3.75	78.6	3.77
4	87.2	4.43 q (4.4)	87.1	4.40 q (4.4)	86.3	4.47 q (4.1)
5	63.1	3.66, 3.58	63.4	3.66, 3.57	62.6	3.71 dd (11.7;3.4); 3.57
						Ara (p) II (C-22)
1	–	–	–	–	102.5	4.73 d (7.7)
2	–	–	–	–	70.5	5.23 dd (8.4, 10.4)
3	–	–	–	–	71.3	5.15 dd (3.3, 10.0)
4	–	–	–	–	68.6	5.31 brs
5	–	–	–	–	64.3	4.05 m, 3.91

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; GlcA, glucuronopyranosic acid; Glc, glucopyranose; Gal, galactopyranose; Ara (p), arabinopyranose; Ara (f), arabinofuranose; Ara (p) II, second arabinopyranose.

H]$^-$ at m/z = 1091.5263 (neg. mode), and as compounds **2** and **1** were poorly separable by HPLC, their structural differences were assumed to be very small. Indeed, only the sugar resonances showed deviations, and no characteristic signals of a β-galactopyranose as found in compound **1** were observed. Instead, chemical shifts for the proton and carbon resonances of a β-glucopyranose unit were assigned [C/H-1: δ_C = 102.6, δ_H = 5.00 (d, J = 7.0); C/H-2: δ_C = 75.9, δ_H = 3.20; C/H-3: δ_C = 77.7, δ_H = 3.38; C/H-4: δ_C = 72.2, δ_H = 3.12; C/H-5: δ_C = 78.0, δ_H = 3.30; C/H-6: δ_C = 63.2, δ_H = 3.58, 3.82 ppm]. This was also supported by acid hydrolysis and subsequent TLC and GC-MS procedures, which gave signals of β-glucuronic acid, β-glucose and α-arabinose. The absolute configurations of sugars were determined as thiazolidine carboxylates, indicating the presence of β-D-glucuronic acid, β-D-glucose and α-L-arabinose. The linkage of the oligosaccharide chain turned out to be the same as in **1**, as HMBC cross peaks between H-1 of the β-glucopyranose and δ_C = 79.8 ppm (GlcA-2), H-1 of the α-arabinopyranose and δ_C = 80.0 ppm (GlcA-3), and H-1 of an α-arabinofuranose and δ_C = 74.6 ppm (GlcA-4) were detected. The novel compound pittangretoside K (**2**) was consequently established as 3β-[β-D-glucopyranosyl-(1→2)]-[α-

L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-15α,16α,22α,28-tetrol.

The ESI mass spectrum of pittangretoside M (**3**) showed quasimolecular ions $[M–H]^-$ at m/z = 1365.5889 (neg. mode) and $[M+2Na]^{2+}$ at m/z= 706.2785 (pos. mode) that were compatible with a molecular formula of $C_{63}H_{98}O_{32}$. In the ^1H NMR spectrum five resonances of anomeric protons were observed at δ_H = 4.57 (d, J = 6.5), 4.73 (d, J = 7.7), 4.92 (d, J = 7.8), 5.03 (d, J = 7.8), and 5.10 (br s) ppm, and the corresponding carbon atom signals were assigned by HMBC correlations at δ_C = 104.6, 102.5, 103.1, 101.9, and 107.4 ppm, respectively (Table 2). Furthermore, an olefinic proton resonance at δ_H = 5.46 ppm, seven signals of tertiary methyl groups at δ_H = 0.89, 0.93, 0.98, 1.01, 1.04, 1.10, and 1.37 ppm, six resonances for oxygenated carbons (Table 1) and comparison with literature data [3] identified compound **3** as a derivative of R_1-barrigenol (olean-12-ene-3β, 15α, 16α, 21β, 22α, 28-hexol). Additionally, the NMR spectroscopic data included characteristic signals of three acetyl residues. On acid hydrolysis, **3** gave evidence for β-glucuronic acid, β-glucose and α-arabinose. The corresponding thiazolidine carboxylates, analyzed by GC-MS, underlined the absolute D configuration for β-glucuronic acid and β-glucose, as well as the L configuration for α-arabinose. Interpretation of two-dimensional NMR spectra, including H-H COSY, HMBC, and HMQC, led to the same oligosaccharide chain as well as the same attachment at C-3 of the aglycone part as in compound **2**, because a long range correlation of one of the anomeric carbon atoms (δ_C = 104.6 ppm) and H-3 (3.23 ppm) was clearly observed. The fifth sugar unit, identified as a second arabinopyranose (Ara II), showed also a long range correlation between its anomeric carbon atom (δ_C = 102.5 ppm) and the H-22 (δ_H = 4.07 ppm) of the aglycone which indicates an attachment at this position. The proton resonances of H-2, H-3, and H-4 of that second arabinopyranose were suspiciously shifted downfield with δ_H = 5.23 (dd, J = 8.4, 10.4), 5.15 (dd, J = 3.3, 10.0), and 5.31 (br s) ppm, respectively. Since HMBC correlations revealed cross peaks between H-2, H-3, and H-4 with a corresponding carbonyl carbon of one of the three acetyl groups, the second sugar attachment was unambiguously identified as a threefold acetylated arabinopyranose. So far, pittangretoside M (**3**) is the first bisdesmosidic triterpene saponin isolated from *P. angustifolium*, and its novel structure was thus elucidated as 3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxy-22α-(2,3,4-triacetyloxy-α-L-arabinofuranosyloxy)-olean-12-ene-15α,16α,21β,28-tetrol.

The ESI mass spectrum of pittangretoside Q (**4**) displayed a quasimolecular ion $[M–H]^-$ at m/z = 1157.5742 (neg. mode) predicting a molecular formula of $C_{57}H_{90}O_{24}$, which meant one oxygen atom less than pittangretosides A and B which were recently isolated from the leaves of *P. angustifolium* [2]. Because the NMR spectra of compound **4** were nearly identical to those of pittangretoside B, again a strong structural similarity was proposed (Tables 3 and 4). Characteristic signals of an angeloyl residue were observed at δ_H = 6.08 (q, J = 7.1) ppm for a methine proton resonance as well as a methyl doublet at δ_H = 1.98 ppm and another methyl singlet at δ_H = 1.91 ppm. The linking position at the aglycone moiety was determined at C-22, because a long range correlation between H-22 (δ_H = 5.46 ppm) and the carbonyl carbon atom of the angeloyl residue has been observed in the HMBC spectrum. The oligosaccharide part turned out to be same as in compound **1** and pittangretoside B [2], consisting of a β-glucuronopyranosic acid, a β-galactopyranose, an α-arabinopyranose, and an α-arabinofuranose, which was also supported by hydrolysis and subsequent TLC and GC-MS analysis. Again, the absolute configuration was determined as D for β-glucuronic acid and β-galactose and L for α-arabinose. In contrast to the similar compound pittangretoside B [2], the proton and carbon resonances of compound **4** at C-15 were shifted upfield (δ_H = 1.34, δ_C = 35.0 ppm), leading to the assumption, that this position was not oxidized as in pittangretoside B. Thus, the aglycone part was identified as camelliagenin A (15-desoxy-A_1-barrigenol), and the structure of pittangretoside Q was established as 22α-angeloyloxy-3β-[β-D-galactopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-16α,28-diol.

Pittangretoside R (**5**) showed in its ESI mass spectrum a quasimolecular ion $[M–H]^-$ at m/z = 1239.5833 (neg. mode), corresponding to a molecular formula of $C_{57}H_{92}O_{29}$. From the ^1H NMR spectrum five anomeric protons were assigned

Table 3. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the aglycone moieties of compounds **4–6** in CD_3OD (J in Hz)[a].

Position	**4**		**5**		**6**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
1	39.9	0.99, 1.64	40.1	0.99, 1.64	39.1	1.00, 1.64
2	25.9	1.73, 1.94	23.8	1.72, 1.92	23.9	1.70, 1.94
3	91.2	3.17 dd (4.1;10.1)	91.8	3.19	90.6	3.20 dd (3.4;12.4)
4	39.7	–	40.3	–	39.4	–
5	56.8	0.80 d (12.0)	56.4	0.79 d (12.0)	55.6	0.81 d (11.8)
6	18.9	1.41, 1.59	19.2	1.36, 1.56	19.0	1.42, 1.60
7	33.8	n. d.	37.2	1.71, 1.76	35.6	1.72, 1.77
8	41.8	–	42.1	–	41.3	–
9	47.5	1.57	48.1	1.59	47.5	1.59
10	36.9	–	36.2	–	36.8	–
11	23.9	1.95	24.5	1.92	24.5	1.93
12	124.4	5.37 br s	126.1	5.41 br s	126.0	5.50 br s
13	143.9	–	144.0	–	142.4	–
14	42.7	–	48.4	–	47.8	–
15	35.0	1.34	68.4	3.79 d	67.8	3.66
16	70.1	4.13 br s	72.8	4.10 d (4.0)	73.5	3.89
17	46.0	–	49.4	–	47.6	–
18	41.2	2.53 brd (15.2)	41.9	2.36 brd (15.0)	40.4	2.66
19	47.8	1.07, 2.47 t (12.5)	47.8	1.04 dd (3.0; 11.9), 2.45 t (13.1)	46.4	1.22 d (10.2), 2.66
20	31.9	–	36.2	–	36.1	–
21	41.6	1.57, 2.28 t (11.6)	77.6	4.03 d (9.5)	78.8	5.97 d (10.7)
22	73.1	5.46 dd (5.2;12.0)	87.4	3.88 d (9.5)	72.9	5.64 d (10.7)
23	28.1	1.10 s	28.2	1.07 s	27.6	1.11 s
24	16.8	0.90 s	16.7	0.87 s	15.9	0.89 s
25	15.8	0.99 s	16.1	0.99 s	15.4	1.00 s
26	17.3	0.97 s	17.5	1.02 s	16.9	1.03 s
27	27.1	1.51 s	20.7	1.36 s	19.8	1.44 s
28	63.9	3.07 d (10.9), 3.29	64.8	2.31, 3.51	62.7	3.06 d (10.7), 3.32
29	32.6	0.93 s	30.1	0.96 s	28.5	0.89 s
30	24.8	1.07 s	19.0	0.91 s	19.1	1.12 s
Acyl		Ang (C-21)				Ang (C-21)
1	169.1	–	–	–	168.4	–
2	129.8	–	–	–	128.3	–
3	137.3	6.08 q (7.1)	–	–	138.2	6.08 q (7.5)
4	15.4	1.98	–	–	15.2	1.94
5	20.9	1.91	–	–	19.6	1.86
						Ang (C-22)
1	–	–	–	–	168.4	–
2	–	–	–	–	128.3	–
3	–	–	–	–	138.2	6.08 q (7.5)
4	–	–	–	–	15.2	1.94
5	–	–	–	–	19.6	1.86

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; Ang, angelic acid.

at $\delta_H = 4.33$ (d, $J = 7.7$), 4.51 (d, $J = 7.6$), 4.91 (d, $J = 7.5$), 5.01 (d, $J = 7.7$), and 5.13 (br s) ppm (Table 4). A detailed look at H-H COSY, HMBC, and HMQC spectra revealed the same bisdesmosidic sugar linkage as in compound **3**. Again, a tetrasaccharide chain was attached at C-3, and a second arabinopyranose (Ara II) was linked to C-22. Thiazolidine carboxylates of a hydrolyzed sugar portion showed peaks for β-D-glucuronic acid, β-D-glucose and α-L-arabinose. The aglycone backbone was also established as R_1-Barrigenol (Table 3). However, compared to compound **3**, the proton resonances of Ara II at H-2 ($\delta_H = 3.61$ ppm), H-3 ($\delta_H = 3.51$ ppm), and H-4 ($\delta_H = 3.81$ ppm) showed a normal shift for

Table 4. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the sugar moieties of compounds **4**–**6** in CD_3OD (J in Hz)[a].

Position	**4**		**5**		**6**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
C-3		GlcA		GlcA		GlcA
1	105.6	4.48 d (7.7)	105.3	4.51 d (7.6)	104.7	4.54 d (7.0)
2	80.0	3.94	79.3	3.92	79.3	3.91
3	79.0	3.90	80.1	3.89	79.9	3.89
4	75.0	3.91	74.8	3.90	73.7	3.82
5	78.2	3.78	77.5	3.78	79.0	3.77
6	n. d.	–	n. d.	–	n. d.	–
		Gal		Glc		Gal
1	103.3	4.88 d (6.5)	102.6	5.01 d (7.7)	102.3	4.88 d (6.8)
2	72.5	3.54	75.9	3.20 dd (7.9; 9.0)	72.2	3.54
3	76.4	3.49	77.8	3.38 t (9.0)	76.0	3.49
4	73.0	3.66	72.3	3.11 t (9.0)	72.5	3.67
5	76.3	3.50	78.2	3.30 m	76.1	3.50
6	62.0	3.66, 3.82	63.2	3.83, 3.65	61.6	3.67, 3.82
		Ara (p)		Ara (p)		Ara (p)
1	103.6	4.94 d (7.5)	103.9	4.91 d (7.5)	102.9	4.94 d (7.6)
2	72.9	3.61	72.8	3.58	72.3	3.59
3	73.0	3.50	72.8	3.51	72.6	3.53
4	69.9	3.75	70.4	3.76	69.7	3.77
5	67.4	3.60, 3,84	67.4	3.56, 3.83	66.8	3.56, 3.84
		Ara (f)		Ara (f)		Ara (f)
1	107.6	5.18 brs	107.9	5.13 brs	106.6	5.14 brs
2	81.7	3.98 brs	81.6	3.96 brs	80.4	3.97 brs
3	79.5	3.76	79.3	3.76	79.0	3.75
4	87.1	4.44 q (4.5)	86.8	4.44 q (4.4)	86.5	4.45 q (4.3)
5	62.6	3.68, 3.57	63.1	3.67, 3.57	63.0	3.66, 3.55
				Ara (p) II (C-22)		
1	–	–	106.3	4.33 d (7.7)	–	–
2	–	–	72.8	3.61	–	–
3	–	–	74.4	3.51	–	–
4	–	–	69.8	3.81	–	–
5	–	–	67.7	3.65, 3.92	–	–

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; GlcA, glucuronopyranosic acid; Glc, glucopyranose; Gal, galactopyranose; Ara (p), arabinopyranose; Ara (f), arabinofuranose; Ara (p) II, second arabinopyranose.

unesterified hydroxyl groups. Furthermore, no signals of acetyl or other acyl groups were observed. This was also underlined by the lack of ATR-IR signals at 1737 and 1228 cm^{-1} that were assigned for the carbonyl part of the threefold acetylated Ara II in compound **3**. The new structure of pittangretoside R was thus determined as 3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxy-22α-L-arabinofuranosyloxyolean-12-ene-15α,16α,21β,28-tetrol.

The ESI mass spectrum of pittangretoside S (**6**) revealed a quasimolecular ion peak $[M–H]^-$ at m/z = 1271.6080 (neg. mode), leading to a molecular formula of $C_{62}H_{96}O_{27}$, which has already been assigned for the known compound **18** [5, 10]. Since the separation of both compounds, **6** and **18**, by HPLC was a challenge, and because of a common ESI-MS fragment ion pattern and almost identical chromatographic behavior, it was presumed that compound **6** could be an isomer of **18**, in which the glucose of the tetrasaccharide chain was again replaced by galactose. This was confirmed by ^{1}H NMR spectra, showing signals of anomeric protons at $\delta_H = 4.54$ (d, $J = 7.0$, GlcA), 4.94 [d, $J = 7.6$, Ara (p)], 5.14 [br s, Ara (f)] and finally 4.88 (d, $J = 6.8$) ppm for the galactose moiety (Table 4),

and, moreover, by TLC and GC-MS analysis. The absolute configuration was determined to be D for β-glucuronic acid and β-galactose and L for α-arabinose. As the remaining part of the chemical structure of compound **6** was identical to that of glycoside **18** (Table 3), the new natural product pittangretoside S was consequently elucidated as 21β-angeloyloxy-22α-angeloyloxy-3β-[β-D-galactopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-15α,16α,28-triol.

Pittangretoside T (**7**) had a molecular formula of $C_{62}H_{96}O_{26}$, deduced from a quasimolecular ion $[M–H]^-$ at m/z = 1255.6174 (neg. mode) in its ESI mass spectrum, just one oxygen atom less than the compounds **6** and **18**. With respect to the NMR spectroscopic data, compound **7** showed a strong structural similarity to glycoside **6**, and resonances for two angeloyl residues as well as for the same oligosaccharide chain as in **6**, consisting of a β-glucuronopyranosic acid, a β-galactopyranose, an α-arabinopyranose, and an α-arabinofuranose, have been assigned (Tables 5 and 6). GC-MS data corroborate an absolute configuration of β-D-glucuronic acid, β-D-galactose and α-L-arabinose. A detailed look at the NMR data revealed again – as already found for compound **4** – a lack of oxidation at C-15 (δ_H = 1.39, 1.70, δ_C = 33.7 ppm) as the only difference between both compounds, **6** and **7**. So, pittangretoside T is the first saponin isolated from *P. angustifolium* possessing a barringtogenol C (15-desoxy-R_1-barrigenol) aglycone. Its new structure was elucidated as 21β-angeloyloxy-22α-angeloyloxy-3β-[β-D-galactopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-16α,28-diol.

The ESI mass spectrum of pittangretoside U (**8**) displayed a quasimolecular ion $[M–H]^-$ at m/z = 1365.5968 (neg. mode) which substantiated a molecular formula of $C_{63}H_{97}O_{32}$, already found for the bisdesmosidic compound **3**. The ^{1}H NMR spectrum of **8** also showed five anomeric protons at δ_H = 4.58 (d, J = 6.8), 4.72 (d, J = 7.8), 4.89 (d, J = 6.8), 4.94 (d, J = 7.5), and 5.11 (br s) ppm, which were assigned to their anomeric carbon atoms at δ_C = 104.7, 102.3, 102.8, 104.0, and 107.4 ppm in the HMQC spectrum (Table 6). Further analysis of two-dimensional NMR spectra revealed that the glucose of the tetrasaccharide chain of compound **3** was again replaced by a galactose moiety in compound **8**. The second arabinopyranose (Ara II) at C-22 was also threefold acetylated as found in **3**, which was supported by NMR data and strong signals in the ATR-IR spectrum at 1736 and 1223 cm^{-1}. On acid hydrolysis, followed by TLC and GC-MS procedures, **8** gave β-D-glucuronic acid, β-D-galactose and α-L-arabinose. Thus, the new compound pittangretoside U was established as 3β-[β-D-galactopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxy-22α-(2,3,4-triacetyloxy-α-L-arabinofuranosyloxy)-olean-12-ene-15α,16α,21β,28-tetrol.

Pittangretoside V (**9**) showed in its ESI mass spectrum a quasimolecular ion peak $[M–H]^-$ at m/z = 1281.5726 (neg. mode), which gave a molecular formula of $C_{59}H_{94}O_{30}$. The NMR data were nearly identical to those of compound **5** (Tables 5 and 6), except for three additional signals at δ_C = 171.6, 20.1 and δ_H = 2.14 (s) ppm corresponding to an acetyl group. Again, a backbone of R_1-barrigenol was deduced from extensive H-H COSY, HMBC, and HMQC experiments, possessing the same branched tetrasaccharide attached to C-3 and a second arabinofuranose (Ara II) at C-22, including identical absolute configurations, as already described for compounds **3**, and **5**. Nevertheless, H-3 of Ara II appeared downfield shifted at δ_H = 4.77 (dd, J = 3.1, 10.0) ppm, and as this proton displayed a long range correlation with the carbonyl carbon atom of the acetyl residue in the HMBC spectrum, the acylation position was unequivocally determined to be at C-3 of Ara II. Consequently, the novel natural product pittangretoside V was elucidated as 3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxy-22α-(3-acetyloxy-α-L-arabinofuranosyloxy)-olean-12-ene-15α,16α,21β,28-tetrol.

Pittangretoside W (**10**) had the same molecular formula as compound **9**, $C_{59}H_{94}O_{30}$, substantiated by its ESI mass spectrum and a quasimolecular ion $[M–H]^-$ at m/z = 1281.5757 (neg. mode). Again, nearly all resonances of recorded NMR spectra were identical to those of compound **9** (Tables 7 and 8), which was isolated from the same purified fraction. This time, no differences between the sugar compositions were observed, since **10** displayed also anomeric protons at δ_H = 4.41 (d, J = 7.7), 4.53 (d, J = 6.5), 4.93 (d,

Table 5. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the aglycone moieties of compounds **7–9** in CD_3OD (J in Hz)[a].

Position	**7**		**8**		**9**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
1	38.9	0.99, 1.66	39.0	0.98, 1.66	38.5	0.99, 1.65
2	23.9	1.74, 1.94	25.7	1.73, 1.88	24.9	1.74, 1.90
3	90.9	3.19 brd (11.4)	90.7	3.20 dd (4.0;12.0)	91.1	3.19
4	39.6	–	39.3	–	39.1	–
5	55.9	0.80 d (11.4)	55.7	0.79 d (11.5)	55.7	0.80 d (11.9)
6	18.8	1.46, 1.60	19.2	1.43, 1.53	19.8	1.45, 1.55
7	32.9	1.71, n. d.	35.8	1.75	35.9	1.76
8	39.9	–	40.8	–	41.4	–
9	47.9	1.56	47.2	1.59	47.4	1.60
10	36.3	–	36.5	–	36.8	–
11	23.8	1.94	23.9	1.92	23.9	1.94
12	124.3	5.41 brs	126.3	5.44 br s	126.0	5.43 br s
13	142.0	–	143.0	–	143.0	–
14	41.6	–	47.6	–	47.4	–
15	33.7	1.39, 1.70	67.7	3.66	67.5	3.81
16	68.7	4.02 brs	73.0	3.65	72.5	4.10 d (4.0)
17	47.2	–	48.9	–	n. d.	–
18	40.4	2.65 brd (15.6)	40.4	2.56 br d (14.4)	41.2	2.39 brd (13.8)
19	46.9	1.22 dd (3.0;11.6), 2.66 t (14.2)	46.5	1.05, 2.46 t (13.1)	46.7	1.07, 2.47 t (13.0)
20	36.0	–	35.3	–	35.3	–
21	78.7	6.01 d (10.1)	76.6	4.04 d (10.0)	77.0	4.06 d (9.6)
22	73.2	5.59 d (10.1)	83.8	4.07 d (10.0)	86.9	3.93 d (9.6)
23	27.4	1.10 s	27.3	1.10 s	27.3	1.10 s
24	16.0	0.89 s	16.3	0.89 s	16.4	0.89 s
25	15.6	1.00 s	15.9	1.00 s	15.8	1.00 s
26	16.3	0.96 s	16.9	1.03 s	17.0	1.03 s
27	26.5	1.51 s	19.7	1.36 s	19.7	1.38 s
28	63.6	2.97 d (10.9), 3.29 d (10.9)	60.8	3.25 d (10.8), 3.66	63.8	3.33, 3.54
29	28.5	0.89 s	28.7	0.98 s	29.4	0.99 s
30	19.2	1.12 s	18.0	0.93 s	18.0	0.93 s
Acyl	Ang (C-21)		Ac [Ara (p) II C-2]		Ac [Ara (p) II C-3]	
1	168.7	–	170.3	–	171.6	–
2	128.5	–	20.0	2.15 s	20.1	2.14 s
3	138.2	6.08 q (7.5)	Ac [Ara (p) II C-3]		–	–
4 (1)	15.0	1.94	170.6	–	–	–
5 (2)	19.5	1.85	19.6	1.98 s	–	–
	Ang (C-22)		Ac [Ara (p) II C-4]			
1	168.7	–	171.0	–	–	–
2	128.5	–	19.5	2.11 s	–	–
3	138.2	6.10 q (7.5)	–	–	–	–
4	15.0	1.94	–	–	–	–
5	19.5	1.85	–	–	–	–

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; Ara (p) II, second arabinopyranose at C-22; Ang, angelic acid, Ac, acetic acid.

$J = 7.5$), 5.04 (d, $J = 7.7$), and 5.15 (br s) ppm, and GC-MS analysis of the corresponding thialzolidine carboxylates confirmed a D configuration for β-glucuronic acid, and β-glucose, and an L configuration for α-arabinose. Instead, the second arabinofuranose (Ara II), whose attachment position to the aglycone has been localized at C-22 by HMBC correlations, showed a clearly downfield shifted resonance of H-4 at $\delta_H = 5.07$ ppm (br s). Further analysis of the HMBC spectrum of compound **10** revealed a cross peak between this proton and the carbonyl carbon atom of an acetyl group. Thus, pittangretosides V and

Table 6. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the sugar moieties of compounds **7–9** in CD_3OD (J in Hz)[a].

Position	**7**		**8**		**9**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
C-3		GlcA		GlcA		GlcA
1	104.3	4.53 d (7.0)	104.7	4.58 d (6.8)	105.1	4.53 d (6.5)
2	79.6	3.91	79.4	3.92	80.3	3.92
3	79.8	3.88	79.6	3.90	78.4	3.91
4	72.7	3.78	72.8	3.79	74.1	3.92
5	79.0	3.78	78.6	3.80	78.7	3.73
6	n.d.	–	n.d.	–	n.d.	–
		Gal		Gal		Glc
1	102.6	4.89 d (6.9)	102.8	4.89 d (6.8)	101.7	5.04 d (7.7)
2	72.0	3.55	72.0	3.56	75.2	3.22 t (8.4)
3	75.8	3.49	75.5	3.48	76.8	3.39 t (9.0)
4	72.2	3.65	72.2	3.65	71.6	3.12 t (9.0)
5	75.8	3.53	75.6	3.52	77.3	3.30
6	61.3	3.67, 3.77	62.1	3.67, 3.81	61.9	3.59, 3.85
		Ara (p)		Ara (p)		Ara (p)
1	102.9	4.94 d (7.7)	103.0	4.94 d (7.5)	102.9	4.93 d (7.5)
2	72.7	3.62	72.5	3.61	72.5	3.57
3	72.3	3.52	72.2	3.53	72.1	3.52
4	69.0	3.78	69.4	3.78	69.6	3.77
5	66.5	3.57, 3.85	66.3	3.57, 3.85	66.8	3.57, 3.85
		Ara (f)		Ara (f)		Ara (f)
1	106.9	5.16 brs	107.4	5.11 brs	107.3	5.15 brs
2	80.6	3.96 brs	80.6	3.97 brs	81.0	3.97 brs
3	79.0	3.77	78.7	3.74	78.1	3.77
4	86.6	4.44 q (4.5)	86.6	4.47 q (3.6)	86.6	4.47
5	63.1	3.66, 3.58	62.0	3.68, 3.58	62.4	3.67 dd (12.1; 4.5), 3.57
				Ara (p) II (C-22)		Ara (p) II (C-22)
1	–	–	102.3	4.72 d (7.8)	105.1	4.45 d (7.7)
2	–	–	70.4	5.24 dd (7.8;10.2)	70.3	3.81
3	–	–	70.9	5.14 dd (3.3;10.2)	76.4	4.77 dd (3.1;10.0)
4	–	–	68.5	5.31 brs	66.9	4.02 brs
5	–	–	64.2	3.89, 4.06	66.6	3.49, 3.93

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; GlcA, glucuronopyranosic acid; Glc, glucopyranose; Gal, galactopyranose; Ara (p), arabinopyranose; Ara (f), arabinofuranose; Ara (p) II, second arabinopyranose.

W differ only in the acetylation position at Ara II, and the new chemical structure of pittangretoside W was determined as 3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl - (1→3)] - [α - L - arabinofuranosyl - (1→4)] - β - D - glucuronopyranosyloxy - 22α - (4 - acetyloxy - α - L - arabinofuranosyloxy) - olean - 12 - ene-15α,16α,21β,28-tetrol.

From the ESI mass spectrum of pittangretoside X (**11**) and a detected quasimolecular ion peak [M–H]$^-$ at m/z = 1349.5986, a molecular formula of $C_{63}H_{98}O_{31}$ was predicted. In contrast to compounds **3** and **8**, this meant the loss of an oxygen atom. A more detailed look at the NMR data (Tables 7 and 8) confirmed the same bisdesmosidic structure as found in compound **8**, with galactose as part of the tetrasaccharide chain at C-3 and a second arabinopyranose (Ara II) linked to C-22. The absolute configuration of the sugars by their corresponding thiazolidine carboxylates revealed D for β-glucuronic acid and β-galactose, and further L for α-arabinose. Since the ATR-IR spectra of compound **11** showed strong signals at 1737 and 1229 cm^{-1}, and the proton resonances of H-2, H-3, and H-4 of Ara II appeared downfield shifted at δ_H = 5.14 (dd, J = 7.7, 10.0), 5.11 (dd, J = 3.0, 10.0), and 5.28, (br s) ppm, respectively, each of those three positions were again

Table 7. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the aglycone moieties of compounds **10**–**12** in CD_3OD (J in Hz)[a].

Position	**10**		**11**		**12**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
1	39.2	0.99, 1.61	39.8	0.97, 1.65	39.1	0.99, 1.62
2	23.7	1.75, 1.91	24.5	1.72, 1.94	25.6	1.74, 1.93
3	91.0	3.21	91.2	3.18	91.5	3.20 dd (4.5;11.0)
4	39.3	–	39.4	–	39.5	–
5	55.6	0.81 d (12.0)	55.8	0.79 d (11.9)	55.4	0.80 d (12.2)
6	18.4	1.43, 1.56	19.2	1.42, 1.55	n.d.	1.43, 1.55
7	35.8	1.76	36.0	1.74	35.4	1.74, 1.77
8	41.1	–	41.1	–	41.2	–
9	47.3	1.56	47.1	1.56	47.4	1.59
10	36.4	–	36.6	–	36.8	–
11	23.5	1.95	23.5	1.93	24.0	1.92
12	125.7	5.43 br s	125.5	5.43 brs	125.0	5.44 brs
13	143.4	–	143.7	–	143.7	–
14	47.2	–	47.6	–	47.7	–
15	67.1	3.81 d (4.4)	67.6	3.84	67.5	3.68 d (4.5)
16	72.3	4.13 d (4.4)	73.3	3.66	73.2	3.74 d (4.5)
17	47.8	–	n.d.	–	n.d.	–
18	41.3	2.38 brd (13.8)	41.1	2.52 brd (14.2)	40.6	2.53 brd (15.0)
19	47.6	1.07, 2.46 t (13.8)	46.8	1.00, 2.36 t (13.0)	46.2	1.02, 2.36 t (13.3)
20	35.3	–	31.4	–	31.4	–
21	77.0	4.06 d (9.7)	43.3	1.70, 2.16 t (13.5)	43.6	1.71, 2.15 t (12.8)
22	86.7	3.90	78.9	4.25 dd (6.5;13.0)	78.9	4.25 dd (6.4;12.8)
23	27.4	1.10 s	27.6	1.10 s	27.4	1.09 s
24	16.5	0.89 s	16.0	0.88 s	15.8	0.89 s
25	15.6	1.00 s	15.9	1.00 s	16.1	1.01 s
26	17.2	1.05 s	16.6	1.04 s	16.6	1.05 s
27	19.5	1.39 s	20.0	1.37 s	19.4	1.37 s
28	63.8	3.33, 3.52	61.5	3.20 d (11.2), 3.30	61.5	3.20 d (12.0), 3.30
29	28.7	0.98 s	32.4	0.90 s	32.6	0.91 s
30	17.9	0.93 s	24.0	0.97 s	24.2	0.98 s
Acyl	Ac [Ara (p) II C-4]		Ac [Ara (p) II C-2]		Ac [Ara (p) II C-2]	
1	171.3	–	170.7	–	170.8	–
2	19.8	2.13 s	19.5	2.15 s	19.8	2.15 s
			Ac [Ara (p) II C-3]		Ac [Ara (p) II C-3]	
1	–	–	170.9	–	170.6	–
2	–	–	20.1	1.99 s	20.3	1.99 s
			Ac [Ara (p) II C-4]		Ac [Ara (p) II C-4]	
1	–	–	170.9	–	170.6	–
2	–	–	19.5	2.10 s	19.8	2.10 s

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; Ara (p) II, second arabinopyranose at C-22; Ac, acetic acid.

acylated with acetic acid. Cross peaks between these protons and the corresponding carbonyl carbon atoms have been detected in the HMBC spectrum. The distinguishing feature to compound **8** (R_1-barrigenol) was found to be at C-21 of compound **11**, showing shifts for a methylene group ($\delta_H = 1.70$, 2.16, $\delta_C = 43.3$ ppm) instead of a hydroxymethin group. Thus, the aglycon part of pittangretoside X (**11**) was determined as A_1-barrigenol, and the structure was elucidated as 3β-[β-D-galactopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxy-22α-(2,3,4-triacetyloxy-α-L-arabinofuranosyloxy)-olean-12-ene-15α,16α,28-triol.

Compared to compound **11**, pittangretoside Y (**12**) possesses the same molecular formula of $C_{63}H_{98}O_{31}$,

Table 8. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the sugar moieties of compounds **10**–**12** in CD_3OD (J in Hz)[a].

Position	**10**		**11**		**12**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
C-3	GlcA		GlcA		GlcA	
1	104.8	4.53 d (6.5)	104.8	4.48 d (7.7)	104.9	4.53 d (7.0)
2	79.2	3.93	79.8	3.95	81.0	3.97
3	79.3	3.90	79.3	3.89	79.2	3.92
4	74.5	3.93	73.0	3.79	74.9	3.90
5	78.6	3.73	78.3	3.79	78.8	3.77
6	n.d.	–	n.d.	–	n.d.	–
	Glc		Gal		Glc	
1	101.6	5.04 d (7.7)	102.6	4.89 d (7.0)	101.6	5.04 d (7.5)
2	75.6	3.23 t (8.5)	72.5	3.54	74.8	3.21
3	77.0	3.39 t (9.0)	75.8	3.48	76.9	3.38 t (9.0)
4	71.6	3.12 t (9.3)	72.4	3.66	71.5	3.13 t (9.5)
5	77.3	3.31	75.6	3.52	77.2	3.30
6	62.2	3.58, 3.85	61.8	3.67, 3.84	61.9	3.58, 3.84
	Ara (p)		Ara (p)		Ara (p)	
1	103.2	4.93 d (7.5)	103.0	4.95 d (7.7)	103.0	4.94 d (7.5)
2	72.0	3.60	72.9	3.64	72.2	3.61
3	72.0	3.533	72.4	3.52	72.9	3.52 dd (3.0;9.5)
4	69.2	3.77	69.5	3.78	69.2	3.78
5	66.5	3.55, 3.89	66.7	3.54, 3.83	66.4	3.56, 3.84
	Ara (f)		Ara (f)		Ara (f)	
1	107.2	5.15 brs	107.2	5.17 brs	107.0	5.16 brs
2	81.2	3.98 brs	90.7	3.98 brs	81.4	3.96 brs
3	78.5	3.79	78.5	3.77	79.2	3.75
4	86.8	4.46 q (4.0)	86.7	4.46 q (4.1)	87.0	4.46 q (4.0)
5	62.3	3.66, 3.58	62.2	3.66, 3.60	62.5	3.68, 3.57
	Ara (p) II (C-22)		Ara (p) II (C-22)		Ara (p) II (C-22)	
1	104.9	4.41 d (7.7)	102.8	4.70 d (7.7)	102.9	4.70 d (7.5)
2	72.4	3.65	70.3	5.14 dd (7.7;10.0)	70.5	5.15 dd (7.5;10.0)
3	72.1	3.72	71.5	5.11 dd (3.0;10.0)	71.4	5.11 dd (3.5;10.0)
4	71.7	5.07 brs	68.6	5.28 brs	68.6	5.29 brs
5	64.8	3.73, 3.99 brd (12.0)	64.0	3.80, 3.99	64.1	3.80, 3.99 brd (14.6)

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; GlcA, glucuronopyranosic acid; Glc, glucopyranose; Gal, galactopyranose; Ara (p), arabinopyranose; Ara (f), arabinofuranose; Ara (p) II, second arabinopyranose.

as deduced from a quasimolecular ion $[M–H]^-$ at $m/z = 1349.5988$. The ATR-IR, NMR (Tables 7 and 8) and HRMS data of **12** were again nearly identical to those of **11**. More detailed studies of the sugar components (NMR, hydrolysis, absolute configuration) verified that galactose in compound **11** was replaced once more by glucose in compound **12**. So far, compounds **1** and **2**, **3** and **8**, **6** and **18**, and **11** and **12** are pairs of isomers, differing only in the presence of glucose or galactose in the tetrasaccharide chain. Consequently, pittangretoside Y (**12**) was finally established as 3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxy-22α-(2,3,4-triacetyloxy-α-L-arabinofuranosyloxy)-olean-12-ene-15α,16α,28-triol.

For pittangretoside Z (**13**), a molecular formula of $C_{57}H_{90}O_{26}$ was deduced from a quasimolecular ion peak $[M–H]^-$ at $m/z = 1189.5634$ from its ESI mass spectrum. The ^{1}H NMR spectrum revealed four anomeric proton resonances at $\delta_H = 4.51$ (d, $J = 6.5$), 4.93 (d, $J = 7.5$), 5.04 (d, $J = 7.7$), and 5.17 (br s) ppm (Table 10), that turned out to belong to a β-glucuronopyranosic acid, a β-glucopyranose, an α-arabinopyranose, and an α-arabinofuranose unit of the common tetrasaccharide chain as in compound **2**.

Table 9. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the aglycone moieties of compounds **13**–**15** in CD_3OD (J in Hz)[a].

Position	**13**		**14**		**15**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
1	38.7	1.00, 1.65	39.3	0.97, 1.62	38.4	0.99, 1.65
2	24.8	1.73, 1.92	24.1	1.71, 1.97	n.d.	1.75, 1.88
3	91.0	3.21	90.3	3.19	91.1	3.20 dd (4.5, 14.0)
4	39.5	–	39.6	–	39.7	–
5	55.6	0.81 d (12.0)	56.3	0.81 d (11.7)	55.8	0.81 d (11.9)
6	n.d.	1.44, 1.56	18.9	1.46, 1.60	19.2	1.46, 1.58
7	35.6	1.77	32.8	n.d.	32.9	1.75
8	41.1	–	40.3	–	40.3	–
9	47.1	1.57	46.9	1.59	47.0	1.58
10	36.6	–	36.3	–	36.5	–
11	23.7	1.92	23.9	1.93	23.7	1.94
12	125.8	5.46 brs	124.0	5.39 brs	123.8	5.40 brs
13	143.0	–	141.9	–	142.0	–
14	47.3	–	41.6	–	41.2	–
15	67.3	3.84	33.7	1.37, 1.74	33.7	1.38, 1.74
16	72.2	3.98	68.3	4.03 brs	68.4	4.04 brs
17	48.2	–	46.4	–	47.4	–
18	41.5	2.47 brd (14.2)	40.5	2.60 brd (13.8)	40.4	2.59 brd (13.5)
19	47.1	1.17 dd (4.0;13.3), 2.59 t 13.3)	46.9	1.20 dd (3.4;13.0), 2.69 t (13.8)	47.0	1.20 dd (3.7;12.7), 2.69 t (13.2)
20	35.5	–	35.3	–	36.0	–
21	81.2	5.61 d (10.2)	78.8	5.90 d (10.0)	78.9	5.90 d (10.3)
22	72.1	4.03 d (10.2)	74.3	5.50 d (10.0)	74.2	5.52 d (10.3)
23	27.5	1.10 s	27.6	1.10 s	27.4	1.11 s
24	16.1	0.89 s	15.8	0.89 s	16.1	0.89 s
25	15.8	1.01 s	15.2	0.99 s	15.4	1.00 s
26	17.1	1.04 s	15.8	0.95 s	15.8	0.96 s
27	19.5	1.41 s	26.7	1.50 s	26.8	1.50 s
28	64.6	3.28, 3.41	63.9	3.01 d (11.0), 3.26 d (11.0)	63.8	3.01 d (11.3), 3.27 d (11.3)
29	28.9	0.87 s	28.3	0.88 s	28.6	0.89 s
30	19.2	1.04 s	19.5	1.09 s	19.2	1.10 s
Acyl		Ang (C-21)		Ang (C-21)		Ang (C-21)
1	169.2	–	168.3	–	168.2	–
2	128.9	–	128.4	–	128.5	–
3	137.2	6.11 q (7.1)	138.1	6.13 q (7.2)	138.0	6.13 q (7.0)
4	15.1	2.00	15.1	1.97	15.2	1.97
5	20.1	1.95	19.7	1.87	19.9	1.87
				Ac (C-22)		Ac (C-22)
1	–	–	172.1	–	172.3	
2	–	–	20.0	1.97 s	20.0	1.97 s

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; Ang, angelic acid; Ac, acetic acid.

Again, GC-MS techniques confirmed the absolute configuration of the corresponding thiazolidine carboxylates of hydrolyzed sugars as D for β-glucuronic acid and β-glucose, and L for α-arabinose. A characteristic proton resonance at 6.11 ppm (q, $J = 7.1$) and two additional methyl signals at $\delta_H = 1.95$ and 2.00 ppm indicated an angeloyl moiety (Table 9). Its carbonyl carbon atom at $\delta_C = 169.2$ ppm showed a long range correlation with H-21 of the R_1-barrigenol aglycone moiety at $\delta_H = 5.61$ (d, $J = 10.2$) ppm, whereby the attachment position of that residue was unambiguously assigned. Comparing compound **13** with the known glycoside **16**, the latter one possesses an additional acylation with acetic acid at C-22. Finally, the new structure of pittangretoside Z (**13**) was established as 21β-angeloyloxy-3β-[β-D-glucopyranosyl-(1→2)]-[α-

Table 10. ^{13}C (125 MHz) and ^{1}H (500 MHz) NMR spectroscopic data (ppm) of the sugar moieties of compounds **13**–**15** in CD_3OD (J in Hz)[a].

Position	**13**		**14**		**15**	
	^{13}C	^{1}H	^{13}C	^{1}H	^{13}C	^{1}H
C-3		GlcA		GlcA		GlcA
1	104.8	4.51 d (6.5)	104.8	4.49 d (6.8)	104.8	4.52 d (6.5)
2	79.0	3.93	79.6	3.90	81.0	3.95
3	79.6	3.90	78.6	3.88	79.1	3.92
4	75.8	3.91	72.7	3.79	73.6	3.95
5	78.5	3.77	79.0	3.77	78.2	3.71
6	n.d.	–	n.d.	–	n.d.	–
		Glc		Gal		Glc
1	101.8	5.04 d (7.7)	102.9	4.90 d (7.0)	101.8	5.04 d (7.8)
2	75.0	3.22 t (8.5)	72.0	3.55	75.5	3.21 t (8.2)
3	77.5	3.38 t (9.0)	75.9	3.48	76.8	3.39 t (9.0)
4	72.2	3.13 t (9.0)	72.4	3.64	71.3	3.13 t (9.5)
5	77.5	3.30	75.7	3.54	77.8	3.29
6	61.9	3.59, 3.85	61.7	3.67, 3.75	62.4	3.67, 3.84 dd (5.3; 12.3)
		Ara (p)		Ara (p)		Ara (p)
1	103.0	4.93 d (7.5)	103.3	4.96 d (7.5)	103.3	4.93 d (7.4)
2	72.1	3.52	72.5	3.63	72.2	3.63
3	73.5	3.50 dd (3.3;9.7)	72.5	3.52 dd (3.0;9.6)	72.5	3.51 dd (3.0;9.5)
4	69.5	3.76	69.2	3.78	69.2	3.77
5	66.3	3.59, 3.76	67.0	3.61, 3.85	66.6	3.54, 3.84
		Ara (f)		Ara (f)		Ara (f)
1	n.d.	5.17 brs	107.1	5.14 brs	107.0	5.14 brs
2	80.7	3.97 brs	80.8	3.98 brs	80.8	3.98 brs
3	78.8	3.78	78.8	3.77	78.8	3.78
4	86.3	4.46 q (4.3)	86.9	4.44 q (4.8)	86.0	4.46 q (4.0)
5	62.4	3.67, 3.58	63.1	3.67, 3.58	63.0	3.66, 3.58

[a] Assignments were made by ^{1}H-^{1}H COSY, HMBC, and HMQC experiments; overlapped ^{1}H resonances are reported without designated multiplicity; n. d., not determined; GlcA, glucuronopyranosic acid; Glc, glucopyranose; Gal, galactopyranose; Ara (p), arabinopyranose; Ara (f), arabinofuranose.

L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-15α,16α,22α,28-tetrol.

In the ESI mass spectrum of pittangretoside A_1 (**14**), a $[M–H]^-$ quasimolecular ion at $m/z = 1215.5881$ substantiated a molecular formula of $C_{59}H_{92}O_{26}$. Again, carefully evaluated one- and two-dimensional NMR spectra revealed a strong structural similarity to the known compound **16**. However, the glycosidic part showed some differences, since distinctive resonances for a galactose moiety were observed [$\delta_H = 4.90$, d (7.0) ppm, H-1; 3.55 ppm, H-2; 3.48 ppm, H-3; 3.63 ppm, H-4, and 3.54 ppm, H-5] (Table 10), instead of a glucose unit as described for compound **16**. This was also supported by TLC and GC-MS procedures, identifying β-D-galactose together with β-D-glucuronic acid, and α-L-arabinose. As another different point, resonances at $\delta_C = 33.7$, and $\delta_H = 1.37$ and 1.74 ppm for C-15 were observed (Table 9), indicating no oxidation at this position and, consequently, a barringtogenol C aglycon moiety as already assigned for pittangretoside T (**7**). Thus, the novel natural product pittangretoside A_1 (**14**) was elucidated as 21β-angeloyloxy-22α-acetyloxy-3β-[β-D-galactopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-16α,28-diol.

For pittangretoside B_1 (**15**), a molecular formula identical to that of compound **14**, $C_{59}H_{92}O_{26}$, was generated by a quasimolecular ion peak $[M–H]^-$ at $m/z = 1215.5864$ in its ESI mass spectrum. Since the chromatographic interactions during the HPLC isolation process were again very similar to those for compound **14**, pittangretoside B_1 (**15**) was proposed to be the glucose isomer of **14**. Indeed, on acid hy-

drolysis and subsequent GC-MS and TLC examinations, **15** gave β-D-glucose, β-D-glucuronic acid, and α-L-arabinose. Except for the NMR data of those exchanged sugars, the remaining data were basically identical (Tables 9 and 10). Resonances for both, an acetyl and an angeloyl residue were observed as well as their linkage at C-22 and C-21, respectively, as assigned by HMBC correlations. C-15 appeared also at $\delta_C = 33.7$, and $\delta_H = 1.38$ and 1.74 ppm as observed for compound **14**, proving that the aglycone structure was barringtogenol C, as well. Pittangretoside B_1 was thus established as 21β-angeloyloxy-22α-acetyloxy-3β-[β-D-glucopyranosyl-(1→2)]-[α-L-arabinopyranosyl-(1→3)]-[α-L-arabinofuranosyl-(1→4)]-β-D-glucuronopyranosyloxyolean-12-ene-16α,28-diol.

In summary, a total of eighteen triterpene saponins, including fifteen new natural products, were isolated from the leaves of *P. angustifolium*. With respect to their structural features, among the elucidated eleven monodesmosidic compounds **1**, **2**, **4**, **6**, **7**, and **13**–**18** as well as the bisdesmosidic ones **3**, **5**, **8**, **9**, **10**, **11**, and **12**, aglycone moieties of A_1-barrigenol (**1**, **2**, **11**, **12**, **17**), R_1-barrigenol (**3**, **5**, **6**, **8**–**10**, **13**, **16**, **18**), camelliagenin A (**4**) and barringtogenol C (**7**, **14**, **15**) were assigned. To the best of our knowledge, the latter two backbones have not been described before as constituents of original triterpene saponins from a *Pittosporum* sp., but have been found in the hydrolyzate of fractions from *P. phillyraeoides* [10] and *P. undulatum* [11]. So far, only two bisdesmosidic triterpene saponins were isolated from the *Pittosporum* genus (*P. senacia* [7]), consisting of oleanolic acid and R_1-barrigenol aglycones, of which the latter one was later isolated from *P. verticillatum* [8] as well, and possesses also a second arabinopyranose linked to C-22, as it is present in all described bisdesmosidic structures of this study. While the second sugar residue in common bisdesmosidic structures is attached mostly at C-28 of the aglycone backbone, the present attachment at C-22 seems to be rarer and was confirmed only for a few saponins isolated from *Glycine max*, *Sophora flavescens* and *Eryngium yuccifolium* [12–14]. Interestingly, a galactopyranose moiety, as it was found in the compounds **1**, **4**, **6**–**8**, **11**, and **14**, has not been described as a part of the common branched tetrasaccharide linked to C-3 in published work on phytochemistry of investigated *Pittosporum* species. Besides five galactose/glucose isomers reported herewith (**1**/**2**, **8**/**3**, **6**/**18**, **11**/**12**, **14**/**15**), the structures of another three pairs of such isomers from *P. angustifolium* have been previously published by us [2]. Furthermore, since esterified R_1- and A_1-barrigenol structures have been characterized as aglycone skeletons of triterpene saponins in a number of *Pittosporum* species, such as *P. tobira* [5], *P. undulatum* [6], *P. senacia* [7], *P. verticillatum* [8], and *P. viridiflorum* [9], their occurrence could be regarded as a chemotaxonomic marker. As a concluding remark, it is mentioned that the present work together with recent results [2, 3] witnesses the isolation and characterization of 28 triterpene saponins from the leaves and another seven from the seeds of *P. angustifolium*, collectively representing 31 different compounds of the class of triterpene glycosides, of which 27 were described for the first time as natural products.

Experimental Section

General

NMR spectra were recorded in CD_3OD on a Bruker DRX 500 (Billerica, MA, USA) instrument. All semipreparative HPLC procedures were carried out on a Shimadzu system (Kyoto, Japan) with a two-channel UV detector by using one of the following columns: RP18 phase with polar endcapping 250 × 10 mm, 4 μm (column I), RP18 phase 250 × 4.6 mm, 5 μm (column II) and 250 × 10 mm, 5 μm (column III) (Phenomenex, Torrance, USA). For GC-MS analysis an Agilent device (gas chromatograph, G1530N; mass selective detector, MSD G2588A; Santa Clara, CA, USA) together with a DB-5MS column (30 m × 0.25 mm × 0.25 μm; J & W Scientific; Folsom, CA, USA) was used under conditions previously described [2]. Detected compounds were assigned by mass spectral data compared with NIST database 2.0 d (National Institute of Standards and Technology, Gaithersburg, MD, USA) and data generated by comparison of retention times of the TIC (total ion chromatograms) of authentic samples of D-glucose (Sigma-Aldrich, St. Louis, MO, USA), D-galactose (Sigma-Aldrich), L-arabinose (Fluka, St. Louis, MO, USA), and D-glucuronic acid (Sigma-Aldrich). All LC-MS measurements were performed on a LC-MS-IT-TOF device (Shimadzu) with a Chromolith SpeedRod RP18 column (50 mm × 4.6 mm, Merck; Darmstadt, Germany) or a Kinetex C18 column (2.6 μm, 100 mm × 3 mm, Phenomenex) and electrospray ionization (ESI). ATR-IR spectra were recorded on a Thermo Scientific Nicolet IR 200 FT-IR spectrometer (Waltham, MA, USA), and optical rotation values were determined on a Perkin Elmer 241 polarimeter (Waltham, MA, USA). Pre-coated silica gel 60 plates (Merck) were applied for thin layer chromatography

analysis, using a mixture of EtOAc-*iso*-PrOH-HOAc-H_2O (4 : 2 : 2 : 1) and a detection reagent [0.25 g Thymol (Sigma-Aldrich), 2.5 mL H_2SO_4, 47.5 mL EtOH] to visualize sugars. Sprayed plates were heated at 135 °C for 5 min. Solid-phase extractions (SPE) were carried out with a vacuum manifold and RP18-cartridges (Strata C18E, 200 g/120 mL, Phenomenex).

Plant material

Leaves of *P. angustifolium* were collected in June 2008 on the grounds of Central Queensland GG foundation (K. A. Amato and the Trustee for Milner Krasser Family Trust) near Mount Morgan, Rockhampton, Queensland, Australia, and were a gift of Dr. Kornelia Krasser and Mr. Klaus von Gliszczynski, Australia. The plant material was authenticated by Dr. Peter König, Curator of the Botanical Garden of Greifswald and a voucher specimen (no. 20110013PA) was deposited at the Institute of Pharmacy, Department of Pharmaceutical Biology, Ernst Moritz Arndt University, Greifswald, Germany.

Extraction and isolation

Pulverized, dried leaves (8.4 g) were defatted with CH_2Cl_2 *via* Soxhlet for 24 h and then extracted three times with 80 % (v/v) EtOH under reflux. 2.8 g of the crude extract were subjected to an open column chromatography using Sephadex LH-20 (Sigma-Aldrich), eluting with MeOH. A saponin-enriched fraction of 1.9 g was then applied for a subsequent fractionation on silica gel (60–40 μm, Merck) using a stepwise gradient of CH_2Cl_2-MeOH-H_2O mixtures as recently reported [2] to yield saponin fractions A_1 (93 mg), A_2 (168 mg), A_3 (107 mg), A_4 (405 mg), A_5 (183 mg). Afterwards, solid-phase extractions were performed, using H_2O as washing solvent for all fractions and the following MeOH concentrations for precleaning and/or fractionation: A_1 (30 %, 100 %), A_2 and A_3 (40 %, 100 %), A_4 (40 %, 60 %), A_5 (30 %, 100 %). Corresponding subfractions were obtained (the MeOH concentration used for elution is represented by additional subscript indices) as $A_{1\_100\,\%}$ (27 mg), $A_{2\_40\,\%}$(26 mg), $A_{2\_100\,\%}$ (63 mg), $A_{3\_40\,\%}$ (24 mg), $A_{3\_100\,\%}$ (69 mg), $A_{4\_40\,\%}$ (139 mg), $A_{4\_60\,\%}$ (231 mg), $A_{5\_30\,\%}$ (64 mg), and $A_{5\_100\,\%}$ (78 mg). HPLC conditions used for isolation: solvent A (H_2O), solvent B (acetonitrile), each with 0.05 % HCOOH, gradient elution or isocratic; flow rate: analytical column (column II) 1 mL min^{-1}, semipreparative columns (columns I and III) 4 mL min^{-1}; detection 206 nm; methods (expressed as time [min]:concentration solvent B [%]: method 1 (0:41, 11:45, 21:46, 28:50, 29:41, 32:41, column I) used for fraction $A_{1\_100\,\%}$ (compound **18**, t_R = 21.73 min), fraction $A_{2\_100\,\%}$ (compound **4**, t_R = 16.56 min; compound **7**, t_R = 29.38 min; compound **16**, t_R = 9.56 min; compound **18**; compounds **14** and **15** were collected unseparated), fraction $A_{3\_100\,\%}$ (compounds **4**, **7**, **16**, compounds **6** and **18** as well as **14** and **15** were collected unseparated and compound **17** was obtained only unpure); method 2 (isocratic 39.5 % B, column III) used for further separation of compounds **14** (t_R = 29.80 min) and **15** (t_R = 31.42 min); method 3 (isocratic 50 % B, column III) used for further separation of compounds **6** (t_R = 13.47 min) and **18** (t_R = 14.54 min); compound **17** (t_R = 17.76 min) was purified by method 4 (isocratic 44 % B, column II); method 5 (0:30, 10:34, 11:36, 22:36, column I) used for fractions $A_{2\_40\,\%}$ and $A_{3\_40\,\%}$ (compound **3**, t_R = 13.79 min; compound **11**, t_R = 25.51 min; compound **12**, t_R = 26.27 min), method 6 (isocratic 28.2 % B, column I) used for fraction $A_{4\_40\,\%}$ (compounds **8**, t_R = 27.09 min and **3**, t_R = 28.84 min); method 7 (0:27, 25:27, 26:80, 27:27, 32:27, column III) used for 90 mg of fraction $A_{4\_60\,\%}$ to isolate compound **13** (t_R = 23.29 min); method 8 (0:22.5, 33:22.5, 37:24, 39:80, 40:22.5, 43:22.5, column I) used for fraction $A_{5\_30\,\%}$ (compound **5**, t_R = 14.07 min; compound **9**, t_R = 29.13 min; compound **10**, t_R = 34.05 min); method 9 (isocratic 27.5 % B, column I) used for fraction $A_{5\_100\,\%}$ (compound **1**, t_R = 21.13 min; compound **2**, t_R = 22.27 min). Total amounts: **1** (2.4 mg), **2** (1.5 mg), **3** (44.1 mg), **4** (3.0 mg), **5** (4.9 mg), **6** (4.1 mg), **7** (4.9 mg), **8** (7.3 mg), **9** (2.3 mg), **10** (2.8 mg), **11** (1.2 mg), **12** (1.7 mg), **13** (2.1 mg), **14** (1.6 mg), **15** (1.9 mg), **16** (10.2 mg), **17** (3.2 mg), **18** (45.2 mg.)

*Compound **1** (pittangretoside J)*

Colorless amorphous powder. $C_{52}H_{84}O_{24}$; $[\alpha]_D^{20} = -54.5$ (c = 0.09, MeOH). – ATR-IR: ν_{max} = 3351, 2945, 1598, 1465, 1390, 1256, 1150, 1072, 1050, 1005 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 1 and 2. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 455.3584 (91.4) [(M+H)-GlcA-Gal-2Ara-2H_2O]$^+$, 437.3428 (38.1) [(M+H)-GlcA-Gal-2Ara-3H_2O]$^+$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1091.5256 (100) (calcd. for $C_{52}H_{83}O_{24}$, 1091.5280, monoisotopic mass, [M–H]$^-$).

*Compound **2** (pittangretoside K)*

Colorless amorphous powder. $C_{52}H_{84}O_{24}$; $[\alpha]_D^{20} = -38.2$ (c = 0.09, MeOH). – ATR-IR: ν_{max} = 3367, 2953, 1605, 1458, 1412, 1359, 1136, 1070, 1039, 1007 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 1 and 2. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 455.3584 (78.3) [(M+H)-GlcA-Glc-2Ara-2H_2O]$^+$, 437.3426 (100) [(M+H)-GlcA-Glc-2Ara-3H_2O]$^+$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1091.5263 (100) (calcd. for $C_{52}H_{83}O_{24}$, 1091.5280, monoisotopic mass, [M–H]$^-$).

*Compound **3** (pittangretoside M)*

Colorless amorphous powder. $C_{63}H_{98}O_{32}$; $[\alpha]_D^{20} = -24.0$ (c = 0.41, MeOH). – ATR-IR: ν_{max} = 3431, 2934, 1737,

1370, 1228, 1073, 1051, 1006 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 1 and 2. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 706.2785 (24.9) $[M+2Na]^{2+}$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1365.5889 (100) (calcd. for $C_{63}H_{97}O_{32}$, 1365.5968, monoisotopic mass, $[M–H]^-$).

*Compound **4** (pittangretoside Q)*

Colorless amorphous powder. $C_{57}H_{90}O_{24}$; $[\alpha]_D^{20} = -8.2$ (c = 0.16, MeOH). – ATR-IR: ν_{max} = 3368, 2920, 1688, 1601, 1426, 1387, 1239, 1147, 1072, 1042 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 3 and 4. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 439.3547 (94.1) [(M+H)-GlcA-Gal-2Ara-Ang-$2H_2O$]$^+$, 421.3446 (100) [(M+H)-GlcA-Gal-2Ara-Ang-$3H_2O$]$^+$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1157.5742 (100) (calcd. for $C_{57}H_{89}O_{24}$, 1157.5749, monoisotopic mass, $[M–H]^-$).

*Compound **5** (pittangretoside R)*

Colorless amorphous powder. $C_{57}H_{92}O_{29}$; $[\alpha]_D^{20} = -12.8$ (c = 0.24, MeOH). – ATR-IR: ν_{max} = 3372, 2931, 1608, 1414, 1373, 1256, 1072, 1043, 1006 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 3 and 4. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 661.3956 (16.5) [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)]$^+$, 643.2797 (68.1) [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)-H_2O]$^+$, 471.3484 (85.0) [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$2H_2O$]$^+$, 453.3400 (100) [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$3H_2O$]$^+$, 435.3293 (87.9) [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$4H_2O$]$^+$, 417.3197 (48.3) [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$5H_2O$]$^+$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1239.5833 (13.9) (calcd. for $C_{57}H_{91}O_{29}$, 1239.5804, monoisotopic mass, $[M–H]^-$).

*Compound **6** (pittangretoside S)*

Colorless amorphous powder. $C_{62}H_{96}O_{27}$; $[\alpha]_D^{20} = -17.5$ (c = 0.33, MeOH). – ATR-IR: ν_{max} = 3364, 2923, 1693, 1591, 1354, 1240, 1155, 1071, 1041, 997 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 3 and 4. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 571.3992 (33.6) [(M+H)-GlcA-Gal-2Ara-Ang-H_2O]$^+$, 553.3849 (100) [(M+H)-GlcA-Gal-2Ara-Ang-$2H_2O$]$^+$, 535.3722 (39.4) [(M+H)-GlcA-Gal-2Ara-Ang-$3H_2O$]$^+$, 453.3331 (17.7) [(M+H)-GlcA-Gal-2Ara-2Ang-$3H_2O$]$^+$, 435.3284 (26.5) [(M+H)-GlcA-Gal-2Ara-2Ang-$4H_2O$]$^+$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1271.6080 (100) (calcd. for $C_{62}H_{95}O_{27}$, 1271.6066, monoisotopic mass, $[M–H]^-$).

*Compound **7** (pittangretoside T)*

Colorless amorphous powder. $C_{62}H_{96}O_{26}$; $[\alpha]_D^{20} = -19.6$ (c = 0.25, MeOH). – ATR-IR: ν_{max} = 3409, 2922, 1701, 1602, 1387, 1239, 1158, 1072, 1042 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 5 and 6. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 537.3941 (4.3) [(M+H)-GlcA-Gal-2Ara-Ang-$2H_2O$]$^+$, 419.3352 (2.0) [(M+H)-GlcA-Gal-2Ara-2Ang-$4H_2O$]$^+$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1255.6174 (100) (calcd. for $C_{62}H_{95}O_{26}$, 1255.6176, monoisotopic mass, $[M–H]^-$).

*Compound **8** (pittangretoside U)*

Colorless amorphous powder. $C_{63}H_{98}O_{32}$; $[\alpha]_D^{20} = -31.1$ (c = 0.22, MeOH). – ATR-IR: ν_{max} = 3402, 2953, 1736, 1619, 1371, 1223, 1049, 1006 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 5 and 6. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 706.2858 (18.6) $[M+2Na]^{2+}$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1365.5895 (2.4) (calcd. for $C_{63}H_{97}O_{32}$, 1365.5968, monoisotopic mass, $[M–H]^-$).

*Compound **9** (pittangretoside V)*

Colorless amorphous powder. $C_{59}H_{94}O_{30}$; $[\alpha]_D^{20} = -11.4$ (c = 0.17, MeOH). – ATR-IR: ν_{max} = 3372, 2923, 1719, 1591, 1377, 1235, 1074, 1041, 1007 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 5 and 6. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 703.4145 (23.9) [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)]$^+$, 664.2858 (100) $[M+2Na]^{2+}$, 471.3440 (20.0) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$2H_2O$]$^+$, 453.3312 (23.8) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$3H_2O$]$^+$, 435.3312 (23.9) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$4H_2O$]$^+$, 417.3183 (2.5) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$5H_2O$]$^+$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1281.5726 (35.6) (calcd. for $C_{59}H_{93}O_{30}$, 1281.5757, monoisotopic mass, $[M–H]^-$).

*Compound **10** (pittangretoside W)*

Colorless amorphous powder. $C_{59}H_{94}O_{30}$; $[\alpha]_D^{20} = -8.7$ (c = 0.21, MeOH). – ATR-IR: ν_{max} = 3355, 2925, 1730, 1600, 1374, 1250, 1073, 1024, 1001 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 7 and 8. – HRMS ((+)-ESI-IT-TOF): m/z(%) = 703.4235 (11,7) [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)]$^+$, 664.2943 (100) $[M+2Na]^{2+}$, 471.3315 (4.9) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$2H_2O$]$^+$, 453.3407 (40.8) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$3H_2O$]$^+$, 435.3409 (27.5) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-$4H_2O$]$^+$. – HRMS ((–)-ESI-IT-TOF): m/z(%) = 1281.5808 (20.0) (calcd. for $C_{59}H_{93}O_{30}$, 1281.5757, monoisotopic mass, $[M–H]^-$).

*Compound **11** (pittangretoside X)*

Colorless amorphous powder. $C_{63}H_{98}O_{31}$; $[\alpha]_D^{20} = -6.5$ (c = 0.09, MeOH). – ATR-IR: ν_{max} = 3390, 2940, 1737,

1602, 1371, 1229, 1066, 1047, 1008 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 7 and 8. – HRMS ((+)-ESI-IT-TOF): $m/z(\%)$ = 771.4248 (16.6) [(M+Na)-GlcA-Gal-Ara(p)-Ara(f)]$^+$, 731.4386 (6.8) [(M+H)-GlcA-Gal-Ara(p)-Ara(f)-H_2O]$^+$, 713.4176 (27.1) [(M+H)-GlcA-Gal-Ara(p)-Ara(f)-2H_2O]$^+$, 698.2894 (100) [M+2Na]$^{2+}$, 455.3557 (14.5) [(M+H)-GlcA-Gal-Ara(p)-Ara(f)-Ara(p)II-2H_2O]$^+$, 437.3466 (30.2) [(M+H)-GlcA-Gal-Ara(p)-Ara(f)-Ara(p)II-3H_2O]$^+$, 419.3355 (16.6) [(M+H)-GlcA-Gal-Ara(p)-Ara(f)-Ara(p)II-4H_2O]$^+$. – HRMS ((–)-ESI-IT-TOF): $m/z(\%)$ = 1349.5986 (100) (calcd. for $C_{63}H_{97}O_{31}$, 1349.6019, monoisotopic mass, [M–H]$^-$).

Compound ***12*** *(pittangretoside Y)*

Colorless amorphous powder. $C_{63}H_{98}O_{31}$; $[\alpha]_D^{20} = -9.0$ (c = 0.13, MeOH). – ATR-IR: ν_{max} = 3423, 2925, 1740, 1609, 1369, 1228, 1050, 1001 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 7 and 8. – HRMS ((+)-ESI-IT-TOF): $m/z(\%)$ = 771.4241 (18.9), [(M+Na)-GlcA-Glc-Ara(p)-Ara(f)]$^+$, 731.4386 (8.2) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-H_2O]$^+$, 713.4177 (31.4), [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-2H_2O]$^+$,698.2890 (100) [M+2Na]$^{2+}$, 455.3557 (17.1) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-2H_2O]$^+$, 437.3466 (35.1) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-3H_2O]$^+$, 419.3354 (19.3) [(M+H)-GlcA-Glc-Ara(p)-Ara(f)-Ara(p)II-4H_2O]$^+$. – HRMS ((–)-ESI-IT-TOF): $m/z(\%)$ = 1349.5988 (100) (calcd. for $C_{63}H_{97}O_{31}$, 1349.6019, monoisotopic mass, [M–H]$^-$).

Compound ***13*** *(pittangretoside Z)*

Colorless amorphous powder. $C_{57}H_{90}O_{26}$; $[\alpha]_D^{20} = -6.0$ (c = 0.17, MeOH). – ATR-IR: ν_{max} = 3346, 2908, 1603, 1387, 1243, 1158, 1075, 1041, 1005 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 9 and 10. – HRMS ((+)-ESI-IT-TOF): $m/z(\%)$ = 647.4031 (13.8) [(M+H)-Glc-2Ara-Ang-2H_2O]$^+$, 629.3905 (2.1) [(M+H)-Glc-2Ara-Ang-3H_2O]$^+$, 471.3552 (74.3) [(M+H)-GlcA-Glc-2Ara-Ang-2H_2O]$^+$, 453.3457 (100) [(M+H)-GlcA-Glc-2Ara-Ang-3H_2O]$^+$, 435.3351 (86.0) [(M+H)-GlcA-Glc-2Ara-Ang-4H_2O]$^+$, 417.3246 (59.3) [(M+H)-GlcA-Glc-2Ara-Ang-5H_2O]$^+$, 399.3108 (4.9) [(M+H)-GlcA-Glc-2Ara-Ang-6H_2O]$^+$. – HRMS ((–)-ESI-IT-TOF): $m/z(\%)$ = 1189.5634 (100) (calcd. for $C_{57}H_{89}O_{26}$, 1189.5648, monoisotopic mass, [M–H]$^-$).

Compound ***14*** *(pittangretoside A$_1$)*

Colorless amorphous powder. $C_{59}H_{92}O_{26}$; $[\alpha]_D^{20} = -12.0$ (c = 0.12, MeOH). – ATR-IR: ν_{max} = 3371, 2929, 1712, 1602, 1372, 1256, 1155, 1072, 1041 cm^{-1}. – ^1H and ^{13}C NMR: see Tables 9 and 10. – HRMS ((–)-ESI-IT-TOF): $m/z(\%)$ = 1215.5881 (24.4) (calcd. for $C_{59}H_{91}O_{26}$, 1215.5804, monoisotopic mass, [M–H]$^-$).

Compound ***15*** *(pittangretoside B$_1$)*

Colorless amorphous powder. $C_{59}H_{92}O_{26}$; $[\alpha]_D^{20} = -14.8$ (c = 0.14, MeOH). – ATR-IR: ν_{max} = 3414, 2945, 1715, 1604, 1369, 1253, 1159, 1074, 1022, cm^{-1}. – ^1H and ^{13}C NMR: see Tables 9 and 10. – HRMS ((–)-ESI-IT-TOF): $m/z(\%)$ = 1215.5864 (15.4) (calcd. for $C_{59}H_{91}O_{26}$, 1215.5804, monoisotopic mass, [M–H]$^-$).

Acidic hydrolysis

0.4 – 1.0 mg of each compound were treated as recently described [3] and applied for TLC and GC-MS procedures. Bisdesmosidic structures, acylated at the second arabinopyranose linked to C-22 of the aglycon moieties (compounds **3**, **8**, **9**–**12**), were pretreated with 1 mL of 1 M NaOH (Merck) at 80 °C for 4 h, then neutralized with 1 M HCl (VWR International, Darmstadt, Germany). The mixture was purified by a solid-phase extraction, using conditioned and equilibrated SPE cartridges (C18E, 500 mg, Phenomenex) by eluting with H_2O. A washing step, using MeOH, was performed for the recovery of deacetylated compounds. The further procedure corresponds to that of the non-acylated compounds. The absolute configuration of sugars was confirmed by their thiazolidine carboxylates [15], comparing obtained GC-MS data with those of authentic samples of L-Ara (t_R = 36.538 min), D-Glc (t_R = 37.647 min), D-Gal (t_R = 40.383 min) and D-GlcA (t_R = 41.109 min).

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