Hydrolytic Reaction of Plant Extracts to Generate Molecular Diversity: New Dammarane Glycosides from the Mild Acid Hydrolysate of Root Saponins of *Panax notoginseng*

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Molecular diversity was generated by hydrolyzing the crude root saponins of *Panax notoginseng* (BURK.) F. H. CHEN under mild acidic condition (AcOH/EtOH 1:1). From the acid hydrolysate, five new dammarane glycosides, named notoginsenoside T_1 (=(3*β*,6*α*,12*β*,20*E*,23*RS*)-24,25-epoxy-6-[(*β*-D-glucopyranosyl)oxy]-dammar-20(22)-ene-3,12,23-triol; **1**), notoginsenoside T_2 (=(3*β*,6*α*,12*β*,20*E*,23*RS*)-24,25-epoxy-6-[(*β*-D-glucopyranosyl)oxy]-23-methoxydammar-20(22)-ene-3,12-diol; **2**), notoginsenoside T_3 (=(3*β*,6*α*,12*β*,20*S*)-6-[(*β*-D-glucopyranosyl)oxy]-20-ethoxydammar-24-ene-3,12-diol; **3**), notoginsenoside T_4 (=(3*β*,6*α*,12*β*,20*S*,22*E*,24*RS*)-6-[(*β*-D-glucopyranosyl)oxy]dammar-22-ene-3,12,20,24,25-pentol; **4**), and notoginsenoside T_5 (=(3*β*,6*α*,12*β*,24*E*)-6-[(*β*-D-xylopyranosyl-(1→2)-*β*-D-glucopyranosyl)oxy]dammara-20(21),24-diene-3,12-diol; **5**), were isolated, together with 15 known dammarane glycosides, and their structures were elucidated on the basis of spectroscopic evidence. Among the known compounds, ginsenosides Rg_3 and Rh_1 were isolated as major constituents, in addition to ginsenosides Rg_5, Rh_4, and a mixture of (20*R*)- and (20*S*)-25-hydroxyginsenoside Rh_1, all of which were obtained from *P. notoginseng* for the first time.

Introduction. – One of the most important aspects in drug discovery is the presentation of a *diverse* selection of compounds to a biological matrix and to look for a response (bioassay). Thus, the structures of natural products should be as diverse as possible. One approach for creating such new natural-product derivatives is combinatorial chemistry. Another approach is to chemically modify the plant extracts, such as by reduction, oxidation, and hydrolytic reactions to afford a whole new range of natural or synthetic 'metabolites' [1].

Panax notoginseng (BURK.) F. H. CHEN (Araliaceae), a famous traditional Chinese medicinal herb indigenous to the southern Yunnan province, has been used for the treatment of cardiovascular diseases, inflammation, and internal and external bleeding due to injury [2]. Extensive studies on this plant led to the identification of dammarane-type saponins named *ginsenosides* or *notoginsenosides*, derivatives of protopanaxadiol and protopanaxatriol [3], as the main bioactive principles [4–9].

The strongly acid-labile nature of the side chains of ginsenosides and notoginsenosides has been reported previously [10–13]. In our study, we hydrolyzed the crude root saponins from *P. notoginseng* under mild acidic condition (AcOH/EtOH 1:1). Preliminary biological testing of the hydrolysate showed that it exhibited medium anticancer and calcium-channel-inhibiting activities.

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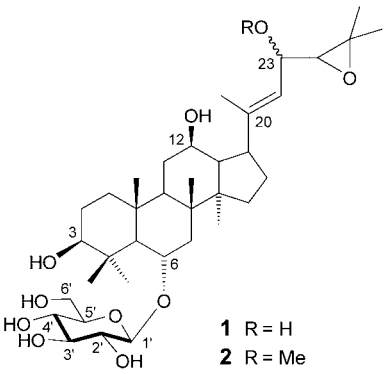
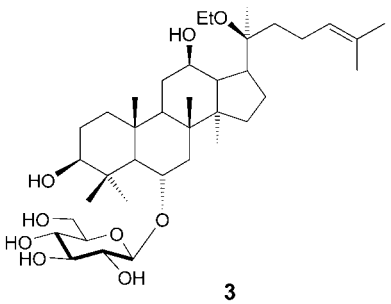
In previous papers, we have reported 15 known dammarane glycosides isolated from this hydrolysate [14–16]: ginsenosides Rg_5, Rg_2, Rg_1, Re, Rh_4, Rd, a mixture of (20*R*)- and (20*S*)-25-hydroxyginsenoside Rh_1, (20*R*)-ginsenoside Rg_3, ginsenoside Rg_3, (20*R*)-ginsenoside Rh_1, ginsenoside Rh_1, notoginsenosides E, R_2, R_1, and gypenoside XVII. In addition, two new dammarane glycosides, notoginsenosides T_1 (**1**) and T_2 (**2**) were partially characterized in a preliminary communication [17]. In the present paper, we report on further studies leading to the isolation of another three new dammarane glycosides, notoginsenosides T_3–T_5 (**3**–**5**), and we report on the isolation and complete structural elucidation of all five new dammarane glycosides.



1 R = H
2 R = Me



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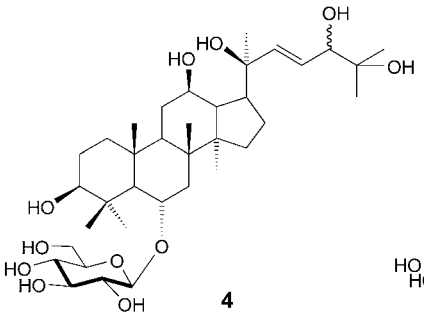

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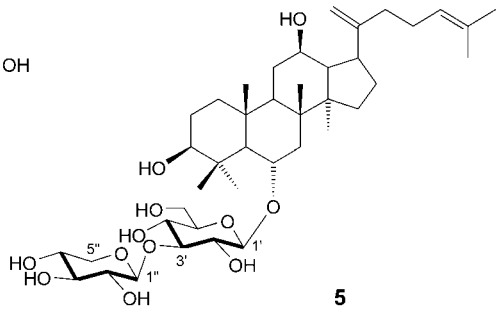

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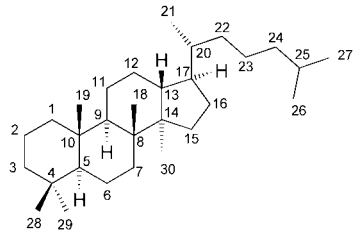
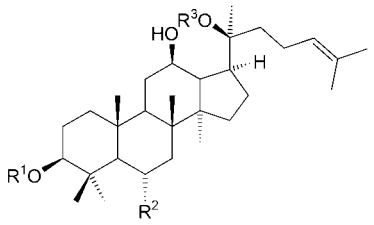


Dammarane



R^2 = H Protopanaxadiol ginsenosides
R^2 = OH, OR Protopanaxatriol ginsenosides

Results and Discussion. – The crude root saponins were hydrolyzed in AcOH/EtOH 1:1 for 6 h at 60°. The hydrolysate was passed through a *Diaion HP-20* column, and then subjected to silica-gel and reversed-phase (*RP-8* and/or *RP-18*) column chromatography to afford the new glycosides **1**–**5**.

For notoginsenoside T_1 (**1**), a molecular formula of $C_{36}H_{60}O_{10}$ was derived by FAB- and HR-FAB-MS (*m/z* 652 (M^-); *m/z* 651.4139 ($[M-H]^-$, calc. 651.4108). Based on 2D-NMR spectra and comparison with NMR data of ginsenoside Rh_4 [18], the structure of notoginsenoside T_1 (**1**) was established as (3*β*,6*α*,12*β*,20*E*,23*RS*)-24,25-epoxy-6-[(*β*-D-glucopyranosyl)oxy]-dammar-20(22)-ene-3,12,23-triol.

The ^{13}C- and ^1H-NMR spectra of **1** (see *Tables 1* and *2*, resp.) showed an anomeric signal each at δ_C 106.1 (C(1′)) and δ_H 5.02 (*d*, *J* = 7.3 Hz, H–C(1′)), respectively, indicating the presence of *one* sugar moiety, in addition to two olefinic signals at δ_C 143.1 and 124.8. The ^1H- and ^{13}C-NMR chemical shifts of **1** were similar to those of ginsenoside Rh_4 [18], except for the side-chain resonances of the aglycone moiety. Furthermore, **1** had seven degrees of unsaturation, as Rh_4, but one C=C bond less than Rh_4, indicating a ring in the side chain of **1**. The structure of the side chain was unambiguously established by 2D-NMR from the direct, vicinal, and long-range H,H and H,C connectivities. From the ^1H,^1H-COSY spectrum of **1**, the signal at δ_H 4.68 (*dd*, *J* = 8.0, 9.6 Hz, H–C(23)) was found to be coupled with both an olefinic H-atom at δ_H 5.89 (*d*, *J* = 9.6 Hz, H–C(22)) and an epoxy resonance at δ_H 3.22 (*d*, *J* = 8.0 Hz, H–C(24)). The HMBC spectrum showed the following long-range correlations: δ_H 1.85 (*s*, H–C(21)) with δ_C 51.0 (C(17)), 124.8 (C(22)), 143.1 (C(20)); δ_H 5.89 (*d*, *J* = 9.6 Hz, H–C(22)) with δ_C 13.8 (C(21)), 51.0 (C(17)), 68.6 (C(24)); δ_H 4.68 (*dd*, *J* = 8.0, 9.6 Hz, H–C(23)) with δ_C 68.6 (C(24)), 124.8 (C(22)), 143.1 (C(20)); δ_H 3.22 (*d*, *J* = 8.0 Hz, H–C(24)) with δ_C 58.6 (C(25)), 68.9 (C(23)); both resonances at δ_H 1.28 (*s*, H–C(26)) and 1.48 (*s*, H–C(27)) with both δ_C 68.6 (C(24)) and 58.6 (C(25)). The configuration of the C=C bond was found to be (*E*), as deduced from the ROESY spectrum, which showed the following NOE correlations: δ_H 1.85 (*s*, H–C(21)) with δ_H 4.68 (*dd*, *J* = 8.0, 9.6 Hz, H–C(23)); δ_H 5.89 (*d*, *J* = 9.6 Hz, H–C(22)) with δ_H 2.81 (*ddd*, *J* = 6.1, 6.7, 10.6 Hz, H–C(17)). This was confirmed by the diagnostic chemical shift of the Me(21) group (δ_C 13.8) compared with those reported in [12] and [18–20].

From the FAB- and HR-FAB mass spectra of **2** (*m/z* 665 ($[M-1]^-$); *m/z* 665.4227 ($[M-1]^-$, calc. 665.4265)), the molecular formula $C_{36}H_{62}O_{10}$ was derived. Comparison of the ^1H- and ^{13}C-NMR spectra of **2** with those of **1** indicated the presence of a MeO group at C(23). Thus, the structure of notoginsenoside T_2 (**2**) was determined as (3*β*,6*α*,12*β*,20*E*,23*RS*)-24,25-epoxy-6-[(*β*-D-glucopyranosyl)oxy]-23-methoxydammar-20(22)-ene-3,12-diol.

The ^1H- and ^{13}C-NMR data of **2** closely resembled those of **1**, except that **2** gave rise to additional resonances at δ_C 55.6 (*s*, MeO) and δ_H 3.42 (*s*, MeO). At the same time, C(23) (δ_C 78.0) was shifted downfield by 9.2 ppm, and C(22) and C(24) (δ_C 120.6 and 66.7, resp.) were shifted upfield by 4.2 and 1.9 ppm, respectively, which suggested that the MeO group was located at C(23). This was further confirmed by a long-range HMBC correlation between the MeO resonances at δ_H 3.42 and C(23) at δ_C 78.0.

The HR-FAB-MS of notoginsenoside T_3 (**3**) showed a quasi-molecular-ion peak at *m/z* 665.4681 ($[M-1]^-$), establishing its molecular formula as $C_{38}H_{66}O_9$ (calc. 665.4629), as confirmed by ^{13}C-NMR (DEPT) and FAB-MS (*m/z* 665 $[M-1]^-$). Comparison of the ^{13}C-NMR chemical shifts of **3** with those of ginsenoside Rh_1 [9] indicated that **3** had an additional EtO group in the side chain. From the spectral data, the structure of notoginsenoside T_3 (**3**) was determined to be (3*β*,6*α*,12*β*,20*S*)-6-[(*β*-D-glucopyranosyl)oxy]-20-ethoxydammar-24-ene-3,12-diol.

Table 1. *^{13}C-NMR Chemical Shifts of Compounds* **1**–**5**. At 125 MHz in C_5D_5N; δ in ppm. Overlapping signals are marked with asterisks (*).

Position	**1**	**2**	**3**	**4**	**5**[a])
1	39.6	39.5	39.5	39.5	39.6
2	28.0	28.0	26.3	28.0	27.8
3	78.6	78.6	78.6	78.7	78.8
4	40.4	40.4	40.4	40.4	40.3
5	61.5	61.5	61.5	61.5	61.4
6	80.1	80.1	80.1	80.1	80.3
7	45.5	45.4	45.3	45.3	45.2
8	41.4	41.4	41.2	41.2	41.3
9	50.8	50.8*	50.0	50.3	52.2
10	39.8	39.8	39.7	39.7	39.8
11	32.8	32.7	31.4	32.3	32.8
12	72.4	72.2	70.6	71.1	72.5
13	50.7	50.7	49.4	50.0	50.6
14	50.9	50.8*	51.7	51.9	51.3
15	31.8	32.6	31.3	31.5	32.8
16	28.7	29.3	28.0	26.6	27.8
17	51.0	51.0	47.4	53.7	52.2
18	17.8	17.8	17.7*	17.6	17.8*
19	17.4	17.4	17.4	17.8	17.8*
20	143.1	146.5	80.0	74.1	155.5
21	13.8	14.1	19.1	29.3	108.2
22	124.8	120.6	36.4	136.4	33.8
23	68.9	78.0	21.8	130.6	30.8
24	68.6	66.7	125.1	80.2	125.4
25	58.6	57.3	131.3	72.9	131.3
26	25.3	25.0	25.8	26.8	25.8
27	20.2	20.0	17.7*	25.7	17.4
28	31.8	31.8	31.8	31.8	31.8
29	16.4	16.4	15.8	16.4	16.7
30	16.8	16.8	16.4	17.2	16.8
MeO	–	55.6	–	–	–
EtO	–	–	15.6	–	–
			56.5		
1′	106.1	106.1	106.1	106.1	103.6
2′	75.5	75.5	75.5	75.5	79.9
3′	79.7	79.7	79.7	79.7	78.1
4′	72.0	71.9	72.0	71.9	71.8
5′	78.2	78.2	78.2	78.2	79.5
6′	63.2	63.2	63.2	63.2	63.0

[a]) Xylosyl resonances of **5** at δ_C 104.9 (C(1″), 75.9 (C(2″), 78.9 (C(3″), 71.3 (C(4″), and 67.3 (C(5″).

The ^{1}H- and ^{13}C-NMR spectra of **3** showed an anomeric resonance at δ_H 5.03 (d, $J = 7.1$ Hz, H–C(1′)) and δ_C 106.1 (C(1′)), respectively, indicating the presence of *one* sugar unit, in addition to two olefinic resonances at δ_C 131.3 and 125.1. The ^{13}C- and ^{1}H-NMR chemical shifts of **3** were very similar to those of ginsenoside Rh_1 [9], except for the side chain. Compound **3** showed an additional EtO group, with signals at δ_C 56.5 ($MeCH_2O$) and 15.6 ($MeCH_2O$), as well as δ_H 3.41 (q, $J = 6.8$ Hz, $MeCH_2O$) and 1.22 (t, $J = 6.8$ Hz, $MeCH_2O$). Moreover, C(20) was shifted downfield to δ_C 80.0 and C(17) upfield to 47.4 ppm. These findings suggested that the EtO group was at C(20), as confirmed by a long-range HMBC correlation between the OCH_2 group at δ_H 3.41 and C(20) at δ_C 80.0.

Table 2. *^{1}H-NMR Chemical Data of Compounds* **1**–**5**. At 500 MHz in C_5D_5N; δ in ppm, J in Hz. Overlapping signals are marked with asterisks (*).

Position	**1**	**2**	**3**	**4**	**5**
1	1.01, 1.68 (2*m*)	1.01, 1.68 (2*m*)	1.05, 1.73 (2*m*)	1.02, 1.68 (2*m*)	0.99, 1.66 (2*m*)
2	1.80, 1.91 (2*m*)	1.81, 1.96 (2*m*)	1.82, 1.92 (2*m*)	1.75, 1.85 (2*m*)	1.80, 1.90 (2*m*)
3	3.51 (*dd*, $J=4.5, 11.5$)	3.52	3.52 (*dd*, $J=4.2, 10.5$)	3.51	3.48
5	1.42	1.42	1.42	1.40	1.39
6	4.42 (*ddd*, $J=3.5, 10.6, 13.5$)	4.44 (*ddd*, $J=3.52, 11.0, 14.2$)	4.43 (*ddd*, $J=3.1, 11.0, 13.0$)	4.40	4.34
7	1.94; 2.52 (*dd*, $J=2.9, 12.8$)	1.94; 2.54	1.94 ($t, J=12.4$); 2.51 (br. *d*, $J=11.6$)	1.90, 2.49	1.94, 2.40
9	1.54	1.57	1.58	1.53	1.52
11	1.42, 2.12	1.45, 2.12	1.45, 2.14	1.52, 2.02	1.41, 2.10
12	3.87 (*dd*, $J=5.8, 10.6$)	3.86	3.78	3.90	3.96
13	1.99	1.99	1.84	1.98	2.01
15	1.14, 1.68	1.16, 1.70	1.11, 1.60	1.12, 1.58	1.19, 1.72
16	1.46, 1.78	1.47, 1.75	1.21, 1.80	1.45, 1.74	1.50, 1.97
17	2.81 (*ddd*, $J=6.1, 6.7, 10.6$)	2.79	2.28	2.31	2.74
18	1.22 (*s*)	1.22 (*s*)	1.16 (*s*)	1.20 (*s*)	1.20 (*s*)
19	1.03 (*s*)	1.04 (*s*)	1.05 (*s*)	1.00 (*s*)	0.97 (*s*)
21	1.85 (*s*)	1.89 (*s*)	1.19 (*s*)	1.53 (*s*)	4.89, 5.11 (2 br. *s*)
22	5.89 ($d, J=9.6$)	5.52 (*d*, $J=10.0$)	1.41, 1.62	6.35 ($d, J=16.1$)	2.26, 2.46
23	4.68 (*dd*, $J=8.0, 9.6$)	4.07 (*dd*, $J=7.8, 10.0$)	1.17, 2.08	6.50 (*dd*, $J=6.7, 16.1$)	2.27; 2.92 ($t, J=7.3$)
24	3.22 ($d, J=8.0$)	3.09 ($d, J=7.8$)	5.19 ($t, J=7.1$)	4.47 ($d, J=6.7$)	5.28 ($t, J=6.5$)
26	1.28 (*s*)	1.26 (*s*)	1.71 (*s*)	1.58* (*s*)	1.66 (*s*)
27	1.48 (*s*)	1.44 (*s*)	1.62 (*s*)	1.58* (*s*)	1.59 (s)
28	2.06 (*s*)	2.07 (*s*)	2.07 (*s*)	2.04 (*s*)	2.06 (*s*)
29	1.60 (*s*)	1.60 (*s*)	1.60 (*s*)	1.59 (*s*)	1.45 (*s*)
30	0.82 (*s*)	0.83 (*s*)	0.84 (*s*)	0.81 (*s*)	0.81 (*s*)
MeO	–	3.42 (*s*)		–	–
EtO	–	–	1.22 ($t, J=6.8$); 3.41 ($q, J=6.8$)	–	–
1′	5.02 ($d, J=8.0$)	5.03 ($d, J=7.3$)	5.03 ($d, J=7.1$)	5.00 ($d, J=7.5$)	4.93 ($d, J=7.0$)
2′	4.09 ($t, J=8.3$)	4.10	4.09 ($t, J=7.6$)	4.07 ($t, J=8.2$)	4.36
3′	4.26 ($t, J=9.0$)	4.26 ($t, J=8.7$)	4.25 ($t, J=8.5$)	4.25 ($t, J=8.6$)	4.31
4′	4.21 ($t, J=8.7$)	4.22 ($t, J=9.2$)	4.20 ($t, J=9.0$)	4.20 ($t, J=8.6$)	4.15
5′	3.95 (*ddd*, $J=2.6, 5.1, 8.3$)	3.96 (*ddd*, $J=3.6, 5.9, 9.2$)	3.95	3.94	3.82
6′	4.36 (*dd*, $J=5.1, 11.5$); 4.53 (*dd*, $J=2.6, 11.5$)	4.37; 4.53 (br. *d*, $J=10.5$)	4.35 (*dd*, $J=5.3, 12.1$); 4.53 (*dd*, $J=3.4, 12.1$)	4.34 (*dd*, $J=5.6, 11.9$); 4.51 (*dd*, $J=3.0, 11.9$)	4.34; 4.46 (br. *d*, $J=11.9$)

[a]) Xylosyl resonances of **5** at δ_H 5.74 ($d, J=6.8$ Hz, H–C(1″)); 4.12 (H–C(2″); 4.14 (H–C(3″); 4.22 (H–C(4″); and 3.63, 4.30 (CH_2(5″)).

The HR-FAB mass spectrum of notoginsenosides T_4 (**4**) showed a quasi-molecular-ion peak at *m/z* 669.4242 ($[M-1]^-$), establishing its molecular formula as $C_{36}H_{62}O_{11}$ (calc. 669.4214), as confirmed by ^{13}C-NMR (DEPT) and FAB-MS (*m/z* 669 ($[M-1]^-$)). Comparison of the ^{13}C-NMR chemical shifts of **4** with those of ginsenoside R_8 [8] showed that one methene C-atom in the side chain of **4** was replaced by a methine C-atom. From the spectral data, the structure of notoginsenosides T_4 (**4**) was elucidated as (3*β*,6*α*,12*β*,20*S*,22*E*,24*RS*)-6-[(*β*-D-glucopyranosyl)oxy]dammar-22-ene-3,12,20,24,25-pentol.

The ^{1}H- and ^{13}C-NMR spectra of **4** showed one anomeric resonance each at δ_H 5.02 (*d*, *J* = 7.5 Hz, H–C(1′)) and δ_C 106.1 (C(1′)), respectively, indicating *one* sugar unit, in addition to two olefinic C-atoms at δ_C 136.4 and 130.6. The ^{13}C-NMR chemical shifts of **4** were similar to those of notoginsenoside R_8 [8], except for the side chain. Compared to notogensinoside R_8, one of the methene C-atoms was replaced by an oxymethine resonance at δ_C 80.23 in **4**, and C(25) was shifted upfield to 72.9 ppm. The above evidence indicated that the oxymethine group (δ_C 80.23) was in C(24) position. This was confirmed by ^{1}H,^{1}H-COSY and HMBC experiments. The ^{1}H,^{1}H-COSY spectrum showed a correlation between δ_H 6.50 (*dd*, *J* = 6.7, 16.1 Hz, H–C(23)) and 4.47 (*d*, *J* = 6.7 Hz, H–C(24)). The HMBC spectrum showed the following correlations: δ_H 6.35 (*d*, *J* = 16.1 Hz, H–C(22)) with δ_C 80.2 (C(24)); δ_H 6.50 (*dd*, *J* = 6.7, 16.1 Hz, H–C(23)) with δ_C 80.2 (C(24)); δ_H 4.47 (*d*, *J* = 6.7 Hz, H–C(24)) with δ_C 25.7 (C(27)), 72.9 (C(25)), 136.4 (C(22)), and 130.6 (C(23)).

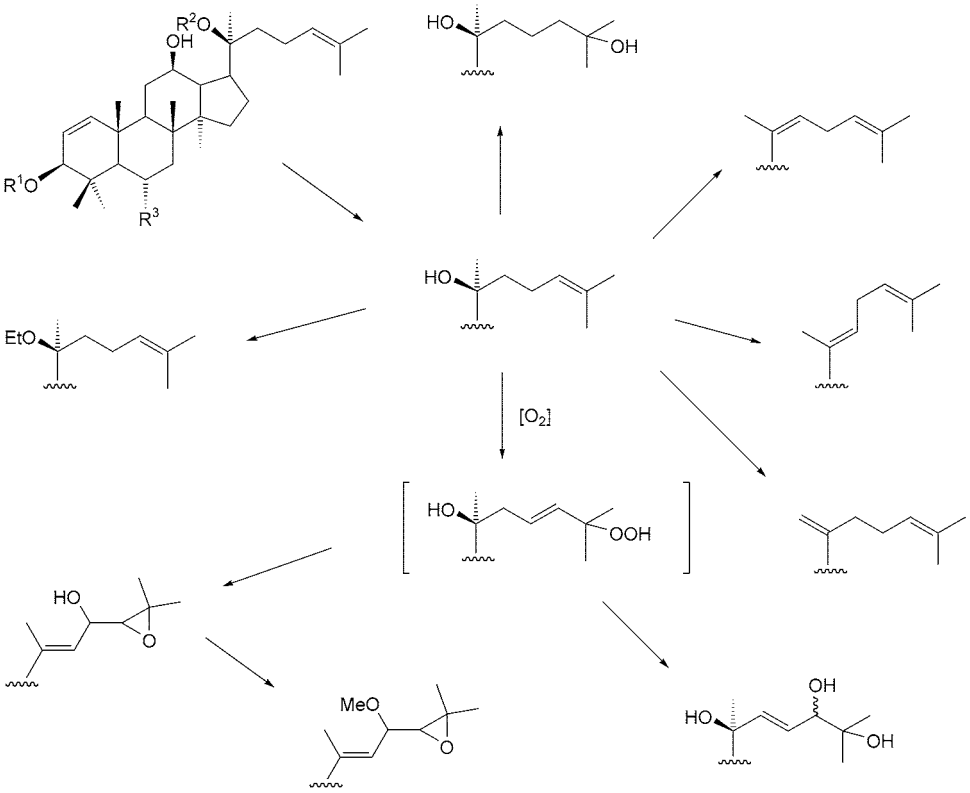
The molecular formula of notoginsenoside T_5 (**5**) was established by FAB-MS (*m/z* 751 ($[M-1]^-$)) and HR-FAB-MS (*m/z* 751.4609 ($[M-1]^-$, calc. 751.4632) as $C_{41}H_{68}O_{12}$, as confirmed by ^{13}C-NMR (DEPT). On the basis of spectral data and comparison with notoginsenoside R_2 [9], the structure of notoginsenosides T_5 (**5**) was determined as (3*β*,6*α*,12*β*,24*E*)-6-[(*β*-D-xylopyranosyl-(1 → 3)-*β*-D-glucopyranosyl)-oxy]dammara-20(21),24-diene-3,12-diol.

In the ^{1}H- and ^{13}C-NMR spectra of **5**, there were two anomeric signals each at δ_H 4.93 (*d*, *J* = 7.0 Hz, H–C(1′)) and 5.74 (*d*, *J* = 6.8 Hz, H–C(1″)), and at δ_C 103.6 (C(1′)) and 104.9 (C(1″)), respectively, which suggested the presence of *two* sugar moieties. Furthermore, the ^{13}C-NMR spectrum of **5** indicated the presence of a disubstituted C=C bond (δ_C 155.5, 108.2), and a trisubstituted C=C bond (δ_C 125.4, 131.3). The ^{13}C- and ^{1}H-NMR chemical shifts of **5** were very similar to those of notoginsenoside R_2 [9], except for the side chain of the aglycone. Based on the NMR data, **5** had to possess an additional (second) C=C bond (instead of a Me group) compared to notoginsenoside R_2. The second C=C bond was, thus, placed between C(20) and C(21), as confirmed by the following HMBC long-range correlations: δ_H 5.11 and 4.89 (*s*, H_a–C(21), H_b–C(21)) with δ_C 48.3 (C(17)), 33.8 (C(22), and 155.5 (C(20); δ_H 2.74 (H–C(17)) with δ_C 108.2 (C(21)) and 155.5 (C(20)). The ^{13}C-NMR chemical shifts of the side chain of **5** were similar to those of dammara-20,24-diene [21], which further confirmed the structural assignment.

From the structures of the new compounds **1**–**5** (and those of the 15 known compounds isolated previously [14–16]), it is evident that hydrolysis reactions mainly occur at the side chain of ginsenosides, since **1**–**5** differed from genuine saponins only in the side chain of the aglycone. Ginsenosides Rg_5, Rh_4, and (20*R*)- and (20*S*)-25-hydroxyginsenoside Rh_1 were isolated from *P. notoginseng* for the first time [22]. Last but not the least, ginsenosides Rg_3 and Rh_1, two promising anticancer agents, were obtained as the major hydrolysis products, while the major components of the crude root saponins were ginsenosides Rg_1 and Rb_1 [22]. Our experiments, thus, have demonstrated that controlled hydrolysis of the crude root saponins of *P. ginseng* increased the molecular diversity of this specific natural pool, which may provide further exploring opportunity or lead compounds for drug discovery.

Possible reaction pathways for the hydrolytic reactions taking place at the ginsenoside/notoginsenoside side chains are shown in the *Scheme*. The glycosyl moiety at C(20) of the dammarane framework tends to be degraded easily under acidic conditions (deglycosylation), which leads to the formation of 20-OH glycosides. These may then lose H_2O (dehydration) to afford different diene derivatives. The 20-OH glycosides can give rise to the 20-*O*-alkylated derivatives, as well as to the formation of 25-OH derivatives *via* hydration of the C=C bond. In the latter case, a hydroperoxy intermediate might be produced by oxidation of the C=C bond, consequently leading to a series of reactions, including dehydration, methylation, and rearrangement (*Scheme*).

Scheme. *Proposed Reaction Pathways for Modifications at the Side Chain of Ginsenosides*



Experimental Part

General. The crude root saponins of *Panax notoginseng* (BURK.) F. H. CHEN were purchased from the *Yuxi Tianqi Weihe Co.*, Yunnan, China. Column chromatography (CC): silica gel (160–200 mesh; *Qingdao Marine Chemical and Industrial Factory*, China) and *RP-18* or *RP-8* gel (40–60 μm, *Merck*). Melting points (m.p.): *Koffler* melting-point apparatus (Sicuan University, China); uncorrected. Optical rotations: *HORIBA SEPA-300* polarimeter; Na-D line. IR Spectra: *Bio-Rad FTS-135* spectrometer; in cm^{-1}. ^1H- and ^{13}C-NMR Spectra: *Bruker DRX-500* spectrometer (500/125 MHz), in C_5D_5N; chemical shifts δ in ppm rel. to C_5H_5N (δ_H 8.71, δ_C

149.9), coupling constants *J* in Hz. 2D-NMR HMBC, HMQC, and ^{1}H,^{1}H-COSY Spectra: *Z*-pulse field gradient; ROESY: spin lock time 300 ms. FAB- and HR-FAB-MS: *VG Autospec-3000* mass spectrometer, in *m*/*z*.

Acid Hydrolysis and Product Isolation. The crude root saponins (400 g), dissolved in EtOH (5 l), were treated with AcOH (5 l). The soln. was heated at 60° for *ca*. 6 h. The alcoholic solvent was evaporated, and the residual was subjected to a *Diaion HP-20* column, washed with plenty of H_2O, and eluted with aq. 80% MeOH to give the crude hydrolysate (200 g). The latter was purified by CC (SiO_2; $CHCl_3$/MeOH/H_2O 10:2.5:0.3): six fractions (Fr.). *Fr. 2* was subjected to CC (SiO_2; $CHCl_3$/MeOH/H_2O 15:1.5:0.15, lower phase): four subfractions. *Fr. 2.2* (1 g of 3 g in total) was purified by CC (*RP-8*; MeOH/H_2O 40:60 → 70:30) to afford compounds **1** (10 mg), **2** (50 mg), ginsenoside Rh_4 (500 mg), and **3** (100 mg). *Fr. 2.4* (100 mg of 2 g in total) was purified by CC (*RP-18*; MeOH/H_2O 40:60 → 60:40) to afford ginsenoside Re (20 mg) and ginsenoside Rg_2 (15 mg). *Fr. 3*, *Fr. 4*, and *Fr. 6* were purified similarly (1. SiO_2; $CHCl_3$/MeOH/H_2O; 2. *RP-8*; aq. MeOH). *Fr. 3* afforded ginsenoside Rh_1 (2 g), (20*R*)-ginsenoside Rh_1 (300 mg), **5** (80 mg), and (20*R*)-ginsenoside Rg_3 (300 mg). *Fr. 4* led to ginsenoside Rd (400 mg), ginsenoside Rg_3 (200 mg), ginsenoside Rg_1 (100 mg), and notoginsenoside R_1 (36 mg). *Fr. 6* gave **4** (35 mg), gypenoside XVII (60 mg), a mixture of (20*R*)- and (20*S*)-25-hydroxyginsenoside Rh_1 (40 mg), ginsenoside Rg_5 (40 mg), notoginsenoside R_2 (17 mg), and ginsenoside Rh_4 (14 mg).

*(3β,6α,12β,20*E*,23*RS*)-24,25-Epoxy-6-[(β-*D*-glucopyranosyl)oxy]-dammar-20(22)-ene-3,12,23-triol* (*notoginsenoside* T_1; **1**). White powder. M.p. 131–133. $[\alpha]_D^{25} = 14.49$ ($c = 0.50$, MeOH). IR (KBr): 3410, 2933, 2877, 1600, 1460, 1383, 1256, 1156, 1076, 1032, 929, 893, 819. ^{1}H- and ^{13}C-NMR: see *Tables 2* and *1*, resp. FAB-MS: 652 (M^-), 593 ([$M - C_3H_7O$]$^-$), 415 ([$M - C_3H_7O - 178$]$^-$). HR-FAB-MS: 651.4139 ([$M - H$]$^-$, $C_{36}H_{59}O_{10}^-$; calc. 651.4108).

*(3β,6α,12β,20*E*,23*RS*)-24,25-Epoxy-6-[(β-*D*-glucopyranosyl)oxy]-23-methoxydammar-20(22)-ene-3,12-diol* (*notoginsenoside* T_2; **2**). White powder. M.p. 155–157. $[\alpha]_D^{25} = 28.38$ ($c = 0.41$, MeOH). IR (KBr): 3435, 2935, 2878, 1655, 1461, 1382, 1255, 1197, 1075, 1031, 929, 899, 812. ^{1}H- and ^{13}C-NMR: see *Tables 2* and *1*, resp. FAB-MS: 665 ([$M - H$]$^-$), 503 [$M - H - 162$]$^-$). HR-FAB-MS: 665.4227 ([$M - H$]$^-$, $C_{37}H_{61}O_{10}^-$; calc. 665.4265).

*(3β,6α,12β,20*S*)-6-[(β-*D*-Glucopyranosyl)oxy]-20-ethoxydammar-24-ene-3,12-diol* (*notoginsenoside* T_3; **3**). White powder. M.p. 154–156. $[\alpha]_D^{28} = 24.95$ ($c = 0.54$, pyridine). IR (KBr): 3393, 2967, 2933, 2877, 1642, 1460, 1386, 1078, 1032, 933, 899. ^{1}H- and ^{13}C-NMR: see *Tables 2* and *1*, resp. FAB-MS: 666 (M^-), 503 ([$M - H - 162$]$^-$). HR-FAB-MS: 665.4681 ([$M - H$]$^-$, $C_{38}H_{65}O_9^-$; calc. 665.4628).

*(3β,6α,12β,20*S*,22*E*,24*RS*)-6-[(β-*D*-Glucopyranosyl)oxy]dammar-22-ene-3,12,20,24,25-pentol* (*notoginsenoside* T_4; **4**). White powder. M.p. > 350°. $[\alpha]_D^{26} = 25.56$ ($c = 0.23$, MeOH). IR (KBr): 3404, 2966, 2934, 2877, 1645, 1464, 1384, 1079, 970, 801. ^{1}H- and ^{13}C-NMR: see *Tables 2* and *1*, resp. FAB-MS: 669 ([$M - H$]$^-$), 507 ([$M - H - 162$]$^-$). HR-FAB-MS: 669.4242 ([$M - H$]$^-$, $C_{36}H_{61}O_{11}^-$; calc. 669.4214).

*(3β,6α,12β,24*E*)-6-[(β-*D*-Xylopyranosyl-(1 → 3)-β-*D*-glucopyranosyl)oxy]dammara-20(21),24-diene-3,12-diol* (*notoginsenoside* T_5; **5**). White powder. M.p. 161–163. $[\alpha]_D^{25} = 5.59$ ($c = 0.31$, MeOH). IR (KBr): 3414, 2963, 2934, 2877, 1731, 1639, 1461, 1376, 1295, 1254, 1077, 1044, 928, 892, 813. ^{1}H- and ^{13}C-NMR: see *Tables 2* and *1*, resp. FAB-MS: 752 (M^-), 519 ([$M - H - 132$]$^-$), 457 ([$M - H - 132 - 162$]$^-$). HR-FAB-MS: 751.4609 ([$M - H$]$^-$, $C_{41}H_{67}O_{12}^-$; calc. 751.4633).

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