*Chemistry of Natural Compounds, Vol. 47, No. 3, July, 2011* [*Russian original No. 3, May-June, 2011*]

# TRITERPENE GLYCOSIDES FROM *Astragalus* AND THEIR GENINS. LXXXIX. ASKENDOSIDE H FROM *Astragalus taschkendicus*

**I. M. Isaev and M. I. Isaev***

UDC 547.918:547.926

*A new triterpene glycoside of the cycloartane series that was called askendoside H was isolated from roots of* Astragalus taschkendicus *Bunge (Leguminosae). Its structure was elucidated based on chemical transformations and spectral data. Askendoside H was a bisdesmoside of cycloorbigenin C, 23R,24R-cycloartan-3β,6α,16β,23,24,25-hexaol 3-O-[(α-L-arabinopyranosyl)(1→2)- β-D-xylopyranoside] 23-O-β-D-glucopyranoside.*

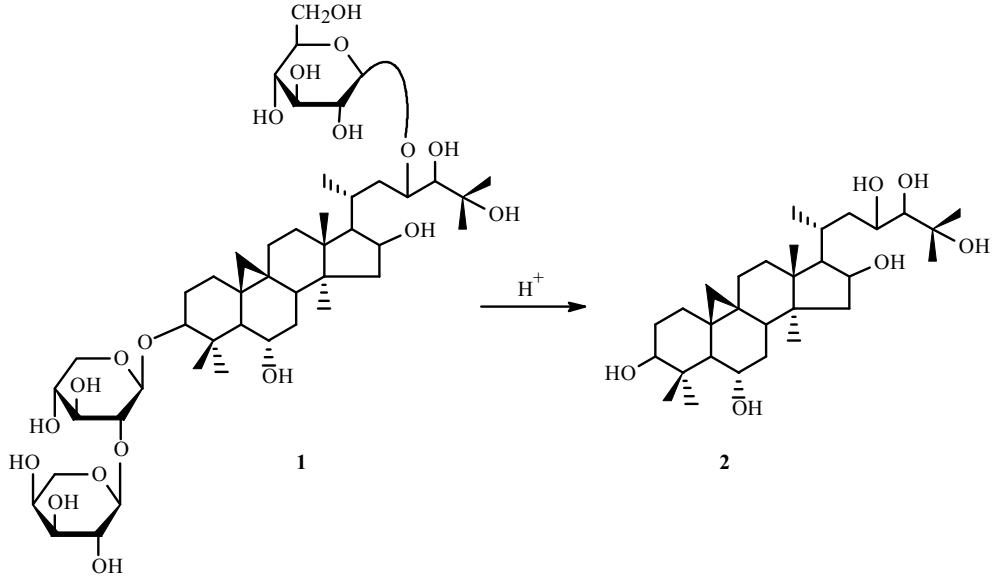
**Keywords:** *Astragalus taschkendicus*, Leguminosae, triterpene glycosides, askendoside H, cycloorbigenin C, PMR, $^{13}$C NMR, DEPT, COSY, Hetcor, and NOE spectra.

In continuation of our chemical research on isoprenoids from plants of the genus *Astragalus* (Leguminosae), we isolated a new triterpene glycoside from roots of *A. taschkendicus* Bunge that we called askendoside H (**1**). Herein we provide proof of the structure of this glycoside.

The PMR spectrum of **1** exhibited at strong field δ 0.12 and 0.45 two 1H resonances for an AX system that belonged to methylene of tetrasubstituted cyclopropane and resonances for seven methyls. This enabled us to classify **1** as a cycloartane series triterpenoid [1–4].

Acid hydrolysis of **1** and subsequent analysis of the carbohydrate part of the hydrolysate by paper chromatography (PC) identified taking into account biogenetic considerations D-xylose, D-glucose, and L-arabinose.

The $^{13}$C NMR spectrum of **1** showed resonances for three anomeric C atoms at δ 105.61, 106.67, and 106.67 (Table 1), thereby showing that **1** contained D-xylose, D-glucose, and L-arabinose in a 1:1:1 ratio, i.e., the new glycoside was a trioside. The chemical shifts of the monosaccharide C atoms as a whole indicated that the monosaccharide units in **1** had the pyranose form.




**1** **2**

S. Yu. Yunusov Institute of the Chemistry of Plant Substances, Academy of Sciences, Republic of Uzbekistan, Tashkent, fax (99871) 120 64 75, e-mail: m_isaev@rambler.ru. Translated from Khimiya Prirodnykh Soedinenii, No. 3, pp. 367–369, May–June, 2011. Original article submitted November 29, 2010.

TABLE 1. Chemical Shifts of C and H Atoms of Askendoside H (**1**) and Chemical Shifts of C Atoms of Cycloorbigenin C (**2**) (δ, ppm, J/Hz, $C_5D_5N$, 0 = TMS for $\delta_C$, 0 = HMDS for $\delta_H$)

| C atom | DEPT | **1** | | **2** |
|---|---|---|---|---|
| | | $\delta_C$ | $\delta_H$ | $\delta_C$ |
| 1 | $CH_2$ | 32.46 | | 32.74 |
| 2 | $CH_2$ | 29.67 | | 31.40 |
| 3 | CH | 88.50 | 3.47 dd (12.3, 4.5) | 78.31 |
| 4 | C | 42.75 | – | 42.42 |
| 5 | CH | 54.02 | 1.63 d (9) | 53.92 |
| 6 | CH | 67.81 | 3.67 td (9, 4) | 68.18 |
| 7 | $CH_2$ | 38.28 | | 38.53 |
| 8 | CH | 46.77 | | 47.09 |
| 9 | C | 21.31 | – | 21.20 |
| 10 | C | 29.15 | – | 29.53 |
| 11 | $CH_2$ | 26.24 | | 26.26 |
| 12 | $CH_2$ | 33.04 | | 32.95 |
| 13 | C | 45.78 | – | 45.61 |
| 14 | C | 46.72 | – | 46.79 |
| 15 | $CH_2$ | 47.86 | 1.53, 1.97 dd (12.9, 8) | 47.68 |
| 16 | CH | 72.09 | 4.53 m | 72.10 |
| 17 | CH | 57.38 | 1.70 | 57.40 |
| 18 | $CH_3$ | 19.66 | 1.30 s | 18.85 |
| 19 | $CH_2$ | 30.36 | 0.12 d (4), 0.45 d (4) | 30.23 |
| 20 | CH | 27.55 | 2.59 m | 27.30 |
| 21 | $CH_3$ | 18.82 | 1.15 d (6.6) | 20.27 |
| 22 | $CH_2$ | 38.95 | 2.45 dd (15, 10) | 42.88 |
| 23 | CH | 82.77 | 4.55 m | 73.09 |
| 24 | CH | 80.66 | 4.50 d (2.8) | 79.08 |
| 25 | C | 72.66 | – | 74.29 |
| 26 | $CH_3$ | 26.35 | 1.52 s | 24.58 |
| 27 | $CH_3$ | 28.41 | 1.54 s | 28.91 |
| 28 | $CH_3$ | 20.08 | 0.88 s | 20.14 |
| 29 | $CH_3$ | 28.58 | 1.86 s | 29.32 |
| 30 | $CH_3$ | 16.22 | 1.28 s | 16.12 |
| | | | *β*-D-Xyl*p* unit | |
| 1 | CH | 105.61 | 4.81 d (7) | |
| 2 | CH | 83.62 | 4.01 dd (9, 6.8) | |
| 3 | CH | 77.64 | 4.06 | |
| 4 | CH | 70.97 | 4.06 td (6.3, 2.3) | |
| 5 | $CH_2$ | 67.01 | 3.50 dd (11, 9.4), 4.16 dd (11, 4.7) | |
| | | | *α*-L-Ara*p* unit | |
| 1 | CH | 106.67[a] | 5.11 d (6.6) | |
| 2 | CH | 73.64 | 4.48 dd (8.7, 6.8) | |
| 3 | CH | 74.29 | 4.11 dd (7.5, 4.7) | |
| 4 | CH | 69.14 | 4.20 td (3.5, 1.9) | |
| 5 | $CH_2$ | 66.60 | 3.70 dd (12.2, 1.9), 4.29 dd (12.2, 3.3) | |
| | | | *β*-D-Glc*p* unit | |
| 1 | CH | 106.67[a] | 5.02 d (7.7) | |
| 2 | CH | 75.89 | 3.87 dd (8.9, 7.7) | |
| 3 | CH | 78.25 | 3.95 dd (9.6, 8.4) | |
| 4 | CH | 72.22 | 4.10 dd (8.4, 7.7) | |
| 5 | CH | 78.22 | 3.88 td (8.9, 2.4) | |
| 6 | $CH_2$ | 63.12 | 4.43 dd (11.5, 2.8), 4.10 dd (12, 9) | |

Resonances marked with the same letter are mutually overlapped.

The anomeric protons of D-xylose, D-glucose, and L-arabinose resonated in the PMR spectrum of **1** as doublets at δ 4.81, 5.02, and 5.11. Their SSCC ($^3J$ = 7, 7.7, 6.6 Hz) also indicated that they had the pyranose form, the $^4C_1$-conformation, and the β-configuration for D-xylose and D-glucose and the α-configuration for L-arabinose.

The genin, which was identified as cycloorbigenin C (**2**), was isolated from the genin part of the acid hydrolysis products [5, 6].

A comparison of $^{13}C$ NMR spectra of cycloorbigenin C and askendoside H showed that C-3 and C-23 of the genin and C-2 of D-xylose experienced a glycosylation effect in the latter. These resonated at δ 88.50, 82.77, and 83.62, respectively. Therefore, the new glycoside **1** was a bisdesmoside glycoside, the carbohydrate constituents of which were located on C-3 and C-23 of cycloorbigenin C and the D-glucose and L-arabinose of which were terminal.

Nuclear Overhauser Effect (NOE) difference measurements with pre-irradiation of the D-glucose anomeric proton revealed an Overhauser effect on the H-23 resonance. This determined unambiguously the location of the D-glucose on C-23. This meant that the biose located on C-3 was (α-L-arabinopyranosyl)(1→2)- β-D-xylopyranose.

The difference PMR spectrum with irradiation of the D-xylose anomeric proton (δ 4.81) revealed a negative NOE on the resonance of the genin H-3 (δ 3.36) [7, 8], which confirmed the conclusion about the attachment of D-xylose to C-3.

Thus, the experimental results led to the conclusion that the new triterpene glycoside **1** was 23*R*,24*R*-cycloartan-3β,6α,16β,23,24,25-hexaol 3-*O*-[(α-L-arabinopyranosyl)(1→2)- β-D-xylopyranoside] 23-*O*-β-D-glucopyranoside.

## EXPERIMENTAL

**General comments** have been published [9]. The following solvent systems were used: $CHCl_3$:MeOH:$H_2O$ (70:23:4, 1), $CHCl_3$:MeOH (10:1, 2), *n*-BuOH:Py:$H_2O$ (6:4:3, 3). NMR spectra were recorded in Py-$d_5$ on a UNITYplus 400 (Varian) spectrometer. $^{13}C$ NMR spectra were obtained with full C–H decoupling and under DEPT conditions. 2D spectra of **1** were recorded using standard Varian programs. Chemical shifts of protons in **1** and **2** are given vs. HMDS. Chemical shifts of C atoms in $^{13}C$ NMR spectra of **1** and **2** are given vs. resonances of the β-C atoms of deuteropyridine (δ 123.493 vs. TMS).

**Isolation and Separation of Triterpenoids from *A. taschkendicus*** [10–12]. Fractions that eluted after askendoside G and contained askendoside H were combined and rechromatographed over a column using system 1 to afford **1** (170 mg, 0.0033% of air-dried raw material).

**Askendoside H (1)**, $C_{46}H_{78}O_{19}$, white non-crystalline compound. Table 1 presents the PMR and $^{13}C$ NMR spectra of **1**.

**Cycloorbigenin C (2) from 1.** Askendoside H (140 mg) was hydrolyzed by methanolic $H_2SO_4$ (15 mL, 0.5%) on a boiling-water bath for 6 h. The mixture was diluted with $H_2O$. The MeOH was evaporated. The resulting precipitate was filtered off, washed with $H_2O$ until neutral, and dried. The dry product was chromatographed over a column with elution by system 2 to afford **2** (25 mg), $C_{30}H_{52}O_6$, mp 256–258°C (MeOH).

PMR spectrum of **2** (400 MHz, $C_5D_5N$, δ, ppm, J/Hz, 0 = HMDS): 0.19 and 0.47 (d, $^2J$ = 4, 2H-19), 0.89 (s, $CH_3$), 1.07 (d, $^3J$ = 6.7, $CH_3$-21), 1.26, 1.27, 1.57, 1.61, 1.79 (s, 5 × $CH_3$), 3.56 (dd, $^3J_1$ = 11.6, $^3J_2$ = 4.7, H-3), 3.65 (d, $^3J$ = 8.5, H-24), 3.69 (td, $^3J_1 = {}^3J_2$ = 9.3, $^3J_3$ = 3.8, H-6), 4.22 (td, $^3J_1 = {}^3J_2$ = 8.7, $^3J_3$ = 2, H-23), 4.59 (td, $^3J_1 = {}^3J_2$ = 7.6, $^3J_3$ = 4.7, H-16).

Table 1 presents the $^{13}C$ NMR spectrum for **2**.

The filtrate was evaporated to a small volume and heated on a water bath for 1 h to destroy methylglycosides. The solution was cooled and neutralized with $BaCO_3$. The precipitate was removed. The solution was evaporated to a small volume. PC using system 3 and comparison with authentic samples detected D-glucose, D-xylose, and L-arabinose. PMR and $^{13}C$ NMR spectra provided evidence that the aforementioned monosaccharides were present in a 1:1:1 ratio in **1**.

## ACKNOWLEDGMENT

The work was supported financially by the Republic of Uzbekistan State Foundation for Basic Research (Grant FA-FZ-T-044) and GNTP (Grant FA-A12-T-101).

**REFERENCES**


1. M. I. Isaev, M. B. Gorovits, and N. K. Abubakirov, *Khim. Prir. Soedin.*, 431 (1985).
2. M. I. Isaev, M. B. Gorovits, and N. K. Abubakirov, *Khim. Prir. Soedin.*, 156 (1989).
3. R. P. Mamedova and M. I. Isaev, *Khim. Prir. Soedin.*, 257 (2004).
4. V. I. Lutskii, A. S. Gromova, E. A. Khamidullina, and N. L. Owen, *Khim. Prir. Soedin.*, 97 (2005).
5. R. P. Mamedova, M. A. Agzamova, and M. I. Isaev, *Khim. Prir. Soedin.*, 384 (2003).
6. I. M. Isaev, R. P. Mamedova, M. A. Agzamova, and M. I. Isaev, *Khim. Prir. Soedin.*, 95 (2007).
7. M. I. Isaev, B. A. Imomnazarov, Yu. M. Fadeev, and P. K. Kintya, *Khim. Prir. Soedin.*, 360 (1992).
8. H. K. Wang, K. He, L. Ji, Y. Tezuka, T. Kikuchi, and I. Kitagawa, *Chem. Pharm. Bull.*, **37**, 2041 (1989).
9. R. P. Mamedova, M. A. Agzamova, and M. I. Isaev, *Khim. Prir. Soedin.*, 453 (2001).
10. M. I. Isaev, M. B. Gorovits, N. D. Abdullaev, and N. K. Abubakirov, *Khim. Prir. Soedin.*, 458 (1982).
11. M. I. Isaev, *Khim. Prir. Soedin.*, 820 (1995).
12. M. I. Isaev, *Khim. Prir. Soedin.*, 723 (1996).