Bioassay Directed Isolation Studies on *Solanum surrattense*

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Summary: Bioassay directed isolation studies on *solanum surrattense* have resulted in the isolation of three steroidal alkaloids namely, acetyl-solasodine (**1**), khasianine (**2**) and solamargine (**3**) from the ethyl acetate soluble fraction. Compounds **1-3** showed moderate inhibitory activity against the enzyme urease. In addition, coumarin (**4**),, cinnamic acid (**5**), lupeol (**6**), betulin (**7**), 3,5,7-trihydroxy-6,4′-dimethoxyflavone (**8**), β-sitosterol 3-O-β-D-glucopyranoside (**9**), 3′,4′,5-trihydroxyflavone 7-O-β-D-glucopyranoside (**10**) have also been isolated, identified by NMR and showed no inhibitory activity against the enzyme urease.

Introduction

Solanum is one of the important genus of family Solanaceae. This family has 84 genera and well over 3000 species, mostly distributed in tropical and temperate regions of both the hemispheres, but chiefly in West and Southern parts of America. *Solanum* [1] has over 1400 species but only 15 have so far been identified in Pakistan [2]. *S. Surrattense* is a thorny more or less prostrate annual herb with star shaped hairs, leaves are 5-10 cm, oblong pinately divided, nerves on both surfaces armed with long strate prickle. Flowers are 2.5 cm in dm, blue, solitary or in small clusters [3]. Seeds are 2 mm in dm, and are smooth. It is found throughout India and Pakistan. It also occurs in South East Asia, Malaysia, Tropical Australia and Polynesian Island. It grows naturally in waste land, road sides, dry places and field boarders in warmer areas of countries [4].

In most of the countries *S. Surrattense* is used as a medicine. The drug is useful in cough, asthma, pain in chest and fever. It possesses diuretic and antibacterial properties. It is also useful in bronchitis, muscular pain, enlargement of liver, spleen, blood cancer, vomiting and controlling of stone in bladder [5].

Results and Discussion

The methanolic extract showed strong toxicity in brime shrimp lethality test [6]. On further fractionation for the extract, the major toxicity was found in the ethyl acetate soluble fraction. As a result of chromatographic resolution, three steroidal alkaloids have been isolated and identified as acetyl-solasodine (**1**), khasianine (**2**) and solamargine (**3**) respectively. All of these showed moderate inhibitory against the enzyme urease.

In addition seven non-alkaloidal constituents have also been isolated and characterized as coumarin (**4**), cinnamic acid (**5**), lupeol (**6**), betulin (**7**), 3,5,7-trihydroxy-6,4′-dimethoxyflavone (**8**), β-sitosterol 3-O-β-D-glucopyranoside (**9**), 3′,4′,5-trihydroxyflavone 7-O-β-D-glucopyranoside (**10**). These were also subjected to urease inhibition assay but showed no activity. Eight compounds have been reported for the first time with already two reported compounds. These compounds were also tested against anti-inflammation using human neutrophil cells which were found to be inactive.

Table-1: Urease inhibition assay of **1-3**.

Compound code	concentration	% inhibition	IC_{50}± S.E.M
1	**0.5 mM**	**56.7 % conc**	**417.93 ± 0.67**
2	**0.5 mM**	**58.5 % conc**	**448.2 ± 1.27**
3	**0.5 mM**	**51.6 % conc**	**546.3 ± 2.37**
Thiourea (Standard)	**0.5 mM**	**100 %**	**21.0 ± 0.11**

S.E.M = STANDARD ERROR MEAN

Acetylation

Acetylation of compounds **1** and **2** were carried out under standard method. In this process 10 mg of each compounds were kept in 4 ml acetic

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anhydride and 3 ml pyridine under anhydrous conditions. Work up were followed by the purification through silica gel column and these products were crystallized by using methanol as a solvent. Acetylation was confirmed by change in melting point, FABMS, IR and ^{1}H-NMR spectroscopy. It confirmed that nitrogen is present in secondary state. These acetylated products were screen against urease inhibition which were inactive against urease inhibition.

Experimental

General

Optical rotations were measured on a JASCO DIP-360 polarimeter. IR spectra were recorded on a 460 Shimadzu spectrometer. EI-MS and HR-FAB-MS were recorded on JMS-HX-110 and JMS-DA 5000 mass spectrometers. The ^{1}H-NMR, ^{13}C-NMR, HMQC, and HMBC spectra were recorded on Bruker spectrometers operating at 400 MHz for ^{1}H- and 100 MHz for ^{13}C-NMR, respectively. The chemical shift values are reported in ppm (δ) units and the coupling constants (*J*) are in Hz. Aluminum sheets precoated with silica gel 60 F_{254} (20 × 20 cm, 0.2 mm thick; E-Merck) were used for TLC and silica gel (230-400 mesh) was used for column chromatography. Visualization of the TLC plates was carried out under UV at 254 and 366 nm by Hitachi UV-3200 spectrometer and by spraying with ceric sulfate reagent solution (with heating).

Plant Material

The whole plant material of *Solamun surrattense* was collected from Bahawalpur Division (Punjab) in June, 2006 and identified by Dr. Muhammad Arshad, Plant Taxonomist, Cholistan Institute of Desert Studies, Islamia University Bahawalpur, Bahawalpur, Pakistan. Where a voucher specimen (CIDS / 01 / 05) has been deposited.

Extraction and Isolation

The shade dried whole plant material (25 kg) was extracted with CH_3OH (3 × 100 L) at room temperature.The combined methanolic extract was evaporated under reduced pressure to obtain a thick gummy mass (600 g). It was suspended in water and successively extracted with *n*-hexane, chloroform, ethyl acetate and *n*-butanol.

The ethyl acetate soluble fraction (85 g) was subjected to column chromatography eluting with $CHCl_3$, $CHCl_3$-CH_3OH and CH_3OH in increasing order of polarity to obtain three fractions (A-C).The Fraction A obtained from ethyl acetate was further purified by column chromatography over silica gel eluting with chloroform and methanol (9.0: 1.0) to afford **9** (12 mg) and (8.5: 1.5) **10** (18 mg). The fraction B obtained from $CHCl_3$-CH_3OH (8.0: 2.0) was a mixture of two components, which were separated by column chromatography using solvent system$CHCl_3$-CH_3OH (7.5.0: 2.5) to afford compounds **2** (8mg).The fraction D obtained from $CHCl_3$-CH_3OH(7.5.0: 2.5) was further purified by column chromatography eluting with $CHCl_3$-CH_3OH (7.3: 2.7) to afford **1** (6 mg).

The chloroform soluble fraction (80 g) was subjected to column chromatography eluting with *n*-hexane, $CHCl_3$, $CHCl_3$-CH_3OH and CH_3OH in increasing order of polarity to obtain five fractions (A-D).The Fraction A obtained from chloroform was further purified by column chromatography over silica gel eluting with *n*-hexane and chloroform (9.0:1.0) to afford **4** (12 mg) and (8.5: 1.5) **5** (16 mg). The fraction B obtained from *n*-hexane-$CHCl_3$ (7.0:3.0) was a mixture of two components, which were separated by column chromatography using solvent system *n*-hexane-$CHCl_3$ (7.5: 2.5) to afford compounds **6** (14 mg) and **7** (10 mg) from the top and the tail fractions, respectively. The fraction C obtained from *n*-hexane-$CHCl_3$ (7.0: 3.0) was further purified by column chromatography eluting with *n*-hexane-$CHCl_3$ (6.8: 3.2) to afford **3** (10 mg). The fraction D obtained from *n*-hexane-$CHCl_3$ (6.0: 4.0) was further purified by column chromatography eluting with *n*-hexane-$CHCl_3$ (5.8: 4.2) to afford **8** (8 mg).

Urease Assay and Inhibition

Reaction mixture s comprising 25 μL of enzyme (jack bean urease) solution and 55 μL of buffers containing 100 mM urea were incubated with 5 μL of test compounds (0.5 mM concentration) at 30 °C for 15 min in 96-well plates. Urease activity was determined by measuring ammonia production using the indophenol method as described by weather burn. Briefly, 45 μl each phenol reagent (1% w/v phenol and 0.005 % w/v sodium nitroprussside) and 70 μL of alkali reagent(0.5 % w/v NaOH and 0.1 % active chloride NaOCl) were added to each well. The increasing absorbance at 630 nm was measured afther 50 min, m using a microplate reader (Molecular Device, USA). All reactions were performed in triplicate in a final volume of 200 μL. The results (change in absorbance per min) were processed by using softMax Pro software (molecular Device, USA). The entire assays were performed at pH 6.8.

Percentage inhibitions were calculated from the formula 100-($OD_{testwell}$ / $OD_{control}$) ×100. Thiourea was used as the standard inhibitor of urease [7, 8].

Acetyl-Solasodine (**1**)

Colorless crystals (8 mg), m.p. 190°C. $[\alpha]_D^{25}$ -119.0 (c = 0.22, CH_3OH); IR (KBr) ν_{max} cm^{-1}: 3390-3385 (OH and NH), 1720 (C=O), 1638 (C=C).; ^{1}H-NMR ($CDCl_3$, 400 MHz); δ 0.83 (3H, s, H-18), 1.04 (3H, s, H-19), 0.86 (3H, d, J = 6.5, H-27) 0.96 (3H, d, J = 7.5, H-21), 1.4-2.34 (many overlapped peaks), 2.7 (2H, m, br, H-26), 2.66 (3H, s, CH_3CO), 4.32 (1H, m, H-16), 5.24(1H, d, J = 4.7, 6-H); C^{13}NMR spectral data (100 MHz, CD_3OD + $CDCl_3$ 1:1): δ 170.4 (C = O), 21.2 (CH_3CO), 37.8 (C-1), 30.4 (C-2), 79.1 (C-3), 40.3 (C-4), 141.0 (C-5), 122.0 (C-6), 32.4 (C-7), 32.0 (C-8), 50.6 (C-9), 37.4 (C-10), 21.2 (C-11), 38.8 (C-12), 41.0 (C-13), 57.0 (C-14), 30.0 (C-15), 79.6 (C-16), 63.0 (C-17), 16.7 (C-18), 19.8 (C-19), 42.1 (C-20), 15.3 (C-21), 98.8 (C-22), 34.3 (C-23), 32.6 (C-24), 31.2 (C-25), 48.6 (C-26), 19.5 (C-27). HRFABMS, *m/z* $[M+H]^+$ 456.3240 (calcd. for $C_{29}H_{46}NO_3$, 456.3293). EIMS *m/z* (rel. Int %): 456 (15), 413 $[M-43]^+$ (25), 399 (30), 396 (100), 396 (22), 381 (58), 329 (40), 303 (65), 275 (25), 114 (90). The physical and spectral data showed complete resemblance with the reported values [11].

Khasianine (**2**)

White crystals, m.p. 253 °C, $[\alpha]_D^{25}$ -95° (c = 0.80, MeOH) IR (KBr) ν_{max} cm^{-1}: 3390-3372 (OH and NH), 1638 (C = C);. ^{1}H NMR (400 MHz, CD_3OD + $CDCl_3$ 1:1): δ 0.83 (3H, s, H-18), 1.04 (3H, s, H-19), 0.86 (3H, d, *J* = 6.5, H-27) 0.97 (3H, d, *J* = 7.5, H-21), 1.28 (3H, m, rhamnosyl CH_3), 1.4-2.36 (many overlapped peaks), 2.5 (2H, m, br, H-26), 3.0-4.0 (overlapped sugar protons), 4.32 (1H, m, H-16), 4.47 (1H, d, *J* = 7.5, H-1′), 4.86 (1H, s, H-1), 5.22, (1H, s, H-1″), 5.36(1H, d, *J* = 4.0, 6-H), C^{13}NMR (100 MHz, CD_3OD + $CDCl_3$ 1:1): δ 37.8 (C-1), 30.6 (C-2), 79.1 (C-3), 40.2 (C-4), 141.0 (C-5), 122.0 (C-6), 32.4 (C-7), 32.0 (C-8), 50.6 (C-9), 37.4 (C-10), 21.3 (C-11), 38.8 (C-12), 41.0 (C-13), 57.0 (C-14), 30.0 (C-15), 79.6 (C-16), 63.1 (C-17), 16.7 (C-18), 19.3 (C-19), 42.5 (C-20), 15.2 (C-21), 98.5 (C-22), 34.3 (C-23), 32.6 (C-24), 31.2 (C-25), 48.6 (C-26), 19.5 (C-27). Sugar moiety: δ 99.8 (C-1′), 79.4 (C-2′), 73.2 (C-3′), 75.6 (C-4′), 78.7 (C-5′), 61.2 (C-6′), 102.2 (C-1″), 71.7 (C-2″), 71.6 (C-3″), 73.2 (C-4″), 69.0 (C-5″), 17.8 (C-6″). FABMS *m/z* (rel.int): HRFABMS, *m/z:* $[M+H]^+$ 722.4455 (calcd for $C_{39}H_{64}NO_{11}$, 722.4401) EIMS *m/z* (rel. int.): 693 (20), 575 (25), 396 (15), 138 (80), 114(100), The physical and spectral data showed complete agreement with those reported in literature [9-10].

Solamargine (**3**)

White crystals, m.p. 284-285 °C, $[\alpha]_D^{20}$ -91 (c = 1.0, MeOH) ; IR (KBr) ν_{max} cm^{-1}: 3395-3374 (OH and NH), 1632 (C=C); ^{1}H NMR spectral data (400 MHz, CD_3OD + $CDCl_3$ 1:1): δ 0.83 (3H, s, H-18), 1.04 (3H, s, H-19), 0.86 (3H, d, J = 6.0, H-27) 0.96 (3H, d, J = 7.0, H-21), 1.28 (6H, m, rhamnosyl CH_3), 1.4-2.37 (many overlapped peaks), 2.7 (2H, m, br, H-26), 3.2-4.0 (overlapped sugar protons), 4.34 (1H, m, H-16), 4.47 (1H, d, *J* = 7.4, H-1′), 4.88 (1H, s, H-1″), 5.22, (1H, s, H-1″), 5.36(1H, d, *J* = 4.1, 6-H); ^{13}CNMR spectral data (100 MHz, CD_3OD + $CDCl_3$ 1:1): δ 37.8 (C-1), 30.4 (C-2), 79.1 (C-3), 40.3 (C-4), 141.2 (C-5), 122.3 (C-6), 32.4 (C-7), 32.0 (C-8), 50.6 (C-9), 37.6 (C-10), 21.5 (C-11), 38.7 (C-12), 41.3 (C-13), 57.4 (C-14), 30.2 (C-15), 79.6 (C-16), 63.3 (C-17), 16.5 (C-18), 19.4 (C-19), 42.2 (C-20), 15.1 (C-21), 98.8 (C-22), 34.3 (C-23), 32.6 (C-24), 31.2 (C-25), 48.6 (C-26), 19.5 (C-27). Sugar moiety: δ 99.8 (C-1′), 79.4 (C-2′), 73.2 (C-3′), 75.6 (C-4′), 78.5 (C-5′),, 61.3 (C-6′), 102.3 (C-1″), 71.7 (C-2″), 71.4 (C-3″), 73.2 (C-4″), 69.0 (C-5″), 17.6 (C-6″), 101.6 (C-1‴), 71.5 (C-2‴), 71.2 (C-3‴), 73.2 (C-4‴), 70.2 (C-5‴), 17.4 (C-6‴). HRFABMS $[M+H]^+$ *m/z*: 868.4958 (calcd for $C_{45}H_{74}NO_{15}$, 868.4980) EIMS *m/z* (rel. int.): 396 $[M-Glu-Rham-Rham]^+$ (5), 378 (20), 282 (30), 215 (15), 138 (80), 114 (100). The physical and spectral data showed complete agreement with those reported in literature [9-10].

Coumarin **(4)**

Amorphous solid (18 mg), m.p. 70-71.5°C; UV (CHCl3) λ_{max} (log ε) nm: 325 (3.7); IR (KBr) ν_{max} cm^{-1}: 1620, 1720,; ^{1}H-NMR ($CDCl_3$, 400 MHz) δ: 7.65 (1H, d, *J* = 9.0 Hz H-4), 7.50 (2H, m, H-6, H-7), 7.48 (1H, d, *J* = 8.5 Hz, H-8), 7.20 (1H, d, *J* = 8.5 Hz, H-5), 6.40 (1H, d, *J* = 9.4 Hz H-3) ; HREIMS, *m/z:* 146.0505 (calcd. for $C_9H_6O_2$, 146.0538), ElMS *m/z* (rel. int.): 146 (30), 118 (100), 90 (60), 83 (22), 63 (75), 50 (44). The physical and spectral data were much closed to the reported values [12].

Cinnamic Acid **(5)**

White solid (15 mg), m.p. 132-132.5 °C; UV ($CHCl_3$) λ_{max} (log ε) nm: 325 (3.8), 284 (3.04); ^{1}H-NMR ($CDCl_3$, 400 MHz) δ: 7.50 (1H, d, *J* = 15.8 Hz, H-1′), 7.38 (m), 7.28 (m), 7.20 (m), 6.24 (1H, d, *J* =15.8 Hz, H-2′); HREIMS, *m/z*: 148.6122 (calcd for $C_9H_8O_2$, 148.6158), ElMS *mlz* (rel. int.): 148 $[M]^+$

(31), 117 (45), 107 (100), 105 (36). The physical and spectral data were in closed agreement to the reported values [13].

Lupeol **(6)**

Colorless needles (13 mg), 214-216 °C; $[\alpha]_D^{25}$ +27° (*c* = 0.10, $CHCl_3$); IR (KBr) ν_{max} cm^{-1}: 3450, 3070, 1640 and 885; ^{1}H-NMR ($CDCl_3$, 400 MHz) *δ*: 4.74, 4.61 (2H, br s, 1H each, H-29), 3.21 (1H, dd, *J* = 9.4, 4.5 Hz, H-3), 1.64 (3H, brs, Me-30), 1.04, 0.96, 0.93, 0.84, 0.78 and 0.76 (3H, each s, Me); ^{13}C-NMR ($CDCl_3$, 100 MHz) *δ*: 150.5 (C-20), 109.1 (C-29), 42.8 (C-17), 78.7 (C-3), 55.2 (C-5), 50.3 (C-9), 48.1 (C-18), 47.6 (C-19), 42.7 (C-14), 40.7 (C-8), 39.7 (C-22), 38.5 (C-1), 38.6 (C-4), 38.0 (C-13), 37.0 (C-10), 35.4 (C-16), 34.3 (C-7), 29.7 (C-21), 28.2 (C-23), 27.4 (C-2), 27.2 (C-15), 20.8 (C-11), 19.1 (C-30).18.2 (C-6), 18.0 (C-28), 16.0 (C-25), 15.8 (C-26), 15.3 (C-24), 14.4 (C-27); HREIMS, *m/z:* 426.3830 (calcd for $C_{30}H_{50}O$, 426.3861), ElMS *mlz* (rel. int.): 426 $[M]^+$ (30), 411 (35), 408 (40), 393 (40), 385 (25), 220 (70), 218 (60), 207 (30), 189 (100), 139 (75). The physical and spectral data corresponded to the reported values [14].

Betulin **(7)**

Colorless crystals (11 mg), m.p. 251-252°C; $[\alpha]_D^{25}$ + 20.3 (c = 0.88, CH_3OH); 1R (KBr) ν_{max} cm^{-1}: 3430, 3075, 1630, 889; ^{1}H-NMR ($CDCl_3$, 400 MHz) *δ*: 4.67 (2H, m, H-29), 3.74 (1H, dd, *J* = 10.5, *J* = 4.0 Hz, H-3), 3.80, 3.40 (1H each, d, *J* = 11.5 Hz, H-28), 1.67 (3H, br. s, CH_3-30), 1.00 (3H, s, CH_3-26), 0.97 (3H, s, CH_3-27), 0.91 (3H, s, CH_3-24), 0.88 (3H, s, CH_3-23), 0.86 (3H, s, CH_3-25); ^{13}C-NMR (100 MHz, $CDCl_3$) *δ*: 150.5 (C-20), 109.5 (C-29), 78.8 (C-3), 60.3 (C-28), 55.1 (C-5), 50.3 (C-9), 48.6 (C-18), 47.8 (C-19), 47.7 (C-17), 42.7 (C-14), 40.7 (C-8), 38.6 (C-4), 38.5 (C-1), 37.3 (C-13), 37.1 (C-10), 34.1 (C-7), 33.8 (C-22), 29.7 (C-21), 29.0 (C-16), 28.0 (C-23), 27.5 (C-2), 27.3 (C-15), 25.5 (C-12), 20.7 (C-11), 19.3 (C-30), 18.2 (C-6), 16.0 (C-25), 16.5 (C-26), 15.8 (C-24), 14.6 (C-27); HREIMS, *m/z*: 442.3800 (calcd for $C_{30}H_{50}O_2$; 442.3810), EIMS (rel. int. %) *m/z*: 442 $[M]^+$ (24), 424 (100), 406 (24), 218 (35), 206 (40), 205 (30), 204 (20), 203 (25), 191 (17). The physical and spectral data showed complete agreement with those reported in the literature [15, 16].

3,5,7-trihydroxy-6,4′-dimethoxyflavone (**8)**

Yellow crystals (15 mg), m.p. 320 °C; UV (CH_3OH) λ_{max} (log *ε*) nm: 272 (3.72), 343 (2.33). IR (KBr) ν_{max} cm^{-1}: 3330 (OH), 1645 (C=O) and 1600(C=C); ^{1}H-NMR (C_5D_5N, 400 MHz) *δ*: 8.13 (2H, d, *J* = 8.7 Hz, H-2′ and H-6′), 6.94 (2H, d, *J* = 8.8 Hz, H-3′and H-5′), 6.68 (1H, s, H-8), 3.91 (3H, s, OMe-6) and 3.76 (3H, s, OMe-4). ^{13}C-NMR (C_5D_5N, 100 MHz) δ : 175.8 (C-4), 158.7 (C-2), 157.4 (C-4′), 152.4 (C-5), 151.4 (C-7), 149.1 (C-9), 136.4 (C-3), 131.3 (C-6), 130.2 (C-2 and C-6′), 121.7 (C-1′), 115.3 (C-3′), 115.3 (C-5′), 105.0 (C-10), 91.7 (C-8), 56.6 and 55.3 (MeO-3 and MeO-4′).; HREIMS, *m/z*: 330.0745 (calcd. for $C_{17}H_{14}O_7$, 330.0739), EIMS *m/z* (rel. int.): 330 (15), 315 (100), 286 (20), 245 (35), 217 (36) and 83 (68) 69 (32). The physical and spectral data showed complete agreement with those reported in literature [17-18].

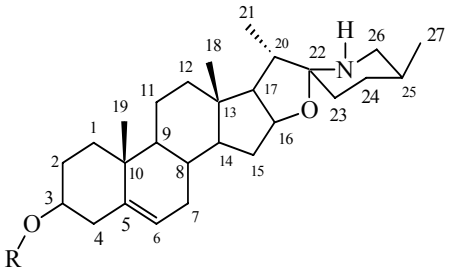
β-sitosterol 3-O-β-D-glucopyranoside (**9**)

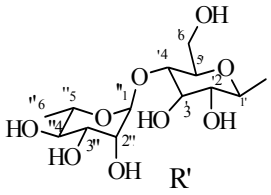
Colorless crystals (12 mg), m.p. 279-280 °C . $[\alpha]_D^{25}$ –15.4 (c = 0.55, CH_3OH); IR (KBr) ν_{max} cm^{-1}: 3450, 3040, 1642, 1610, 1590-1551. ^{1}H-NMR (C_5D_5N, 400 MHz) δ: 5.32 (1H, d, *J* = 7.0 Hz, H-1′), 5.11 (1H, br d, *J* = 5.2 Hz, H-6), 3.84 (1H, m, H-3), 3.81-4.43 (m, Glu-H), 1.00(3H, s, Me-19), 0.90 (3H, d, *J* = 6.0 Hz, Me-21), 0.83 (3H, t, *J* = 7.1 Hz, Me-29), 0.82 (3H, d, *J* = 6.4 Hz, Me-26), 0.80 (3H, d, *J* = 6.4 Hz, Me-27), 0.67 (3H, s, Me-18). HRFABMS, *m/z* 576.4350 (calcd. for $C_{35}H_{60}O_6$, 576.4389), EIMS *m/z* (rel. Int %): 414 $[M\text{-}Glu]^+$ (72), 399 (25), 396 (100), 381 (48), 329 (35), 303 (67), 275 (30), 255 (35). The physical and spectral data showed complete resemblance with the reported values [19].

5,3′,4′-Trihydroxyflavone 7-O-β-D-glucopyranoside **(10)**

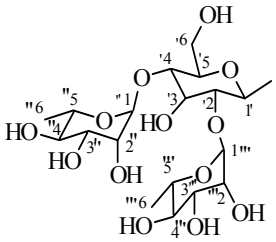
Yellow crystals (11 mg), m.p. 229-231°C; UV ($CHCl_3$) λ_{max} (log *ε*) nm: 255(3.65), 265 (3.61), 360 (4.11) and 305 (4.40). IR (KBr) ν_{max} cm^{-1}: 3300 (OH), 1660 (C=O) and 1590; ^{1}H-NMR (C_5D_5N, 400 MHz) *δ*: 6.81 (1H, d, *J* = 1.4 Hz, H-6), 6.90 (1H, d, *J* = 1.5 Hz, H-8), 6.78 (1H, s, H-3), 7.86 (1H, dd *J* = 7.0, 2.0 Hz, H-6'), 7.23 (1H, d, *J* = 7.1 Hz, H-5'), 7.51 (1H, d, *J* = 2.0 Hz, H-2'), 13.60 (1H, br s, 5-OH), g1ucose moiety *δ*: 4.55 (d, *J* = 7.7 Hz, H-1″), 4.14 (dd, *J* = 9.5, 7.4 Hz, H-2″), 4.32 (d, *J* = 9.4 Hz, H-3″), 4.45 (t, *J* = 9.5 Hz, H-4″), 4.17 (dt, *J* = 9.5, 2.4 Hz, H-5″) and 4.41 (dd, *J* = 10.1, 2.7 Hz, H-6″); ^{13}C-NMR (C_5D_5N, 100 MHz) *δ* : 181.8 (C-4), 166.8 (C-7), 157.7 (C-5), 101.7 (C-9), 162.5 (C-2), 114.6 (C-3), 150.3 (C-3′), 151.8 (C-4′), 147.7 (C-6′), 116.7 (C-1′), 119.3 (C-5′), 122.6 (C-2′), 164.5 (C-10), 100.5 (C 1″), 106.4 (C-6), 79.3 (C-3″), 78.4 (C-5″) , 74.7 (C-2″), 71.0 (C-4″), and 62.2 (C-6″); HREIMS, *m/z:* 448.3745 (calcd for $C_{21}H_{20}O_{11}$, 448.3771), EIMS *m/z*

(rel. int.): 448 (25), 314 (35), 286 (100), 270 (50), 153 (38), 69 (32). The physical and spectral data showed complete agreement with those reported in literature [20].



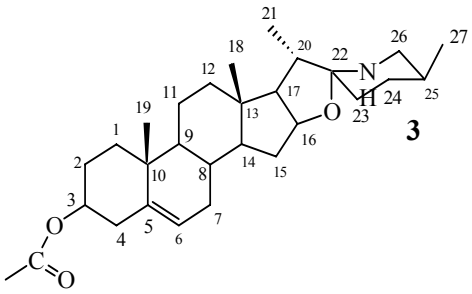


R'



R"

Solamargine **(1)** R' = Rham-Glc-Rham
Khasianine **(2)** R" = Rham-Glc



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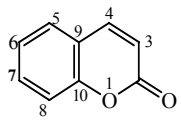

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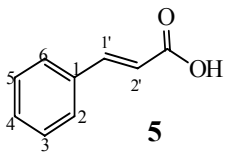

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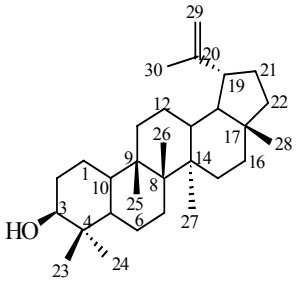

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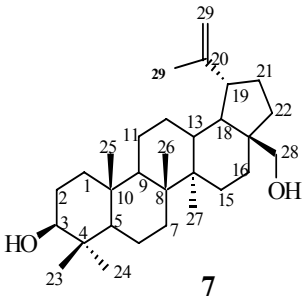

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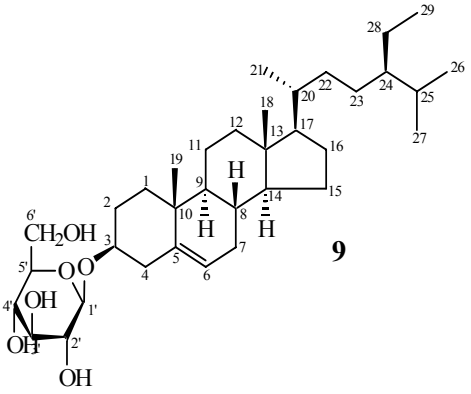

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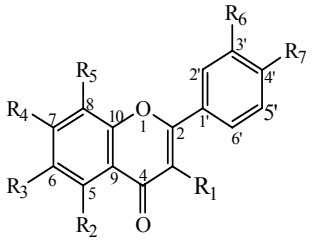

8. R_1, R_2, R_4 = OH, R_3,R_7 = OCH_3

10. R_1, R_3, R_5 = H, R_2,R_6,R_7 = OH, R_4 = 7-*O*-β-*D*-glucopyraniside.

Fig 1: Compounds isolated from *S. surrattense.*

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