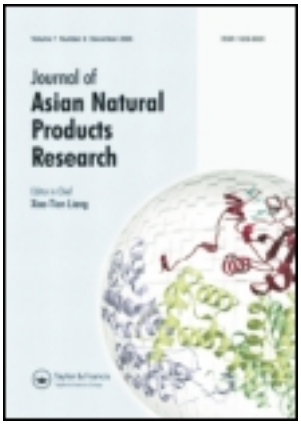

Journal of Asian Natural Products Research

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/ganp20

Phenolic acid glucosides from the seeds of *Entada phaseoloides* Merill

Onkar Singh [a], Mohd Ali [a] & Nida Akhtar [b]

[a] Department of Pharmacognosy and Phytochemistry, Faculty of Pharmacy, Jamia Hamdard (Hamdard University), New Delhi, 110 062, India

[b] Department of Chemistry, Faculty of Science, Jamia Hamdard (Hamdard University), New Delhi, 110 062, India

Available online: 13 Jul 2011

To cite this article: Onkar Singh, Mohd Ali & Nida Akhtar (2011): Phenolic acid glucosides from the seeds of *Entada phaseoloides* Merill, Journal of Asian Natural Products Research, 13:8, 682-687

To link to this article: http://dx.doi.org/10.1080/10286020.2011.584444

Journal of Asian Natural Products Research
Vol. 13, No. 8, August 2011, 682–687

Phenolic acid glucosides from the seeds of *Entada phaseoloides* Merill

Onkar Singh[a], Mohd Ali[a]* and Nida Akhtar[b]

[a]*Department of Pharmacognosy and Phytochemistry, Faculty of Pharmacy, Jamia Hamdard (Hamdard University), New Delhi 110 062, India;* [b]*Department of Chemistry, Faculty of Science, Jamia Hamdard (Hamdard University), New Delhi 110 062, India*

(*Received 21 July 2010; final version received 21 April 2011*)

Phytochemical investigation of the defatted seeds of *Entada phaseoloides* Merill. (Mimosaceae) led to the isolation of three new phenolic acid glucosides, which were characterized as 2-hydroxy-5-methylbenzoyl-β-L-glucopyranoside (*p*-cresotyl glucoside, **1**), 2-hydroxy-5-methylbenzoyl-β-L-glucopyranosyl (2 → 1)-β-L-glucopyranosyl (2 → 1)-β-L-glucopyranoside (*p*-cresotyl triglucoside, **2**), and 2-hydroxybenzoyl-β-L-glucopyranosyl (2 → 1)-β-L-glucopyranosyl (2 → 1)-β-L-glucopyranosyl (2 → 1)-β-L-glucopyranoside (salicylic acid tetraglucoside, **5**), along with sucrose and triglucoside. The structures of these phytoconstituents have been established on the basis of spectral data analysis and chemical reactions.

Keywords: *Entada phaseoloides*; Mimosaceae; seeds; phenolic acid glucosides

1. Introduction

Entada phaseoloides, Merrill; syn. *Entada scandens* Benth and *Entada pursaetha* DC (Mimosaceae), is a gigantic climber with a twisted and angled stem. It is distributed in central and eastern Himalayas up to 1300 m, along the seacoasts of southern India, Andaman Islands, Burma, Sri Lanka, Philippines, and Malacca [1,2]. The seeds possess anthelmintic, narcotic, antiperiodic, emetic, febrifuge, and tonic properties and are used to treat glandular swellings, chest and joint pains, debility, cerebral hemorrhage, as fish poison, and hair wash. The kernel powder with some spices is taken to relieve body pain and cold; roasted seeds are substituted for coffee and as purgative [3]. The seeds contain entadamides [4–7], fatty acids [8], amino acids [9], phenylacetic acid derivatives and entamide A-glucoside [10], phaseoloidin [11], and triterpenoid sapogenins [12]. This paper describes the isolation and characterization of three new benzoyl glycosides along with sucrose [13] and triglucosides [14] from the seeds of *E. phaseoloides* (Figure 1).

2. Results and discussion

Compound **1**, named *p*-cresotyl glucoside, was obtained as a buff white amorphous powder from chloroform–methanol (19:1) eluants. It gave positive test for glycosides and phenols. Its IR spectrum displayed absorption bands for hydroxyl groups (3386, 3250 cm^{-1}), an ester group (1725 cm^{-1}), and an aromatic ring (1640, 1530, 1047 cm^{-1}). The FAB mass analysis of **1** showed a molecular ion peak at *m/z* 314 corresponding to the molecular formula $C_{14}H_{18}O_8$. Its ^{1}H NMR spectrum exhibited two one-proton doublets at δ 6.96 ($J = 8.1$ Hz) and 6.57 ($J = 3.0$ Hz) assigned to *ortho*-coupled H-3 and *meta*-coupled H-6, respectively, and one-proton double doublet at δ 6.60 ($J = 8.1, 3.0$ Hz) attributed to *ortho*-, *meta*-coupled H-4

*Corresponding author. Email: maliphyto@gmail.com

ISSN 1028-6020 print/ISSN 1477-2213 online

DOI: 10.1080/10286020.2011.584444
http://www.informaworld.com

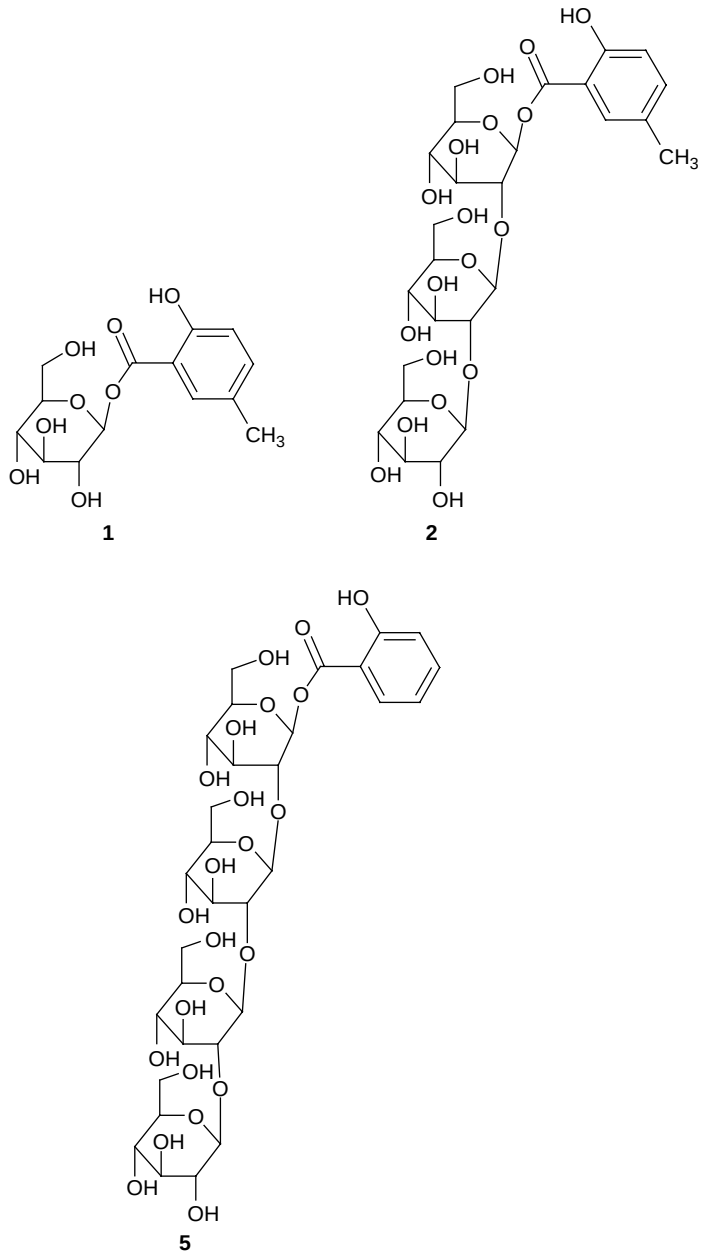

Figure 1. Chemical structures of compounds **1**, **2**, and **5**.

proton. A one-proton doublet at δ 5.04 ($J = 7.1$ Hz) was ascribed to anomeric H-1′ proton. The remaining sugar protons resonated from δ 4.54 to 3.20. A three-proton broad signal at δ 2.30 was accounted to C-8 methyl protons attached to aromatic ring. The ^{13}C NMR spectrum of **1** exhibited signals for ester carbon at δ 173.2 (C-7), aromatic carbons from δ 154.2 to 117.3, anomeric carbon at δ 103.1 (C-1′), other sugar carbons between δ 77.1 and 61.0 and methyl carbon at δ 21.1. The HMBC spectrum of **1** showed correlations of C-7 with H-1′; C-1 with H-6; C-2 with H-3 and

H-4; and C-5 with H_3-8, H-4, and H-6. Acid hydrolysis of **1** yielded D-glucose and *p*-cresolic acid (mp 150°C). On the basis of these evidence, the structure of **1** has been elucidated as 2-hydroxy-5-methyl-benzoyl-β-L-glucopyranoside. This is a new *p*-cresotic acid glucoside.

Compound **2**, named *p*-cresotyl triglucoside, was obtained as a buff white amorphous mass from chloroform–methanol (9:1) eluants. It responded to chemical test of glycosides and phenols and exhibited IR absorption bands for hydroxyl groups (3410, 3350, 3275 cm^{-1}) and an ester group (1725 cm^{-1}). On the basis of FAB–MS and ^{13}C NMR spectra, the molecular weight of **2** was determined at *m/z* 638 consistent with the molecular formula $C_{26}H_{38}O_{18}$. The ^{1}H NMR spectrum of **2** showed two one-proton doublets at δ 6.95 ($J = 8.4$ Hz) and 6.55 ($J = 2.8$ Hz) and a one-proton double doublet at δ 6.59 ($J = 2.8$, 8.4 Hz) assigned to aromatic protons H-3, H-6, and H-4, respectively, three one-proton doublets at δ 5.17 ($J = 7.1$ Hz), 5.06 ($J = 7.0$ Hz), and 4.98 ($J = 7.1$ Hz) due to anomeric H-1′, H-1″, and H-1‴ protons, respectively, and other sugar protons at δ 4.52–3.09. A three-proton broad signal at δ 2.30 was ascribed to C-8 methyl protons linked to the aromatic ring. The ^{13}C NMR spectrum of **2** exhibited signals for ester carbon at δ 173.2 (C-7), aromatic carbons from δ 152.1 to 113.9, sugar carbons from δ 104.0 to 60.4, and methyl carbon at δ 22.1. The appearance of the sugar protons in deshielded region at δ 3.87 (H-2′) and 3.77 (H-2″) in the ^{1}H NMR spectrum and ^{13}C NMR values at δ 82.5 (C-2′) and 76.9 (C-2″) suggested 2 → 1 linkage of the sugar moieties. The HMBC spectrum of **2** exhibited interactions of C-7 with H-6 and H-1′; C-2 with H-3, H-4, and H-6; C-5 with H-4 and H-6; C-2′ with H-1′, H-3′, and H-1″; and C-2″ with H-1″, H-3″, and H-1‴. Acid hydrolysis of **2** yielded *p*-cresotic acid (mp 150°C) and D-glucose (co-TLC comparable). On the basis of spectral data analysis and chemical reactions, the structure of **2** was formulated as 2-hydroxy-5-methyl benzoyl-β-L-glucopyranosyl (2 → 1)-β-L-glucopyranosyl (2 → 1)-β-L-glucopyranoside.

Compound **5**, designated as salicyl tetraglucoside, was obtained as a pale yellow powder from chloroform–methanol (3:1) eluants. It gave positive tests for glycosides and phenols. Its IR spectrum exhibited characteristic absorption bands for hydroxyl (3465, 3364, 3290 cm^{-1}) and ester (1735 cm^{-1}) groups. On the basis of FAB–MS and ^{13}C NMR spectra, the molecular weight of **5** was established at *m/z* 786 consistent to the molecular formula $C_{31}H_{46}O_{23}$. The ^{1}H NMR spectrum of **5** showed two one-proton double doublets at δ 6.98 ($J = 8.9$, 2.7 Hz) and 6.90 ($J = 8.7$, 3.0 Hz) assigned to *ortho*-, *meta*-coupled aromatic H-3 and H-6 protons and two one-proton multiplets at δ 6.60 and 6.49 due to H-4 and H-5 protons, respectively. Four one-proton doublets at δ 5.19 ($J = 7.2$ Hz), 5.01 ($J = 7.1$ Hz), 5.21 ($J = 7.1$ Hz), and 4.90 ($J = 7.0$ Hz) were ascribed to anomeric protons H-1′, H-1″, H-1‴, and H-1⁗, respectively. The other sugar protons resonated from δ 4.65 to 3.11.

The ^{13}C NMR spectrum of **5** exhibited signals for ester carbon at δ 173.0, aromatic carbons at δ 152.2–112.9, four anomeric carbons at δ 104.1 (C-1′), 103.8 (C-1″), 98.6 (C-1‴), and 91.8 (C-1⁗) and other sugar carbons from δ 82.4 to 60.6. The shifting of C-2′, C-2″, and C-2‴ carbons in the deshielded region at δ 82.4, 77.1, and 77.1 suggested the attachment of sugar units through (2 → 1) linkages. The HMBC spectrum of **5** showed interactions of C-7 with H-6 and H-1′; C-1 with H-3 and H-6; C-2 with H-3, H-4, and H-6; C-2′ with H-1′, H-3′, and H-1″; C-1‴ with H-2″; and C-1⁗ with H-2‴ and H-2⁗. Acid hydrolysis of **5** yielded salicylic acid (mp 157–158°C) and L-glucose (TLC comparable). On the basis of the foregoing evidences, the structure of **5** has been determined as 2-hydroxybenzoyl-β-L-glu-

copyranosyl (2 → 1)-β-L-glucopyranosyl (2 → 1)-β-L-glucopyranosyl (2 → 1) β-L-glucopyranoside.

Compounds **3** and **4** have been identified as β-D-fructofuranosyl-α-D-glucopyranoside (sucrose) and β-D-glucopyranosyl (1 → 6)-β-D-glucopyranosyl (1 → 2)-β-D-glucopyranoside by comparing their spectral data with those reported in the literature [13,14].

3. Experimental

3.1 General experimental procedures

Melting points were determined on a Perfit melting point apparatus (Ambala, Haryana, India) and are uncorrected. IR spectra were recorded on KBr discs, using a Bio-Rad FT-IR 5000 spectrometer (FTS 135, Hongkong, China). UV spectra were measured with a Lambda Bio 20 spectrophotometer (Perkin-Elmer, Schwerzenbach, Switzerland) in methanol. ^{1}H and ^{13}C NMR spectra were obtained using Bruker Advance DRY 400 spectrospin and Bruker Advance DRY 100 spectrospin instruments (Karlsruhe, Germany), respectively, in DMSO-d_6 and TMS as an internal standard. FAB–MS spectra were obtained using JEOL-JMS-DX 303 spectrometer (Bruker Daltonics, Billerica, MA, USA). Column chromatography was performed on silica gel 60–120 mesh. TLC was run on silica gel G (Qualigens, Mumbai, India). Spots were visualized by exposure to iodine vapors, UV radiation, and by spraying reagents.

3.2 Plant material

The seeds of *E. phaseoloides* were purchased from Khari Baoli, a local market of Delhi and authenticated by Dr M.P. Sharma, Prof. and Taxonomist, Department of Botany, Jamia Hamdard, New Delhi. A voucher specimen, No. PRL/JH/03/23, is deposited in the herbarium section of the Phytochemical Research Laboratory, Faculty of Pharmacy, Jamia Hamdard, New Delhi.

3.3 Extraction and isolation

Air-dried seeds (2 kg) were coarsely powdered, defatted with petroleum ether, and then exhaustively extracted with ethanol (95%). The combined extracts were then concentrated on a steam bath and dried under reduced pressure to get 150 g (7.5% yield) of viscous dark brown mass. It was dissolved in a small quantity of methanol and adsorbed on silica gel (60–120 mesh) for the preparation of slurry. It was dried in air and chromatographed over silica gel column packed in petroleum ether. The column was eluted with petroleum ether, chloroform, and methanol successively in the order of increasing polarity to isolate compounds **1**–**5**. Elution of the column with $CHCl_3$–MeOH (4:1) and recrystallization in CH_3OH, respectively, afforded colorless amorphous powder of **3** (310 mg, 0.0155%) and white amorphous powder of **4** (275 mg, 0.01375%).

3.3.1 *p-Cresotyl glucoside (**1**)*

Elution of the column with $CHCl_3$–MeOH (19:1) furnished buff white amorphous mass of **1**, recrystallized from acetone–MeOH (1:1), 210 mg (0.0105%); R_f: 0.80 (acetone–chloroform; 8.5:1.5); mp: 196–198°C; UV λ_{max} (MeOH): 292 nm (log ε 5.6); IR ν_{max} (KBr): 3386, 3250, 2920, 2850, 1725, 1640, 1530, 1047 cm^{-1}; ^{1}H NMR (DMSO-d_6): δ 6.96 (1H, d, $J = 8.1$ Hz, H-3), 6.60 (1H, dd, $J = 8.1$, 3.0 Hz, H-4), 6.57 (1H, d, $J = 3.0$ Hz, H-6), 5.04 (1H, d, $J = 7.1$ Hz, H-1′), 4.54 (1H, m, H-5′), 3.71 (1H, dd, $J = 7.1$, 6.9 Hz, H-2′), 3.60 (1H, m, H-3′), 3.54 (1H, m, H-4′), 3.20 (2H, br s, H_2-6′), 2.30 (3H, br s, Me-8); ^{13}C NMR (DMSO-d_6): δ 148.7 (C-1), 154.2 (C-2), 117.6 (C-3), 117.3 (C-4), 140.5 (C-5), 114.2 (C-6), 173.2 (C-7), 21.0 (C-8), 103.0 (C-1′), 76.6 (C-2′), 71.7 (C-3′), 69.9 (C-4′), 77.0 (C-5′), 61.0 (C-6′); + ve FAB–MS *m/z*: 314 $[M]^+$; HR–FAB–MS: *m/z* 314.1002 $[M]^+$ (calcd for $C_{14}H_{18}O_8$, 314.1002).

3.3.2 *p-Cresotyl triglucoside (**2**)*

Elution of the column with $CHCl_3$–MeOH (9:1) gave buff white amorphous mass of **2**, recrystallized from methanol, 450 mg (0.0225%); R_f: 0.55 ($CHCl_3$–MeOH; 8:2); mp: 174–175°C; UV λ_{max} (MeOH): 287 nm (log ε 5.6); IR ν_{max} (KBr): 3410, 3350, 3275, 2921, 2855, 1725, 1640, 1545, 1384, 1044 cm^{-1}; ^1H NMR (DMSO-d_6): Table 1; ^{13}C NMR (DMSO-d_6): Table 1; + ve FAB–MS *m/z* 638 $[M]^+$; HR–FAB–MS: *m/z* 638.2058 $[M]^+$ (calcd for $C_{26}H_{38}O_{18}$, 638.2058).

3.3.3 *Salicyl tetraglucoside (**5**)*

Elution of the column with $CHCl_3$–MeOH (3:1) gave pale yellow powder of **5**, recrystallized from methanol, 1 g (0.05%); R_f: 0.75 ($CHCl_3$–MeOH–NH_3; 8:4:1); mp: 207–209°C; UV λ_{max} (MeOH): 286 nm (log ε 5.1); IR ν_{max} (KBr): 3465, 3364, 3290, 2960, 2845, 1735, 1640, 1558, 1370, 1044 cm^{-1}; ^1H NMR (DMSO-d_6): Table 1; ^{13}C NMR (DMSO-d_6): Table 1; + ve FAB–MS *m/z*: 786 $[M]^+$; HR–FAB–MS: *m/z* 786.2430 $[M]^+$ (calcd for $C_{31}H_{46}O_{23}$, 786.2430).

Table 1. ^1H and ^{13}C NMR spectral data for compounds **2** and **5** (400 and 100 MHz, DMSO-d_6 solvent).

	2		**5**	
Position	δ_H	δ_C	δ_H	δ_C
1	–	148.6	–	148.9
2	–	152.1	–	152.2
3	6.95 d (8.4)	117.5	6.98 dd (8.9, 2.7)	117.4
4	6.59 dd (8.4, 2.8)	113.9	6.60 m	112.9
5	–	126.1	6.49 m	116.2
6	6.55d (2.8)	117.1	6.90 dd (8.7, 3.0)	129.6
7	–	173.2	–	173.0
8	2.30 br s	22.1	–	–
1′	5.17 d (7.1)	104.0	5.19 d (7.2)	104.1
2′	3.87 m	82.5	3.86 m	82.4
3′	3.55 m	72.8	3.70 m	70.2
4′	3.66 m	71.6	3.51 m	68.8
5′	4.52 m	74.2	4.49 m	74.3
6′	3.18 br s	61.1	3.23 br s	61.0
1″	5.06 d (7.0)	103.0	5.01 d (7.1)	103.8
2″	3.77 m	76.9	3.84 m	77.1
3″	3.52 m	72.8	3.70 m	69.7
4″	3.62 m	69.8	3.51 m	68.5
5″	4.40 m	74.7	4.65 m	73.6
6″	3.12 br s	60.8	3.21 br s	61.1
1‴	4.98 d (7.1)	91.7	5.21 d (7.1)	98.6
2‴	3.66 m	74.1	3.86 m	77.1
3‴	3.46 m	72.8	3.50 m	69.4
4‴	3.70 m	69.8	3.41 m	68.6
5‴	4.51 m	74.5	4.50 m	72.9
6‴	3.09 br s	60.4	3.17 br s	60.6
1⁗	–	–	4.90 d (7.0)	91.8
2⁗	–	–	3.73 m	74.2
3⁗	–	–	3.50 m	69.7
4⁗	–	–	3.41 m	66.5
5⁗	–	–	4.65 m	74.3
6⁗	–	–	3.11 br s	61.1

Note: Coupling constants in Hertz are provided in parentheses.

Acknowledgment

The authors are thankful to the Head, Sophisticated Analytical Instrumentation Centre, Central Drug Research Institute, Lucknow, for recording spectral data of the compounds.

References

[1] Anonymous, *The Wealth of India, A Dictionary of Indian Raw Materials and Industrial Products* (NISCAIR, CSIR, New Delhi, 2003), Vol. III, pp. 174–175.
[2] R.N. Chopra, S.L. Nayar, and I.C. Chopra, *Glossary of Indian Medicinal Plants* (CSIR, New Delhi, 1986), p. 55.
[3] K.R. Kirtikar and B.D. Basu, *Indian Medicinal Plants* (Lalit Mohan Basu Prakashan, Allahabad, 2000), Vol. II, pp. 1267–1270.
[4] F. Ikegami, S. Isao, O. Shigeru, R. Nijsiri, and M. Isamu, *Chem. Pharm. Bull.* **31**, 5153 (1985).
[5] F. Ikegami, S. Ohmiya, N. Ruamgrungsi, S.I. Sakai, and I. Murakoshi, *Phytochemistry* **26**, 1525 (1987).
[6] F. Ikegami, T. Sekine, S. Duangteraprecha, N. Matsushita, N. Matsuda, N. Ruangrungsi, and I. Murakoshi, *Phytochemistry* **28**, 881 (1989).
[7] F. Ikegami, T. Sekine, M. Aburada, Y. Fuji, Y. Komatsu, and I. Murakoshi, *Chem. Pharm. Bull.* **37**, 1932 (1989).
[8] D.N. Grindley, E.H.W.J. Burdan, and A.A. Akour, *J. Sci. Food Agric.* **5**, 278 (2006).
[9] V.R. Mohan and K. Janardhanan, *Int. J. Food Sci. Nutr.* **44**, 47 (1993).
[10] J. Dai, L.B.S. Kardono, S. Tsouri, K. Padmawinata, J.M. Pezzuto, and A.D. Kinghom, *Phytochemistry* **30**, 3749 (1991).
[11] A.K. Barua, M. Chakraborty, P.K. Dutta, and S. Ray, *Phytochemistry* **27**, 3259 (1988).
[12] A.K. Barua, *Tetrahedron* **23**, 1499 (1967).
[13] P. Bagri, M. Ali, S. Sultana, and V. Aeri, *Chem. Nat. Compd* **46**, 201 (2010).
[14] K. Ishimaru, M. Osabe, L. Yan, T. Fujioka, K. Mihashi, and N. Tanaka, *Phytochemistry* **62**, 643 (2003).