6. Chaudhuri, R. K., Zymalkowski, F. and Frahm, A. W. (1978) *Tetrahedron* **34**, 1837.
7. Frahm, A. W. and Chaudhuri, R. K. (1979) *Tetrahedron* **35**, 2035.
8. Hambloch, H. and Frahm, A. W. (1980) *Tetrahedron* **36**, 3273.
9. Weigert, F. J., Husar, J. and Roberts, J. D. (1973) *J. Org. Chem.* **38**, 1313.
10. Bohlmann, F., Zeisberg, R. and Klein, E. (1975) *Org. Magn. Resonance* **7**, 426.
11. Breitmaier, E. and Voelter, W. (1978) ^{13}C *NMR Spectroscopy* 2nd edn, p. 222. Verlag Chemie, Weinheim.

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PHENOLICS FROM THE SEEDS OF *ARGEMONE MEXICANA*

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Key Word Index—*Argemone mexicana*; Papaveraceae; 5,7,2',6'-tetrahydroxyflavone; argemexitin; 5,7-dihydroxychromone 7-neohesperidoside.

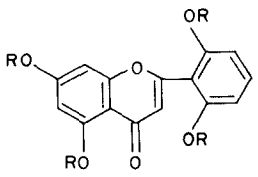
Abstract—Two new phenolic compounds, 5, 7, 2', 6'-tetrahydroxyflavone and 5, 7-dihydroxychromone 7-neohesperidoside have been characterized from the seeds of *Argemone mexicana*.

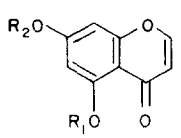
Argemone mexicana (Papaveraceae) a spiny herbaceous annual is reported to possess a number of medicinal properties [1, 2]. Earlier chemical investigations [3–8] of various parts of this plant have revealed a number of alkaloids, fatty acids, amino acids and carbohydrates, but phenolic components [6] have been reported only from the flowers. The present communication reports the isolation and characterization of two new phenolic compounds, 5, 7, 2', 6'-tetrahydroxyflavone (argemexitin) (**1**) and 5, 7-dihydroxychromone 7-neohesperidoside (**2**), from the seeds.

The colour reactions and UV spectral data (λ_{max}^{MeOH} nm: 270, 343) of **1** suggested that it was a flavonoid. Strong IR absorptions at 3350 and 1640 cm^{-1} indicated the presence of –OH and chelated >CO functions, respectively. Its UV spectrum shifted bathochromically on addition of both sodium acetate and aluminium chloride indicating the presence of a free hydroxyl at C-7 and a chelated hydroxyl at the C-5 position, respectively. The solubility of **1** in 10% sodium carbonate also supported the presence of a hydroxyl at C-7. Negative Quastel [9] as well as Gossypetone [10] tests indicated the absence of *ortho*- and *para*-dihydroxy groupings, respectively. Methylation of **1** yielded a tetramethyl ether (**1a**). The ^1H NMR spectrum of **1a** in deuterochloroform showed the presence of four methoxyl functions (two of identical nature) along with the signals for six aromatic protons. The broad singlet at δ 6.6 integrating for two protons was attributed to the aromatic protons at C-3 and C-6 positions whereas the one proton *meta*-coupled doublet at δ 6.86 was considered to be due to a C-8 proton. The remaining two methoxyls in **1a** were therefore located in ring B. The most appropriate positions for these methoxyls appeared to be at the C-2' and C-6' positions because this explained the presence of two identical methoxyl functions and also the presence of two *ortho*-coupled doublets ($J = 9$ Hz) in the aromatic region. Thus the doublet at δ 6.75 integrating for two protons was assigned to the protons at C-3' and C-5' positions whereas the one at δ 7.05, integrating for one proton, to the proton at C-4'. **1a** was therefore identified as 5, 7, 2', 6'-tetramethoxyflavone.

The ^1H NMR spectrum of the acetate (**1b**) of **1**, showing signals for four acetoxyl functions and six aromatic protons, also supported the placements of the four oxygen functions, at the C-5, C-7, C-2' and C-6' positions. The signals attributed to protons at C-6, C-8, C-3' and C-5' were shifted markedly downfield compared to their relative values in **1a**, whereas the signals attributed to the C-3 and C-4' protons remained almost unchanged. The downfield shifts of the signals could be due to the deshielding effect of the acetoxyl functions on the adjacent aromatic protons. Hence **1** was identified as 5, 7, 2', 6'-tetrahydroxyflavone.

The colour reactions and UV spectrum of **2** showed





1 R = H
1a R = Me
1b R = COMe

2 R_1 = H; R_2 = Neohesperidose
2a R_1 = R_2 = H
2b R_1 = R_2 = COMe
2c R_1 = Me; R_2 = H
2d R_1 = COMe; R_2 = Neohesperidose-*O*-acetyl
2e R_1 = H; R_2 = C_7H_7
2f R_1 = Me; R_2 = C_7H_7

it to be a chromone glycoside. Strong IR absorptions at 1620 and 3420 cm^{-1} indicated the presence of chelated >CO and -OH functions. The UV spectrum of **2** (λ_{max}^{MeOH} nm: 253, 285) shifted bathochromically with aluminium chloride showing the presence of a chelated hydroxyl function. On acid hydrolysis **2** yielded an aglycone (**2a**), rhamnose and glucose. Acetylation of the aglycone yielded a diacetate (**2b**). The ^1H NMR spectrum of **2b** had signals for two acetoxyls along with the signals for four aromatic protons; two of which appeared as *ortho*-coupled doublets (J = 7 Hz) at δ 6.1 and 7.65 and two as *meta*-coupled doublets (J = 2 Hz) at δ 6.78 and 7.1. On direct comparison **2a** and **2b** were found to be identical with authentic samples of 5, 7-dihydroxychromone and its acetate, respectively.

Methylation of **2** followed by acid hydrolysis yielded the aglycone **2c** which was found to be soluble in 10% sodium carbonate indicating a free hydroxyl at C-7. **2c** was found to be identical with 5-methoxy-7-hydroxychromone, synthesized using 5,7-dihydroxychromone as the starting material. Thus **2** was identified as 5, 7-dihydroxychromone 7-O-diglycoside.

The mode of attachment of sugar moieties in **2** was determined from the ^1H NMR spectrum of the acetate (**2d**). Appearance of a doublet (J = 6 Hz) at δ 1.22 integrating for three protons and another doublet (J = 2 Hz) at δ 4.8 integrating for one proton, characteristic [11] of rhamnose C-Me and H-1, respectively, showed the sugar unit to be 2-*O*-α-L-rhamnosyl-D-glucose (neohesperidose). Hence **2** was formulated as 5, 7-dihydroxychromone 7-neohesperidoside.

EXPERIMENTAL

Seeds of *Argemone mexicana* L. (12 kg) were collected in May, 1978 in the suburbs of Delhi and identified by Dr. G. S. Paliwal, Reader, Department of Botany, Delhi University. The seeds were air-dried, ground and successively extracted with petrol, C_6H_6, EtOH and aq. EtOH. The solvent-free EtOH extract was separated into glycosidic and non-glycosidic components by extractions with Et_2O and EtOAc. Et_2O and EtOAc extracts containing the same components were combined (fraction 1). The Et_2O and EtOAc insoluble parts of the EtOH extract and the solvent-free aq. EtOH extract, found to contain the same glycosidic components were also combined (fraction 2). Fraction 1 (40 g) was chromatographed over a column of Si gel and eluted with petrol, C_6H_6, EtOAc and MeOH in varying proportions of increasing polarity. Elution of the column with C_6H_6–EtOAc (1:4) and C_6H_6–EtOAc (1:9) yielded **1**. Fraction 2 (30 g) was also subjected to chromatographic separation. Elution of the column with EtOAc–MeOH (9:1) and EtOAc–MeOH (4:1) gave **2**.

1: yellow needles from EtOH, mp > 330°. (Found: C, 62.6; H, 4.0. $C_{15}H_{10}O_6$ requires C, 62.94; H, 3.52%.) It developed a yellow colouration on reduction with Mg–HCl and a green colouration with $FeCl_3$–EtOH; was soluble in 10% Na_2CO_3 but did not give any colouration with ammonium molybdate in HOAc (Quastel test) or with benzoquinone in EtOH (Gossypetone test). IR ν_{max}^{KBr} cm^{-1}: 3350, 2918, 1640, 1600, 1582, 1545, 1450, 1415, 1375, 1330, 1280, 1255, 1190, 1122, 1102, 1070, 1010, 950, 835, 730, 680. UV λ_{max}^{MeOH} nm: 270, 343; +NaOAc: 280, 343; +$AlCl_3$: 278, 415. Methylation of **1** with Me_2SO_4–K_2CO_3 in Me_2CO gave **1a**: colourless needles from EtOAc–petrol, mp 214–215° (Found: C, 66.5; H, 5.0. $C_{19}H_{18}O_6$ requires C, 66.66; H, 5.30%.) ^1H NMR (δ, $CDCl_3$): 3.66 (3H, s, -OMe), 3.85 (6H, *s*, 2×-OMe), 4.09 (3H, *s*, -OMe), 6.6 (2H, *br s*, H-3, H-6), 6.75 (2H, *d*, J = 9 Hz, H-3', H-5'), 6.86 (1H, *d*, J = 2 Hz, H-8), 7.05 (1H, *d*, J = 9 Hz, H-4'). Acetylation of **1** with Ac_2O–pyridine gave the acetate **1b**: colourless needles from $CHCl_3$–petrol, mp 188–190°. (Found: C, 61.2; H, 4.2. $C_{23}H_{18}O_{10}$ requires C, 60.79; H, 3.99%.) ^1H NMR (δ, $CDCl_3$): 2.07 (3H, *s*, -OAc), 2.25 (6H, *s*, 2×-OAc), 2.45 (3H, *s*, -OAc), 6.55 (1H, *s*, H-3), 7.01 (2H, *m*, H-8, H-4'), 7.19 (3H, *m*, H-6, H-3', H-5').

2: colourless needles from MeOH, mp 227–228°. (Found: C, 51.5; H, 5.8. $C_{21}H_{26}O_{13}$ requires C, 51.85; H, 5.35%.) It gave a yellow colouration on reduction with Mg–HCl, a violet colouration with $FeCl_3$–EtOH, a positive Molisch's test for glycosides but a negative Quastel test. IR ν_{max}^{KBr} cm^{-1}: 3420, 1655, 1620, 1570, 1500, 1440, 1420, 1390, 1290, 1240, 1200, 1175, 1060, 980, 845, 800, 780, 680. UV λ_{max}^{MeOH} nm: 253, 285; +NaOAc: 255, 285; +NaOAc + H_3BO_3: 255, 285; +$AlCl_3$: 260, 300, 370; +$AlCl_3$ + HCl: 260, 300, 365. Hydrolysis of **2** with H_2SO_4–MeOH (7%) gave **2c**: colourless needles from EtOAc–petrol, mp 272°, glucose and rhamnose. Acetylation of **2a** with Ac_2O–pyridine gave an acetate **2b**: colourless needles from $CHCl_3$–petrol, mp 113–114°. ^1H NMR (δ, $CDCl_3$): 2.25 (3H, *s*, -OAc), 2.35 (3H, *s*, -OAc), 6.1 (1H, *d*, J = 7 Hz, H-3), 6.78 (1H, *d*, J = 2 Hz, H-8), 7.1 (1H, *d*, J = 2 Hz, H-6), 7.65 (1H, *d*, J = 7 Hz, H-2). Methylation of **2** with Me_2SO_4–K_2CO_3 in dry Me_2CO followed by hydrolysis with H_2SO_4–MeOH (7%) gave **2c**: colourless needles from EtOAc–petrol, mp 215–220°, identical with synthetic 5-methoxy-7-hydroxychromone. Acetylation of **2** with Ac_2O–pyridine gave **2d**: colourless needles from $CHCl_3$–petrol, mp 123–125°. (Found: C, 53.6; H, 5.5. $C_{35}H_{40}O_{20}$ requires C, 53.85; H, 5.13%.) ^1H NMR (δ, $CDCl_3$): 1.22 (3H, *d*, J = 6 Hz, Rha-Me), 2.02–2.10 (18H, 6×-OAc, alcoholic), 2.43 (3H, *s*, -OAc, phenolic), 3.7–3.8 (4H, *m*, sugar protons), 4.8 (1H, *d*, J = 2 Hz, Rha C_1-H), 5.1–5.5 (7H, *m*, sugar protons), 6.24

(1H, *d*, *J* = 7 Hz, H-3), 6.71 (1H, *d*, *J* = 2 Hz, H-8), 7.01 (1H, *d*, *J* = 2 Hz, H-6), 7.84 (1H, *d*, *J* = 7 Hz, H-2).

5-Methoxy-7-hydroxychromone (**2c**). 5,7-Dihydroxychromone (200 mg), $PhCH_2Cl$ (0.13 ml) and K_2CO_3 (300 mg) in dry Me_2CO (200 ml) were refluxed for 8 hr to give **2e**: colourless needles from EtOAc–petrol, mp 141–142°. ^1H NMR (δ, $CDCl_3$): 5.08 (2H, *s*, $-O\underline{CH_2}Ph$), 6.15 (1H, *d*, *J* = 7 Hz, H-3), 6.4 (2H, *s*, H-6, H-8), 7.35 (5H, *m*, $-OCH_2\underline{Ph}$), 7.67 (1H, *d*, *J* = 7 Hz, H-2), 12.64 (1H, *s*, OH-5). Methylation of **2e** gave **2f**: colourless needles from $CHCl_3$–petrol, mp 170–171°. ^1H NMR (δ, $CDCl_3$): 3.9 (3H, *s*, –OMe), 5.07 (2H, *s*, $-O\underline{CH_2}Ph$), 6.14 (1H, *d*, *J* = 7 Hz, H-3), 6.45 (2H, *m*, H-6, H-8), 7.35 (5H, *m*, $-OCH_2\underline{Ph}$), 7.54 (1H, *d*, *J* = 7 Hz, H-2). Debenzylation of **2f** (100 mg) with HCl (0.5 ml) in HOAc (3 ml) afforded 7-hydroxy-5-methoxychromone (**2c**) which crystallized from EtOAc–petrol as colourless needles (50 mg), mp 218–220°.

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REFERENCES

1. Dastur, J. F. (1962) *Medicinal Plants of India and Pakistan* p. 22. D. B. Taraporevala Sons, Bombay.
2. Chopra, R. N., Badhwar, R. L. and Ghosh, S. (1965) *Poisonous Plants of India* p. 143. I.C.A.R., New Delhi.
3. Krishnamachari, V. S. and Ramachandra, B. V. (1954) *J. Sci. Ind. Res. Sect. B* **13**, 815.
4. Misra, P. S., Bhakuni, D. S., Sharma, V. N. and Kaul, K. N. (1961) *J. Sci. Ind. Res. Sect. B* **20**, 186.
5. Misra, P. S., Bhakuni, D. S., Sharma, V. N. and Kaul, K. N. (1961) *Vighyan Pragati* **10**, 215.
6. Krishnamurti, M., Ramanathan, J. D., Seshadri, T. R. and Shankaran, P. R. (1965) *Indian J. Chem.* **3**, 270.
7. Badami, R. C. and Gunstone, F. D. (1962) *J. Sci. Food Agric.* **13**, 258.
8. Santra, D. K. and Saoji, A. N. (1971) *Curr. Sci.* **40**, 548.
9. Quastel, J. H. (1931) *Analyst* **56**, 311.
10. Perkin, A. G. (1913) *J. Chem. Soc.* 650.
11. Mabry, T. J., Markham, K. R. and Thomas, M. B. (1970) *The Systematic Identification of Flavonoids* p. 269. Springer, New York.

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QUERCETIN 3-(6″-CAFFEOYLGALACTOSIDE) FROM *HYDROCOTYLE SIBTHORPIOIDES*

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Key Word Index—*Hydrocotyle sibthorpioides*; Umbelliferae; quercetin 3-galactoside; isorhamnetin; quercetin 3-(6″-caffeoylgalactoside).

Abstract—In addition to quercetin, quercetin 3-galactoside and isorhamnetin, a new caffeoylgalactoside has been isolated from *Hydrocotyle sibthorpioides* and identified by chemical and spectral data as quercetin 3-*O*-β-D-(6″-caffeoylgalactoside).

Hydrocotyle sibthorpioides Lam. (Umbelliferae) is a common weed in Japan. No flavonoid constituents have been reported from this plant but from another species, *H. wilfordi* Maxim. quercetin 3-galactoside has been isolated [1].

Fractionation of the methanolic extract of whole plants of *H. sibthorpioides* with organic solvents followed by repeated CC on Sephadex LH-20 afforded quercetin, methyl caffeate, quercetin 3-galactoside, and its caffeoyl ester (**1**). The ^1H and ^{13}C NMR chemical shift values of **1** suggested that it is composed of quercetin 3-galactoside and caffeic acid. This was confirmed by a mild hydrolysis [2, 3] of **1** with sodium carbonate, which also afforded quercetin 3-galactoside and caffeic acid. The FD-mass spectrum of **1** showed an ion peak indicating the presence of the ester molecule. In the ^{13}C NMR spectrum of **1**

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