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ORIGINAL ARTICLE

Phytochemical investigation of *Calotropis procera*

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Abstract Several sterols were isolated from *Calotropis procera*. Analysis of their spectral data (UV, IR, MS, ^{1}H, ^{13}C NMR experiments) confirmed their structures as urs-19(29)-en-3-yl acetate, β-sitosterol and stigmasterol, multiflorenol, urs-19(29)-en-3-β-ol and 3β,27-dihydroxy-urs-18-en-13,28-olide.

1. Introduction

Calotropis procera Linn., also known as *Alarka*, *Surya*, *Suuryaahvya*, *Vikirna*, *Vasuka*, *Tapana*, *Tuulaphala*, *Kshirparna*, *Arkaparna*, *Aasphota Aakh, Madaar, Ashar* in India (Khare, 2007), belongs to the Asclepiadaceae family and grows in tropical region and most abundant in Bangladesh, India, Burma, Pakistan and in the subHimalayan tract (CSIR, 1992; Kritikar and Basu, 1999). This plant was used first time as a medicinal plant by *Ved Sushruta,* which is about 800–900 AD. *C. procera* is used from very ancient period in folk beliefs as well as a drug of choice for different ailments. Its different formulas are found in old *Vedic* book of India, "*Sushruta Samhita*". In the old days, Hindus used it at the time of worship of sun in the period of *Ved.* Therefore, the name of "*Arkaputra*" or "*Arka prana*" was awarded to this plant, which means such leaf or shining leaf.

Different parts of the plant have been used in Indian traditional system of medicine for the treatment of leprosy, ulcers, tumors, piles and diseases of spleen, liver and abdomen (Kritikar and Basu, 1999).

The water, ethanol, acetone and some other organic solvent extracts of this plant possess insecticidal (Moursy, 1997), larvicidal (Markouk et al., 2000), anti-bacterial and anti-parasitic (Larhsini et al., 1999) activities.

The milky white latex obtained from the plant exhibits potent anti-inflammatory activity in various animal models that is comparable to standard anti-inflammatory drugs (Sangraula et al., 2002). The latex of *C. procera* has also been tested in guinea pigs and found to have a good efficacy in healing of dermal wounds (Rasik et al., 1999). The aqueous extract of the latex has been shown to inhibit cellular infiltration and afford protection against development of neoplastic changes in the transgenic mouse model of hepatocellular carcinoma (Choedon et al., 2006). The chloroform extract of the root has been shown to exhibit protective activity against carbon tetrachloride induced liver damage (Basu et al., 1992). An ethanol extract of the flower is reported to have anti-microbial,

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anti-inflammatory, antipyretic, analgesics (Mascolo et al., 1988), anticancerous (Smit et al., 1995) and antimalarial (Sharma and Sharma, 1999) activities.

The different parts of the plant have been investigated by several workers and found to contain pentacyclic triterpenes (Ansari and Ali, 1999, 2000; Gupta et al., 2003; Khan et al., 1988), alkaloid (Israili and Israr, 1978), cardinolides (Seiber et al., 1982), phytosterols (Khan and Malik, 1989; Lal et al., 1985) and triterpenoid saponins (Gupta et al., 2000).

2. Experimental

2.1. Plant collection

Fresh or dried plant material can be used as a source for the extraction of secondary plant components. Freshly harvested and dried material is more commonly used since old dried material stored for a long period may undergo some qualitative changes. Latex of *C. Procera* was collected from the rural areas of Rewa and Bhopal districts in different month of 2009. These plants were identified by the Botany Department of Bareily College, Bareily. The pooled frozen latex was thawed a fortnight after collection when the serum separated and the solid part of the latex could easily be removed and dried between filter paper sheets.

2.2. Extraction

The dried latex of plant (~5 kg) was soaked in with ethanol at room temperature for two weeks. The filtrate was evaporated under reduced pressure to obtain a dark brown gunmy material (750 g), partitioned between *n*-hexane, chloroform, acetone, ethyl acetate and *n*-butanol, yielding petrol–ether soluble (80 g), chloroform soluble (285 g), ethyl acetate soluble (112 g) and *n*-butanol soluble (159 g) fractions. The petrol–ether soluble fraction was evaporated under vacuum on rotatory evaporator below 50 °C temperature to yield 60 gm of petrol–ether extract. A well-stirred suspension of silica gel (100–150 g in petrol–ether 60–80 °C) was poured into column (150 cm long and 50 mm in diameter). When the absorbent was well settled, the excess of petrol–ether was allowed to pass through column. Slurry was made to petrol–ether extract with 5 g of silica gel in petrol–ether and was digested to well settle column. The column was successively eluted with the solvents and solvent mixtures of increasing polarity. Elution with chloroform/ethyl acetate (3:5) yielded compound **CP-1**. Whereas on elution with ethyl acetone/acetate (1:1) afforded compound **CP-2**. Elution with chloroform/EtOAc (3:2) provided compound **CP-3**. Similarly the acetone soluble fraction was also subjected to column chromatography. The column was successively eluted with the solvents and their mixtures of increasing polarity. Elution with petrol/acetone (8:2) yielded compound **CP-4**. The ethyl acetate soluble thus obtained was successively eluted with the solvents and their mixtures of increasing polarity. Elution with benzene/ethyl acetate (8:2) afforded **CP-5** (Table 1).

2.3. Compound *CP-1*

White powder; m.p.: 198–201 °C; IR ($CHCl_3$) λ_{max}: 1720, 1320, 1635 and 885 cm^{-1} Mass spectra *m/z*: 468 $[M]^+$, 453 $[M-Me]^+$, 408 $[M-AcOH]^+$, 393 $[M-Me-AcOH]^+$, 272 $[M-C_{12}H_{20}O_2]^+$, 257 $[M-C_{13}H_{23}O_2]^+$, 249 $[M-C_{16}H_{27}]^+$, 218 $[M-C_{16}H_{25}O_2]^+$, 189 $[M-C_{16}H_{27}-AcOH]^+$; ^1H NMR (300 MHz, $CDCl_3$): δ 4.52 (1H, dd, $J_{ax,ax}$ = 9.8, $J_{ax,eq}$ = 4.5 Hz, H-3), 2.12 (3H, s, O–C–Me), 1.04 (3H, s, 27-Me) 1.02 (3H, s, 26-Me), 1.01 (3H, s, 23-Me), 0.91 (3H, d, J = 6.6 Hz 30-Me), 0.83(3H, s, 25-Me), 0.82 (6H, s, 24,28-Me), 4.67–4.62 (2H, br s H-29).

2.4. Compound *CP-2*

Colorless needles; R_f (silica gel): 0.38 ($MeOH–CH_2Cl_2$) 1:9; MS *m/z* (rel. int.%): 414 (76), 412 (92), 396 (22), 369 (17), 351 (14), 329 (10), 300 (43), 283 (10), 271 (43), 255 (47), 231 (18), 213 (22), 69 (87), 51 (100); ^1H NMR ($CDCl_3$): δ 5.01 (1H, dd, J = 8.4, 15.2 Hz, H-23), 5.15 (1H, dd, J = 8.4, 15.2 Hz, H-22), 5.35 (2H, d, J = 5.2 Hz, H-6), 3.51 (2H, m, H-3), 1.00 (3H, s, 19-Me), 0.69 (3H, s, 18-Me), 1.03 (3H, d, J = 6.60 Hz, 21-Me), 0.85 (3H, d, J = 6.60 26-Me), 0.81 (3H, d, J = 7.53 Hz, 29-Me), 0.80 (3H, d, J = 6.41 Hz, 27-Me).

2.5. Compound *CP-3*

White crystals; m.p.: 189–191 °C; IR ($CHCl_3$) λ_{max}: 3510 (hydroxyl group), 3045, 1640, 811 (tri substituted double bond) cm^{-1}; MS *m/z* (rel. int.%): 426 $[M+H]^+$, (10), 411 $[C_{30}H_{50}O-CH_3]^+$ (15), 408 $[C_{30}H_{50}O-H_2O]^+$ (25), 393 $[C_{30}H_{50}O-CH_3-H_2O]^+$ (30), 273 [a]$^+$ (42), 259 [b]$^+$ (35), 247 [c (R.D. fragment)]$^+$ (42), 220 [d]$^+$ (50), 205 [e]$^+$ (100).

2.6. Compound *CP-4*

Colorless shining needles; m.p.: 167–168 °C; IR ν_{max} ($CHC1_3$): 3420, 3075, 1645, 890 cm^{-1}; MS *m/z* (rel. int.%): 426 (30) $[M]^+$, 411 (10) $[M-Me]^+$, 408 (16) $[M-H_20]^+$, 393 (8) $[M-Me-H_20]^+$, 272 (10) [ion d]$^+$, 257 (20) [ion c]$^+$, l218 (6) [ion b]$^+$, 207 (100) [ion a]$^+$, 189 (62) [ion a$-H_20$]$^+$.

2.7. Compound *CP-5*

Colorless crystals; m.p.:165–166 °C; IR ν_{max}: 3400 (OH), 2980 (CH), 1760 (five-membered lactone ring) and 1570 (C=C) cm^{-1}; MS *m/z* (rel. int.%): 470 $[M+H]^+$ (29), 452 $[M-H_20]^+$ (I2.0), 263 [$C_{16}H_{23}O_3$; fragment a] (23.6), 249 [$C_{15}H_{21}O_3$; fragment b] (8.8), 235 [$C_{16}H_{23}O$; (fragment a)$-CO$] (18.5), 220 [$C_{15}11_{24}O$; (fragment a)$-CO_2+H$] (13.3), 207 [$C_{14}H_{23}O$; fragment c] (19.5), 205 [$C_{14}H_{21}O$; (fragment b)$-CO_2$] (22.0), 189 [$C_{14}H_{21}$; (fragment c)$-H_2O$] (33.4) and 187 [$C_{14}H_{19}$; (fragment b)$-CO_2-H_2O$] (12.6); ^1H NMR ($CDC1_3$, 300 MHz) 3.18 (H-3, 1H,dd, J = 11.4, 4.8 Hz), 3.55–3.73 (each 1H, d, J = 11.0 Hz, H-27a, 27b), 1.05 (3H, d, J = 7.1 Hz, H-30), 0.99 (3H, s, 23-Me), 0.97 (3H, s, 24-Me), 0.94 (3H, s, 25-Me), 0.88 (3H, s, 29-Me).

3. Results and discussion

3.1. Compound *CP-1*

Compound **CP-1** was isolated as white needle, m.p.: 198–201 °C, on elution with chloroform/ethyl acetate (3:5). Mass spectrum showed $[M]^+$ ion at *m/z* 468 corresponding to molecular formula $C_{32}H_{52}O_2$. In the ^1H NMR of **CP-1** a pair of broad singlets observed at δ 4.67 and 4.62 was indicative of

Table 1

Assignment/(δ_C, ppm)	CP-1	CP-2	CP-3	CP-4	CP-5
C-1	38.51	37.3	38.50	38.49	37.7
C-2	23.73	31.7	27.09	27.00	27.3
C-3	81.89	71.8	77.87	79.05	78.9
C-4	37.86	42.2	37.81	38.80	32.8
C-5	55.50	140.8	55.91	55.47	55.2
C-6	18.27	121.7	33.71	18.42	18.2
C-7	34.09	31.9	122.78	33.95	33.1
C-8	41.00	31.9	141.72	154.37	38.1
C-9	89.78	50.2	56.09	39.40	47.6
C-10	37.55	36.5	38.07	30.99	36.4
C-11	21.55	21.1	35.72	37.85	23.8
C-12	25.67	39.7, 39.8	23.70	25.55	28.1
C-13	39.29	42.3	32.20	39.26	88.1
C-14	42.10	56.8, 56.9	34.92	42.20	46.8
C-15	26.21	24.3, 24.4	43.12	25.95	26.7
C-16	28.69	28.2, 28.9	52.42	28.01	25.7
C-17	37.57	55.9, 56.1	18.21	18.42	48.2
C-18	59.42	11.8, 12.0	29.86	33.95	136.7
C-19	154.56	19.4	37.20	40.95	24.8
C-20	39.43	36.1, 40.5	18.30	48.76	37.1
C-21	31.21	18.9, 21.1	35.01	30.99	30.7
C-22	38.98	33.9, 138.3	12.92	37.85	31.4
C-23	28.00	26.1, 129.3	140.97	28.00	28.1
C-24	16.55	45.8, 51.2	37.10	16.38	15.5
C-25	15.98	29.2, 31.9	21.61	15.69	16.1
C-26	16.35	19.8, 21.2	21.60	16.73	17.1
C-27	25.55	19.0	13.50	24.94	62.5
C-28	28.11	23.1, 25.4	19.36	28.01	178.9
C-29	107.25	12.2	25.59	107.72	19.5
C-30	19.55		14.09	19.00	20.7
CH_3–*C*–O–	170.85				
CH_3–C–O–	21.23				

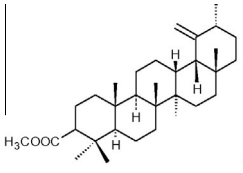
the exomethylene group. The acetoxy group was placed at C-3 on the basis of biogenetic consideration. Furthermore a two proton double doublet observed at δ 4.52 (1H, s, J = 9.8, 4.5 Hz, H-3) was attributed to the proton attached to the carbon bonded to the acetoxy group, showing α- and axial configuration, thus confirming that acetoxy group must be β- and equatorial at the usual C-3 position (Alam and Ali, 2009).

The ^{1}H NMR of **CP-1** revealed signals for six singlet methyl at δ 1.04 (s, H-27), 1.01 (s, H-23), 1.02 (s, H-26), 0.83 (s, H-25) and 0.82 (s, H-24 and H-28), a doublet methyl at δ 0.91 (d, J = 6.6 Hz, H-30), A signal observed at δ 1.12 assignable to the methyl attached to the acetoxy group. The ^{13}C NMR spectrum of this compound showed 32 signals clearly indicating that the compound contains eight methyl, eleven methylene, six methine groups and seven quaternary carbons. On the basis of these spectral data, the compound **CP-1** was identified as urs-19(29)-en-3-yl acetate. All these spectral data were in good concurrence with those reported in the literature for calotropenyl acetate (Ali, 2001).

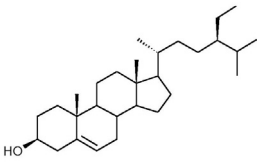
3.2. Compound *CP-2*

Compound **CP-2** was isolated as colorless needles, on elution with ethyl acetone/acetate (1:1). This compound gave purple coloration upon spraying with anisaldehyde reagent. The sample showed a dark green colour with Liebermann–Burchard reagent, which is typical of a steroid. In the ^{1}H NMR spectrum, the signal at δ 5.01 (1H, dd, J = 8.4, 15.2 Hz), 5.15 (1H, dd, J = 8.4, 15.2 Hz) and 5.35 (2H, d, J = 5.2 Hz) could be assigned to H-23 and H-22 of stigmasterol and H-6 of β-sitosterol and stigmasterol, respectively. The integration value for H-6 was twice those of H-22 or H-23. Therefore, it could be deduced that the compound was a 1:1 mixture of β-sitosterol and stigmasterol. The ^{13}C NMR spectrum of this compound displayed 43 signals.

The compound **CP-2** in its EI-MS showed two major molecular ion peaks at m/z 412 and 414. The fragmentation pattern with significant peaks at m/z 396, 369, 351, 329, 300, 273, 271, 231 and 213 was reminiscent of a sterol mixture of stigmasterol and β-sitosterol. The NMR data of this compound were in full agreement with the published valued mixture of β-sitosterol and stigmasterol (Wright et al., 1978).



CP-1



CP-2

3.3. Compound **CP-3**

The compound was isolated as white crystals, m.p.: 189–191 °C, by eluting the column with chloroform/EtOAc (3:2). The molecular ion peak of this compound in mass spectra m/z at 426 corresponds to the molecular formula $C_{30}H_{50}O$. The IR spectrum of the compound showed absorption bands at 3450 (hydroxyl group) and 3045, 1640, 811 (trisubstituted double bond). The mass spectrum of this compound showed characteristic peaks of bauerene and multiflorene type triterpenes having double bond at 7(8) (Budzikiewicz et al., 1963). An eminent fragment ion at m/z 247 arises by retro Diels Alder decomposition of ring; 'c' proceeding the initial rearrangement of C-9 proton (Budzikiewicz et al., 1963). The ^{1}H NMR spectrum provided signals at δ 0.85 (3H), 0.93 (3H), 0.97 (6H), 0.95 (3H), 1.05 (3H), 1.07 (3H) and 1.02 (3H) were assignable for eight tertiary methyl groups. In the ^{1}H NMR a pair of doublets at δ 3.21($J_{ax,ax}$ = 9.7 Hz, $J_{ax,eq}$ = 4.2 Hz) were assigned to α-axial proton germinal to hydroxyl group, one proton doublet observed at δ 5.47 (d, J = 8.9, 4.1 Hz) assignable for olefinic proton. The ^{13}C NMR spectrum exhibited 30 carbon signals for different type carbon atoms. Survey of the literature showed that this compound is similar to multiflorenol (Bhandari and Rastogi, 1984).

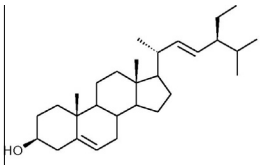
3.4. Compound **CP-4**

The compound was isolated, from acetone extract by eluting column with petrol/acetone (8:2). In its electro spray mass spectrum the molecular ion peak of the compound at m/z 426 $[M+H]^+$ corresponded to the molecular formula $C_{30}H_{50}O$. This compound responded to Liebermann–Burchard test and showed violet coloration with ceric sulphate, suggesting its triterpenic nature. The IR spectrum showed absorption bands at 3420, 3075, 1645 and 890 cm^{-1} assignable for the hydroxyl and a methylene groups, respectively.

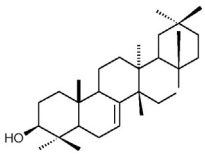
The ^{1}H NMR spectrum showed signals for vinylic proton at δ 4.65–4.70 (2H, br s) and a proton geminal to the hydroxyl group at δ 3.23. In addition there were signals at δ 0.93 (3H, d, J = 6.2 Hz) allocated to one secondary and at δ 1.05 (3H), 1.02 (3H), 0.99 (3H), 0.84 (3H) and 0.82 (6H) six tertiary methyl groups (Rahman, 1986). The mass spectrum of this compound was characteristic of pentacyclic triterpene of the ursane series with saturation in rings A, B, C and D. The major ions 'a' and 'b' were generated by the transfer of hydrogen from C-26 to C-11 accompanied by cleavage of 9–11 and 8–14 bonds with charge retention on either of the resulting fragments (Benn and Gunther, 1983; Karline and Djerassi, 1966; Barton and de Mayo, 1954). Oxidation of **CP-4** with Jones reagent gave calotropenone (**CP-4a**). The latter gave positive Zimmermann test indicating the presence of 3-oxo group (Panosyan et al., 1977). The $NaBH_4$ reduction of **CP-4a** regenerated the parent alcohol **CP-4**, confirming the equatorial orientation of the hydroxyl group in **CP-4**. The configuration of the hydroxyl group in **CP-4** was also indicated by the characteristic downfield shift of the 3-α-H which appeared as double doublets at $J_{ax,ax}$ = 9.8 Hz and $J_{ax,eq}$ = 4.7 Hz owing to coupling with proton at C-2. The catalytic reduction of **CP-4** afforded the corresponding dihydro derivative calotropanol (**CP-4b**) which was identified as 3-hydroxy ursane through comparison of spectral data with these reported in the literature (Domingelez et al., 1974; Hassall and Reyle, 1959). Its ^{1}H NMR spectrum showed the absence of olefinic proton but gave a signal at δ 0.86 (3H, d, J = 6.9 Hz) assignable for an additional secondary methyl group. The reduction of the methylene group was expected to form a methyl group with α- or β-orientation. However, the former was formed in trace amount due to steric crowding of the β-oriented methyl group at C-8. These observations left only one position for the vinylic double bond, i.e., C-19 (Khan et al., 1988). In the ^{13}C NMR spectrum the signals of C-3, C-12, C-13, C-21, C-29 and C-30 could easily be correlated with the corresponding protons in the ^{1}H NMR spectrum. The ^{13}C NMR spectrum of the compound showed signals for 23 carbons atoms, which revealed the presence of seven methyl, 11 methylene and six methine carbon atoms. On the basis of these spectral data the compound was identified as urs-19(29)-en-3-β-ol. Its acetate was been isolated from the same plant source reported in literature (Kojima and Ogura, 1989).

3.5. Compound **CP-5**

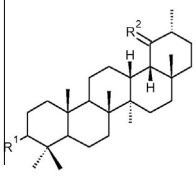
The compound (m.p.: 165–166 °C) was isolated from ethyl acetate soluble fraction by eluting the column with benzene/ethyl acetate (8:2). The mass spectrum exhibited a molecular ion peak at m/z 470 which consistent with the molecular formula $C_{30}H_{46}O_4$. The IR absorption at 1760 cm^{-1} and a carbon signal at δ_C 178.9 suggested the presence of a lactone moiety. The IR spectrum showed absorptions at 1760 cm^{-1} suggesting the presence of a five membered lactone ring and a band at 1620 cm^{-1} for C=C. The molecular formula showed eight double bond equivalents, five of which were accounted for by the pentacyclic ring nucleus, two by the lactone ring and one by a C=C. These features suggested that the remaining two oxygen atoms are present as hydroxyl groups (3400 cm^{-1}). The NMR spectrum showed a double doublet at δ 3.18 (J = 11.4 and 4.8 Hz) and δ_C 78.9 attributable to the proton germinal to the hydroxyl group placed at C-3 on biogenetic consideration (Jacobs and Hoffman, 1928). The coupling constants and the chemical shifts of H-3 favored the β-orientation (equatorial) of the hydroxyl group at C-3 (Siddiqui et al., 1990). The ^{1}H NMR spectrum further showed two AB doublets at δ 3.73 and 3.55 each with a geminal coupling of 11.0 Hz (δ_C 62.5), indicating a methylene carbon linked to an oxygen function. Further, in the ^{1}H NMR spectrum six methyl signals were observed, five as singlets at δ 0.78, 0.94, 0.97, 0.99 and 1.88 and one as a doublet at δ 1.05 (J = 7.1 Hz). These data pointed out that compound **CP-5** belongs to the ursane series of triterpcnes, with two methyls functionalized, one involved in the lactone ring and the other forming a $-CH_2OH$ function. The chemical shift of one of the methyls (δ 1.88) indicated that it is vinylic and presence of only one methyl as a doublet (δ 1.05) along with absence of any vinylic proton led to place a double bond at C-18 (δ_C 136.7). In mass spectra the fragments a–c exemplified that the lactone ring and the remaining hydroxyl group are present in rings C and D. The ^{13}C NMR spectrum also favors the presence of lactone ring between C-13 and C-28 and the hydroxyl group at C-27. Thus on the basis of the above spectral evidences, the structure of the isolated compound CP-5 was finally concluded to be 3β,27-dihydroxy-urs-18-en-13,28-olide, good concurrence with those reported in the literature (Kang, 1987; Numata et al., 1990; Siddiqui et al., 1992; Begum et al., 1993).



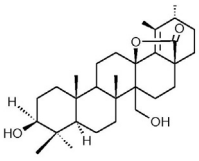
CP-2



CP-3



CP-4: R_1= αH, βOH R_2= CH_2
CP-4a: R_1= O R_2= CH_2
CP-4b: R_1= αH, βOH R_2= αH, βMe



CP-5

4. Conclusion

We believe this is the first report describing the isolation of 3β,27-dihydroxy-urs-18-en-13,28-olide from *C. procera*.

References

Alam, P., Ali, M., 2009. Indian J. Chem. 48B, 443–446.
Ali, M., 2001. Techniques in terpenoidal identification. Birla Publishers, Delhi, p. 407.
Ansari, S.H., Ali, M., 1999. Indian J. Med. Arom. Plant Sci. 21, 978.
Ansari, S.H., Ali, M., 2000. Indian J Chem 39 (B), 287.
Barton, D.H.R., de Mayo, P., 1954. J. Chem. Soc., 887.
Basu, A., Sen, T., Ray, R.N., Nag Chaudhuri, A.K., 1992. Fitoterapia 63, 507–514.
Begum, S., Adil, Q., Siddiqui, B.S., Siddiqui, S., 1993. J. Nat. Prod. 56, 613.
Benn, R., Gunther, H., 1983. Angew. Chem. Int. Ed. Engl. 22, 350.
Bhandari, P., Rastogi, R.P., 1984. Phytochemistry 23 (9), 2085.
Budzikiewicz, H., Wilson, J.M., Djerassi, C., 1963. J. Am. Chem. Soc. 85, 3688.
Choedon, T., Mathan, G., Arya, S., Kumar, V.L., Kumar, V., 2006. World J. Gastroenterol. 12, 2517–2522.
CSIR, 1992. The Wealth of India, Raw Materials, vol. 1. Publications and Information Directorate, CSIR, New Delhi, 1992, p. 78.
Domingelez, A., Gonzales, J.A., Quintanilla, Paulino Rojas, M., 1974. Phytochemistry 13 (3), 673.
Gupta, A., Siddiqui, I.R., Singh, J., 2000. Indian J. Chem. 39 (B), 941.
Gupta, A., Singh, R., Purwar, C., Chauhan, D., Singh, J., 2003. Indian J. Chem. 42 (B), 2030.
Hassall, K., Reyle, K., 1959. J. Chem. Soc. 85.
Israili A, A.H., Israr, R.K., 1978. J. Res. Indian Med. Yoga Homeo. 13 (3), 120.
Jacobs, W.A., Hoffman, A., 1928. J. Biol. Chem. 79, 519.
Kang, S.S., 1987. Saengyak Hakhoechi 18, 151, CA: 108, 132,061 (1988).
Karline, J., Djerassi, C., 1966. J. Org. Chem. 31, 1945.
Khan, A.Q., Malik, A., 1989. Photochemistry 28 (10), 2859.
Khan, A.Q., Ahmad, Z., Kazmi S N, H., Malik, A., 1988. J Nat. Prod. 51 (5), 925.
Khan, A.Q., Ahmad, Z., Kazmi, S.N.H., Malik, A., 1988. J. Nat. Prod. 51, 925.
Khare, C.P. (Ed.), 2007. Indian Medicinal Plants, an Illustrated Dictionary. Springer Science, Springer Verlag, Berlin/Heidelberg, Germany, pp. 113–114.
Kojima, H., Ogura, H., 1989. Phytochemistry 28, 1703.
Kritikar, K.R., Basu, B.D., 1999, 2nd ed.. In: Indian Medicinal Plants, vol. 3 International Book Distributors, Dehradun, India, p. 1610.
Lal, S.D., Kumar, P., Pannu, D.S., 1985. J. Sci. Res. 7, 141.
Larhsini, M., Oumoulid, L., Lazrek, H.B., Wataleb, S., Bousaid, M., Markouk, M., Jana, M., 1999. Therapia 54 (6), 763.
Markouk, M., Bekkouche, K., Larhsini, M., Bousaid, M., Lazrek, H.M., Jana, M., 2000. J. Ethnopharmacol. 73 (1-2), 293.
Mascolo, N., Sharma, R., Jain, S.C., 1988. J. Ethnopharmacol. 22 (2), 211.
Moursy, L.E., 1997. J. Egypt. Soc. Parasitol. 2, 505.
Numata, A., Takahashi, C., Miyamoto, T., Yoneda, M., Yang, P., 1990. Chem. Pharm. Bull. 38, 942.
Panosyan, A.G., Mnatsa, V.A., Kanyan, 1977. Khim. Prir. Soedin. 1, 59.
Rahman, A., 1986. Nuclear Magnetic Resonance. Springer Verlag, New York, p. 262.
Rasik, A.M., Raghubir, R., Gupta, A., Shukla, A., Dubey, M.P., Srivastava, S., Jain, H.K., Kulshresth, D.K., 1999. J. Ellrnopharmacol 6 (8), 261–266.
Sangraula, H., Dewan, S., Kumar, V.L., 2002. Inflammopharmacol 9, 257–264.
Seiber, J.N., Nelson, C.J., Lee M, S., 1982. Phytochemistry 21, 2343.
Sharma, P., Sharma, J.D., 1999. J. Ethnopharmacol. 68 (1–3), 83.
Siddiqui, S., Siddiqui, B.S., Naeed, A., Begum, S., 1990. J. Nat. Prod. 53, 1332.
Siddiqui, S., Siddiqui, B.S., Naeed, A., Begum, S., 1992. Phytochemistry 31, 4279.
Smit, H.F., Woerdenbag, H.J., Singh, R.H., Meulenbeld, G.J., Labadie, R.P., Zwaving, J.H., 1995. J. Ethnopharmacol. 47 (2), 75.
Wright, J.L.C., Mclnnes, A.G., Shimizu, S.D., Smith, G., Walter, J.A., 1978. Can. J. Chem. 56, 1898–1903.