

**PHYTOCHEMICAL EXAMINATION OF *CORCHORUS OLITORIUS*  
(*TILIACEAE*) CAPSULES**

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**ABSTRACT**

From the capsule extract of *Corchorus olitorius* L, stigmasterol,  $\beta$ -sitosterol, betulinic acid, kaempferol, quercetin, and corchoroside-A were isolated and characterized by IR, HNMR spectroscopy.

**Key words:** *Corchorus olitorius* Capsules,  $\beta$ -sitosterol, Lupeol, Betulin, 2-Methyl anthraquinone, Scopoletin and Corchoroside-A

**INTRODUCTION:**

*Corchorus olitorius* is a *Tiliaceae* member is an erect to procurement of annuval herb grow up to 20 cm long . Capsules are 1.5-2.7 cm long. Several important bioactive molecules were reported which includes cardiac glycosides, their aglycones and polysaccharides, triterpenoids, phenolics, sterols and fatty acids [1-96]. Biologically *Corchorus* species are used as diuretic, chronic cystitis, gonorrhoea and dysuria antihistaminic, anti-inflammatory, and

antimicrobial, cardiotoxic, and also to increase the viscosity of the seminal fluid [97-98].

**MATERIALS AND METHODS:**

**Plant material collection:** The plant material, *Corchorus olitorius* capsules was collected from Warangal in September 2007(2kg). The plant was authenticated by Prof. V.S. Raju , Department of Botany, Kakatiya University, Warangal. A specimen was deposited in the herbarium (Voucher specimen number (COC/07)

capsules were collected from the plant and dried under shade.

#### Extraction of Isolated Compounds:

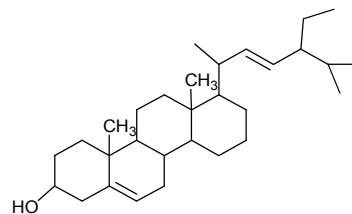
The capsules (2kg) of *Corchorus olitorius* were air dried and coarsely powdered in a Wiley mill and successively extracted with petroleum ether (3×3 l), chloroform (3×3 l) and methanol (3×3 l) and concentrated under reduced pressure. The petroleum ether, chloroform extracts of *Corchorus olitorius* capsules shown similar spots on TLC (1:1 Benzene : Chloroform) and hence combined and column chromatographed over silica gel (Acme 100 mesh), which afforded three compounds designated as COC-1, COC-2, and COC-3. The methanolic extracts showed positive tests for terpenoids and cardiac glycosides. On column chromatography the methanolic extract gave three compounds COC-4, COC-5, and COC-6.

#### Characterization of Compounds:

##### COC-01(Stigmasterol):

It was obtained in petroleum ether:benzene (90;10) fraction of the column and crystallized from benzene, hexane mixture has colorless feathery needles of m.p. 136-138<sup>0</sup>C. It showed positive result to L.B.test for sterols. Fractions were collected and monitored

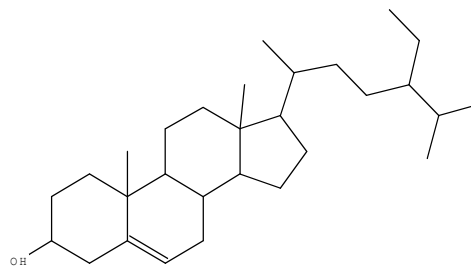
over Silica gel TLC. Based on TLC studies the fractions 32-54 were found to be similar.



##### Stigmasterol

##### COC-02 ( $\beta$ -sitosterol):

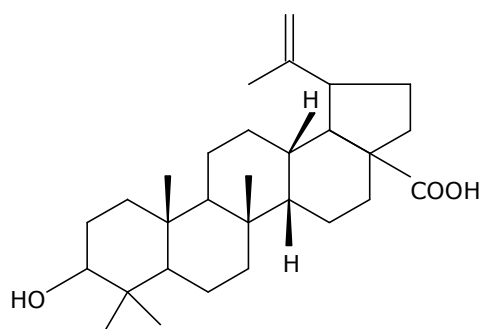
It was obtained in petroleum ether:benzene(70:30) fraction of the column and It was crystallized from petroleum ether as colourless needles of m.p. 134-136<sup>0</sup>C. It showed positive result to L.B.test for sterols. Fractions were collected and monitored over Silica gel TLC. Based on TLC studies the fractions 70-111 were found to be similar.



##### $\beta$ -sitosterol

**COC-03 (Betulinic Acid):** It was obtained in petroleum ether:benzene(30:70) fraction of the column and it was crystallized from

petroleum ether as colourless needles of m.p. 134-136<sup>0</sup>C. It showed positive result to L.B.test for sterols. Fractions were collected and monitored over Silica gel TLC. Based on TLC studies the fractions 210-221 were found to be similar and hence combined as crystallized from chloroform-methanol as shining silky needles, melting point 294 - 296<sup>0</sup>C, ( $\alpha$ )<sup>30</sup><sub>D</sub>+8.6<sup>0</sup>C (chloroform) and analyzed for the formula C<sub>30</sub>H<sub>48</sub>O<sub>3</sub>. It gave pink color in L.B test suggesting that compound is a triterpenoid. IR showed absorption at 3460 (-OH) 1382, 1690 (carbonyl of COOH) and at 1380 and 1390 cm<sup>-1</sup> (gem dimethyl), 1640 (double bond).It formed monoacetate, melting point 287 - 290<sup>0</sup>C, ( $\alpha$ )<sup>30</sup><sub>D</sub> +12.2<sup>0</sup>C (chloroform) with AC<sub>2</sub>O/PY, a mono ester with diazomethane, melting point 220 - 221<sup>0</sup>C, ( $\alpha$ )<sup>30</sup><sub>D</sub>+8.5<sup>0</sup>C (chloroform), a methyl ester acetate with AC<sub>2</sub>O/PY, melting point 195 - 198<sup>0</sup>C, ( $\alpha$ )<sup>30</sup><sub>D</sub>+14.2<sup>0</sup>C (chloroform).



**Betulinic Acid**

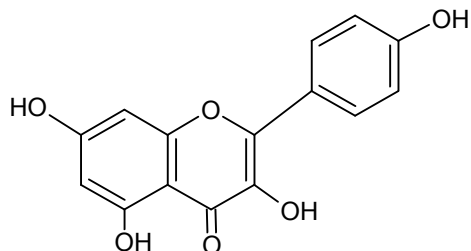
#### **COC-04: (Kaempferol):**

It was obtained in methanol:chloroform(1:97) fraction of the column and crystallized from methanol as yellow needles, m.p. 279-280<sup>0</sup> and analyzed for the formula C<sub>15</sub>H<sub>10</sub>O<sub>6</sub>. In U.V light, it showed a single yellow spot and on exposing to ammonia it turned to bright yellow. It gave positive colour reactions characteristic for flavonols. An orange red precipitate with neutral lead acetate and an yellow colour with Wilson's citric -boric acid reagent confirmed the presence of a free 3-hydroxyl and 5- hydroxyl groupings respectively.

It formed a tetra acetate, m.p. 186-188<sup>0</sup> and a tetra methyl ether, m.p. 163-164<sup>0</sup>. The ultra violet spectrum in methanol had absorptions at  $\lambda_{max}^{MeOH}$  253sh, 265, 294sh, 322sh, 365nm. Sodium acetate gave 10 nm bathochromic shift in Band II indicating the presence of a free 7- hydroxyl. With aluminium chloride / HCl, it formed a complex and showed a shift of 55 nm in Band I which further confirmed the presence of 3-OH group. NaOAC / H<sub>3</sub>BO<sub>3</sub> reagent did not give any pronounced shift which suggested the absence of a free ortho-dihydroxy system.

From the above properties compound COC- 04 was identified as Kaempferol

and the identity was confirmed by comparison with authentic sample (mmp and co-PC).

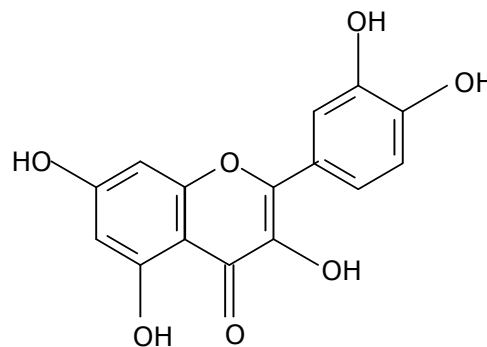


### Kaempferol

#### COC-05 (Quercetin):

It was obtained in methanol:chloroform(3:97)fraction of the column and crystallized from methanol as yellow crystalline solid, melting point 312-314°C. It showed positive result to Shinoda test for flavonoids. Fractions were collected and monitored over Silica gel TLC. Based on TLC studies the fractions 351-365 were found to be similar. The compound was obtained from methanol-chloroform mixture and was crystallized from methanol as yellow crystalline solid, melting point 312-314°C and analyzed for the formula  $C_{15}H_{10}O_7$ . On paper chromatography it was yellow and intense yellow under U.V/ $NH_3$ , with ferric chloride it gave green colour, characteristic for flavonoids, an orange red precipitate, with neutral lead acetate indicating the presence of free 3-hydroxyl group. U.V spectrum in ethanol shows

absorptions at  $\lambda_{\max}^{EtOH}$  255, 267sh, 301sh, 374 nm. Gave 10nm bathochromic shift in band-II indicating the presence of a free 7-hydroxy group, with aluminum chloride, it formed a complex and showed a shift of 55nm band-I further confirmed the presence of free 3-hydroxyl group. Sodium acetate, boric acid reagent showed a bathochromic shift of 20nm indicating the presence of free 3', 4' - di hydroxyl groups (ortho dihydroxy system). <sup>1</sup>HNMR exhibits peaks at  $\delta$  7.60 (d, 6'H) and  $\delta$  7.75 (d, 2'H). From the above properties COC-5 was identified as quercetin and its identity was confirmed by comparison with authentic sample (m.m.p and Co - T.L.C).



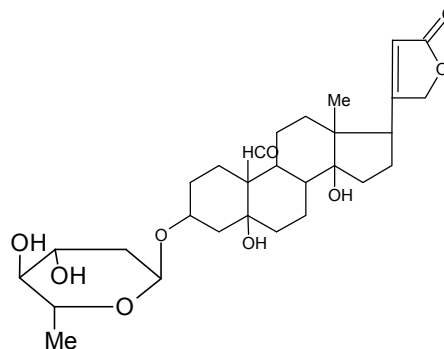
### Quercetin

#### COC-06 (Corchoroside A):

It was obtained in methanol in chloroform (7:93) fraction of the column and it was crystallized from methanol-ether as colour less prisms. It showed positive result to Shinoda test for flavonoids. Fractions were

collected and monitored over Silica gel TLC. Based on TLC studies the fractions 480-503 were found to be similar. It crystallized from methanol-ether as colourless prisms, with one molecule of water, m.p. 164-168°C;  $[\alpha]_D^{20} = +19.7^\circ$  (methanol), it showed positive Kedde and Legal reactions indicating the cardenolide nature of the compound. The U.V. spectrum (ethanol) showed two maxima at 218 nm and 298 nm confirming the presence of  $\alpha, \beta$  unsaturated  $\gamma$ -lactone group and a carbonyl group. It analyzed for the formula ( $C_{29}H_{42}O_9$ ) and formed a diacetate. Keller-Killini reaction was positive suggesting the presence of a 2-deoxy sugar. Mild acid hydrolysis according to the method of Rangaswami and Reichstein gave an aglycone which crystallized from acetone-ether or methanol-ether with half molecule of water as colourless prisms, m.p. it showed U.V. absorption maxima (in ethanol) at 218 and 300 nm mixed m.p with authentic strophanthidin was undepressed. All the properties described

above for the aglycone and its derivatives agreed well with those reported for strophanthidin and its derivatives.



### Corchoroside A

### RESULTS AND DISCUSSION:

The chemical Examination of the capsules of *Corchorus olitorius* on conventional extraction and a sequence of chromatographic methods afforded six compounds. These are characterized stigmasterol,  $\beta$ -sitosterol, betulinic acid, kaempferol, quercetin, and corchoroside-A. Out of these compounds stigmasterol, kaempferol and quercetin were reported first time from *Corchorus olitorius* capsules.

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