

**PHYTOCHEMICAL INVESTIGATION AND MEDICINAL IMPORTANCE OF  
*CYATHOCLINE LYRATA***

Joshi Amit\*, Baghel Vijay, Pathak A.K., Tailang M.  
Department of Pharmacy, Barkatullah University, Bhopal (M.P.) India

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**ABSTRACT**

In this study of new sesquiterpene alcohol, Cythoclol has been isolated from the essential oil of both *Cyathocline purpurea* and *C. lyrata*. Two new sesquiterpene lactones, Eudesmanolide and Guianolide have also been isolated from *C.purpurea*. Chemical investigations were done to determine its molecular formula and structure. The essential oil has been biologically screened and was found to possess antimicrobial and antifungal activity.

**KEYWORDS:** *C.lyrata*, sesquiterpene alcohol, Cythoclol

**\*Correspondence**

Amit Joshi

Lecturer

Department of Pharmacy, Barkatullah University, Bhopal

E-mail:- amit\_joshi\_4580@yahoo.co.in

Mob.no.- +91-9425094311

## INTRODUCTION

*C. lyrata* (Don) Kuntze belongs to the family Compositae. Its vernacular name is Phultar and Mukki. It is commonly found in Balaghat area. Generally, the tribal people use its roots for the treatment of stomach pain. Essential oils are volatile chemical products formed by odoriferous substances isolated from a great variety of plants. These are water insoluble but highly soluble in alcohol, vegetable and mineral oils and ether. They are categorized according to their main volatile components.<sup>1,2,3</sup> *C. lyrata* is indigenously found near Lonavala in Maharashtra<sup>4</sup>. Aerial parts of the plant *C. lyrata* are used for the isolation of essential oil and alcohol. Biological screen includes tests for antibacterial, antiprotozoal, antiviral, antifungal, anti fertility and pharmacological activities<sup>5</sup>. Variance in the proportion of the volatile components within different species of the same genus attributes to different toxic and medicinal properties.<sup>6</sup>

## CHEMICAL PROFILE

Only one species of the genus *Cyathocline* had been chemically investigated and identified for the presence of volatile components. Recent research has led to the isolation of two new sesquiterpene lactones- Eudesmanolide, Isoivangustin and the guaianolide – 6 $\alpha$ - hydroxy- 4,10- guaianadien- 8 $\alpha$  , 12-olide from *C. lyrata*. Signals of Alantolactone , Ivangustin and lactone which was epimeric at C-8 were found to be analogous to the H NMR spectral data of Isoivangustin and its hydrogenated product. This corroborated the presence of Eudesmanolides. Additionally, H NMR spectral data of hydrogenated product, Alantolactone were similar to those corresponding 1- deoxy compound . The presence of 1 $\beta$  - hydroxyl group was established by spin decoupling. A critical comparison of the H NMR spectral data with those of ivangustin revealed that , which has an olefinic bond at C-3, different from the latter in the position of the double bond isomer of ivangustin. The second lactone was isolated from the complex mixture as its acetate. The H NMR spectral data of the latter showed that it was guaianolide. Careful decoupling, allowed the assignment of all signals which were similar to those of Chrysostamalide 5 , although some signals were typically different. The coupling of J7,8 and J8,9 as well as the chemical shifts of H-6 and H-9 $\beta$  which suggested the presence of a 8,12-cis lactone ring system. The negative cotton effect supported this assumption. This isolation is interesting as the compounds of this type have not been isolated so far.

The non-phenolic portion of the essential oil of *C.lyrata* has yielded a new sesquiterpene alcohol (50%). It exists both in the free form (57%) and in the form of acetic ester (43%). It is found to have b.p. 130-132<sup>0</sup>C/ 10mm, specific gravity of 0.9611 at 27<sup>0</sup>C, n<sub>D</sub><sup>27</sup> 1.4950, optical rotation- racemic 0<sup>0</sup>C (in chloroform) and molecular refraction of 67.3. Thus, it is supposed to be a bicyclic sesquiterpene alcohol. It contains one double bond adjacent to one of the two menthyl groups. Presence of one double bond was confirmed by titration with perbenzoic acid and its position was determined by Ozonolysis of alcohol. On Ozonolysis, alcohol gave non-volatile products which gave colour reaction of methyl ketone and showed the presence of aldehyde group. Molecular formula was found to be C<sub>15</sub>H<sub>26</sub>O; C: 80.98%, H: 11.78%, requires C:81.08% and H: 11.71%. Cythocin gave an acetate on acetylation; this and the subsequent saponification of the acetyl derivative showed the presence of one hydroxyl group; Specific gravity: 0.9719 at 32<sup>0</sup>C and n<sub>D</sub><sup>32</sup> of 1.4880. It neither reacted with phenyl urethane nor with phthalic anhydride at 110<sup>0</sup>C suggesting its tertiary nature. Cythocin benzoate has a m.p. of 114-115<sup>0</sup>C and is obtained by refluxing alcohol dissolved in pyridine with Benzoyl chloride for 1hr. Treatment with ice cold dilute hydrochloric acid yielded solid benzoate. It was washed with dilute sodium hydroxide and water; on recrystallisation with ethanol, it gave pale yellow needles. Dehydrogenation with Sulphur; Dehydrogenated product was isolated by distillation. Redistillation over Sodium yielded picrate which on recrystallisation with alcohol gave Melting point and mixed m.p. of 115<sup>0</sup>C. Ozonolysis: Presence of aldehyde group was confirmed as the alcohol reduced Tollen's reagent and Fehling's solution. Also, methyl ketone group was confirmed as it gave a positive iodoform test.

## PHARMACOLOGICAL PROPERTIES

Aerial part of the plant has prominent anti-microbial action. Essential oil obtained by hydro-distillation gave an yield of 0.28%. Antibacterial and anti-fungal activities, were tested by screening the oil in pure state and at the dilutions of 1:10, 1:100 and 1:1000 in ethylene glycol. Oxoid nutrient broth for culturing bacteria and Sabouraud's dextrose media for cultivating fungi. Sterile paper discs of 6mm and Whatman No.1 paper were moistened in oil and different dilutions, placed on seeded Petri-dishes and incubated at 37°C±0.5°C for 24 hrs-antibacterial and at 28°C for 72hrs-antifungal activity.

Activity was calculated by measuring the zones of inhibition. Experiments repeated in triplicate. Penicillin G and Streptomycin were taken as standards for antibacterial and Hamycin for comparing antimicrobial activity. Antimicrobial activity was seen at very high dilutions as well (1% ethylene glycol). Gram positive bacteria were found to be sensitive and the growth of a Gram negative bacteria-*P. aeruginosa* was checked. Also, oil exhibits antifungal activity against the strains of *Cryptococcus neoformans*, *Helminthosporium rostratum*, *Fusarium oxysporium*.<sup>8</sup>

Antihelminthic activity against Earthworms and Tapeworms: Earth worms (*Pheritima posthuma*) were obtained from intestines of sheep, goats and pigs. Tapeworm-*Taenia solium*. An emulsion of Essential oil in Tween -20 (1%) was prepared and diluted to 0.5%, 1% and 2%. In addition to this Piperazine phosphate dilutions of 0.5%, 1% and 2%. were made in water. 2 ml of essential and Piperazine phosphate solution were diluted to 10 ml separately with saline and poured into petridishes. Six earthworms and tapeworms were placed in two different sets and checked for death and paralysis in accordance with Gaind *et al.*<sup>9</sup> The plant also found to be active against Hookworms (*Bunostomum trigonocephalum*) and Nodularworms (*Oesophagostomum columbianum*). Ringer's solution was used in place of Normal saline. The activity was compared against hexyl resorcinol.<sup>10</sup>

## FUTURE PROSPECTIVES

The varied Sesquiterpene lactones (SLs) in medicinal plants have been found to possess anti-helminthic, anti-inflammatory antioxidant activity and antibacterial activity. Eidesmanolide type of Sesquiterpene lactones are potent non-antioxidant inhibitors of NF-Kappa B. Plants with these compounds show anti-inflammatory activity by giving a delay in onset of capillary reactions of the allantois membrane in a physiological model. Variants of SLs have powerful antioxidant properties too that can neutralize harmful free radicals. Alantolactone is supposed to be responsible for anti-cancer activity. Structural activity of SLs is responsible for in-vitro cytotoxicity against the tumour cells.<sup>11</sup> Of the series, Guaianolides are purported to possess antifeedant, anti-tumoral, anti-viral and growth regulating property.<sup>12</sup> Consequently, SLs of the *Cyathocline* sp. could be analysed for anti-inflammatory activity and utilized for treating pathological states like Arthritis. Its Alantolactone constituent could be checked for antitumour activity due to its potent antioxidant property. Apart from this sesquiterpene alcohol have antifeeding property against larva.<sup>13</sup> These long chain alcohols have also shown antibacterial activity against *Streptococcus mutans*. Thus, "Cyathoclo" could be screened for insect-repelling properties and be used to treat dental problems like plaques, caries and periodontal gingivitis caused by bacterial action<sup>14</sup>.

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