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Research Paper

COMPARATIVE STUDY OF LYCORINE IN TWO MEDICINAL SPECIES OF *Crinum* L.

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Abstract

Plants are treasure house of many biomolecules useful to humankind. Species of *Crinum* L. are also like that. Amongst a host of such medicinally important biomolecules being available in them lycorine has been found to have the merit of curing many a malady. The anti-nociceptive, anti-inflammatory, hepatoprotective, hypotensive properties are remarkable for this biomolecule. The present study has displayed the difference in the amount of occurrence of lycorine in two congeneric species *C. asiaticum* L. and *C. latifolium* L., growing in Medinipur. It also shows the species producing greater amount of the chemical per plant, in consideration of their biomass.

Key words: *Crinum L., Crinum asiaticum, Crinum latifolium, Lycorine. HPLC*.

INTRODUCTION

The genus *Crinum* belongs to the family *Amaryllidaceae* and comprises 85 genera and 1100 species of tropical and subtropical region of the world in Asia, Australia, Africa and America [1]. The family is a rich source of pharmacologically active bases of the crinine, galanthamine and lycorine series, which are known for their anticholinesterase, anticancerous, and expectorant properties. The genus *Crinum* L. have commercial, economical and medicinal importance also due to various active components. Approximately 180 types of alkaloids have been isolated and identified from different *Crinum* sp. and out of them 120 bases belong to crinine and lycorine types, like as -

caranine, crinamine, crinine, galanthamine, galanthine, haemanthamine, hippestrine, lycorine, narciclasine, augustamine etc.The representative alkaloids are: norbelladine, lycorine, homolycorine, crinine, haemanthamine, narciclasine, tazettine, montanine and galanthamine [2]. These secondary metabolites constitute a tool of great interest in chemotaxonomic studies of the Amaryllidaceae family. Lycorine, which is one of the most frequently occurring alkaloids in Amaryllidaceae plants, has been found in all *Lycoris* species. Other studies of lycorine have reported potential anti-nociceptive, anti-inflammatory, hepatoprotective, hypotensive activities [3, 4] and also shows antifungal activity against *Candida albicans* [5, 6].

In the light of growing global demand for natural pharmaceuticals, searching of prospective active components from natural sources is getting much attention. In that respect, different species of *Crinum* L. emerge as sources of multifarious bioactive principles, especially alkaloids.

Crinum asiaticum has long been used to treatments such as anti-inflammation, anti-allergic and anticancer [7]. The major biological active compounds of *Crinum asiaticum* L. is Lycorine which shows strong effect on cancer cells [7] *Crinum latifolium* L. has also been reported to contain lycorine [8, 9]. The present study presents a comparative account of active principle - Lycorine in both the population of *Crinum asisaticum* L. and *Crinum latifolium* L., collected from the wild areas of Paschim Medinipur of West Bengal.

MATERIALS AND METHODS

The species *Crinum asiaticum* L. and *Crinum latifolium* L. both havebeen collected from three different sites of Midnapore forest range of West Bengal, India (Table 1).

Table no 1: Name of the studied species and their location.

Name of Species	Location	Latitude (ºN)	Longitude(ºE)
Crinum asiaticum L.	Midnapore forest area of West Bengal	22.430889	87.321491
Crinum latifolium L.	Midnapore forest area of West Bengal	22.430889	87.321491

Study of Biomass

After harvesting, fresh single mature bulbs of both species were weighed and respective dry weight was taken after drying the bulbs in hot air oven at 40 ^{0}C.

Extraction of Lycorine

The fresh 100 gm of individual bulbs of both species were cut into small pieces and crushed properly into a mortar pestle with 50% glacial acetic acid and kept in a sealed beaker for 72 hours. Then it was filtered through Whatman filter and pouredin water. Thenthe material acidified with H_2SO_4 to pH 3-4, and was extracted with petroleum ether and diethyl ether to remove lipophilic, acidic and neutral materials. After basifying the aqueous solution to pH 9-10 with NH_4OH, it was extracted with chloroform, the extract washed with distilled water to neutral pH, dried with a rotary evaporator under reduced pressure at 40 °C. The extract was, then, concentrated under reduced pressure to obtain lycorine. Afterwards the concentrated plant material was kept in 80% methanol for further study.

Quantitative estimation of Lycorine

The extract was quantified by High Performance Liquid Chromatography (HPLC). The analysis of Lycorine was done through HPLC (Agilent) with UV detector. Crude extracts (10 mg) were dissolved in methanol (1 mL) and injected 20 µL to high performance liquid chromatography (HPLC) using C18 column, eluted with CH_3OH : H_2O (95:5) at

flow rate 1.0 mL/min and detected UV at 340 nm. Quantitative determination was carried out by the external standard method based on peak area [10].

Percentage of lycorine

Percentage of lycorine was calculated by getting the weight of lycorine from initial 10 g of bulb powder. Then it was calibrated in terms of the weight of lycorine in 100 g of bulb powder.

$$Lycorine(\%) = \frac{\text{Lycorine present in 10 g of tissue}}{\text{Initial weight of tissue (10 g)}} \times 100$$

Average of total lycorine

The amount of total lycorine available in each species and obtained after each treatment have been measured and expressed as the dry weight of each sample.

Average of total lycorine (g) = dry weight (g) of specific treated bulbs × percentage of lycorine obtained from that respective species was calculated.

Statistical analysis of experimental data

The experimental data were statistically analyzed with SPSS software. The level of significance used in F test was P = 0.05. The means followed by the same lower case letters do not differ significantly at the 0.05 probability level.

RESULTS

Two different species haveshown the presence of different amountof lycorine. Amongthem *Crinum latifoliunm* was noted to contain greater amount of lycorine (Table 03).Average biomass of individual bulb was also recorded to be greater in *Crinum latifolium* (Table 02).

Table 02: Biomass of bulbs of *C. asiaticum* L. and *C. latifolium* L.

Name of species	Fresh weight (g)	Dry weight (g)
C. asiaticum L.	52.39± 02.45	5.26± 01.55
C. latifolium L.	89.41± 03.25	12.68± 02.15

Table 03: Percentage and amount of lycorine present in *C. asiaticum* L. and *C.latifolium* L.

Name of species	Percentage of lycorine (%)	Lycorine (g)/ g weight of bulb
C. asiaticum L.	0.065	0.342
C. latifolium L.	0.095	1.205

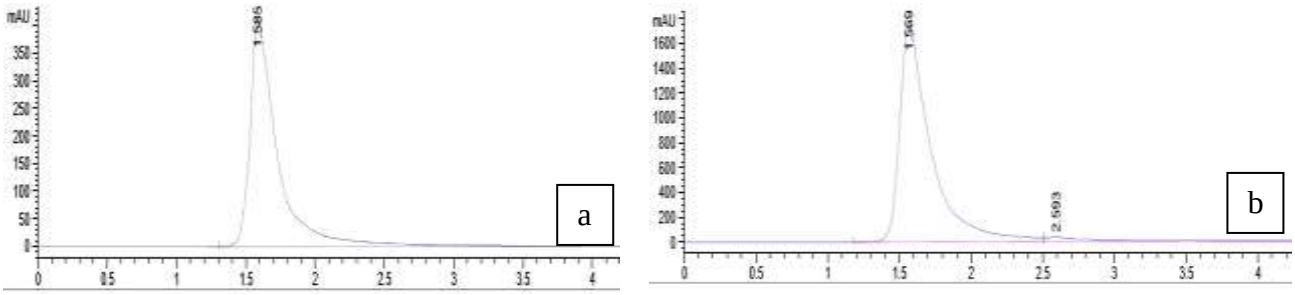

Figure 01: HPLC peak of Lycorine of *C. asiaticum* (a) and *C. latifolium* (b).

DISCUSSION

Lycorine as an important bioactive constituent has been extracted and estimated with the aid of HPLC for two species of *Crinum* L., *C. asiaticum* and *C. latifolium* in the present study. Though the presence of this medicinally important biomolecule has been reported earlier [7, 8, 11, 12], present study was intended to record its availability and amount in the locally available individuals of the species and the relative abundance among two species. The record shows *C. latifolium* to have greater amount of lycorine per gram of tissue and also more biomass of bulb. Thus, in consideration of productivity per plant the amount of lycorine is revealed to be quite greater in *C. latifolium* in comparison to *C. asiaticum*.

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