BIOLOGY OF *CENTELLA ASIATICA* (LINN.) URB., A MEDICINALLY IMPORTANT CLONAL PERENNIAL

By

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THESIS
SUBMITTED
IN
FULFILMENT OF THE REQUIREMENT OF THE DEGREE OF
DOCTOR OF PHILOSOPHY IN BOTANY

NORTH-EASTERN HILL UNIVERSITY
SHILLONG, INDIA

OCTOBER, 1987

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I certify that the thesis entitled "Biology of Centella asiatica (Linn.) Urb. - a medicinally important clonal perennial", submitted by Miss Borabell Wankhar, M.Sc., for the Degree of Doctor of Philosophy of the North Eastern Hill University, Shillong, embodies the record of original investigation carried out by her under my supervision. She has been duly registered and the thesis presented is worthy of being considered for the award of the Ph.D. Degree. The work has not been submitted for any Degree of any other University.

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CHAPTER I

General Introduction and Review of Literature

Centella asiatica (Linn.) Urb. is among the most important medicinal plants of India. For a long time it has been used in the indigenous system of medicine. Perusal of the literature reveals a long list of diseases cured by C. asiatica, of which the most remarkable is the treatment of leprosy. The plant grows wild under a wide range of climatic conditions and thrives well in presence of other weedy species. It forms localized patches as a result of clonal multiplication wherever it grows. The clonal growth habit of the plant is one of the interesting aspects which attracts one's attention.

Clonal perennials have, by defination, ramets derived from a single genet. The clonal growth of a rhizomatous plant involves the continued reduplication of discrete modular units, the 'ramets' - the sum of these units representing the 'genet' or product of a single zygote (Harper & White 1974, Kays & Harper 1974, Harper 1977). The ability of a single genotype to form fragmented phenotypes is just one of the variants in the life-history patterns of modular organisms (Harper & Bell 1979). A unique feature of clonal plants is that rooting at nodes of individual shoots may create physiologically distinct plants with independent fates. This developmental feature has important physiological and ecological implications because it affects how the plant functions as a single physiological unit and interacts with its environment (Bazzaz *et al.* 1987).

Clonal plants exhibit a range of strategies, from a very conservative with short internodes and overlapping rosette, as in *Soliva valdiviana*, to highly exploratory as in *Ranunculus repens* (Ginzo & Lovell 1973,a,b, Lovett Doust 1981 a). The two categories have been called 'phalanx' and 'guerrilla' by Lovett Doust (1981 a). In phalanx type for example, *Bellis* sp., genets expand slowly and modules (rosettes) are tightly aggregated within genets, whereas in guerrilla type for example, *Prunella* sp., clonal expansion is fast and modules are loosely aggregated within genets (Schmid & Harper 1985). Different growth forms require different energy expenditures (Lovell & Lovell 1985). They argued that a prostrate habit permits maximum ground cover at minimum cost as the requirement for supporting tissue is reduced, while Pickett & Kempf (1980) have commented that the clonal form of some shrubs may permit exploitation of a broad horizontal area relatively cheaply. Furthermore, Lovell & White (1986) argued that the prostrate habit (plagiotropic shoots, psuedo - rhizomes, stolons, runners and rhizomes) encourages vegetative spread because these organs are prolific producers of adventitious roots essential for the establishment of independent ramets. However, the pattern of growth may be greatly affected by environmental conditions (Lovell & Lovell 1985).

In clonal plants, persistence of clonal connections and extent of resource sharing after establishment of ramets vary greatly among species (Antos & Zobel 1984, Callaghan 1984).In some species (e.g., Aster acuminatus and Clintonia borealis) connections among ramets decay or cease transport soon after new ramets establish, which might reflect selection against continued resource sharing under certain conditions (Ashmun et al. 1982, Pitelka & Ashmun 1985), while other species e.g., Solidago canadensis,Agave salmiana and Agave deserti exhibit facultative integration in which reduced photosynthetic capacity or increased demand for carbon induces import of photosynthates from other connected ramets (Hartnett & Bazzaz 1983, Pitelka & Ashmun 1985, Martinez-Morales & Meyer 1985, Raphael & Nobel 1986). Several studies have shown that connection among ramets can increase total growth of clonal groups for example, Ranunculus repens, Fragaria chiloensis and Agave deserti (Lovett Doust 1981 b, Salzman & Parker 1985, Alpert & Mooney 1986, Raphael & Nobel 1986) or at least of individual established ramets within groups e.g., Carex arenaria and Solidago canadensis (Noble & Marshall 1983, Hartnett & Bazzaz 1983,1985 b).Salzman & Parker(1985) measured growth of pair of connected established ramets of a rhizomatous herb when one, both or neither of the pair was subjected to high salinity.When just one was stressed,connection appeared to increase growth of the stressed ramet and to a lesser degree, to decrease growth of the unstressed ramet.

In the case of clonal plants, the major advantage appears to be the potential for physiological integration of connected rosettes via translocation of resources such as water, minerals and photosynthates as pointed out by Hartnett & Bazzaz (1983),Noble & Marshall (1983), Callaghan (1984), Pitelka & Ashmun (1985 , 1986),Salzman & Parker (1985), and Alpert & Mooney (1986).Added costs of continued integration might include maintenance of connections and long term support of injured, abnormal or poorly situated ramets. Current studies suggest that benefits may be high in habitats where resource availability varies on the scale of individual ramets, such that sharing can compensate for localized resource scarcity, salinity or other potential stress (Hartnett & Bazzaz 1983, 1985 b, Pitelka & Ashmun 1985, Salzman & Parker 1985).

The relative investment into reproduction and clonal growth under different environmental conditions has been one of the main problems in life history studies of clonal plants(Tripathi & Harper 1973, Ogden 1974, Abrahamson 1975, Holler & Abrahamson 1977, Harper 1978,Douglas 1981, Armstrong 1982, 1984). Generally these two functions have been regarded as antagonistic (Salisbury 1942,Harper

1977, Abrahamson 1980). However, Armstrong (1982,1984) has proposed that allocations to reproduction and clonal growth depend on the growth form of the plant, and are not expected to be inversely related. Watson (1984) has recently presented an elegant study of **reproduction** and clonal growth in water hyacinth (Eichhornia crassipes), in which she showed that the ramet production is constrained by the development of inflorescences. This was not the result of resource limitation, but instead it was due to the developmental morphology of the shoot. Kingsolver (1986) reported that the blooming rosette of Yucca glauca apparently expends a fixed effort on sexual reproduction and then allocates its remaining resources to vegetative growth. Reproduction and clonal growth in Potentilla anserina was studied by Erikson (1985), who emphasized that in the investigation of resource allocation in clonal plants growth form should also be considered. Variation in biomass allocation whether it is genetic or environmentally induced, is usually presumed to reflect adaptive response of individual to variation in environmental factors (Ashmun et al. 1985). Bazzaz et al. (1987) emphasised that the allocation pattern of a plant defines its ecological roles and is, therefore, an important factor in understanding plant distribution and adaptation. Hence, studies of biomass allocation patterns are likely to be very useful in understanding the ecology of a species.

Centella asiatica is a clonal perennial plant which is widely spread in Meghalaya under a variety of conditions. Although the plant reproduces both through vegetative and sexual means, the latter mode of reproduction is negligible. Local patch expansion occurs primarily as a result of clonal growth. Growth period extends from March to November and the plant carpets the ground completely during rainy season. Throughout the altitudinal range it occupies a variety of habitats e.g., wasteland, garden, roadside, streamside, fallows, open forest canopy, relatively dense forest canopy etc. The environment of the individual patches varied widely in terms of light intensity, soil moisture, texture and nutrients and accordingly, there are differences in population density (Table 1.1) which is a function of clonal growth in this species. Patches of *Centella asiatica* flourish under high light whereas in closed habitats plants were observed to have thin and smaller leaves, and also longer petiole. It can be described as a variable species, tolerant of a wide range of climatic and edaphic conditions, however, it prefers comparatively open and moist habitats.

The plant bears a well defined rosettes on above ground stolons, which are borne in the axils of older leaves, and they give rise to rosettes at every node. Connections between rosettes persist throughout the growing

Table 1.1. Features of a few selected habitats of *Centella asiatica* in Meghalaya, alongwith its density on these habitats.

Habitat	Density of *C. asiatica* /m^{-2}	Light intensity (Lux)	Nutrient level: Organic matter content (%)	N (%)	P (Kg/ha)	K (kg/ha)	p^H	Soil moisture content (%)	Soil texture
Open waste-land	53	55000 - 61000	1.77	0.15	2.24	61.60	5.6	25.4	Clay loam soil
Shaded wasteland	40	31700 - 33500	1.80	0.15	2.24	51.60	5.5	27.7	Clay loam soil
Cropland	26	34600 - 37100	1.22	0.18	2.69	168.0	6.2	25.8	Sandy loam soil
Jhum fallow	42	39200 - 41000	2.37	0.18	3.20	122.0	5.2	27.5	Sandy loam soil
Open forest canopy	39	30200 - 34000	2.20	0.16	7.85	226.0	6.5	26.6	Clay loam soil
Relatively dense forest canopy	22	20100 - 21000	2.29	0.16	7.85	226.0	6.5	28.4	Clay loam soil
Stream side	43	40800 - 42500	1.85	0.17	27.35	61.60	5.8	34.5	Sandy loam soil

period except in winter when the above ground plant part is killed due to cold. A rosette produced at each node serves as unit of both perennation and multiplication (clonal growth). The sprouting of shoot from plants that were apparently dried up during winter, begins in March. The plant initiates stolon production in spring and this continues throughout the growing season. Actual ages cannot be determined in nature, although overall trend in ramet number or density may be easily determined. The underlying pattern of mortality and clonal growth in nature are also not easily measured.

As an important medicinal plant *Centella asiatica* has engaged the attention of several workers in the past. Its medicinal applications have been recorded in many books and journals. A glycoside called asiaticoside isolated from the plant has been shown to possess activity against leprosy. In fact every part of this plant is used in Indian medicine (See 'Review of literature'). The tribals dwelling in the interior places in Meghalaya use this plant for curing stomachache, dysentery and also as blood purifier and nerve tonic.

In India ecological studies on several medicinal plants have been made. Of these, the works of Biswas (1964) on *Rauvolfia tetraphylla* and *R. serpentina*, Sah (1966) on

Bacopa monnieri, Pathak (1967) on *Tribulus terrestris*, Shetty (1967) on *Biophytum sensitivium*, and Singh *et al*. (1982) on *Silybum marianum* are worth mentioning.

The clonal habit of *C. asiatica* and its high medicinal value make this species an attractive material for detailed ecological study. Several reports have been published on its chemical composition and few reports on the pharmacological action, however, the ecology of this important medicinal plant has not been touched so far. The present study on the ecology of *C. asiatica* fills the gap.

The study covers the following aspects of the ecology of *C. asiatica* :

(i) Seed germination.

(ii) Phenology and growth of *C. asiatica* in relation to altitude, season and associated species.

(iii) Population dynamics on jhum fallows of different ages.

(iv) Competitive interaction between individuals raised from seeds and cuttings.

(v) Growth behaviour in relation to light regime, NPK level, soil moisture and soil texture under controlled conditions.

The 'General Introduction' outlines the objectives of the present study. It is followed by the 'Review of Literature'. Description of the study sites and general biology of *Centella asiatica* are given in Chapter II. The experimental data on various aspects mentioned above have been presented in Chapter II to VII. The results of each chapter have been discussed separately, however, the major findings of the thesis have been discussed under 'General Discussion' in an integrated manner.

Clonal growth habit is one of the most widespread form of asexual reproduction in plants (Cook 1983, Hartnett & Bazzaz 1983).Salisbury (1942) showed that many widespread species often have vegetative multiplication as an alternative and argued that these species have a considerable range of tolerance of climatic conditions and efficient reproduction under suboptimal conditions. Clausen et al. (1947) suggested that plants with efficient vegetative reproduction are better equipped to tolerate extreme environmental conditions. Vegetative spread and its possible adaptive significance has been discussed by Bell (1974, 1976), Harper & White (1974), Harper (1977), Harper & Bell 1979, Bell et al. (1979) and Bell & Tomlinson (1980). Recruitment by vegetative means is independent of external pollination and a single plant is completely equipped to colonize a given area. Ramet population can expand or contract depending on the prevailing environmental conditions (Sarukhan 1976). Another advantage of vegetative reproduction is the increased longevity and perennation of the genet. However, there are of course, inherent costs such as the lack of genetic recombinations and reduced ability to disperse to far away places (Schellner et al. 1982).

Harper (1967) has suggested that sexual and asexual reproduction are competitive within the plant. Because sexual and vegetative reproduction are dependent on the same limited resources, and since physiological conditions best favourable to one are not identical with those most favourable to the other, the two modes appear competitive (Salisbury 1942). However, it is more useful to view the two modes of reproduction as constituting an essentially inseparable system in the life history of the plants (Kawano 1975). The sexual reproduction produces new genetic recombinations and allows dispersal to new sites, while asexual reproduction perpetuates the successful genotypes in the local populations (Abrahamson 1980) and the optimal balance between the two modes of reproduction depends on a variety of abiotic and biotic factors.

Despite dominance of ramet individuals (cloning plants) in aquatic, forest floor, grassland communities etc., there is little information available on the population biology of ramets. There are numerous problems associated with such plants, for example, the occurrence of overlapping generations (Sagar & Mortimer 1976), the difficulty of defining the individual plant (Jansen 1977), the existence of both vegetative and seed reproduction (Thomas & Dale 1975), the long pre-reproductive period (Tamm 1956) and the non-appearance in some years of above-ground parts (Well 1967). Further, acquiring ramet data from natural populations of cloning plants is difficult. First, ramet linkages are frequently underground, secondly ramet linkages usually decay in a relatively short time (Dickerman & Wetzel 1985). The demographic analysis of clonal perennial plants growing in natural habitats has been done by some workers, for example, Sarukhan & Harper (1973) on Ranunculus spp., Noble et al. (1979) on Carex arenaria, Solbrig et al. (1980) on Viola sororia, Barkham (1980) on Narcissus sp., Lovett Doust (1981 a,b,c,) on Ranunculus repens, Newell et al. (1981) on Viola blanda and V. pallens, Schellner et al. (1982) on Viola spp., Pitelka et al. (1984) on Clintonia borealis, Raphael & Noble (1986) on Agave deserti and few others. However, wide gaps in our knowledge remain.

Based on his study on the population dynamics and local specialization in Ranunculus repens, Lovett Doust (1981 b) reported that germination and establishment of the new genets was rare in grassland and mixed deciduous woodland. Further, the birth - rate of ramets was apparently density - independent, but death-rate of the rosettes was density - dependent. Poor recruitment of seedlings in many other species was also reported by Anderson & Loucks (1973), Whigham (1974), Raynal (1979), Barkham (1980) and Pitelka et al. (1984). Populations maintaining their numbers by vegetative propagation typically show Deevy Type II survivorship curve (Sarukhan 1976). The effects of hoeing and rototilling on population dynamics of pure stands of Portulaca oleracea were studied by Miyanishi & Cavers (1981).

They found that soil disturbances resulted in decreased dry matter yield per unit area. Froud - Williams *et al.* (1984) studied the effects of seed burial and soil disturbance on emergence and survival of arable weeds and observed that such disturbances increased the total number of seedlings which emerged. Jurik (1985) observed that the production of runners and plantlets in strawberry population was greatest in open and recently disturbed habitats than in closed habitats.

The studies made by various workers (e. g. Sarukhan & Harper 1973, Hawthorn & Cavers 1976, Yadav & Tripathi 1981) on several plant species revealed that the plant mortality is maximum in the active growing season of population. William (1970), Bazzaz & Harper (1976), Yadav & Tripathi (1981), Silvertown & Dickie (1981), Weiss (1981), Law (1981) and Rai & Tripathi (1984) reported that heavy mortality occurs during the seedling state, while Canfield (1957) found high mortality during old age. Subsequent studies (Cook 1979, Meagher & Antonovics 1982) have shown that mortality is frequently more closely associated with size than the age of the plant. Regehr & Bazzaz (1979) worked on the population dynamics of *Erigeron canadensis* and observed 16-86% plant mortality due to frost heaving. Yadav & Tripathi (1981) also observed some mortality during winter for both seedling and adult populations of *Eupato-*

rium odoratum, and they ascribed it to the prevailing low temperature and high moisture stress.

Late emerging plants have to face severe competition from established adult plants of their own or of other associated species. Many workers (e.g. Tamm 1956, Cavers & Harper 1967, Putwain & Harper 1970, Friedmann 1971, Kushwaha et al. 1981, Yadav & Tripathi 1983, Rai & Tripathi 1985, Tripathi 1985) have reported that seedling establishment was relatively poor in established community. Tripathi & Dwivedi (1978) suggested that the fundamental niche of a species may be restricted to a smaller hyper volume due to the interference caused by the presence of associated species. Sagar (1970) reported a better vegetative and reproductive growth of Plantago lanceolata when the associated vegetation was removed. Titus & Stephen (1983) found that total dry weight, leaf length, stolon length, rosette number and winterbud production in Vallisneria americana had higher mean values for individuals in a monospecific stand of Vallisneria than at sites dominated by Potamogeton amplifolius or Chara vulgaris. The effects of experimental removal of species was also studied by Raynal & Bazzaz (1975) and Fowler (1981).

Studies on the growth of mixed populations established from tillers and from seeds were made by Tripathi & Harper (1973) and Kushwaha et al. (1983 a). They found that plants established from tillers were more vigorous and aggressive than their counterparts produced from seeds. Genetic variation and plasticity between and within populations of *Bellis perennis* and *Prunella vulgaris* was studied by Schmid (1985 b). Clay and Levin (1986) studied the relative success of six populations of *Phlox drummondii* raised under conditions of intra-specific conditions and observed that pure stands had higher means for flower production than mixed stands.

Werner (1975) studied the fate of teasel (*Dipsacus fullosum*) raised from the rosettes of different size and reported that the probability that an individual will die, remain vegetative or flower during a particular growing season is highly correlated with the size of its vegetative rosette. He also found that the rosette size provides better predictions of fate than age. Similar study was made by Gross (1981). Many workers (e.g. Cook 1980, Pitelka *et al.* 1980, Bradbury 1981, Solbrig 1981, Thompson & Beattie 1981) have shown that plant or ramet size proved to be an important component of fitness in many plants. Brown *et al.* (1985) studied variation in vegetative phenology in two forest herbs, *Aster acuminatus* and *Clintonia borealis*, and reported that large ramets tended to emerge earlier, mature more slowly, live longer and die later. The relationship between seasonal variation in light intensity, ramet size and sexual reproduction in natural and experimental populations of *Aster acuminatus* was studied by Pitelka *et al*. (1985). The results provide new insight into the general phenomenon of size - dependent reproduction in herbs.

The differences between plant populations along an elevational gradient were studied by Mooney (1963), Ward (1969) and Rochow (1970). Reduction in total biomass with increasing altitude has been reported by Kira & Shidei (1967), Yoda (1968) and Maruyama (1971). The functional dynamics of *Artemisia vulgaris* at different altitudes was studied by Todaria & Purohit (1979) who observed a significant reduction in the extension growth and leaf area expansion with increasing altitude. Jolls (1980) studied the pattern of biomass allocation in *Sedum lanceolatum* at four elevation sites and found a significant difference in biomass allocation to asexual and sexual tissues amongst populations. The balance between vegetative and sexual reproduction of *Mimulus primuloides* at different altitudes in California was studied by Douglas (1981) who argued that increased allocation to reproduction at high altitude may be an adaptation for greater potential dispersal or seedling establishment.

Studies on seasonal productivity and growth dynamics in response to the prevailing climate may provide useful indication about the time at which the harvest would be maximum. The seasonal changes in biomass allocation in eight winter annuals of Majove desert was studied by Bell et al. (1979). They found that there was no rapid change in biomass allocation during the life cycle. In the case of Chilean shrubs, Montenegro et al.(1982) found a significant seasonal change in biomass allocation. Seasonal change in biomass was also studied by Westlake (1966) and Hussey & Long (1982).The productivity, caloric content, and chemical composition of a population of Spartina alterniflora in relation to change in season were studied by Squiers & Good (1974), and they found that the amount of nitrogen that the plant accumulated in its aboveground parts early in the growing season was directly related to the peak of dry matter standing crop.

The effect of light intensity on the growth of purslane, Portulaca oleracea, was studied by Zimmermann (1976) who found that the higher the light intensity, the greater the fresh weight yields. The number of capsules produced was also positively correlated with the amounts of light received. Dennis et al. (1970) showed that high light intensity increased flowering but had no effect on vegetative reproduction in cultivated strawberries.Paterson(1980) studied the partitioning of dry matter in Cogon grass (Imperata cylindrica) from shaded and exposed habitats and found that allocation to reproductive activities was greater under exposed situation. Light-induced variations in the growth and dynamics of transplanted ramets of the understorey herb, Aster acuminatus were studied by Ashmun & Pitelka (1984) who observed that ramet growth and flowering were positively correlated with the level of light but not at all with soil moisture. The effects of light and nitrogen on dry matter allocation in Chaparral shrub, Diplacus aurantiacus have been studied by Gulmon & Chu (1981). Growth and reproductive strategies of two annual weeds as affected by soil nitrogen and density levels was studied by Trivedi & Tripathi (1982 a). Tripathi & Yadav (1982) studied the effect of population density, soil nitrogen and light intensity on population regulation of Eupatorium adenophorum and E. riparium and found that both species were favoured by soil nitrogen, however, nitrogen requirement was reduced under low light intensity. The growth of Elymus repens and Agrostis gigantia at different light intensities was studied by Skuterud (1984). Both species responded to reduced light by increased stem length and decrease in number of aerial shoots. The effect of shade on Crotolaria juncea and C. sericea was studied by Pandey & Sinha (1977), on Oxalis acetosella by Packham & Willis (1977) and on Acchillea millefolium by Bourdot et al. (1984). Rajeswara Rao et al. (1983) studied the effect of nitrogen, phosphorus, and potassium on pyrethrum (Chrysanthemum cinerariifolium) and found that pyrethrin concentration was not influenced by various rates of nitrogen, phosphorus, and potassium applications and sources of phosphorus.

Interacting influence of soil types and soil moisture plays an important role in determining the growth of the plant. Cunningham et al. (1979) studied the aboveground production and biomass allocation to reproduction in Larrea tridentata and found that enhanced soil moisture during the period of active growth reduces the allocation of biomass to reproductive structures, while, Abrahamson & Hershey (1977) found that reproductive allocation in Impatiens capensis remained unchanged due to soil moisture. However, Werner and Platt (1976) reported that in Solidago there is an increase in allocation of biomass to vegetative propagation with increased soil moisture. Differences in root and shoot development of tomato (Lycopersicum esculentum) varieties across contrasting soil environments were studied by Gulmon & Turner(1978).Trivedi & Tripathi(1982 b) studied the effect of soil texture and moisture on reproductive strategies of Spergula arvensis and Plantago major and found that daily watering on sandy soil gave the greatest dry weight in S. arvensis while, fortnightly watering gave the greatest in P. major. Similar studies were made by Rai & Tripathi (1983) on Galinsoga species and Davies (1984) on Erica species. The effect of moisture stress on stolon and adventitious root development in white clover (*Trifolium repens*) was studied by Stevenson & Laidlaw (1985) who found that growth of young roots and nitrogen fixation were adversely affected by drought.McIntyre (1976) emphasized the importance of water availability and showed that in *Agropyron repens* the degree of inhibition of buds on the rhizomes is inversely related to internal water content of the segment of rhizome bearing buds. When the supply of water to individual rhizome was increased, vigorous root production occurred at the nodes. The examples given illustrate the importance of the environmental factors in determining the plant growth and development.

Work done on *C. asiatica* :

Centella asiatica enjoys considerable reputation in Indian system of medicine and its virtues have been recorded in many books and journals on medicinal plants (Dymock *et al*. 1891, Kirtikar & Basu 1935, Sastri 1950, Chopra *et al*. 1956, Chopra *et al*. 1958,Basu & Rastogi 1967, Chopra *et al*. 1969, Heywood 1971, Grieve 1974, Dey 1980, Kharkongor & Joseph 1981,Bhattacharjee *et al*. 1981, Rastogi & Dhawan 1982,Rao & Jamir 1982, Biswas & Chopra 1982,Chopra *et al*. 1984, Ambasta 1986).

A great variety of medicinal application has been recorded for *C. asiatica*. While some of the uses are localized, others have been employed around the world. For example, in India (Malabar coast) and Madagascar, the plant is used for treating leprosy (Dymock et al. 1891, Kirtikar & Basu 1935, Sastri 1950). In Coromandel coast, the leaves are used for blows and bruises, in Java and Indo-China it is used as diuretic and in Konkan, juice is applied for skin eruptions (Dymock et al. 1891). In Ceylon it is used against digestive disorder, nervousness, dysentery and as a tonic and blood purifier (Kirtikar & Basu 1935). In Meghalaya, the plant is used for curing stomachache, dysentery and as a blood purifier. The plant is widely used against these ailments even today by the people in rural areas where modern medical facilities have not yet reached. The medicinal uses of C. asiatica are summarised in Table 1.2.

As recorded in Pharmacographia Indica (1891), the chemical screening of Centella asiatica began with the work of Lepine who found a substance named vellarine which is obtained from the dried plant to the extent of 0.8-1%. Beside this, resin fat (8.9%), tannin and sugar (24.5%), gum and salts (11.5%), albuminoid matter (12.5%) and ash (2.4%) were also present. Since then, many investigators have studied the chemical constituents of C. asiatica and many compounds have been charactesised. Wali and Katli (1937) reported the presence of an essential oil, sitosterol, tannin, glucose, a large amount of resinous material and

Table 1.2. Summary of the medicinal uses of *C. asiatica*.

Ailment/Uses		Reference No.
Anaemia	-	12
Loss of Appetite	-	2,12
Asthma	-	2,12
Blood disease	-	2,4,5,6,7,11,12
Boils	-	11
Blows & Bruises	-	1,8
Bowel complaints & Diarrhoea	-	7,8,9
Brain tonic	-	2,3,12
Bronchitis	-	12
Cholera	-	12
Cough	-	7,11
Dysentery	-	7,8,11,13
Eczima	-	1,5
Eye lesion	-	5,7,14
Fever	-	2,8,9,12
Gonorrhea	-	8
Headache	-	2,12
Inflammation	-	2,12
Jaundice	-	8
Leucoderma	-	2,12
Leprosy	-	3,4,5,7,8,9,10,15
Lupus	-	15

Weak memory	-	2,5,7,8,9,10,12
Nervous disease	-	1,4,5,6,7,12
Piles	-	1
Rheumatism	-	1,6,9
Smallpox	-	12
Spleen trouble	-	2,12
Skin diseases	-	1,3,4,5,6,7,8,10,12,15
Syphilitic ulcer	-	1,5,9
Stomachache	-	3,13
Tumour	-	11
Urinary discharge	-	2,8,11

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1 - Atal & Kapur 1977, 2 - Babu 1977, 3 - Biswas & Chopra 1982, 4 - Chopra _et al_. 1956, 5 - Chopra _et al_. 1958 6 - Chopra _et al_. 1984, 7 - Dey 1980, 8 - Dymock _et al_. 1891, 9 - Grieve 1974, 10 - Heywood 1971, 11 - Kharkongor & Joseph 1981, 12 - Kirtikar & Basu 1935, 13 - Rao & Jamir 1982, 14 - Sastri 1950, 15 - William Boericke 1971.

oleic, linolic, linolenic, palmitic, stearic acids.An alkaloid, hydrocotyline ($C_{22}H_{33}NO_8$) has been isolated from the dried plant by Basu & Lamsal (1947).

Bontems (1941) obtained from the fresh leaves a glycoside, named asiaticoside consisting of a colourless needle (m.p. 232°C), the yield being 0.7 to 1.2 g per Kg of the fresh leaves. Crystalline form of asiaticoside has been obtained from specimens grown in Madagascar. This glycoside is a rhamnose - glucose derivative of a triterpene acid- aciatic acid. The 'oxidised form' of asiaticoside may be present in the fresh plant. Melting points, optical rotations, chemical and physical behaviour of different derivatives of asiaticoside show greater similarity with those of triterpene acids of the β- amyrin group (Boiteau _et al_. 1949).Boiteau _et al_. (1949) **noted the chemical constitution** of asiaticoside ($C_{51}H_{88}O_{23}$).Singh & Rastogi (1969) investigated the asiaticoside from _C. asiatica_ growing in India and Rahandraha _et al_. (1963) determined anthrone asiaticoside from the plant.

Bhattacharyya & Lythgoe (1949) could not isolate the glycoside from the dried specimen, instead, three amorphous triterpene acids were obtained in pure condition. These are centoic acid ($C_{30}H_{48}O_6$, m.p. 245-61°C decomp.), centellic acid ($C_{30}H_{48}O_6$, m.p. 160-95°C decomp.), and centic acid ($C_{30}H_{48}O_5$, m.p. 230-55°C decomp.). Lythgoe & Trippet (1949) examined the fresh plant from Ceylon and found that it contained a pale yellow glycoside named centelloside, which resembles asiaticoside. On hydrolysis it yields centellic acid, glucose and fructose in the proportion of 1 : 10 : 2 (mol.). The chemical constituents of the Ceylonese material was analysed by Bhattacharyya (1956 a) and it contained centic, centoic and centellic acids and centelloside. Reducing sugar, salts, pectin and essential oils were also present. The plant of the Indian origin was, however, found to contain salts, sugars, essential oils, pectins and a mixture of triterpene acids. The major triterpenic constituents are indocentoic acid which appears to be isomeric with centoic acid and a water soluble glycoside identified as 'indocentelloside' (Bhattacharyya 1956 b, 1956 c).

Rastogi et al. (1960) found that the alcoholic extract of 10 kg air-dried plant powder gave two triterpene acids (9.7 g brahmic acid- $C_{30}H_{48}O_6$, m.p. 293° and 9.0 g isobrahmic acid- $C_{30}H_{48}O_6$, m.p. 263°), two saponins (37.5 g brahmoside m 242° decomp., and 16.0 g brahminoside m 223° decomp.), betulic acid and stigmasterol.

Malhotra et al. (1961) reported the presence of alkaloids, glycosides, sterols, tannins and sugar. Triterpene glycoside, thankuniside and terpenic acid, thankunic acid were reported by Dutta & Basu (1962). The brahmosides and brahminosides are said to be tri- and tetra-glycosides of brahmic acid (Rastogi & Dhar 1963). Chemical formula of brahmic acid- $C_{30}H_{48}O_6$, was given by Singh & Rastogi (1968). The triterpenoid saponins and sapogenins were isolated by Basu & Rastogi (1967). Rastogi & Dhawan (1982) concluded that saponins from this plant proved to be rich sources of new triterpenoids.

Medicinal uses of C. asiatica are many and several compounds have been isolated from the plant but so far, very few clinical tests have been conducted. As mentioned in the Pharmacographia Indica (1891), the use of the plant in the treatment of leprosy was first brought to notice by Boiteau (1859). Dr. A. Hunter tried it in Madras Leper Hospital and concluded that it is useful in ameliorating the symptoms and improving the general health of the patient. Dr. Daruty observed that the application of this drug to leper causes at first a sensation of warmth and pricking to the skin especially of the hands and feet, followed by a general sensation of warmth, sometimes unbearable. The capillary circulation is accelerated and after about a week the appetite improves, in time the skin becomes softer, throws off the thickened epidermis and recovers its transpiratory function. In the Pharmocopoeia of India it has been made official and it is described as an alternative, tonic and local stimulant more specially useful in syphilitic skin diseases in which it may be used both as an internal and local remedy. Directions for making a powder and poultice are given. Reports from Europe in 1885 confirmed this, and there has been some enquiry for the drug in Bombay which has led to its cultivation on a small scale.

Drs. Boiteau & Grime quoted by Chopra et al. (1958) isolated glycoside from the Madagascar variety and they found that in doses nearing a toxic level, it gave encouraging results in leprosy. They are of the opinion that the glycoside acts as solvent on the waxy covering of the bacillus leprea which becomes softened and could be easily destroyed by the tissue or an adjuvant drug. Results from clinical trials include softening, breaking down of nodules followed by cicatrization proved its powerful action against leprosy. Healing of whitlows and perforating ulcers and gradual improvement of anaesthesia and muscular atropy are also said to occur. Still more remarkable is the claim that eye lesions are rapidly cured if treated before the posterior chamber is involved. The most hopeful features of the present account of the work is the success in breaking down leprosy nodules, and still more the clearing up of hitherto intractable eye lesions. However, further work needs to be undertaken on modern scientific line.