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Study of pollination mechanism on *Leucas aspera* (Wild) link family lamiaceae

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Abstract

Leucas aspera is commonly known as Goma. The plants flower once in a year during September – February. The flowers are white sessile small zygomorphic, bilabiate, gullet shaped produced in verticils of terminal or axillary whorls and oriented horizontally to the axis. The flowers open each day during 6.00-7.30 hours. The open flowers are abundantly visited by honey bees, ants and butterflies for pollen and/or nectar. The honey bees *Apis florea*, *A. C. indica*, the digger bee *Amegilla* sp. *Megachile* sp. *Anthophora zonata* are the principal pollinators. During foraging visits of bees gains contact by their notaeum with essential organs and thus promote nototribic pollination. The small bodied *Trigona*, *Ceratina*, *Apis florea* collected pollen sternotribically.

Keywords: *Leucas aspera*, pollination, mechanism, lamiaceae

Introduction

Lamiaceae, known as mint family, comprising ca.210 genera and 3500 species is cosmopolitan in distribution abundant in Mediterranean. The lower lip or labia is characteristic and responsible for conserved family name, labiates. The family is represented in India by 64 genera and 350 species.

Different modes of pollination are correlated with complex syndromes of floral features. The functional aspects of these features related to the plant family Lamiaceae have been reviewed and discussed by Vander pijil. The available pollination literature of the family documents that different genera and different species with in a particular genus radiated with regard to their pollination relationships. *Leucas aspera* is an annual weed and is commonly known as the Goma. It is almost distributed throughout India. During growing seasons, it adds considerable biomass to the soil. The paper describes the breeding and pollination mechanisms of *Leucas aspera* in relation to its weed status.

Materials and Methods

Leucas aspera occurs in waste lands, agricultural fields and road sides in Rewa, Madhya Pradesh. The studies were carried out in 2021-22 through systematic and regular visits to three sites, the flowering of three different populations were recorded. Flower production on a daily basis was recorded on 25 randomly selected inflorescences. The time of daily anthesis and anther dehiscence in relation to prevailing weather were also recorded. Pollen number per anther was determined from 10 flowers distributed over different plants. Pollen-ovule ratio was computed after Cruden (1977) ^[1]. The sugars in the nectar were differentiated by paper chromatography (Harborne 1973) ^[2]. The presence of amino acids and proteins in nectar was demonstrated as described by Baker and Baker (1973) ^[3]. Hand pollinations were used to ascertain the prevalent mode (s) of reproduction. Also tests through emasculation were performed for apomixes. Five hundred flowers were observed for fruit development after their natural pollination, and the resulting fruit set, seed set and fecundity were calculated following Cruden (2000) ^[4].

Results and Discussion

Flowering phenology: *Leucas aspera* plants show up during September – February, grow vegetatively for a month, then flower for three months; sometimes even up to mid-March provided the soil is sufficiently wet. A plant flowers for an average period of 70 days (Range 60-110). Average number of flowers produced per day in one plant is 67 (R31 -128) during flowering season and flowers produced per plant in a one flowering season is 1569 (Range 545-2,572).

Inflorescence phenology: In *Leucas aspera*, the verticillate inflorescence lasts for 16- 30 days, producing an average 53 (range 20-112) flowers. Each day the number of newly opened flowers of an inflorescence varies from 1-6 flowers (average 3 flowers). Flower production ceases in February /March then the plants starts to dry up and wither.

Verticells produced at each node: 80 % of the nodes consist of one verticel, 9.3% of the nodes consist of set of two verticells without spacing, 9.6% of the nodes consist of set of two verticells with a distance of 5-10 cm and 0.2% of nodes consist of three verticells.

Flower morphology: The flowers white, sessile small, produced in verticils of terminal or axillary whorls and oriented horizontally to the axis. Flowers 20 mm long, gullet shaped. Calyx is green, tubular 8-12 mm; tube curved. Corolla is 13- 18 mm long, tube 5mm long and pubescent above, annulate in the middle. Upper lip 5 mm long, densely white wooly, the lower lip is broader, obviates, and rounded. The stamens are epipetalous, didynamous and almost appressed to the inner surface of the upper corolla lip. The ovary is superior, bicarpellary, syncarpous and tetralocular with four ovules. The style is gynobasic with a bifid stigma.

Anther dehiscence and anthesis: Flowers open each day during 600-7.30 hours with a peak at 6.30 hours. Anthers dehiscence by longitudinal slits in the bud stage about 30 minutes before anthesis. In association with a temperature range of 19-25 °C and RH 70- 85%.

Pollen characters: Pollen grains are oval / spheroidal, white in colour, powdery, tricolpate with a smooth exine. The size of the grain is 16.6 µm -24.9 µm. Output of grains per anther 1572 ± 150 (n=10) and the pollen ovule ratio is approximated to 1179 pollen output per flower 4716.

Flower life time: The stigma attains receptivity soon after anthesis and remains receptive for 30 hours. Thereafter, within 2-3 hours, the corolla, stamens and stigma drop off. The pollen grains remain viable for 48 hours after anther dehiscence.

Breeding behaviour: The results of breeding tests show that the plant is not apomitic, but reproduces through xenogamy, geitonogamy and autogamy; the first mode being most fruitful (Table-4).

Natural fruiting: The natural fruiting varies from 60- 71 % (Average 64 %). 40 % of fruits contain 4seeds, 12.6% of the fruits have 3 seeds, 9.1% of the fruits have 2 seeds, 3% of the fruits have only one seed and 36% of fruits have no seed.

Nectar characters: Nectar production begins with anthesis of the flower. A single flower produces 1µl. In bagged flowers with a total sugar concentration of 25-30%. The nectar sugar analysis indicates the presence glucose, sucrose and fructose.

Flower visitors: We recorded 18 species of flower visitors on *Leucas aspera* (Table 2 &3). The flower visitors include bee's ants and butterflies. The bees were digger bee, honey bees and

other small bees. Of the total spectrum of foragers some bee species are rather regular to their visits to *Leucas aspera*. *Amegilla* sp. *Apis florea*, *Apis cerana indica*, *Megachile* sp. *Anthophora* sp. are the common foragers throughout the flowering season. The bees made 96%, ants 1% and the butterflies 3% of the total visits. Of the total bee foraging visits, the *Amegilla* sp. constituted 52%, followed by honey bees (*A. florea*, *A.C indica*) 25.7% *Megachile* sp. 6.5 % *Anthophora* sp. 5.86 % *Apis dorsata* 2.2%, *Ceratina* and *Trigona* 3.2%. Of these visitors honey bees visited for pollen and nectar. *Trigona* and *Ceratina* foraged for pollen only. *Amegilla* Camponotus, *Anthophora* visited for nectar only. All these visitors were found to be active during day time from 7.00-1600 hr. The bees *Amegilla* sp. and *Apis florea* were more mobile in that they spent less time per flower and visited more flowers per unit time.

Foraging behavior: The lower corolla lip served as the landing place. When *Amegilla* sp., honey bees and *Anthophora* sp. probed the flowers for nectar, the reproductive parts brushed against their dorsal surface. *Apis florea*, *Ceratina*, *Trigona* and *Megachile* sp. while collecting pollen and /nectar, alight on the lower side of the reproductive parts contacting them with their ventral body surfaces. The ant camponotus probed the flower from side or front without contacting stigma or anthers. The flower visitors were most active and vigorous between 800- 1200 hr.

Observations on the behavior of different foragers suggested that except for butterflies, all other visitors' effect pollination to a greater or lesser degree. Correlation and integration of the data on the number of flowers visited by different flower visitors per unit time and length of time they spent at the flowers. The number of pollen grains deposited on the stigma in a single visit and successive visits and pollen deposition on stigmas under the forager supported by observations on the foraging behavior of different foragers proved that the bees *Amegilla* sp., *Apis florea*, *Apis cerana indica*, *Megachile* sp. could be effective pollinators of *Leucas aspera*. *Amegilla* sp and honey bees are the major pollinators to use the terminology of Baker *et al.* (1971) [5]. The little bee *Apis florea* may also reckoned as the major pollinator. The digger bee *Amegilla* sp. is very effective in bringing about pollen transfer, it constituted 52% of the total visits.

The medium sized *Apis florea*, *Amegilla* sp. and *Megachile* sp. mediated nototriby while probing for nectar and sternetriby when collecting pollen. Such foraging behaviour with bumble-bees was reported in *Pedicularis sudetica* and *P. grayi* by Macior (1973) [6], who attributed it to the flower behaviour and forager's body as well as to the kind of forage collected. The small bodied *Trigona*, *Ceratina* being unable to reach at the concealed nectar because of short tongues, collected pollen by inverting their bodies; they thus transferred pollen sternotribically.

The plant species of *Leucas aspera* studied occur in temporary habitats. The adaptation of their floral mechanism for pollination by nectar and pollen collecting insects, together with the flexibility in breeding systems with a provision for viable seed production through self (Spontaneous) and cross pollinations were assure their survival in changing environments where the availability of insects is unreliable.

Table 1: Flowering phenology of *Leucas aspera*

Date	Place	No. of plants observed	No. of inflorescences (verticells)	Total no. of flower on the day	Total no. of flowers produced during the flowering one season
14/10/22	Kuthulia	10	279	644; Avg. 64	14268; Avg. 1426
19/12/22	Saman	10	119	316; Avg. 31.	5452; Avg. 545
04/01/23	University campus	22	457	876; Avg. 40	31990; Avg. 1454
04/02/23	Agriculture College campus	8	299	1014; Avg. 128	20583; Avg. 2572
10/02/23	Chorhata	15	417	1077; Avg. 72	27,735; Avg. 1849

Table 2: Spectrum and census of flower visitors on *Leucas aspera*

Name o Visitor	Kuthulia 16/10/22	Saman 23/11/22	University campus 04/11/22	Agriculture College campus 14/01/23
Bees				
<i>Amegilla</i> sp.	1552	1408	539	1299
<i>Apis cerana indica</i>	164	248	300	249
<i>Apis florea</i>	356	409	477	173
<i>Apis dorsata</i>	70	96	54	-
<i>Megachile</i> sp.	214	156	100	136
<i>Anthophora</i> sp.	134	147	120	140
<i>Polistes</i> sp.	-	-	33	62
<i>Junonia almana</i>	88	-	-	109
<i>Trigona</i> sp.	18	57	40	49
<i>Ceratina</i> sp.	27	23	30	54
Un identified	47	-	-	50
Ants				
<i>Camponotus</i> sp.	21	29	38	22
Butterflies				
<i>Danaus chrysippus</i>	16	15	07	45
<i>D. limniace</i>	-	-	05	-
<i>Atrophaneura aristolochiae</i>	-	22	07	26
<i>Papilio Polytes</i>	36	-	-	-
<i>Eupleocore</i>	24	34	10	-
<i>Acraea violae</i>	-	-	-	11
<i>Borbo bevani</i>	-	20	-	-
Total visits	2767	2644	1760	2425

Table 3: Flower visitors on *Leucas aspera*, their forage type and body parts of pollen deposit

Name of the forager	Forage type		Region of pollen deposit
	Pollen	Nector	
Hymenoptera			
APIDAE			
<i>Apis cerana indica</i>	+	+	Dorsal thorax
<i>Apis florea</i>	+	+	Head, legs and ventral side
<i>Apis dorsata</i>	+	+	Dorsal thorax
<i>Trigona</i> sp.	+	-	Head, legs and ventral side
<i>Camponotus</i> sp.	-	+	-
<i>Ceratina</i> sp.	+	-	Head, legs and ventral side
<i>Amegilla</i> sp.	-	+	Dorsal thorax
<i>Megachile</i> sp.	-	+	Dorsal thorax
<i>Anthophora</i> sp.	-	+	Dorsal thorax
<i>Junonia almana</i>	+	+	Head, legs and Ventral side
<i>Polistes</i> sp.	+	+	Head, legs and Ventral side
Lepidoptera			
<i>Danaus chrysippus</i>	-	+	Proboscis
<i>Danaus limniace</i>	-	+	Proboscis
<i>Atrophaneura aristolochiae</i>	-	+	Proboscis
<i>Atrophaneura aristolochiae</i>	-	+	Proboscis
<i>Epleocare</i>	-	+	Proboscis
<i>Acreoviola</i>	-	+	Proboscis

Table 5: Number of flower visits per unit time and length of a visit at *Leucas aspera* flowers by different flower foragers

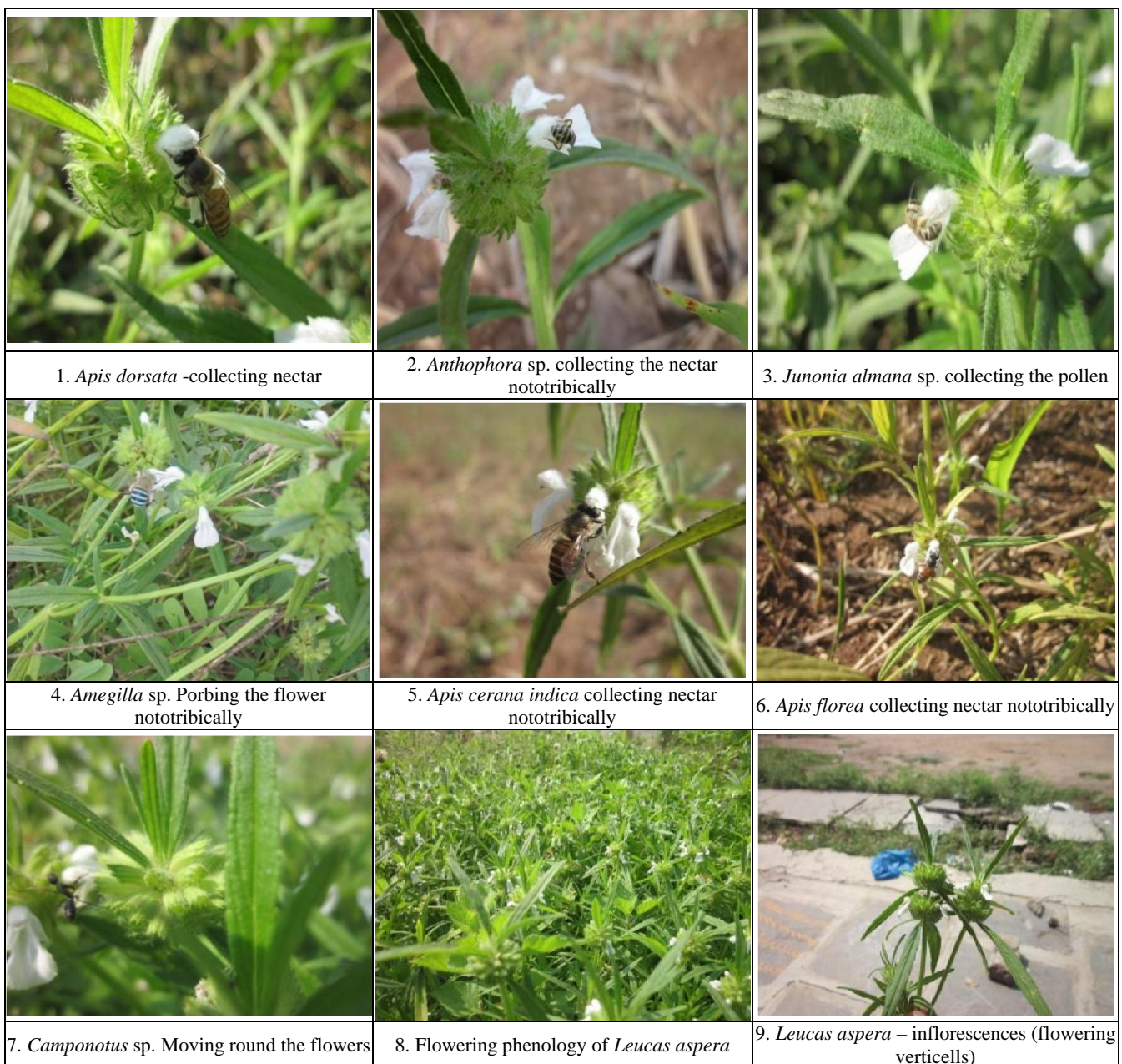
Species	Length of a visit (in seconds)				No of flower visits/minute			
	n	R	X	S.D	N	R	X	S.D
<i>Apis cerana indica</i>	10	5-9	6	1.41	10	6-12	9.5	2.32
<i>Apis florea</i>	10	4-6	4.3	1.19	10	6-10	7.4	1.17
<i>Apis dorsata</i>	10	5-8	6.3	0.94	10	7-12	9.2	1.76
<i>Trigona</i> sp.	10	5-7	6.1	0.87	10	8-15	10.7	2.49
<i>Ceratina</i> sp.	10	5-7	6.1	0.87	10	8-15	10.7	2.49

<i>Amegilla</i> sp.	10	2-5	3.2	1.05	10	12-30	16.5	5.39
<i>Megachile</i> sp.	10	4-7	6.1	1.33	10	8-20	11.7	3.91
<i>Anthophora</i> sp.	10	4-7	5.5	2.04	10	7-20	12.1	4.50
<i>Junonia almana</i>	10	5-8	6.3	2.56	10	7-12	8.9	2.43
<i>Polistes</i> sp.	10	5-8	6.3	0.95	10	8-14	10.5	2.34
<i>Danaus chrysippus</i>	10	3-6	4.2	1.03	10	7-13	9.6	1.83
<i>Danaus limniace</i>	10	3-6	4.3	0.94	10	6-14	9.9	1.91
<i>Atrophaneura aristolochiae</i>	10	4-7	5.2	0.94	10	9-18	12.1	2.89
<i>Papilio polytes</i>	10	5-7	5.8	0.78	10	8-17	12.3	2.59
<i>Eupleocore</i>	10	4-7	5.7	0.94	10	8-15	11.3	2.57

Table 6: Deposition of number of pollen grains on stigmas in first visit by different flower visitors on *Leucas aspera*

Name of the Species	Mean number of pollen on a stigma after the visit
<i>Amegilla</i> sp.	96
<i>Apis florea</i>	65
<i>Apis cerana indica</i>	106
<i>Megachile</i> sp.	58
<i>Anthophora</i> sp.	70
<i>Junonia almana</i>	64

Number of flowers sampled = 05



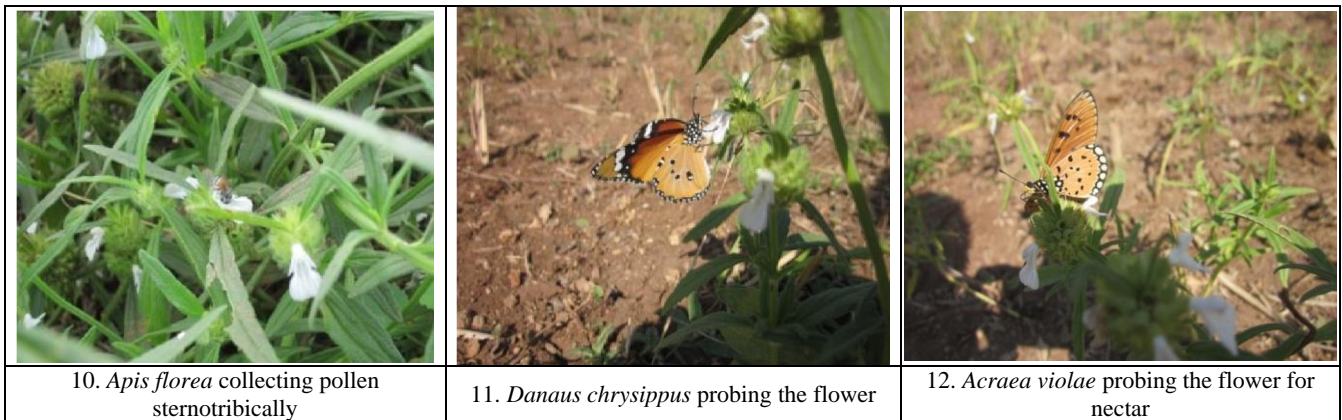


Fig 1-12: Photographs of foraging insects on flowers of *Leucas aspera*

Conclusion

Reproductive ecology of *Leucas aspera* was studied along with the morphology of the plant and the flower. Butterflies and bees play a major role in the pollination of *Leucas aspera*. Further research is to be conducted for chemical constituents and medicinal uses of the plant.

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