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Floral biology and pollination ecology of onion (*Allium cepa* L.)

SJ Kavitha and PV Rami Reddy

Abstract
Onion, an entomophily cross pollinated vegetable crop. The present study reports the details of structures, behaviour and biochemical aspects of onion flower. It takes 64-67 days to flower from the date of bulb sowing, remain flowering for next 40-45 days and 88-91 days to produce maximum number of open flowers. Anthesis occurs at 06-07 am and anther dehiscence continues from 07 am to 05 pm. Each anther produces an average of 1609 pollen grains. Floral nectaries located at lower part of ovary, produces an average of 1.80 µl nectar per floret. As a reward, onion flowers produce surplus quantity of pollen and nectar. All the four honeybee species acts as major pollinators in onion ecosystem. Maximum pollinator activity was observed at temperature 28˚c to 32˚c. So thorough understanding of flower morphology, floral biology and phenology helps to have perspective about onion ecosystem services. In this paper, an attempt has been made in this point of view.

Keywords: *Allium cepa*, onion, floral biology, nectar characters, pollinators, honeybee

Introduction
Onion (*Allium cepa* Linn.) is one of the oldest and most widely used vegetable known to human beings and is cosmopolitan in its distribution. The genus *Allium* is the second largest genus of monocotyledons in the family Amaryllidaceae.
In genus *Allium*, seven species are in cultivation out of 750 species that are known (Yousaf *et al*., 2004) [15]. Although, onion is native to south western part of the Asia, it is cultivated throughout the world, predominantly in temperate regions (Thompson and Kelly, 1957) [14].
Onion crop has two growth cycles; it is biannual for the seed production and herbaceous annual for bulb production. Propagation of onion happens through seeds, so seed production is a prerequisite for its multiplication. As onions are protandrous in nature, the flowers cannot fertilize themselves since pollens are shed by anthers even before the stigma becomes receptive (Delaplane and Mayer, 2000) [3]; hence Geitonogamy (pollination between different inflorescences of same plant) or allogamy (cross pollination between the different inflorescences belonging to different plants) becomes mandatory for seed production. Onion pollens are sticky and wet. Because of this reason, wind cannot act as a mediator and help in their transfer; hence the insect pollinators are the only option.
In current available literature, there is scanty information on many aspects of the floral biology of onion, even though it is one of the widely cultivated and studied vegetable. A systematic study on floral biology is lacking; therefore it is crucial to generate information on flowering biology and phenology and pollinator associated and which might also help in planning seed production programme. Hence the present studies were carried out to assess and understand in detail the floral biology and phenology of onion.

Materials and methods
Studies were conducted during 2015-17 at the ICAR– Indian Institute of Horticultural Research, (IIHR), Bengaluru (13°58′ N, 77°35′ E), India with an objective to understand the floral biology of onion from perspective of pollinator activity.
The popular onion cultivar Arka Pragathi was used in the study. The bulbs were planted on beds of size 3m x 3m at a spacing of 30 cm during November. Three replications were maintained in randomized complete block design. Standard package of practices was followed. Application of insecticide was completely avoided to encourage pollinator activity.
An extensive field observation has been done to find out the floral characteristics of the onion. The flowering period, flowering duration, days to first flower bud initiation, number of days for maximum flowering, number of flowering stalks per bulb were some of the characteristics that were studied. Number of florets per umbel, life of single floret, flower colour and odour were recorded.

Certain parameters like pollen size, lenghth of stigma etc. were measured in the laboratory with the help of a stereo-microscope (Olympus SZX7). Receptivity of the stigma was analyzed by α- naphthyl acetate test in which the excised stigma was dipped into two drops of α- naphthyl solution. This preparation was incubated for 10-20 minutes in a humid chamber and was observed under the microscope. The number of pollen grains per flower was calculated as per Shivanna and Rangaswamy (1992) [9].

Volume of the nectar was determined by measuring the length of the nectar column from 20 florets with a constant-bore micropipette. Total soluble solids (TSS) of nectar were assessed by Bellingham and Stanley 40-85% sugar hand refractometer. Each Component of nectar sugars was analyzed by using LC-MS/MS (Waters UPLC H class system fitted with TQD MS/MS system).

Pollinators in ten randomly selected umbels were recorded for five minutes during different hours of a day and field temperature was recorded each time. The visitors were collected using insect net, killed using ethyl acetate or ethanol and observed under stereo microscope for pollen load on the body parts. Representative samples were preserved and sent for identification.

The data were analysed for statistical significance by analysis of variance (ANOVA) and the means were compared through the Fisher least significant difference test by using SPSS software. The results were significant at $P<0.05$ unless specified otherwise.

Results and Discussion

Onion being a biannual crop with two crop cycles, i.e., seed-bulb-seed. Seed is the primary propagation material to produce bulbs.

In our study, onion plants started flowering in January which was approximately 64-67 days after bulb sowing (Table.1).The onion flowers were produced on a single elongated scape which is hollow inside. Each plant produced 6.72±0.01 scapes and each scapes holds an umbel consists of 479±19.06 florets (Figure.1). All the plants that belong to genus *Allium* produce umbelliferous inflorescence (Zuraw *et al*., 2009 and Zuraw *et al*., 2010) [16, 17]. The small flowers called florets are bound in a membranous white coloured spathe (2 to 3 in number). The number of florets per umbel can differ depending on the type of the species, plant genotype, time of the planting, size and storage conditions of the mother bulb (Sunita Devi *et al*., 2015) [13]. Due to the pressure created by such growing flower buds the spathe splits open. The life span of each floret was found to be approximately six days.

The colour of the flowers varies among the species. *Allium afatunense* bears violet coloured floret which turn into purple later (Zuraw *et al*., 2009) [16] and *Allium giganteum* produce purple red cluster (Zuraw *et al*., 2010) [17]. Sunita Devi *et al*. (2015) [13] reported that the florets of onion were of white to bluish in colour. But in our study, the florets were of dull white colour with green stripes and this is probably due to varietal difference.

Onion flowers possess very strong odour which is characteristic and is created by a chemical alteration of a sequence of volatile secondary metabolites like sulphur containing compounds (Jones *et al*., 2004) [5].

The maximum number of open flowers was found on 88-91 days. Only a few flowers open on the umbel at the beginning of flowering, which gradually increases until more florets open in one day at full bloom. Umbels were in bloom for a period of 30 days as the florets continue to open over a 2 to 3 week period. Initiation of the flowering depends on various physical factors like temperature, photoperiod, number of leaves and bulb development. This shows that photo-thermal conditions have a strong influence on the reproductive response of the onion plants (Branca and Ruggeri. 1994) [1].

Anthesis takes place during early morning hours, usually between 06-07 am. Anthesis occurs in succession from outer layer of florets in umbel and goes to central. Pollen fertility and stigma receptivity was found to be highest on the day of anthesis in our study. Both the perianth and stamens were six in number and are arranged in two whorls. Stamen was found to be 5.33 mm in length and consists of bilocular anthers which split longitudinally to release pollen. Anther dehiscence occurred between 07 am to 05 pm. Anthers produced pollen grain of size 0.04 x 0.02 mm in length. Each anther produced on an average of 1609.58 pollens which sums up to 9657.41 pollen grains per flower. Florets produced elongated pistil having length of 4.56 mm and a superior ovary (hypogynous) (Figure.2).

Living organisms have mutualistic inter-species relationships which help in pollination (Bronstein *et al*., 2006) [2]. It is a well-known fact that nectar is offered as a food-reward, by animal-pollinated plants to their pollen vectors (Simpson and Neff, 1981) [12]. Plants are attached to the ground hence have restricted movement. By mutualistic interaction with animals, especially with insects, the plants receive many benefits. The benefits of ability to cover distances by the insects play a vital role in pollen transfer in the plant and insects collect reward for themselves in the form of nectar (Schoonhoven *et al*., 2005; Rico-Gray and Oliveira, 2007) [7, 6].

The flowers of *Allium* are described as bowl shaped flowers in morphological terms with hidden nectarines. Generally they are in under part of the ovary. Onion flowers are a good source of floral nectar and pollen. The nectar produced from the nectaries was found to be collected in three cups between the lower ovarian wall and in inner whorl of stamens. Undisturbed nectar on florets was easily visible like a small sparkling bubble in the sunlight (Fig.2) thus attracts the pollinators. Onion flowers produced nectar volume of 1.80µl/floret with average total soluble solids 59.73 ± 2.73 in our study. LCMS sugar analysis data showed that onion floral nectar was rich in fructose (20.339 mg/g), glucose (10.99 mg/g) and sucrose (0.151 mg/g) sugars (Table.2). The onion flowers produced nectar which was rich source of nectar thus acts as a good food reward and attracts wide range of pollinators belongs to different family. Among them, family Apidae including honeybees *viz*., *Apis cerena*, *Apis florea*, *Apis dorsata* and *Tetragonula irridipennis* were major pollinators whose activity also depended on temperature. Since nectarines are located at shallow depth, short tongued bees were attracted more. So we can say onion flowers and bees are coevolved. As the time of the day progressed temperature kept increased and number of pollinators decreased. Maximum pollinator activity was observed between 9.00 am and 12.00 pm and at temperature 28˚c to 32˚c (Figure. 3). Similar results were obtained by Sunita Devi *et al*., (2015) [13] where four *Apis* species *viz. A. dorsata*, *A. cerana*, *A. mellifera* and *A. florea* were recorded as major foragers on onion umbels as pollen and nectar gatherers.

Onion is a cross pollinated and entomophilic crop. A deeper insight into its flower morphology, floral biology, phenology and pollination syndromes helps in understanding natural pollinator complex and plan effective seed production programs. Even though lot of variation exists in plant

behaviour and phenology among different cultivars, in different region, at different agro climatic zones, effort has been made to understand the fundamental aspects of floral biology of the crop.

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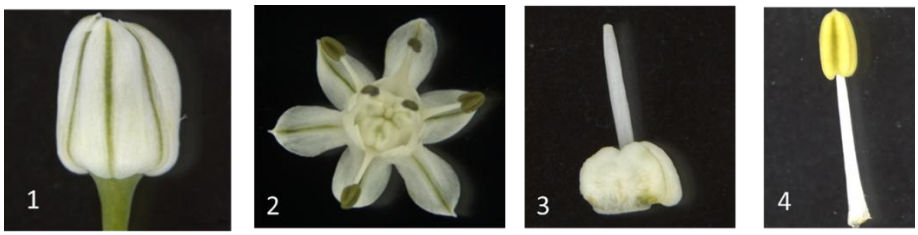
Table 1: Onion floral characters and the observations

S.N	Floral Characteristics	Observations
1	Days from planting to 1st flower bud opening	64-67 days
2	Flowering duration	40-45 days
3	No. of days for maximum plants in flowering	88-91 days
4	Number of flowering stalks per bulb	6.72 ± 0.01
5	Inflorescence type	Umbel
6	Number of florets per umbel	479±19.06
7	Life of single floret	6 days
8	Odour	strong odour
9	Flower colour	Dull white with light green tinge
10	Length of pistil	4.56 ± 0.58 mm
11	Length of Style	2.78 ± 0.67 mm
12	Ovary	Superior
13	Stamen length	5.33 ± 0.50 mm
14	No. of anther / flower	6, rarely 7
15	Anthers	Bilocular
16	Anther dehiscence mode	Longitudinal
17	Anthesis time	6-7 am
18	Anther dehiscence	7am to 5pm
19	Pollen shape	Oval/ oblate
20	Pollen type	Wet and Sticky
21	Pollen grain size	0.04x0.02mm
22	Number of pollen grain/floret	9657.41
23	Pollination type	Cross pollination
24	Mode of pollination	Entamophily
25	Self incompatibility	Protandrous
26	Nectaries	Present
27	Position of nectaries	Lower part of the ovary

Table 2: Composition of onion nectar

Parameters	Quantity
Nectar quantity	1.80 µl/floret
Total soluble solids in nectar	59.73 ± 2.73
Fructose	20.339 mg/g
Glucose	10.99 mg/g
Sucrose	0.151 mg/g

Fig 1: Inflorescences of *Allium cepa*



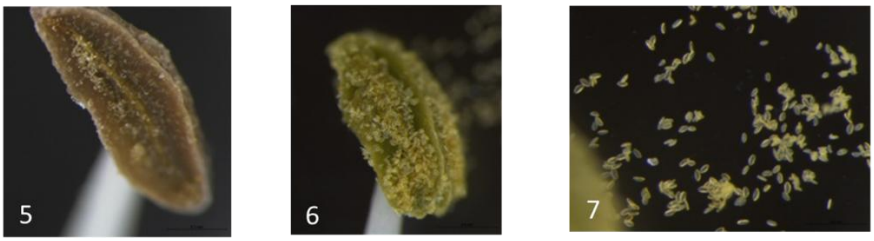


Fig 1, 2-unopened and opened floret
Fig 3- Pistil
Fig 4- Stamen
Fig 5, 6- Anther dehiscence
Fig 7- Pollen

Fig 2: Onion floral parts

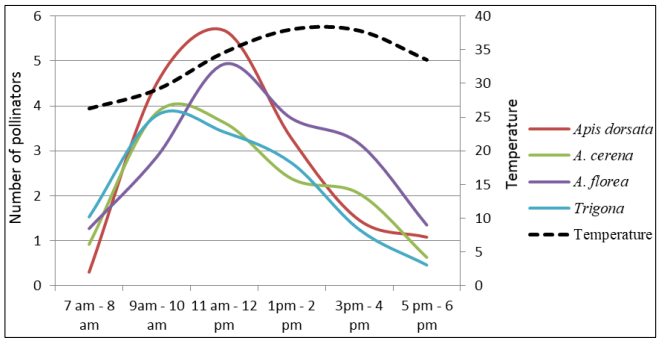


Fig 3: Activity of bee pollinators on *A. cepa* umbels at different hours of the day

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