Article Type: Research Article J Name: Modern Phytomorphology Short name: MP ISSN: ISSN 2226-3063/eISSN 2227-9555 Year: 2023 Volume: 17 Page numbers: 109-112 DOI: 10.5281/zenodo.200121 (10.5281/zenodo.200121 (10.5281/zenodo.Year-Volume-PDFNo.) Short Title: Comparative flower morphology in Allium sativum L. (Amaryllidaceae)



RESEARCH ARTICLE

Comparative flower morphology in Allium sativum L. (Amaryllidaceae)

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Received: 30.08.2023, Manuscript No.: mp-23-112947 Editor Assigned: 10.09.2023, Pre-QC No. mp-23-112947(PQ) Reviewed: 28.09.2023, QC No. mp-23-112947(Q) Revised: 02.10.2023, Manuscript No. mp-23-112947(R) Accepted: 09.10.2023 Published: 16.10.2023

Abstract

The vascular anatomy and morphology structure of the *Allium sativum* flower were studied. New morphological features were investigated for taxonomy in *Amaryllidaceae*. 15 flowers of *Allium sativum* were sectioned using standard methods of Paraplast embedding and serial sectioning at 20 mm thickness. Sections were stained with Safranin and Astra Blau and mounted in Eukitt. We find three vertical zones in the *Allium sativum* gynoecium: synascidiate, symplicate and hemisymplicate. The micromorphology and vascular flower anatomy were described by using flowers transverse sections. The *Allium sativum* peduncle contains 6 vascular bundles - three large and three small ones, which branch out and form a circle of vascular bundles - a vascular cylinder, from which at the level of the peduncle they depart six pairs of vascular bundles, traces of tepals, traces of stamens and above - traces of dorsal carpel bundles. Stamens depart from the ovary wall.

In the center of the ovary, there remains a circle of small vascular bundles- the roots of the ventral complex, which were reorganized above, with the appearance of locules, into ventral carpel bundles, which supplied ovules and end blindly. In the same way, the dorsal carpel bundles also end blindly. The trace of the ovules was single-bundle. The traces of the dorsal carpel bundles were single-bundled. There were 6 ovules, 2 ovules in each locule. Traces of stamens and traces of inner and outer tepals were single-bundled. The study of new features of vascular anatomy and flower morphology will make a significant contribution to the taxonomy of the Amaryllidaceae family.

Keywords: Allium sativum, Ovary, Vertical zones, Vascular bundle.

Introduction

The study of the *Amaryllidaceae* family, in particular, the location of genera in tribes and subfamilies is quite relevant (Chase et al. 2016; Jang et al. 2023). The internal structure features of the flower and the gynoecium of the family have not been sufficiently studied (Fishchuk 2021; Fishchuk 2021). That's why we examine *Allium sativum* vascular anatomy and flower morphology.

The family *Amaryllidaceae* consists of 3 subfamilies and 14 tribes (Chase et al. 2016). The subfamily *Allioideae* includes genus *Allium* (tribe *Allieae*, monogeneric, *Allium*). Scientists from Bangladesh studied the representative's morphology of the genus *Allium*. Analysis of 25 germplasms variance showed that garlic germplasm of different origins differed significantly in all characteristics (Salahuddin et al. 2019). Biochemical and botanical aspects of *Allium sativum* L. were studied in connection with the assessment of yield and suitability of selected varieties of Iranian garlic (*A. sativum* L.) (Ammarellou et al. 2022). In accordance with the agrotechnical requirements of garlic sowing, the morphology of garlic is studied and a dosing mechanism for garlic seeds with excellent sowing productivity is developed (Guo et al. 2021).

Materials and Methods

Plant material *Allium sativum* was collected in the agricultural station of Lesya Ukrainka Volyn National University of Lutsk and fixed in 70% alcohol. Fifteen flower buds were dehydrated in t-butanol series (20%, 30%, 50%, 70%, 100% - 2 h each, the last one - 24 h) and stored in 100% t-butanol and paraplast in the ratio 1:1. Infiltration was performed in Paraplast according to standard instructions Transverse sections of 20 µm thickness were obtained with a manual rotary microtome and according to staining protocol (Soukup and Tylová, 2019) stained in Safranin and Astra Blue. Slides were mounted in "Eukitt®". Images were obtained with an AMSCOPE 10MP digital camera attached to an AMSCOPE T490B-10M (USA) microscope. For the

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morphological analysis, measurements were made on at least 15 fresh flowers. We used the concept of gynoecium vertical zonality by W. Leinfellner (Leinfellner 1950) to analyze the gynoecium's internal structure. The height of the zones of gynoecium was measured according to the number of cross-sections. Vascular anatomy was studied on the series of flower cross sections.

Results

Allium sativum flowers were 0.2 cm-0.4 cm long, actinomorphic, white (Fig. 1. A). The peduncle was 50 cm-65 cm long, 1.2 cm in diameter at the base and 0.5 cm at the top, the inflorescence was a spherical umbrella with a large number of small flowers. There were two bracts, they were pyramidal, pointed, about 0.5 cm at the base and 2.5 cm wide, leathery, light brown. Pedicel up to 0.4 cm long, about 0.05 cm in diameter. The *A. sativum* perianth had six free tepals, the floral tube was absent, the outer tepals were 0.4 cm long and 0.1 cm wide, and the inner tepals were 0.2 cm long and 0.15 cm wide. *A. sativum* had six stamens, they were free. Internal stamen filaments, 0.025 cm long and 0.15 cm in diameter, external stamen filaments 0.025 cm long and 0.15 cm in diameter. Anthers of outer stamens were 0.125 cm long and 0.05 cm in diameter, anthers of inner stamens were 0.125 cm long and 0.075 cm in diameter. Anthers were attached to the stamen thread below the middle of the height of the anther. The *A. sativum* gynoecium was 0.05 cm long. The ovary was upper, 0.05 cm long and 0.075 cm in diameter, spherical, three-sided, white, which turned into a short white style 0.045 cm long and 0.013 cm in diameter. Stigma was cephalic, three-lobed, 0.005 cm long and 0.012 cm in diameter.



Figure 1. Ascending series of transversal sections of the flower *Allium sativum*. Scale bar 500 µm. A–B – pedicel; C–D – inferior ovary; E – synascidiate zone; F – symplicate zone; G – hemysymplicate zone; H – free tepals, anthers and style; an – anther; dv – dorsal vein; fi – filament; lo – ovary locule; ov – ovule; sn – septal nectary; st – style; te-tepal; vb – vascular bundle; vv – ventral vein

In the *Allium sativum* ovary there were three structural ovary zones: at the base of the locules there was a synascidiate zone of 100 μ m (Fig. 1. E), a symplicate structural zone of 100 μ m, which contains ovules (Fig. 1. F). above was the hemisymplicate zone, which occupies the upper part of the locules in height. This zone is the longest in the ovary - about 120 microns (Fig. 1. G). the asymplicate ovary zone was absent. The septal nectary appeared below the ovary locules and opened with septal slits at the base of the style, its total height was 380 μ m and was characterized by the presence of three septal nectary zones - the

separate zone, the "lilioid" united nectary zone and the external nectary zone. The ovary base was 220 microns. The ovary roof was 220 microns. The *A. sativum* peduncle contained 6 vascular bundles- 3 large and three small ones (Fig. 1. A-B), which branched out and form a circle of vascular bundles - a vascular cylinder (Fig. 1. C), from which at the level of the peduncle they departed six pairs of vascular bundles, traces of tepals, traces of stamens and above - traces of dorsal carpel bundles (Fig. 1. D). Stamens departed from the ovary wall.

In the center of the ovary, there remains a circle of small vascular bundles - the roots of the ventral complex, which were reorganized above, with the appearance of locules, into ventral vein (Fig. 1. E), which supplied ovules and end blindly. In the same way, the dorsal carpel bundles also end blindly. The trace of the ovules was single-bundle (Fig. 1.E). The traces of the dorsal carpel bundles were single-bundled. There were 6 ovules, 2 ovules in each locule (Fig. 1. G). Traces of stamens and traces of inner and outer tepals were single-bundled (Fig. 1.H).

Discussion

The flowers in the *genus Allium* had 2 ovules in each locule. Comparing the gynoecium and androecium of male-fertile and male-sterile *Allium cepa* lines, and analysis of the three-dimensional structure of the floral nectary were presented (Gonzalez et al. 2023). In *Allium sativum* gynoecium presence synascidiate, symplicate and hemisymplicate zones. In *Allium cepa* gynoecium were found synascidiate, symplicate and hemisymplicate zones (Fishchuk 2022). Scientists examined the taxonomic relationships between selected flower characteristics and a phylogenetic tree based on ITS sequences (Jang et al. 2023).

Conclusion

In the *Allium sativum* ovary there were three structural ovary zones: synascidiate zone, symplicate, hemisymplicate. The asymplicate ovary zone was absent. The septal nectary appeared below the ovary locules and opened with septal slits at the base of the style, and was characterized by the presence of three septal nectary zones- the separate zone, the "lilioid" united nectary zone and the external nectary zone. The *Allium sativum* peduncle contained 6 vascular bundles. In the center of the ovary was formed ventral carpel bundles which supplied ovules and end blindly. The traces of the dorsal carpel bundles were single-bundled. Traces of stamens and traces of inner and outer tepals were single-bundled. Our results confirm that floral morphology provides key taxonomic information to assess species delimitation in *Amaryllidaceae*.

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