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Floral Ontogeny in *Lathyrus latifolius* (*Fabaceae-Vicieae*)

By

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With 4 Figures

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Summary

PRENNER G. 2003. Floral ontogeny in *Lathyrus latifolius* (*Fabaceae-Vicieae*). – Phyton (Horn, Austria) 43 (2): 392–400, with 4 figures. – English with German summary.

Floral ontogeny of *Lathyrus latifolius* was studied by the use of scanning electron microscopy (SEM). No bracteoles are formed. The order of organ initiation differs from the acropetal mode in that, that the members of the calyx, corolla and outer stamen whorl overlap at the time of initiation. Furthermore, the carpel is precocious and arises together with the petal whorl.

The sepal whorl and both of the stamen whorls vary from the unidirectional mode, in that tendencies towards whorled organ initiation occur. In the calyx whorl the abaxial and the two lateral sepals are formed simultaneously, followed by the two adaxial sepals. Petal formation is unidirectional from the abaxial towards the adaxial side of the flower. In the episepalous stamen whorl the abaxial and the two lateral stamens are formed simultaneously, followed by the two adaxial stamens. The formation of the epipetalous stamen whorl starts after a developmental pause, in which the sepals, episepalous stamens and the carpel enlarge. The two abaxial epipetalous stamens are formed first, followed simultaneously by the two lateral and the adaxial stamen. All organs are exactly alternating with the previous organs, and the young floral bud is monosymmetric.

The remarkable asymmetry of the flower, the formation of two basal nectar windows, and the formation of the nectary on the abaxial side of the receptacle are consequences of the late floral ontogeny. The tendencies towards whorled organ formation in the sepal whorl and in both stamen whorls are seen as the result of harmonisation of the plastochrons within the whorls.

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Zusammenfassung

PRENNER G. 2003. Die Blütenentwicklung von *Lathyrus latifolius* (*Fabaceae-Vicieae*). – Phyton (Horn, Austria) 43 (2): 392–400, mit 12 Abbildungen. – Englisch mit deutscher Zusammenfassung.

Die Blütenentwicklung von *Lathyrus latifolius* wurde mittels Rasterelektronenmikroskopie (REM) untersucht. Es werden keine Vorblätter angelegt. Die Anlagenfolge der einzelnen Blütenorgane weicht vom akropetalen Modus ab, in dem die Organe des Kelches, der Krone und des äußeren Staubblattkreises überlappend angelegt werden. Desweiteren wird der Fruchtknoten simultan mit den episepalen Staubblättern angelegt.

Im Kelch und in beiden Staubblattkreisen weicht die Organanlage von der unidirektionalen Anlage ab. Hier wurden Tendenzen zu wirteliger Anlage gefunden. Im Kelch wird das abaxiale und die beiden lateralen Sepalen synchron angelegt. Die beiden adaxialen Sepalen werden wenig später sichtbar. Die Anlage der Krone erfolgt unidirektional von abaxial nach adaxial. Im äußeren Staubblattkreis wird das abaxiale und die beiden lateralen Staubblätter simultan angelegt. Die beiden adaxialen Stamen folgen kurze Zeit später. Die Anlage des inneren, epipetalen Staubblattkreises erfolgt erst nach einer Pause, in der sich die Kelchblattanlagen, die episepalten Stamen und die Anlage des Gynoeceum vergrößern. Die beiden abaxialen Stamen werden zuerst angelegt. Die lateralen und das adaxiale Stamen folgen simultan. Alle Organe werden exakt alternierend mit den vorangegangenen Organen angelegt, und die junge Blütenknospe weist dorsiventrale Symmetrie auf.

Die Ausbildung der deutlichen Asymmetrie der Blüten, der beiden Nektarfenster am Grund des Androeceums und die Bildung des Nektariums an der abaxialen Seite des Rezeptaculums erfolgen erst in der späten Blütenentwicklung. Die Tendenzen zur wirteligen Organanlage des Kelches und der beiden Staubblattkreise werden als das Resultat der Harmonisierung der Plastochrone in diesen Wirteln gedeutet.

1. Introduction

Lathyrus latifolius L. is a perennial climbing herb, distributed from central to southern and western Europe. *L. latifolius* has asymmetric flowers, which is characteristic for the section *Lathyrus* to which this species belongs (KUPICHA 1983, ASMUSSEN & LISTON 1998).

The asymmetric flowers of *Lathyrus* species drew attention in context to its pollination ecology: TEPPNER 1988 showed in *L. grandiflorus* specialised adaptations for the pollination by Xylocopa violacea, and WESTERKAMP 1993 showed in *L. latifolius* the influence of asymmetry in respect of pollination mainly by Megachile ericetorum.

Concerning ontogenetic aspects of the floral asymmetry, SCHÜEPP 1911 studied late developmental stages in *L. latifolius* flower buds. This study was supplemented by WESTERKAMP 1993, who also refers only to growth processes during the late floral ontogeny. TUCKER 1999 quotes that asymmetry within *Fabaceae* is a result of the late floral development. But a complete ontogenetic SEM-study of an asymmetric member of *Fabaceae* is still lacking. PAYER's 1857 studies on *L. sylvestris* are interpreted by TUCKER 1987 as unidirectional initiation of petals and whorled formation of stamens. SCHUMANN 1890 quotes the following order of organ initiation for different species of *Lathyrus*: sepals (unidirectional), followed by the episepalous stamens (unidirectional) plus carpel, and finally petals plus inner stamen whorl. *L. grandiflorus* is alleged to have unidirectional organ initiation in all whorls (TUCKER 1987), which is generally held to be the rule in *Fabaceae* (TUCKER 1984, 1987).

The aim of this study is to prove the above cited disagreements concerning the order of organ initiation, and to define the appearance of asymmetry in the floral ontogeny.

2. Material and Methods

Floral buds of different size of *Lathyrus latifolius* were collected on 14th of May, 2001 in the Mediterranean quarter of the Botanical Garden Graz (origin of the material unknown). The specimens were immediately fixed in FAA (5 parts formalin – 5 parts acetic acid – 90 parts 70% ethanol) and stored in 70% ethanol. Floral parts were dissected under a Leica MZ6 stereomicroscope in alcohol of the same concentration. For scanning electron microscopy (SEM) the specimen were dehydrated in formalindimethylacetal (FDA) for at least 24 hours, and afterwards critical-point dried with liquid CO_2 in a Polaron 7010 CPD. The dried specimens were mounted on aluminium stubs with nail polish. The buds were coated with gold in an Agar sputter coater. SEM studies and micrographs were taken in a Philips XL 30 ESEM at 20 kV at the Institute of Plant Physiology, Karl-Franzens-University Graz. All photographs show the buds with the abaxial subtending bract at the bottom of the image. In total 88 micrographs were taken and analysed.

3. Results

3.1. Organ Initiation

No bracteoles are formed, and the order of organ formation within a flower varies from the acropetal mode, in that the organs of the calyx, corolla and outer stamen whorl overlap in their initiation. The organs arise in very short intervals. The bare floral primordium is roundish first (Fig. 1a), and after abaxial and lateral enlargement, the initiation of sepals starts with synchronous formation of the abaxial and the two lateral sepals (Fig. 1b–c). After a short pause, in which the already formed sepals enlarge (Fig. 1d), the two adaxial sepals arise synchronously (Fig. 1e). When all five sepals are visible the floral diameter is about 260×240 µm.

Formation of petals is unidirectional from the abaxial towards the adaxial side of the flower: Parallel to the last formed sepals, the abaxial keel petals arise simultaneously (Fig. 1e). The two lateral wing petals are initiated shortly afterwards (Fig. 1e), and finally the adaxial standard is formed (Fig. 1f). At a floral diameter of about 280 µm all petal primordia are visible. Simultaneous to the initiation of the last petal, the primordium of the gynoecium becomes visible as a shallow bulge in the centre of the flower (Fig. 1f).

The initiation of the episepalous stamen whorl starts after all petal primordia and the carpel are visible. At first the abaxial and the two lateral stamens are formed synchronously (Fig. 1f). Immediately after them

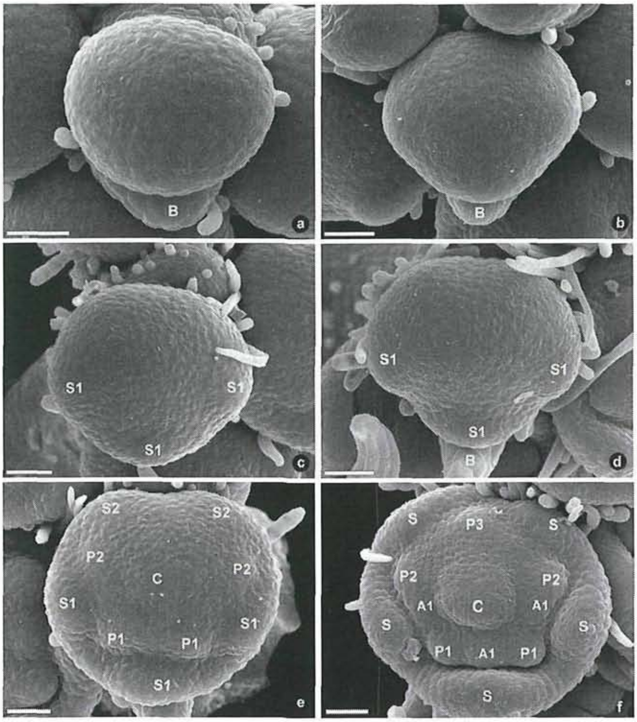

Fig. 1. *Lathyrus latifolius*, early floral ontogeny: a. Bare roundish floral primordium in the axil of a bract (B). b. Abaxial and lateral enlargement of the primordium. The floral bract remains small and no bracteoles are visible. c. Synchronous initiation of the abaxial sepal and the lateral pair of sepals (S1). d. Enlargement of the initiated sepals (S1). e. Synchronous initiation of the adaxial pair of sepals (S2) and the abaxial keel petals (P1). Immediately after the keel petals the lateral wing petals (P2) are formed synchronously, and the primordium of the carpel (C) becomes visible in the floral centre. f. Synchronous formation of the adaxial standard (P3) together with the abaxial and the lateral episepalous stamens (A1). – Scale bar = 50 µm.

the two adaxial stamens arise (Fig. 2a). At a floral diameter of c. 315 µm all episepalous stamens are visible.

The initiation of the epipetalous stamen whorl starts after a developmental pause, in which the sepals, the episepalous stamens and the gynoecium enlarge (Fig. 2b–c). This complicates the further observation, because the enlarged organs mask the epipetalous stamens. At first the two abaxial stamens arise synchronously (Fig. 2d). Shortly later the two lateral stamens and the adaxial stamen are formed synchronously too (Fig. 2e). At a floral diameter of about 370 µm all floral organs are visible. They are exactly alternating with the former organs, and the young floral bud is zygomorphic (Fig. 2e).

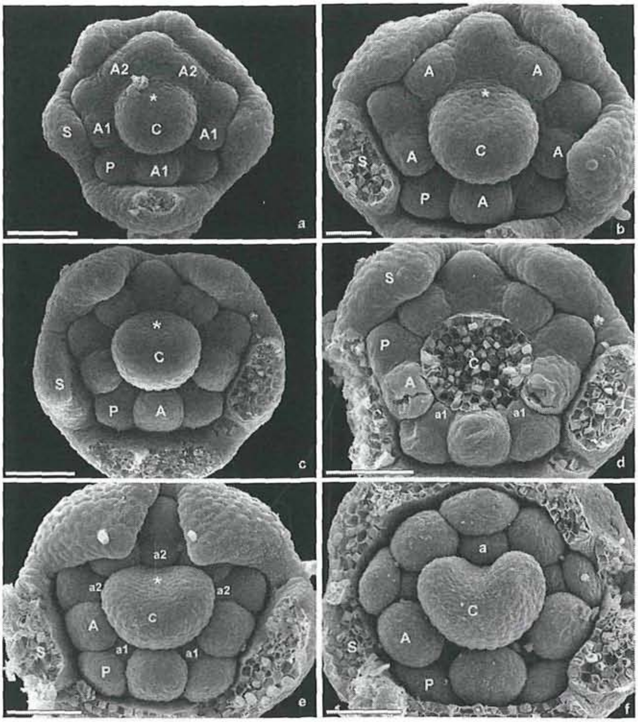

Fig. 2. *Lathyrus latifolius*, early floral ontogeny and developmental midstage: a. Simultaneous initiation of the last two episepalous stamens (A2). Note the adaxial depression on the carpellary primordium (asterisk). b. Developmental pause, in which the already initiated organs enlarge. c. Ongoing organ enlargement, particularly of the gynoecium and the episepalous stamens (abaxial and one lateral sepal removed). d. Same as c., gynoecium removed. Synchronous initiation of the abaxial epipetalous stamens (a1, abaxial and lateral sepals removed). e. Initiation of the lateral and the adaxial epipetalous stamens (a2). Note the distinct adaxial flattening of the carpellary primordium (asterisk, abaxial and lateral sepals removed). f. Enlargement of floral organs. The lateral margin of the carpellary primordium begins to bend upwards (sepals removed). – Scale bar = 50 µm in a–b, 100 µm in c–f.

3.2. Organ Enlargement and Differentiation

The enlargement of sepals does not take place very fast, so that the initiation of the petals and episepalous stamens is observable without additional preparations (Fig. 1e–f, 2a). Due to pronounced abaxial growth, the abaxial and the two lateral sepal lobes have to be removed for the observation of the abaxial epipetalous stamens (Fig. 2d).

The growth of the petal primordia is slow, and they soon are overtopped by the enlarging episepalous stamens (Fig. 2b–c). This organs also mask the inner stamen whorl, which therefore is a little bit hard to observe

(Fig. 2d–e, 3a–b). Late in floral ontogeny nine stamens fuse to an adaxially open sheath, to which the adaxial epipetalous stamen is tightly appressed (Fig. 3c, e–f). To the left and right of this stamen the filament sheath curves outwards basally, and two nectar windows are formed, through which nectar is accessible. The nectary is formed late in floral ontogeny, and is confined to the abaxial side of the receptacle, where nectar stomata are distributed symmetrically (Fig. 4a–b).

The carpel primordium enlarges fast. Parallel to the formation of the two adaxial episepalous stamens the formation of the adaxial cleft becomes visible as a shallow depression in the median plane (Fig. 2a). Due to decreased abaxial and increased adaxial growth, this depression enlarges and becomes remarkably broad during floral development (Fig. 2e–f, 3a). Later on, it forms the ventral suture of the pod (Fig. 3d), which is oriented exactly in the median plane. Finally an ovally shaped, papillate stigma is formed and the style becomes adaxially pilose.

The floral bud remains monosymmetric for a long period (Fig. 3a, c, e). Only at a length of about 6 mm the apical part of the androecium and gynoecium begins to bend towards one side, and the flower becomes more and more asymmetric (Fig. 3f).

4. Discussion

4.1. Overlapping Organ Initiation

The vertical order of organ initiation differs from the acropetal mode, which is frequently found in *Fabaceae* (TUCKER 1987): The two adaxial sepals are formed almost simultaneously with the abaxial petals, and the adaxial banner is formed together with three episepalous stamens. This overlapping of organ initiation between whorls is a common phenomenon of *Fabaceae* (TUCKER 1989, personal observations). Within *Vicieae* it has been found in *Lathyrus grandiflorus* and *Pisum sativum* (TUCKER 1989).

4.2. Modifications from the Unidirectional Organ Formation

Since the publications of TUCKER 1984 and 1987, unidirectional organ initiation from the abaxial towards the adaxial side of the flower is seen as the norm in *Fabaceae*. Exceptions have been documented rarely: TUCKER & STIRTON 1991 showed helical sepal formation in *Psoralea pinnata*, and KLITGAARD 1999 documented "modified helical initiation of sepals" in *Dalbergia brasiliensis*, and in *Pterocarpus rotundifolius* he found bidirectional formation of the epipetalous stamens. Unidirectional organ initiation sensu TUCKER 1984 means the stepwise initiation of organs or organ pairs within a whorl from the abaxial towards the adaxial side of the flower. In *L. latifolius* this mode of organ formation is only found in the petal whorl. In the calyx and in the outer stamen whorl the abaxial and the two lateral organs are formed simultaneously. In the inner stamen whorl

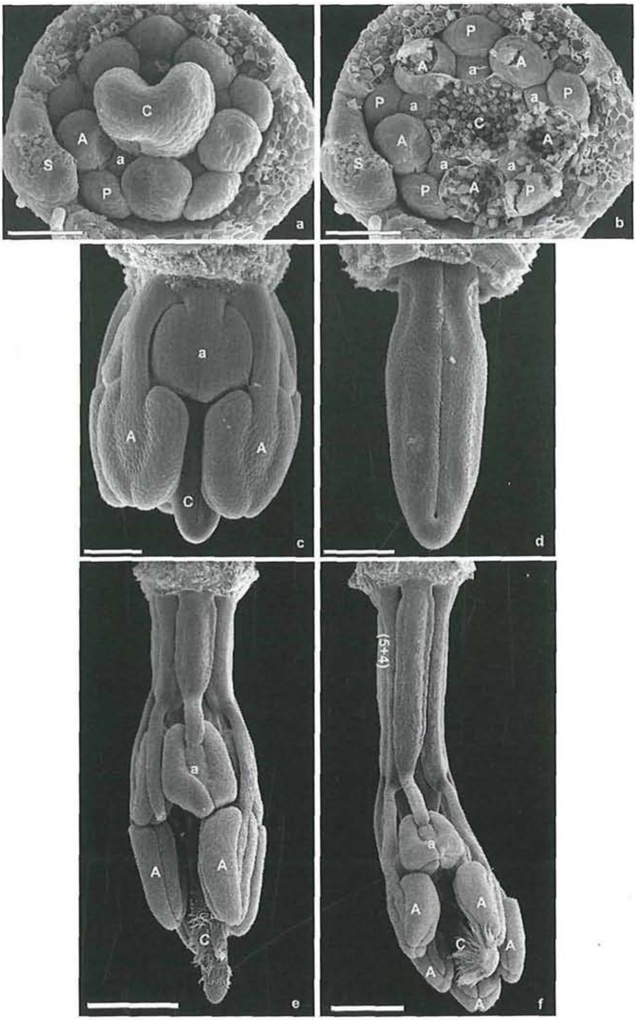

Fig. 3. *Lathyrus latifolius*, late floral development: a. Ongoing enlargement of floral organs. The carpellary cleft begins to close (sepals removed). b. Same as a., carpel removed in order to show the small primordia of the epipetalous stamens (a). c. Androecium and gynoecium out of a young floral bud. In the inner (a) and outer (A) stamen whorl anthers and filaments are formed, and the apex of the young carpel is visible (C). d. Same as c., stamens removed. The adaxial cleft of the carpel is closed and lies exactly in the median plane. e. Monosymmetric androecium and gynoecium out of a young floral bud, showing the free epipetalous stamen (a) in adaxial position (anther of laterally left stamen removed). f. Androecium and gynoecium out of an older floral bud. Asymmetry becomes evident due to bending to the right of the filaments and bending to the left of the style. – Scale bar = 100 µm in a–b, 200 µm in c–d, 1 mm in e–f.

the two lateral stamens and the adaxial stamen are initiated synchronously. These patterns are seen as tendencies towards whorled organ formation, and as a result of the harmonisation of the plastochrons within the organs of a whorl. While TUCKER 1989 stated that "simultaneous initiation is a condition rare or absent in papilionoids", I found tendencies towards whorled organ formation as well as whorled organ formation in itself frequently within *Fabaceae* (personal observations).

4.3. Symmetric Initials in an Asymmetric Flower

While in asymmetric flowers of *Caesalpiniaceae* asymmetry is manifested early in floral ontogeny (TUCKER 1999), I could show that the distinct asymmetry in the flowers of *L. latifolius* is solely a result of the latest floral ontogeny. The floral organs of *L. latifolius* are initiated strictly alternating, and the young floral bud is monosymmetric. During early developmental stages no evidence could be found for the highly asymmetric flower. Asymmetry emerges late in floral ontogeny, when the bud is about half the length of the anthetic flower. Basically the same holds true for *Vigna caracalla* (personal observation). With this results TUCKER's 1999 suggestion on the late development of asymmetry in *Fabaceae* can be proved.

4.4. Asymmetric Nectar Emission Versus Symmetric Nectary

According to WESTERKAMP 1993 the nectar windows at the androecium's base are of different relative dimensions, and the nectar droplet is often found only below the right access hole. This does not reflect the condition of the nectary, where nectar stomata are distributed symmetrically on the abaxial side of the floral receptacle.

5. Acknowledgments

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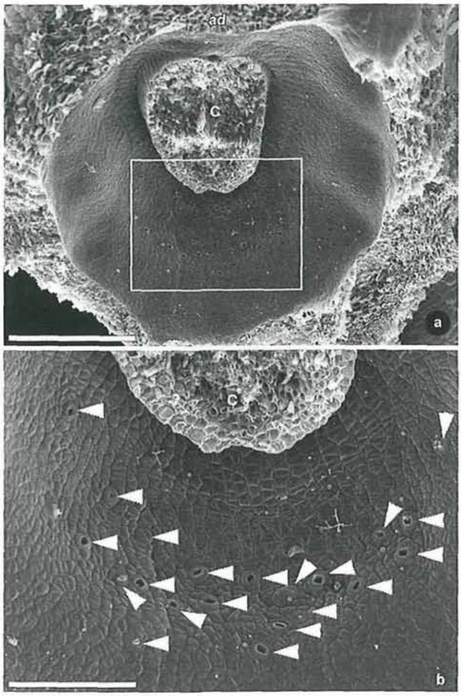

Fig. 4. *Lathyrus latifolius*, receptacle and nectary out of an anthetic flower: a. Overview of the receptacle. The rectangle marks the region, where nectar stomata are located. b. Marked rectangle out of a, showing the field of nectar stomata in abaxial position (arrowheads). Scale bar = 500 μm in a, 200 μm in b.

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