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On the Epidermal Elements of *Origanum calcaratum* JUSS. (*Labiatae*)

By

Theodoros G. VRACHNAKIS*)

With 5 figures

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Summary

VRACHNAKIS Th. G. 2002. On the epidermal elements of *Origanum calcaratum* JUSS. (*Labiatae*). – Phyton (Horn Austria) 42 (1): 39–67, with 5 figures. – English with German summary.

In the present work the aerial surface of *Origanum calcaratum* during its life-cycle is investigated. The morphology and distribution of the epidermal elements on the different organs are studied by light and scanning electron microscopy. One type of non-glandular hair and four types of glandular hair (one peltate and three capitate) with different sites, and functions are distinguished. The distinct peltate hairs are in abundance, hiding in the leafy stem, considered as the "back line" of defence. The morphological distinction between the capitate hairs is not clear, but in this study the following types are considered: Trichome-hydathodes, floral glandular trichomes, and stalked glandular hairs. Trichome-hydathodes are abundant in the early stage and in immature tissues, suggesting that they serve as active hydathodes. Floral glandular trichomes suggested as attractants predominate at the reproductive organs. The various stalked glandular hairs are the most common trichome type with a suggested multifunction. The non-glandular hairs are covering the plant tissues and particularly protect the peltate hairs. Besides the suggested specific functions of the trichomes their role in the epidermal cell fate is postulated. The differentiation of the cuticle may distinguish the pavement cell from the papillate-cell, however the universality of the cuticle in all the epidermal elements may contribute to the totipotency of the epidermal cell of *Origanum calcaratum*.

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*) Theodoros G. VRACHNAKIS, Institute of Plant Physiology, 8010 GRAZ, AUSTRIA. Present Address: School of Agricultural Technology, TEI of Crete Stavromenos, P.O. Box 140, GR-71500 Heraklion-Crete, Greece. e-mail: tvrachnakis@steg.teiher.gr

Zusammenfassung

VRACHNAKIS Th. G. 2002. Epidermis und Trichome von *Origanum calcaratum* Juss. (*Labiatae*) – Phyton (Horn, Austria) 42 (1): 39–67, mit 5 Abbildungen. – Englisch mit deutscher Zusammenfassung.

Die vorliegenden Studie befasst sich mit Oberflächenstrukturen auf oberirdischen Organen von *Origanum calcaratum* im Laufe der Entwicklung der Pflanze. Die Morphologie und Verteilung epidermaler Elemente wird mit Licht- und Rasterelektronenmikroskopie untersucht. Ein Haartyp ohne Drüsenfunktion und vier verschiedene Drüsenhaartypen (eine Drüsenschuppe und drei Drüsenhaare mit Köpfchen) mit unterschiedlicher Verteilung und Bedeutung werden beobachtet. Die reichlich vorhandenen Drüsenschuppen sind am Sproß verdeckt angeordnet und dienen grundsätzlich der Abwehr. Die Köpfchenhaare können anhand ihrer Morphologie allein nicht eindeutig zugeordnet werden, in dieser Studie werden jedoch folgende Typen unterschieden: Trichomhydathoden, florale Köpfchenhaare und gestielte Drüsenhaare. Trichomhydathoden sind im frühen Entwicklungsstadium und auf immaturen Geweben reichlich vorhanden, was nahe legt, dass es sich hiebei um aktive Hydathoden handelt. Florale Drüsenhaare, die der Anlockung dienen, überwiegen auf den Organen, die der Reproduktion dienen. Der häufigste Trichomtyp sind die verschiedenen gestielten Drüsenhaare, die mehrere Funktionen ausüben sollen. Haare ohne Drüsenfunktion dienen dem Schutz insbesondere der Drüsenschuppen. Neben den spezifischen Aufgaben der Trichome wird ihre Bedeutung in der Entwicklung der Epidermis als Einheit postuliert. Die Differenzierung der Cuticula kann eine gewöhnliche Epidermiszelle von einer Papillenzelle unterscheiden, aber das Vorhandensein einer Cuticula in allen epidermalen Elementen kann zur Totipotenz der Epidermiszelle von *Origanum calcaratum* beitragen.

Introduction

Origanum calcaratum JUSS. [syn: *O. tournefortii* AITON (TUTIN & al. 1972), *Amaracus tournefortii* (RECHINGER 1949), *Beltokon tourneforti(i)* (AITON) RAFINESQUE, *O. tournefortii* AITON var. *barbatum* VOGEL (cit. IETSWAART 1980)] is an aromatic subshrub of *Labiatae*. Nomenclature in this paper follows TURLAND & al. 1993. After a long period of being considered an endemic species of the island of Amorgos (Cyclades, S. Aegean, Greece), it has been found on Crete and other Cycladic islands (IETSWAART 1980). Later on, it is reported on the island of Khalki (Dodecanese, SE. Aegean) by CARLSTRÖM 1984. It belongs to the section Amaracus according to the monograph of the genus *Origanum* by IETSWAART 1980, closely related to "Cretan Dittany", *O. dictamnus* L. (endemic to Crete). Within this section (in a wider sense), it resembles other endemic species in this area: *O. symes* A. CARLSTRÖM on Symi (SE. Aegean), *O. vetteri* BRIQUET & BARBEY on Karpathos (Cretan area) and *O. scabrum* BOISS & HELDR. on Euboea and in the Peloponnese (CARLSTRÖM 1984). Due to its membranous-bracted character, it forms a natural assemblage with *O. amanum* POST, *O. rotundifolium* BOISS , *O. sipyleum* L., *O. libanoticum* BOISS and other *Amaracus* members (PATON 1994), floristic elements of the continental East Mediterranean.

The habit of *O. calcaratum* is confined in fissures on bare calcareous rock cliffs, alt. 0–500 m, as an obligated chasmophyte (KYPRIOTAKIS 1998). In Amorgos it is under the name "κεφαλοχορτο", i.e. head-herb (HELDREICH & MILIARAKIS 1909), due to its healing properties for headaches. It can be found in rock gardens, treated as an alpine house plant, by the name "Dittany of Amorgos" (ELLIOT 1966). It is commercially available in the U.S.A, cultivated along the Pacific Coast, where a typical Mediterranean climate prevails (TUCKER & ROLLINS 1989). Two artificial hybrids are known: *O. calcaratum* x *O. dictamnus* (IETSWAART 1980) and *Origanum* "Barbara Tingey": *O. calcaratum* x *O. rotundifolium* (STARLING 1981).

Origanum plants are prized for their odours and flavours and are of commercial interest because of the essential oils they produce (OHLOFF 1992, TUCKER & MACIARELLO 1994, LANGE & SCHIPPMANN 1997). Essential oils are secreted by the glandular trichomes and their morphology, structure, histochemistry and ecophysiological roles are under intense research (reviews by UPHOF 1962, RODRIGUEZ & al. 1984, HALLAHAN & GRAY 2000). Glandular and non-glandular trichomes are an integral element of the plant surface. All outgrowths from the epidermis are termed "trichomes" (ESAU 1953, JOHNSON 1975, FAHN 1979, WERKER 2000). The plant epidermis is a multifunctional tissue playing a central role to the health of the plant, controlling the access to the interior whilst also balancing interactions between the plant and its environment (GLOVER 2000).

The composition of the essential oils of aromatic *Origanum* species have been widely studied (LAWRENCE 1984, KOKKINI & VOKOU 1989, BASER & al. 1993, SKOULA & al. 1999), but much less information is available on the morphology and development of the trichomes on the different organs of these species (WERKER 1993, KAROUSOU 1995). BOSABALIDIS & TSEKOS 1984, investigated the glandular scale formation of *O. tournefortii* (*O. calcaratum*) and related *Origanum* species. BOSABALIDIS & TSEKOS 1982, BOSABALIDIS 1987, 1990, studied the leaf epidermis of *O. dictamnus*. MATTERN & VOGEL 1994 compared calyx with leaf glands of *Amaracus dictamnus* (*O. dictamnus*) and other Labiatae. These studies are limited to mature leaves, focused on the peltate glandular trichomes, characteristic within the Labiatae genera as the (main) site of the essential oils.

The present study deals with the aerial surface of *O. calcaratum* from seed germination until seed formation. The ecophysiological role of the trichomes as mediators between plant and environment and the factors affecting the fate, form, position and function of these epidermal elements are discussed.

Materials and Methods

Plant material

Flowering branches of *O. calcaratum* were collected from the most reliable sites of its distribution: Amorgos, near the village Langada on calcareous rock facing the sea, alt. 150 m and E. Crete: Sitia, above Roussa Ekklisia in fissures of the calcareous rock "Γαιτανα" (Figs. 1a, d), facing the sea, alt. 450 m (35°10′05″ N, 26°09′431″ E, GPS II Plus) in September 1999. Both sites confirmed the "epitheton" *calcaratum*. Seeds (nutlets) were extracted from their calyces and were germinated in containers under 12 h light/dark, 20 °C/15 °C, 70% RH, 180 μmol m^{-2} s^{-1} PAR (THANOS & DOUSI 1995) in growth chamber (Heraeus Vötsch) and in greenhouse conditions (Institut für Pflanzenphysiologie, University of Graz, Austria) on 1 December 1999. The conditions in the growth chamber were shifted to: 17 h light/7 h dark, 23 °C/17 °C, 55% RH on 30 March 2000. Containers from growth chamber and greenhouse were placed outside of the greenhouse area (47°04′862″ N & 15°27′391″ E, alt. 380 m) on 14 April 2000. A potting mix based on sand was used and irrigation was the only agriculture practice to the growing plants. For a comprehensive study, herbarium material (Amorgos, E. Crete origin) was provided by Dr. Z. Kypriotakis (T.E.I. of Crete), and commercial samples (E. Crete origin) from a local nursery: Iraklion, Crete. Voucher specimens are deposited in the author's herbarium (Institut für Pflanzenphysiologie, Graz), and plants can be found in *Hortus Botanicus Graecensis*.

Light microscopy (LM)

For light microscopy, free-hand sections of all plant's parts were observed under a Zeiss Axioplan Photomicroscope. Paraffin oil was used for hydrophilic secretions (HEINRICH 1973a) and Sudan III for the localisation of lipophilic substances (GREEN 1991).

Scanning electron microscopy (SEM)

Plant parts, fixed with 2.5% glutaraldehyde in 0.05 M cacodylate buffer at pH 7.2 for 4 h, were dehydrated in a graded acetone series, critical point dried with CO_2 and the specimens sputter-coated with a thin layer of gold (Agar Sputter Coater B7340). Observations were carried out on a Philips XL 30 ESEM, operating at 20 kV. Unfixed, natural dried material was observed using the environmental modus (ESEM).

Measurements

Measurements took place during the life-cycle of the plants. The number of the epidermal elements per mm^2 surface area was counted on SEM images and directly on LM at 400 × magnification. The size of cells and trichomes was measured and the number of trichome cells was determined on LM (1000 ×). Micromeasurements were confirmed using an image analysing system (Optimas 5.2 for Microsoft Windows, Optimas Corporation). Measurements took place at cotyledons, leaves and bracts. Data represent mean values of randomly chosen plant material of different origin (herbarium, wild, cultivated). Observations took place in vivo and under a Zeiss SV8 Stereomicroscope.

Results

Plant growth and development

The growth and development of *O. calcaratum* consists of: the seedling stage, followed by the (main) vegetative stage which is converted into the reproductive stage. The stages are not always distinct and overlapping is common to plants not growing in their natural environment: e.g. in humid Austrian summers.

The early stage is considered from seed germination until the appearance of the first order branching, on the first nodes (Fig. 1b). As the seedling emerges on germination, the hypocotyl and cotyledons, although looking glabrous, are pubescent (Fig. 2a). In hypocotyl, trichomes and trichomes initials (Fig. 2j) are distributed in a gradient from its base (root), up to the petioles of the cotyledons (Figs. 2f, g, h). At its base, first appear the capitate hairs (stalked glandular trichomes and trichome-hydathodes) (Fig. 2f). Later on, few non-glandular trichomes as well as peltate glandular trichomes and stomata, as few as one, can be found at hypocotyl top (Figs. 2e, g, i). The pavement cells are rectangular in shape, elongated parallel to the direction of hypocotyl elongation (Figs. 2f, g, i). As the cotyledons come out from the nutlet, with the seed coat attached to the cotyledons tips, trichomes distribution and development starts at the cotyledon's base, extending up to the cotyledon's tip (Figs. 2a, h). Capitate trichomes and stomata can be found in the abaxial (lower) cotyledon surface, while peltate glandular and non-glandular trichomes are absent (Figs. 2a, b, d). In the adaxial (upper) side, all types of trichomes are present, while stomata are very rare or absent (Figs. 2a, c and Table 1). Trichomes are present with undeveloped pavement cells. As the cotyledon matures, the rectangular pavement cells of young cotyledons develop to cells looking like the interlocking pieces of a jigsaw puzzle, simultaneous

Table 1.
Density of the epidermal elements on a mature cotyledon (ca. 12 mm^2), fully-expanded leaf (ca. 3 cm^2), and bract (ca. 0.6 cm^2) of *O. calcaratum* (per mm^2 area, n = 60).

	Peltate	Capitate	Non-glandular	Stomata
Lower cotyl.	–	60–80	–	140–200
Upper cotyl.-	2.8–3.2	80–110	70–96	–
Lower leaf	2.2–3.4	30–85	7–47	140–210
Upper leaf	2.2–3.4	42–96	12–60	60–120
Lower bract	–	120–240	–	30–80
Upper bract	0.6–0.9	120–240	–	0–12

Variation in the values is partly caused by the inhomogenous distribution of the epidermal elements on the plant surfaces (see also text).

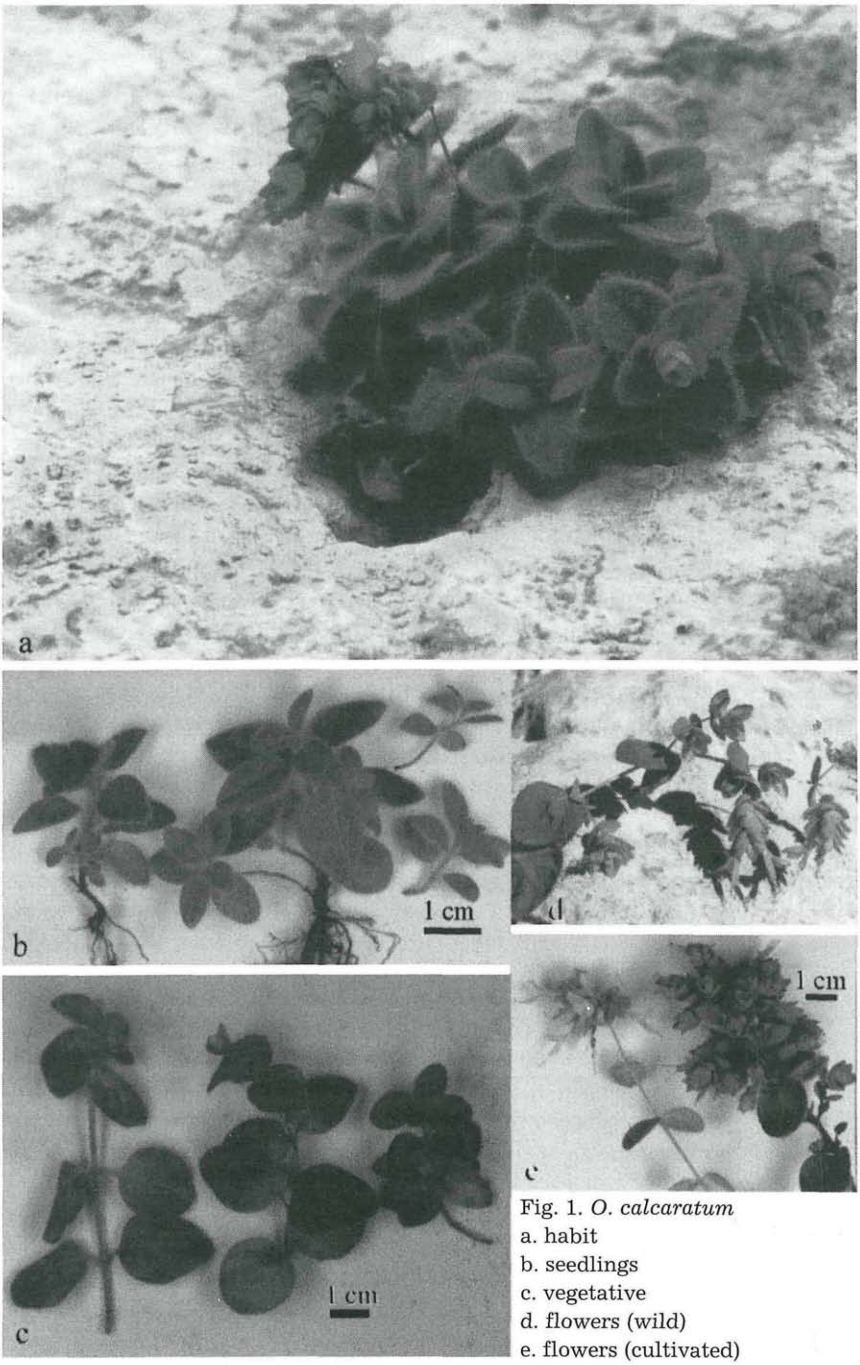

Fig. 1. *O. calcaratum*
a. habit
b. seedlings
c. vegetative
d. flowers (wild)
e. flowers (cultivated)

with stomata and peltate hairs (Figs. 2b, c, d). At the cotyledon tip an epithem-hydathode can be found. The size of the cotyledon varies, from that of an opened nutlet: 1.4 mm × 1.3 mm (*0.74) = 1.4 mm^2, to a big mature one: 4.6 × 5 (*0.75) = 17 mm^2. The presence of anthocyanins gives a bright pink-red appearance of the hypocotyl and the cotyledons (intense on abaxial side).

Within the *vegetative stage* a simultaneous growth of the main stem and the first order branches as individual stems, builds the bulk of the plant vegetation. A cylindrical form of the leafy stem is resulting from the *decussate* type phyllotaxy, with the leaf size increasing up to the middle of the stem (radical), and then decreasing upwards to the top (cauline) (Figs. 1a, b, c). As the apical meristem is differentiated to leaf primordia, (glandular, non-glandular) hairs and undeveloped pavement cells constitute the young leaf epidermis. With the leaf expanding, the trichomes initiate in a gradient of development starting at the leaf-apex and extends downwards to the leaf-base (Fig. 3a). In the abaxial side capitate hairs (mainly trichome-hydathodes) are concentrated on the base of the raised veins, petiole. Peltate hairs are located at the periphery of the lamina and on the veins sides, while in young leaves different stages of these hairs can be found at the leaf-base. The non-glandular hairs are protruding from the veins, leaf-margin and petiole. Stomata are abundant on the leaf-blade, although they are rare upon the veins and on the petiole (Figs. 3a, b and Table 1). In the adaxial side, with the veins and the petiole are not raised, all types of hairs and few stomata are uniformly-distributed on the leaf blade and the petiole. In young leaves the most upper part is devoid of peltate hairs, and different stages of these trichomes co-exist at the base (Figs. 3a, c, f and Table 1).

The leaf size depends on its age and position on the stem, reaching its maximum at the middle of the stem (ca. 6th node): 4.1 cm × 3.2 cm(*0.75) = 9.84 cm^2 (commercial), 3.2 × 2.4 = 5.76 cm^2 (wild), 2.6 × 2.2 = 4.29 cm^2 (experimental). In these "big", sessile leaves the indumentum of both sides is almost equal, and the dominance of the abaxial stomata is also minimised. The number of the epidermal elements per leaf varies as a function of the leaf level (order). Since this number increases up to stem's middle, and decrease up to the stem's tip, all the epidermal elements are differentiated on fully-expanded leaves. The pavement cells on the leaf blade are like in the cotyledons and those of elongated or protruding tissues (internodes, petioles, veins), as in the hypocotyl. In young leaves, an epithem hydathode can be found at the ending of the main vein and the leaf-margin is pigmented by anthocyanins. The indumentum of the internodes is similar to that of the leaves, predisposed the gradient of trichomes development at the upper node leaf. A replacement of the capitate trichomes by another "type" of trichomes is detectable in "old" radical leaves, is visible in the cauline leaves and it is manifested in the reproductive organs.

The reproductive stage

After six months of cultivation, the stem's tip is transformed to a spike (inflorescence), and from the nodes of the cauline stem a branching appears, forming a spike, or remaining as pairs of small leaves/bracts. The inflorescence, separated from the vegetative parts by an extended internode, comprises successive pairs of bracts subtending two flowers per verticillaster, attached with pedicel (stalk of individual flower) to the peduncle (main axis of the inflorescence). The form of a spike is like a leafy stem, with the bracts in the roles of the leaves (Figs. 1a, d, e). Within this stage all types of hairs occur, while the obvious change in the "type" of the capitate hairs, allows the name: floral glandular trichomes (Figs. 4h, i and 5c, d). Papillate-cells and a remarkable variegation characterise the floral organs. The development pattern of the epidermal elements (at least on bracts) is similar to that of the leaves.

In spite of the leaf like character of the bract, its indumentum resembles that of the cotyledon. The abaxial side lacks peltate and non-glandular trichomes, while it bears stomata. In the adaxial side stomata are rare, and few peltate trichomes can be found at the middle, downwards the bract-base. The "floral" trichomes in abundance on both sides, are concentrated at the bract-base, "replacing" the other types of capitate hairs (Table 1). The acute tip of the bract posses an epithem-hydathode, at the ending of the not-raised veins. The pavement cells are like in the leaves. Anthocyanin gives a brilliant red-purple colour to the bract-apex with the remainder part opalescent green. The bract has a membranous appearance with a distinct venation and its size, can be: 0.27–0.94 cm^2. The leaf/bract indumentum is intermediate of a cauline leaf and a bract.

In the outer calyx the following elements are observed: peltate trichomes on the sides of the raised veins, distributed from the middle downwards to the base (pedicel); few stalked glandular, non-glandular trichomes along the margin of the acute lips, and many "floral" trichomes,

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Fig. 2. The very early stage

a. abaxial, adaxial cotyledon, bar: 1 mm

b. abaxial, arrow: capitate, ar.head: peltate, bar: 50 µm

c. adaxial, arrow: initial, ar.head: peltate, bar: 100 µm

d. detail of b., arrow: stoma, bar: 10 µm

e. hypocotyl, arrow: non-glandular, thick arrow: capitate, ar.head: peltate, bar: 20 µm

f. hypocotyl base, bar: 200 µm

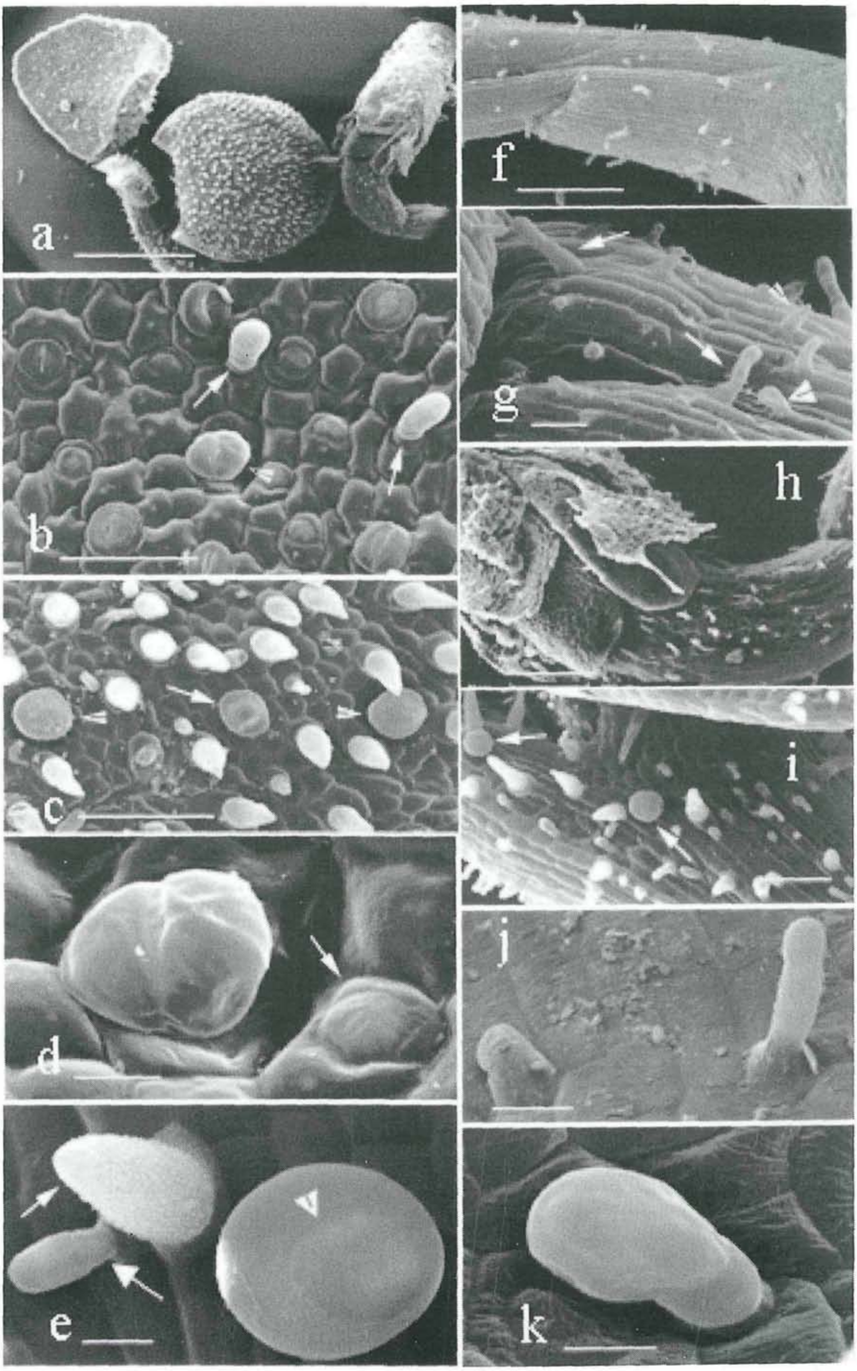
g. hypocotyl middle, arrow: capitate, ar.head: trichome- hydathode, thick ar.head: non-glandular, bar: 50 µm

h. cotyledon comes out from nutlet, bar: 200 µm

i. cotyledon petiole, arrow:peltate, bar: 100 µm

j. hypocotyl trichomes initials, bar: 20 µm

k. trichome-hydathode, bar: 10 µm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dense towards the base and in the pedicel (Figs. 4a, c, f and 5c). The inner calyx is characterised by a hairy throat of long 7–10celled non-glandular trichomes. It lacks peltate trichomes, while towards the base the "floral" trichomes are abundant (Figs. 4a, b, c and 5a). The acute tip(s) of the calyx posses epithem-hydathode(s). Stomata, pavement cells, as well as colour of the calyx is like in bracts. The pattern of development of the calyx itself and its epidermis revealed some discrepancies. Calyces with fused and not fused margins in the same verticillaster (floral node) and discord extending of the upper lips of the calyx were observed. Moreover, the development of the trichomes and the enclosed corollas in the calyces was found unclear.

When the corolla comes out of the calyx and opens out, it reveals a sparingly-pubescent epidermis characterised by the shape and variegation of all its components (Figs. 4d, e and 5a, n). On the outer corolla, capitate trichomes integrated with non-glandular trichomes are protruding from the upper lip lobes (Fig. 4m). Their clear location is presented only on closed corollas, where the upper lip lobes cover the lower ones (Fig. 5a). The remainder outer epidermis is hairless. The rectangular pavement cells of the corolla-tube, are transformed gradually to puzzle-like cells, up to the corolla lobes (Fig. 4m). In the sac-like protuberance (Fig.5a), the puzzle-like cells are more indented, while in the next located lobes the pavement cells are papillate. On the inner corolla, a number of integrated trichome types are distributed near the insertions of the staminate filaments and papillate-cells are present at the corolla lobes. The slightly elevated veins, distinct on the inner corolla, are ending as epithem-hydathodes or at the trichomes of the outer corolla lobes. Stomata are not observed on the corolla and its colour is pinkish to purple, intense in the papillate-cells.

The phanerantherous stamens, (two pairs of 2-lobed anthers) are ascending under the upper lip of the corolla (Figs. 4e and 5a). The anther lobes, joined by the connective tissue on the dorsal surface of the hairless filaments, present a glabrous epidermis. Capitate trichomes on the underside of the connective and one or two trichomes protruding from the edges of the pollen sacs are the only trichomes to be found on the anthers (Figs. 4e, n). Pavement cells of different forms is a characteristic feature of the stamen. They are rectangular-elongated on the filaments; rectangular

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Fig. 3. The leafy stage

a. abaxial, adaxial leaf, bar: 1mm

b. abaxial base, arrow:long non-glandular, ar.head: short non-glandular, bar: 100 μm

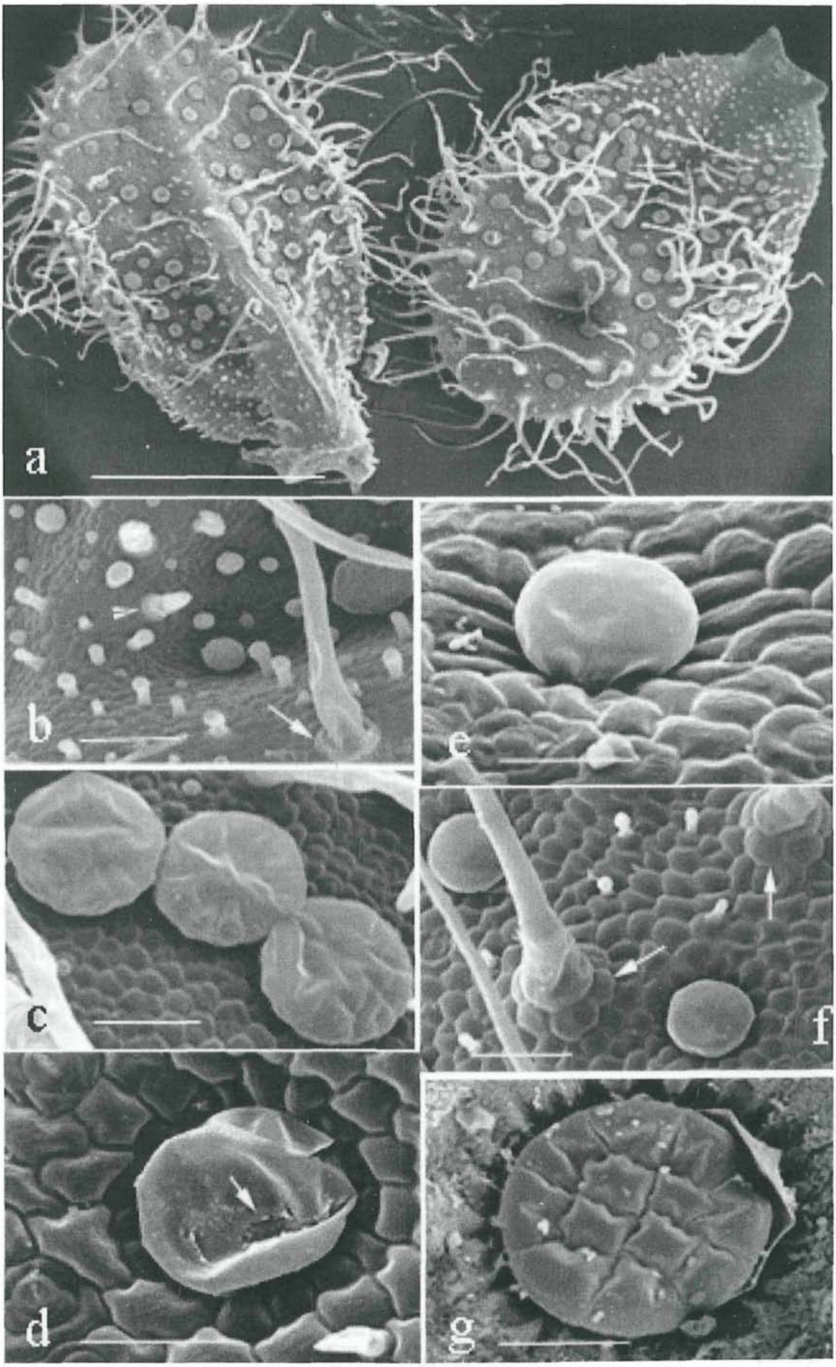
c. adaxial, three peltate, bar: 50 μm

d. abaxial, arrow: secreted material, bar: 50 μm

e. abaxial, sunken swollen peltate, bar: 50 μm

f. adaxial, arrow: pedestal of non-glandular trichome, bar: 100 μm

g. adaxial old laef, 12-celled(in turgor) peltate: bar: 50 μm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on the connective, differentiated to conical-papillate cells towards the anther stomium (*loculus*), where they are elongated further, forming a "toothed" ring around it (Figs. 4d, e, n and 5m, n). The style, which can protrude far beyond the stamens, lacks trichomes (Figs. 1d, e and 5a). The rectangular-elongated pavement cells of the style are differentiated to conical-papillate cells up to the stigmatic lobes, and then are transformed to smaller receptive papillae on the inner lips of the lobes (Fig. 5n). The papillosous ovary lacks trichomes, and papillae are not found on the ovules, unless the observations were unclear, due to the concentrated solutes (nectar?), at the basal portion of the style. The colour varies: deep purple for the conical-papillate cells, pinkish to opalescent for the rectangular-elongated cells, and opalescent to pale-brown for the stigmatic receptive papillae, anther connective. The pollen-grains are distinctly white, contrasting with the deep purple anther. The length of the style is noticeable, occasionally up to 4-fold the size of the approximately 10 mm long stamen. The epidermis of the peduncle, similar to that of the pedicel, is densely covered by "floral trichomes". The development of the corolla was found to be complex. At the same floral node, full opened corolla and closed corolla occurs from calyces of the same development. Inside corollae, the development of stamens varies among them, or compared with that of the style. Occasionally, two styles are present instead of the "normal" one. Cross-sections of closed corollas reveal that pollen grains are released from the pollen sacks and the stigmatic lobes bear receptive papillae, while a solute is apparent, concentrated around the opened *loculus* and the pollen-grains.

The epidermis as a unit, marks the changes between the stages of *O. calcaratum*. The changes are depicted as well as at the epidermal elements individually.

Fig. 4: The reproductive stage

a. calyx, arrow: hairy throat, ar. head: corolla, bar: 1mm

b. inner calyx, arrow:„floral“ trichome, bar: 500 µm

c. outer calyx, corolla, arrow: hairy throat, bar: 1000 µm

d. anther, arrow: connective, ar.head: papillate-cell, bar: 300 µm

e. pollen sacks, arrow:2 glandular tr., thick ar.: papillate-cell, ar.head: pollen, bar: 200 µm

f. calyx base, arrow: peltate, ar.head: "floral" trichome, bar: 200 µm

g. "floral" trichome, arrow: base cell, bar: 10 µm

h. ESEM: "floral" trichome; arrow: cuticle, ar.head: secretion, bar: 20 µm

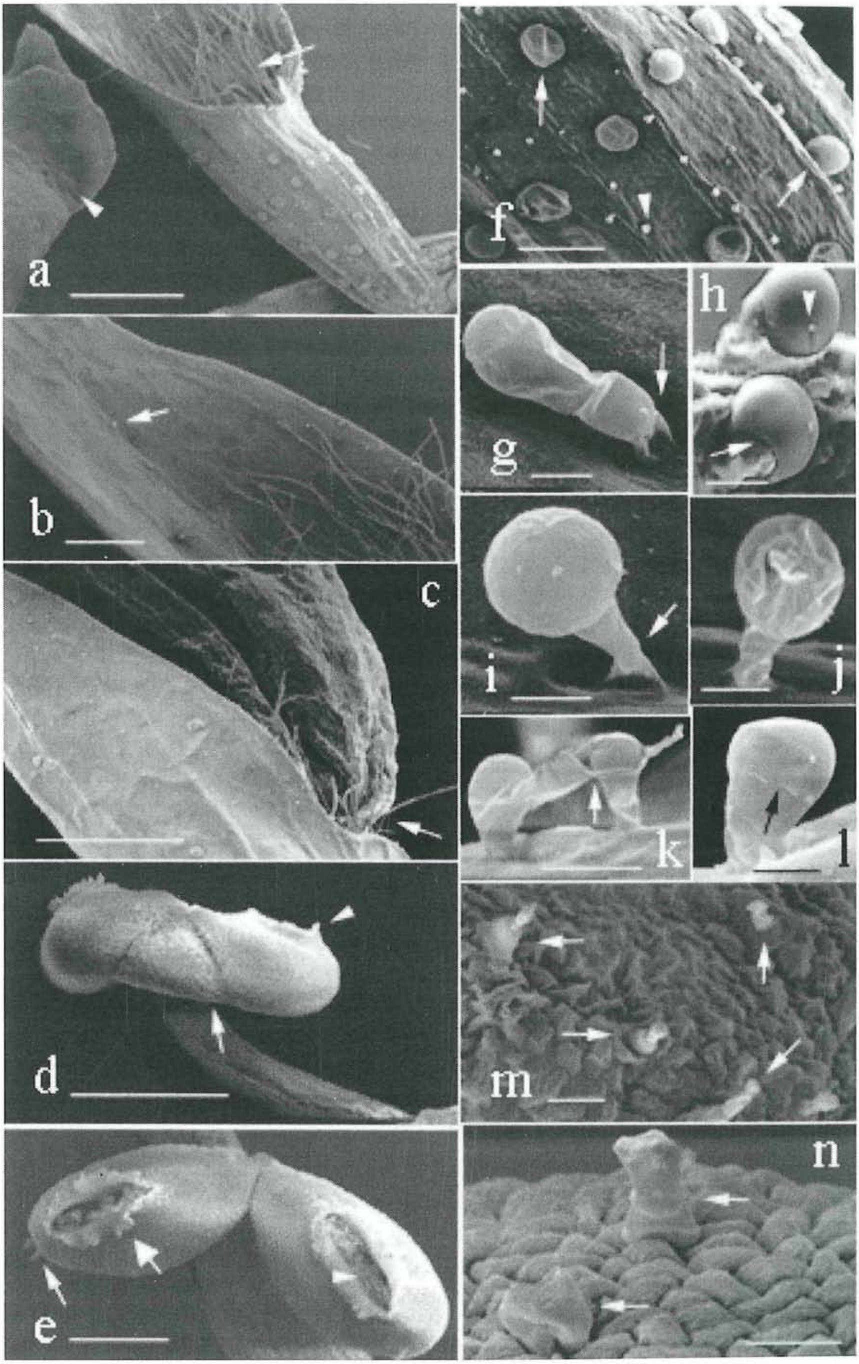
i. "floral" trichome, arrow: base cell, bar: 20 µm

j. collapsed "floral" trichome, bar: 10 µm

k., l. "floral" trichomes, arrow: cuticle detachment, bar: 20, 10 µm

m. outer corolla lobes, pappilate-cells, arrow: trichomes, bar: 50 µm

n. trichomes on papillosus anther sack, bar: 20 µm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Epidermal elements

The Glandular trichomes of *O. calcaratum* are distinguished between peltate and capitate.

Peltate glandular trichomes consist of a basal cell, a very short stalk cell and a large round head of up to twenty secretory cells arranged in two concentric circles. The diameter at the head of a 12–16 celled head trichome is 90 µm (± 10) – 110 µm (± 10) (Figs. 3g and 5b, h, k). In SEM micrographs, the heads show a smooth or wrinkled surface, revealing the presence of a large subcuticular space formed by detachment of the cuticle together with an outer part of the cell wall, or emphasizes the head-cells outlines, due to close attachment of the cuticle to the upper cell walls (Figs. 2d, e and 3c, e, f). Peltate hairs with torn cuticle reveals either collapsed or turgid head cells (Figs. 3d, g). Under LM, the head is opaque to hyaline, with the subcuticular space either homogeneously filled with the material, or emulsified by numerous coloured droplets constituting another phase (Figs. 5b, h). With Sudan III, pale-pinkish droplets in few cases can be seen in long time immersed material (Fig. 5k). When using paraffin oil, occasionally small hydrophilic droplets were observed to be secreted out of the head (Fig. 5e). Under the stereoscope or with unaided-eye the colour of the head varies from opalescent/pale yellow to orange/red-brown (the latter is common to herbarium material). The colour is not always related with the size or cell number of the head or the location of the trichome. The peltate hairs are often sunken to various degrees into epidermis, forming a ring of half-sunken peribasal cells (Fig 3e, d). The degree of sinking, as well the number of the peribasal cells depends on the form, size of the neighbouring epidermal cells and/or the underneath tissue. On the leaf blade, peltate hairs are sunken, "forming" more peribasal cells than on non flattened tissues (vein sides, adaxial petiole, internodes and calyces), where the peltate hairs are not sunken (Figs. 2d, e, i; 3c, d, e, f and 4f). A peltate hair on the abaxial leaf-side can be surrounded by more peribasal cells than its "counterpart" on the adaxial one. Nevertheless, on a flat tissue a 12–16 celled-head peltate hair can be surrounded by equal in

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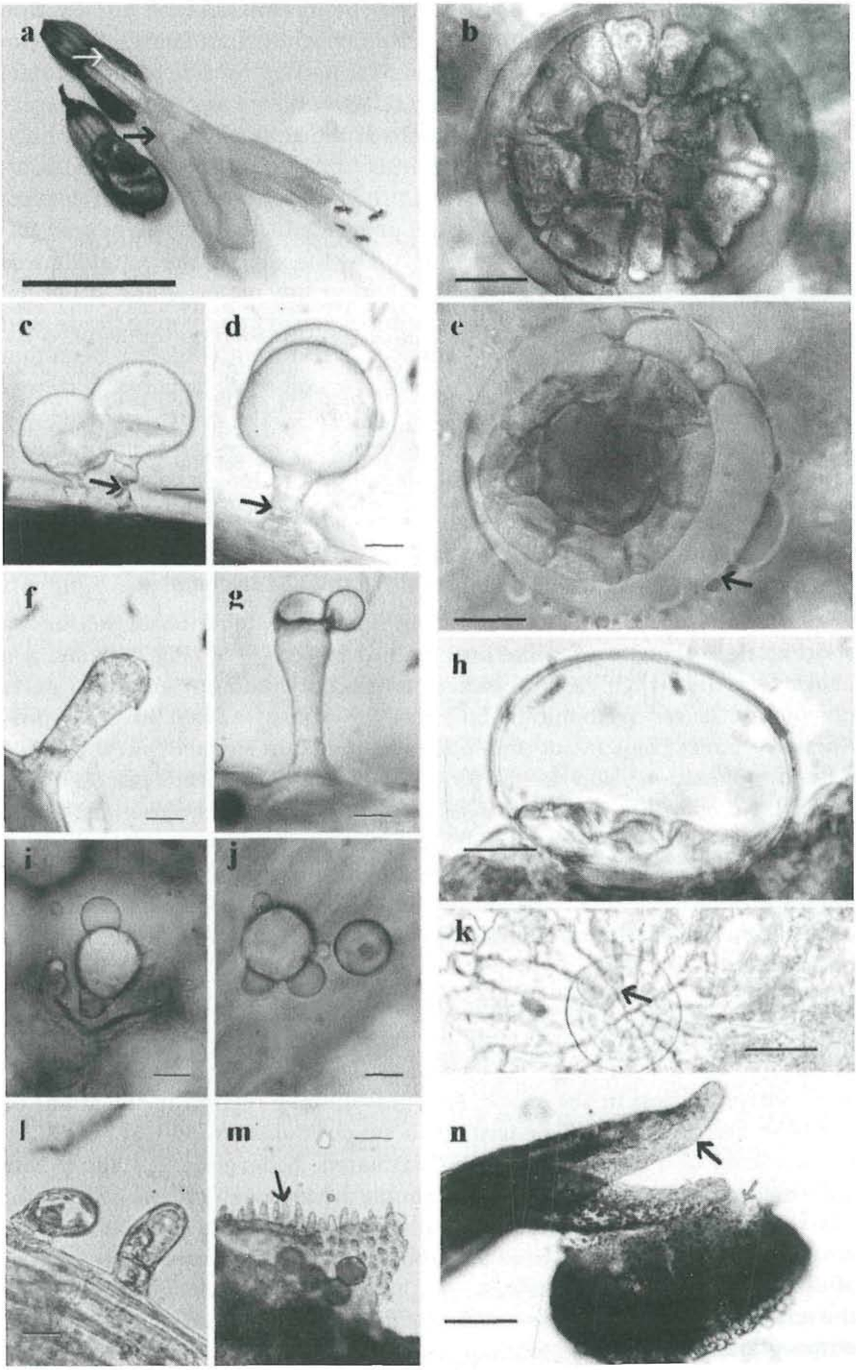
Fig. 5. a. closed, opened corolla, black arrow: "sack", white arr.: hairy throat, bar: 1 cm
b. 16-celled peltate, bar: 25 µm
c., d. „"floral" trichomes, arrow: base, bar: 10 µm
e. peltate, arrow: droplets, bar: 25 µm
f., g, stalked glandular trichomes, bar: 10 µm
h. "hyaline" peltate, bar: 25 µm
i., j. trichome-hydathodes secreting, bar: 10 µm
k. 14-celled peltate, arrow: Sudan III, bar: 50 µm
l. capitate glandular trichomes (tr.-hydathode, stalked glandular tr.), bar: 10 µm
m. conical papillate-cells on the pollen sack, bar: 10 µm
n. inner stigmatic papillae (black arrow), conical papillae (white arr.), bar: 100 µm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number or more peribasal cells. Apart from being sunken, their distribution on the plant surface exhibits a character for protection. Peltate hairs can be found at sheltered locations: grooves formed by infolds of the abaxial leaf margins, sides of protruded veins, flatly petiole, inner bract. They are rare on protruding tissues: veins, petiole of the abaxial leaf side and hairless tissues: abaxial cotyledon, outer bract, and corolla-complex (Table 1). Their presence on displayed tissues: indernodes, and adaxial leaves/ petioles is always associated with the presence of the non-glandular trichomes (Figs. 2a, b, c, e, i; 3a, f and 4a, f). The life span of the peltate hair is unknown, since, even in 12 year old herbarium material the spherical head, due to accumulated material, is similar to that of a young tissue, and the broken cuticle reveals the secretory cells in turgor (Fig. 3g). Secretion of lipophilic material out of the "cuticular" membrane, or pores on it, was not observed. The occasional ruptured or torn of the cuticular layer was caused by artificial damage. Undamaged peltate hairs can even be found after a heavy rain. Cuticular disruption can occur along a line of apparent weakness in the connection of the stalk and the head (Fig. 3g).

The capitate glandular trichomes are treated as: trichome-hydathodes, stalked glandular trichomes and "floral" glandular trichomes.

Trichome-hydathodes consist of an almost invisible basal cell and a short stalk bearing a pear-like head cell of 15 µm (± 5) (Fig. 2k); and are characterised by their early presence on hypocotyl and on "growing" parts of young tissues. Trichome-hydathodes are almost absent in the reproductive organs. They do not protrude from the plant surface (but they have a bending position), and do not form a subcuticular space (Figs. 2b, k; 3b and 5l). No coloration was observed under the LM, in any case (unstained or using Sudan III). Its unique character is showed with the use of paraffin oil, with distinct droplets of aqueous/hydrophilic material secreted in almost any case, obviously in well watered plants (Figs. 5i,j). The life span of the trichome-hydathodes is short, and their detection in dry material is not easy.

Under the name *stalked glandular trichomes* several forms of glandular trichomes with a distinct stalk are included (Figs. 2g, e, j and 5f, g, l). The size of the basal cell varies, as well as the number and size of the stalk cells. The variation in the shape, size (absolute or related to the stalk) of the head-cell, either with or without a subcuticular space, did not allow the maturity of a stalked hair to be estimated. A developing stalked hair does not differ from a developing trichome-hydathode, and the difference of a stalked hair forming subcuticular space from a "floral" one is only the size of the "head". At the same location of the plant tissue different forms of stalked hairs can be found. Inside the corolla tube and in the edges of the anthers, the variation of a stalked trichome makes the distinction from a non-glandular trichome without a difference (Fig. 4m). The stalked trichomes are the most common trichomes (not always the most numerous), and are protruding from all plant surfaces. Under LM no specific coloration was detected, with the "head" opaque to hyaline, occasionally with a "foamy" appearance of an emulsion with different size droplets (Fig. 5f). The reaction with Sudan III and paraffin oil is similar to that of the peltate trichomes (Fig. 5g). The short life span is evident due to collapsed (not always) stalked trichomes in dry material.

"Floral" glandular trichomes are characterised by the big "cuticular" sphere of the head-cell, which can reach 35 μm in diameter (Figs. 4i, h and 5c). In surface view, a "floral" trichome resembles a small peltate one, due to a short, very narrow stalk cell and to an "invisible" basal cell (Figs. 4i and 5c, d). This impression is caused by the optical "emptiness" of the basal cell in trichomes with swollen head. Thus, the whole trichome looks suspended from the tissue and movable (Figs. 4g, i, h and 5c, d). SEM micrographs of successive developmental stages reveal a "pump-like" nature of the "floral" hairs. The basal cell collapses with a simultaneous swelling of the stalk cell (Fig. 4g). In succession, the subcuticular space of the head cell is filled with the material and a simultaneous collapsing of the subtending cells occurs (Fig. 4i). The secreted material in the subcuticular space seems to be sucked and transported to the head cell via the stalk cell, at the expense of the basal cell. "Floral" trichomes can often be seen entirely collapsed (Fig. 4j) or with the cuticle unstuck (Fig. 4l), revealing the temporariness of the accumulated secretion in the "head". Cuticle detachment occurs only at the upper region of the glandular head, while towards the base the cuticle adheres closely to the cell wall (Figs. 4h, k, l). If the cuticle detached, in SEM micrographs the globose head cell with a narrow basal part is evident, with the whole floral trichome resembling a trichome-hydathode (Fig. 4l). The distribution of the floral trichomes on the different tissues did not exhibit a character for protection. Under LM, the head seems to be filled with a homogenous (rather than emulsified) opalescent to pale-yellow secretion. The reaction with Sudan III or paraffin oil was not so clear due to the "fragile" character of the trichome and the interference of the secretion with solutes of other origin (nectar). Occasionally, Sudan III intensified the peripheral "cuticular" line, while, using paraffin oil, solutes in the size of the head-sphere were secreted and interfered with it (Fig. 5d). The short life span of the "flower" trichome is evident, whilst the sphere is "shining" in carefully dried material.

The *non-glandular trichomes* are considered of one type. In cotyledons one-celled hairs appear as conical papillae, due to their form, size, and to the rough ornamented surface with cuticular warts (Figs. 2c, e). The non-glandular trichome of *O. calcaratum* is unbranched, uniseriate, unicellular to multicellular, pointed, distinctly articulated between cells, erect or leaning from the plant organs. The surface is covered with cuticular ornamentation, while on long, 8–14 celled hairs the surface appears smooth (Figs. 3b, f and 4a). The length of a 14-celled hair can reach 1.7 mm. The multicellular hairs are supported by a cellular pedestal (socket cells) formed from elevated epidermal cells arranged in a circle around the base (Fig. 3f). The number and the degree of elevation of pedestal cells follows the rules of the sunken peribasal cells of the peltate hair. A 10-celled hair can be surrounded by equal in number (or more) pedestal cells. The non-glandular hairs do not terminate in a gland and their development is actually a growth of almost equal size (length) cells. Therefore, predictions of maturation are relative. In the early plant development, the number of hair-cells (hence the size) is increasing with increasing order leaves. Later on, this rule is disturbed, since non-glandular hairs with the same number of cells, but extremely different in size (5-fold), can be found at the leaf base. This phenomenon is associated with the presence of peltate hairs of various development (Fig. 3b). In the reproductive stage the trichomes in the calyx throat are uniform (Figs. 4a, b, c), but those at the corolla are out of rules. Non-glandular hairs are in abundance on the leafy stem, are present in specific places on reproductive organs, and their presence is related with the presence of glandular hairs (Figs. 2a, c, e; 3a, b, c; 4b, m and Table1). Their rules of distribution on the different plant organs, timing and spacing are similar to those of the peltate hairs, indicating the relation between these two different trichome types. In young tissues, the vacuole of the basal-cell of the hyaline (opalescence) non-glandular trichome is often coloured by anthocyanins. The unclear life span of the non-glandular hairs seems to be not important for the plant itself, since even dead, they remain on the leaf and can be functional as a protection.

The epidermis of *O. calcaratum*, except the hypocotyl, adaxial cotyledon and corolla complex, is interrupted by *stomata* of *paracytic* type (Fig 2b, c, f, i; 3d, e and Table 1). They are in abundance on the abaxial sides, either elevated (leaf), or not (bract, calyx). They appeared later than the trichomes and are developed simultaneously with the pavement cells, peltate hairs, and non-glandular hairs (Figs. 2b, d). On young tissues, an increasing number of stomata is related with a decreasing number of other "space consuming" epidermal elements: peltate and non-glandular hairs. Opened and closed stomata, on the same leaf side, are observed under LM, SEM.

Epithem-hydathodes (passive-hydathodes) were observed in most of the plant's organs, as vein terminations. Under LM they can be observed on the abaxial side, obviously on young tissues. Guttation was not observed in well watered plants, and the use of paraffin oil did not exhibit any secretion.

Although *papillae* can been classified as trichomes, they are treated in this study as *papillate-cells*, differentiations of ordinary epidermal cells, confined to the reproductive organs.

The *pavement cell* (ordinary cell) is the most frequently occurring cell type in the epidermis of *O. calcaratum*. In surface view a developed one is either puzzle-like for flat surfaces, or rectangular elongated for elongated tissues (Figs. 2b, g and 3d, e, f). This is the rule in the vegetative stage, with the pavement cell of the abaxial side smaller and more indented than its "counterpart" of the adaxial one (ca. 1485 μm^2 and ca. 2503 μm^2, respectively). In the floral organs, pavement cells with thick cuticular striations (papillate-cells) are apparent (Figs. 4m, n and 5m, n). This change in the cell shape is to be seen also as change in coloration and was observed to be massive (as a group of cells), served specific needs. Pollen grains, land on and adhere only to the inner lips of the stigmatic lobes, where the papillate cells are different in form and colour from those of the outer lips (Fig. 5n). The rectangular cells of the anther's connective (Fig. 4d), are suspected to be involved in the observed in vivo motion of the anther. Disagreements were observed in the gradient of pavement cells differentiation. The puzzle-like cells of the sac-like corolla's protuberance were in "discontinuity" with the neighbouring papillate-cells of the outer corolla lobes, since papillate-cells are uncommon on the outer corolla. Field observations of open corollae showed that the enfolded lobes are functioning as inner corolla epidermis (where papillate-cells is the rule). The sack-like protuberance can serve as a landing site for a pollinator, thus a strong tissue (puzzle-like cells) is needed. The uniform "papillosous" epidermis of the pollen sacks can be interrupted from 1 or 2 distinctly protruding trichomes (Fig. 4e, n). Papillate-cells are able for secretion, as the receptive papillae of the stigmatic lobes and the papillate cells of the *tapetum*.

All the epidermal elements exhibit an "individual" character, sensitive to any change in the life cycle of *O. calcaratum*, with one common feature, the presence of the cuticle.

Discussion

Besides the evident small size of *O. calcaratum* and despite the stability of its characteristics in all the examined ecotypes, the observed gross phenology partly agrees with IETSWAART 1980. Branching from the lower stem nodes is not mentioned by this author, while vegetative branching in the upper stem (inflorescences in this study) is mentioned. Field observations reveal the role of the lower branches as reserve stems, in cases of pruning or browsing from goats. The 35 pairs of leaves per stem mentioned by the same author are in disagreement with our study in which approximately 12 pairs of leaves per stem were observed in *O. calcaratum* and related *Origanum* species: *O. dictamnus, O. saccatum, O. rotundifolium, O. scabrum, O. vetteri, O. symes* (IETSWAART 1980, CARLSTRÖM 1984), as well as with the limited vegetative growth period of 5-6 months in nature of the *xerophytes* (MARGARIS & VOKOU 1982). The observed hairy calyx throat is in agreement with VOGEL 1841 (cit. IETSWAART 1980), CARLSTRÖM 1984, TUCKER & ROLLINS 1989 (used it as a taxonomic marker). However partly disagrees with IETSWAART 1980, who does not recognise this characteristic for *O. calcaratum*.

It is noteworthy that in the early stage of the plant development trichome-hydathodes appear earlier in the hypocotyl than the non-glandular hairs and stomata. The early formation and function of the hydathodes in immature organs is reported by FAHN 1979. SMALL & BROOKES 1986 suggest a defensive role (against insects) for the same morphologically trichomes on the otherwise hairless cotyledonary petioles of some *Leguminosae*. In *O. calcaratum* the potential protective non-glandular and peltate glandular hairs appear later. The present capitate hairs are considered unable to produce defensive monoterpenes as reported for the young cotyledons of *Monarda fistulosa* (HEINRICH & al. 1983), as well as at an early stage of *Mentha x piperita* by BRUN & al. 1991, and in thyme seedlings by YAMAURA & al. 1992. Trichome-hydathodes are a common type of capitate glandular trichomes within *Labiatae*: ZIEGENSPECK 1949 reported the presence of "Trichom hydathoden" in all the 59 *Labiatae* investigated. They correspond to the capitate hairs type I (short stalked) and salt glands (WERKER & al. 1985 and WERKER 2000). The observed hydrophilic nature of their secretion agrees with that of: *Monarda fistulosa*, *Mentha piperita* (HEINRICH 1973a, 1977); *Teucrium* (BINI-MALECI & SERVETTAZ 1991); *Scutellaria altissima* (THALER & al. 1992); *Melissa officinalis* (SCHULTZE & al. 1992); *Nepeta racemosa* (BOURETT & al. 1994) and *Salvia officinalis* (CORSI & BOTTEGA 1999). This finding disagrees with earlier report for lipophilic secretion in *Mentha piperita* (AMELUNXEN 1964), and both lipophilic+hydrophilic) secretion in some *Labiatae* (WERKER & al. 1985). This type of capitate hairs must be related with the water status, essential in young growing tissues explaining their presence in the hypocotyl and immature tissues, before the development of stomata.

The absence of the non-glandular hairs from the hypocotyl and abaxial cotyledon suggests that protection must be attributed to the other mechanism of the young plant. CHIEN & SUSSEX 1996 reported the absence of non-glandular protective hairs on the hypocotyl, cotyledons and abaxial sides of the first-formed leaves of the otherwise hairy *Arabidopsis thaliana*. The presence of anthocyanins, intense in the (almost) glabrous abaxial cotyledon, can be the protective mechanism of young seedlings. The red colour of a tissue is less attractive than a chlorophyllous green one for the potential herbivores (LAMBERS & al. 1998), and anthocyanin can be a feeding deterrent (HOLTON & CORNISH 1995).

The leaf indumentum and its development in *O. calcaratum* is similar to that of other *Labiatae*. Increasing "hairiness" with increasing leaf size up to the middle-stem and successive decreasing up to the stem's tip is reported also for: *Monarda fistulosa* (KLINK 1971, HEINRICH 1973), *Mentha* *x piperita* (BRUN & al. 1991), and *Plectrannthus ornatus* (ASCENSAO & al. 1999). The presence of trichomes on leaf primordia and their gradient of development starting at the leaf-apex extending down to the leaf-base is the rule in *Labiatae*: *Mentha x piperita* (BRUN & al. 1991); *Ocimum basilicum* (WERKER & al. 1993); *Nepeta racemosa* (BOURETT & al. 1994); *Leonotis leonorus, Plectrannthus ornatus* (ASCENSAO & al. 1995, 1999). The trichome types of the leaf epidermis in *O. calcaratum* are identical to those reported for *Monarda fistulosa* (HEINRICH 1973), *O. syriacum* (DUDAI & al. 1988), and *O. vulgare* (WERKER & al. 1985a). Peltate glandular trichomes were found with the head consisting of up to 20 cells, while BOSABALIDIS & TSEKOS 1984 reported 12 celled heads. The density of the peltate hairs on developed leaves agrees with that of IETSWAART 1980. The head coloration irrespective of the development or the location of the peltate hairs and the occasionally "foamy" appearance of its contents integrated with the reaction with paraffin oil and Sudan III manifest the individuality of each trichome and the different nature of the secreted material. WERKER 2000 points out that structurally similar trichomes can produce different materials as for instance, the peltate hairs in *O. x intercedens* (BOSABALIDIS & SKOULA 1998) and multicellular trichomes function as miniature organs (GLOVER & MARTIN 2000). Lipophilic nature of the secreted material is reported by SKOULA & al. 1999 and hydrophylic one by HUSAIN & al. 1982. BOSABALIDIS & al. 1998 reported the same "foamy" appearance of the heads in peltate hairs of *O. x intercedens*. The character for protection of the peltate hairs as well as their long (unknown) life-span and the absence of evident secretion of lipophilic material out of the cuticular membrane is common in *Labiatae* (the latest controversial, see WERKER 2000 and references therein). The potential defensive role of the peltate hairs is restricted against those herbivores that can reach the (plant surface) covered by the non glandular hairs and they are able to penetrate the cuticular membrane. For these trichomes DUKE 1994 points out that there is not evidence for a direct function as a physical impedance to insects. Due to the phytoxicity of their contents (BROWN & al. 1987, DUKE 1994), the peltate hairs seem to be not the "first line of defence" (KELSEY & al. 1984) against the phytophagous enemies, but the "back line" or the rearguard, since they are lethal for the plant bearing them. Under this scope the hidden character of the peltate hairs and their relation with the non-glandular ones can be explained. The "peculiar" co-existence of variously developed non-glandular trichomes in the leaf-base where variously developed peltate hairs also co-exist can be attributed to their function in providing protection to the leaf from its own peltate hairs. Besides the "traditional" function of the non-glandular hairs to protect the leaf (covering hairs: PETERSON & VERMEER 1984) from the environment, the protection of the peltate hairs by the non-glandular ones is also suggested by WERKER 2000.

The observed variability of the stalked glandular trichomes in the matter of shape, location or secreted material suggesting their multifunctional role is reported in all *Labiatae* so far investigated (KELSEY & al. 1984, WERKER 1993).

The early presence of the trichomes irrespective of the pattern of their development (cotyledon versus leaf) can be attributed rather to their role as receptors of environmental signals than to their protective role. The cuticle, common to all these specialised cells, further supports the "signalling" role of trichomes, since the cuticle is an essential barrier in epidermal cell functions (LOLLE & PRUITT 1999, GLOVER 2000). The variability in trichome density as a result of environmental factors (EHLERINGER 1984, KOKKINI & al. 1994, BOSABALIDIS & KOKKINI 1997) can be seen in *O. calcaratum* if it is concerning its architecture and its natural habit. The increae followed by an "equivalent" decrease of the leaf size compined with the location were this chasmophyte is found on vertical cliffs (KYPRIOTAKIS 1998), results in different amounts of radiation received by the leaves and thus affecting its leaf-pubescence. This can be a proper explanation for the almost equal (in hairiness) sides of the middle stem leaves, due to the perpendicular orientation of the leaves and the resulting equal display of both leaf-sides to environment. In young leaves, reverse density between peltate trichomes and stomatal-complex is also reported for *O. vulgare* (BOSABALIDIS & KOKKINI 1997), *O. x intercedens* (BOSABALIDIS & SKOULA 1998), and (stomata versus hair density) in *Arabidopsis thaliana* (GLOVER 2000). This suggests the competitive interactions between "space consuming" epidermal elements. The high density of trichomes on young leaf-base, reported by all authors who studied the trichome development on the leaf, integrated with the also universally observed non-differentiation of peribasal, socket, and neighbouring of stomatal-complex cells, indicates the role of epidermal cell interactions in their differentiation (fate) and "spacing". This is the case for the unicellular non-glandular hairy *Arabidopsis thaliana* (LOLLE & PRUITT 1999, GLOVER 2000). More complex interactions (due to the „complexity“ of the trichomes), influenced by environmental factors, can also affect the fate and spacing of the epidermal cells of *O. calcaratum*. Presence of anthocyanins in the apex of young leaves is also reported for the "hairy" *Platanus orientalis* (NTEFIDOU & MANETAS 1996), confirming their presence in a wide range of plant organs, masked by chlorophylls, carotenoids, or pubescence (HARBORNE 1967, NEILL & GOULD 1999). Their presence in the "hairless" adaxial side of young leaves (contrary to the "hairy" mature ones, and to the "hairy" counterpart abaxial side) further suggest their protective role as proposed for the glabrous early stage of *O. calcaratum*.

The reduced pubescence of the reproductive organs (as compared to leaves) and in particular their remarkable coloration, and their peculiarities can be related to the pollination ecology of *O. calcaratum*, as in the *Lamiaceae* generally (HUCK 1992, MEEUSE 1992). The observed "low" density of peltate hairs on the bracts (with identical degree of reduction indeed), a reduction in stomata density and the presence of anthocyanins at bract-apex (which is devoid of peltate hairs) is also reported for all 3 subspecies of *O. vulgare* occurring in Greece (KOKKINI & al. 1994). The outer calyx surface of *O. calcaratum,* is relatively hairy in contrast to the glabrous inner one. A hairy outer calyx is common within *Labiatae* associated with the leaf hairiness (EL-GAZZAR & WATSON 1970, IETSWAART 1980, CANTINO & SANDERS 1986, WERKER 1993, MATTERN & VOGEL 1994, KOKKINI & al. 1994). Its role is the protection of the enclosed parts of the corolla (WERKER 1993) or the production of floral fragrances, by means of its dense and exposed glandular trichomes (MEEUSE 1992, MATTERN & VOGEL 1994). The former seems not to be the case for *O. calcaratum*, since the upper calyx lacks trichomes and the enclosed corolla is not covered by the calyx-tube. The latter can be a proper role for the numerous "floral" hairs of the calyx-tube which, due to their "fragileness" they are "ready" to release the accumulated material inside the spherical head even with a light sea-breeze. The proposed alluring function of the "floral" hairs agrees with the reaction of their secretion with paraffin oil (and the interference with the nectar indeed) and with their "pump-like" character. The "pump - like" character of similar anatomical floral attractants (trichome-nectaries) is reported also by FAHN 1979 and the rhythmicity of fragrance emission in flowers by MATILE & ALTENBURGER 1998. LUNAU 2000 suggests the signalling function of yellow colour abundant structures of reproductive organs in entomophily and ALDASORO & al. 2000 suggest that shining spherical structures on petals attract insects. The non-glandular hairs of the calyx-throat combined with the hairs of the outer corolla lobes can be the proper protection for the exposed (not covered by the calyx) glabrous corolla. The highly differentiated hairs on the insertions of the staminate filaments inside the corolla is common within *Labiatae,* serving as a guiding arrangement for nectar -searching insects (SCHULTZE & al. 1992), or as attractants (WERKER 2000). The peculiar 1 or 2 protruding trichomes from the pollen sacs and the trichomes on the underside of the anthers connective can be related with a signaling action (visual and/or tactile) proposed by LUNAU 2000. Trichomes on the underside of the connective are reported also within *Labiatae* without a clear function (see WERKER 2000). LUNAU 2000 suggests a signalling function "by an expansion of connectives", in species other than of *Labiatae*. The observed movement of the anther (versatile anther) can be regarded as a morphological adaptation to pollination biology within *Labiatae* (HUCK 1992, PETANIDOU 1996). The observed cleistogamy (release of pollen on receptive stigmatic papillae inside closed corollas), can be related to the humid Austrian summers. In absence of polinators or in case of adverse climate, evolution to autogamy (cleistogamy) may occur in *Labiatae* (HUCK 1992). The floral coloration of *O. calcaratum*, like most of *Labiatae*, have a high proportion of red to violet-purple colour (to the human eye), caused by anthocyanins (HARBORNE 1992). Although bees (main pollinators for *phryganic Labiatae*: PETANIDOU 1996) are insensitive to red colours they can receive a visual signal by the contrasting colours against the green leafy background (HARBORNE 1993). The white pollen-grains contrasting with the deep purple anther, the differential distribution of pigments in the flower tissue, as well as the distinct venation of the membranous bracts of *O. calcaratum* can act as visual signal and honey guides for the pollinators according to HARBORNE 1993, LUNAU 2000.

The interlocking form of the pavement cells for flat surfaces, and the elongated one with the long axis parallel to the direction of elongation for elongated tissues of *O. calcaratum* is common in dicots providing material strength (GLOVER & MARTIN 2000). The smaller sized and more indented (sinuous) pavement cell of the abaxial side compared to that of the adaxial one, agree with findings for *O. dictamnus* (BOSABALIDIS 1987). A proper explanation can be when one considers the spongy mesophyll with large air spaces underneath the abaxial side opposed to the denser palisade one of the adaxial side, and thus the need for a stronger "covering" structure on the abaxial side. The pavement cells were also found to develop different cuticular patterns during the life-cycle of *O.calcaratum*. In the early stage small hairs are covered by thick cuticular striations, but the thickness and complexity of the cuticle increase in the reproductive stage, where pavement cells with thick cuticular striations (papillate-cells) is the rule, a generality in reproductive organs (GLOVER & MARTIN 2000). The association of the papillate-cells with the "hairless" of the epidermis and their coloration can be explained by the ability of the papillate-cell (the cuticular striations), to enhance light absorption and focus/reflect the light into pigment-containing areas (GORTON & VOGELMANN 1996). The above, integrated with the ability of the stigmatic papillate-cells for secretion, as well as their diversity in shape, suggested as tactile cue by bees (KEVAN & LANE 1985), makes the distinction between papillate-cell and trichomes in *O.calcaratum* a difficult task. The findings in *O. calcaratum* are in accordance to "all epidermal outgrowths are termed trichomes" (ESAU 1953, FAHN 1979, WERKER 2000), or in other words "that every epidermal cell has the potential of forming trichome" (UPHOF 1962, JOHNSON 1975, GLOVER & MARTIN 2000).

Concluding remarks

As a plant, isolated in a harsh environment, *O. calcaratum* exhibits a plasticity in the behaviour, necessary for better adaptation (survival). This plasticity is depicted in the epidermis, which is the barrier to the "unfriendly" outside, but also, may act as the receptor for potential beneficial signals. The results of these partly conflicting needs, are the multiforming functional epidermal elements, and particularly the outgrowths from the epidermis, the trichomes. Indications and proposals of the ecological significance of plant trichomes were made a long time ago, but comparative studies from different disciplines will allow us to understand better these aromatic plants which thrive in coastal areas where a sea-breeze blows.

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