Atti Accademia Nazionale dei Lincei

Classe Scienze Fisiche Matematiche Naturali

Rendiconti

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Studies in plant morphology by scanning electron microscopy and applications to plant species of pharmaceutical interest. Inflorescences of Calendula officinalis L. and Anthemis tinctoria L.

Atti della Accademia Nazionale dei Lincei. Classe di Scienze Fisiche, Matematiche e Naturali. Rendiconti, Serie 8, Vol. **66** (1979), n.1, p. 51–56.

Accademia Nazionale dei Lincei

<http://www.bdim.eu/item?id=RLINA_1979_8_66_1_51_0>

Atti della Accademia Nazionale dei Lincei. Classe di Scienze Fisiche, Matematiche e Naturali. Rendiconti, Accademia Nazionale dei Lincei, 1979.

SEZIONE III

(Botanica, zoologia, fisiologia e patologia)

Botanica. — *Studies in plant morphology by scanning electron microscopy and applications to plant species of pharmaceutical interest. Inflorescences of* Calendula officinalis *L. and* Anthemis tinctoria *L.* Nota di ELSA MARIELLA CAPPELLETTI (*), presentata (**) dal Socio C. CAPPELLETTI.

RIASSUNTO. — Vengono studiati i microcaratteri delle superfici epidermiche delle infiorescenze di una specie di interesse farmaceutico, *Calendula officinalis* L., e di *Anthemis tinctoria* L., entrambe sofisticanti di *Arnica montana* L. Viene sottolineato il contributo offerto dal SEM nell'evidenziare alcune particolarità morfologiche degli acheni, suscettibili di possibili utilizzazioni a fini tassonomici.

INTRODUCTION

The results of previous works [3, 10] have evidenced the great diagnostic value of the surface microcharacters of epidermal cells for the botanical identification of some powdered vegetable drugs.

In a previous paper [2] the results of a SEM investigation on *Arnica* flower heads were reported.

The surface microcharacters of the inflorescences of *Calendula officinalis* L. (the flower heads of which have pharmaceutical use) and of *Anthemis tinctoria* L., both adulterant species of *Arnica montana* L. [4, 5, 11, 12], are here described.

In a subsequent paper the cell distortion phenomena occurring in the air-dried inflorescences of *Arnica*, *Calendula* and *Anthemis*, and the possibility of identification of the three species by surface scanning of the powdered drug, will be discussed.

MATERIAL AND METHODS

The inflorescences of *Calendula officinalis* L. and of *Anthemis tinctoria* L. came from specimens cultivated in the Botanical Garden of the University of Padua.

The following parts of the flower heads have been considered: bracts of the involucre, ligulate ray-florets, tubulate disk-florets and, for *Anthemis*, receptacular paleae.

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(**) Nella seduta del 16 dicembre 1978.

The samples were fixed, dehydrated and metallized following the procedure previously described [2], and observed with a Jeol Scanning Electron Microscope JSM-U_3 at the accelerating voltage of 25 kV, at the « Centro Universitario Grandi Apparecchiature Scientifiche » (CUGAS) of the University of Padua.

Results

Involucral bracts

On the abaxial bracts of *Calendula*, subpolygonal epidermal cells with smooth convex outer walls, unrisen stomata, biseriate covering trichomes along the margins (Pl. I, Fig. 1) and conical glandular hairs on the bract body (Pl. I, Fig. 2) are present.

The bracts of *Anthemis tinctoria* differ from those of *Calendula* due to the presence of scarious margins showing on the abaxial surface smooth rectangular epidermal cells, as found in both Pyrethrum and *Leucanthemum vulgare* [3]. Inside the scarious margins, striated epidermal cells and numerous stomata are present (Pl. I, Fig. 3). Uniseriate pluricellular covering trichomes, and not pedunculate glands, are also found on abaxial bracts (Pl .I, Fig. 4).

In the adaxial bracts of *Anthemis* the entire page is covered with smooth rectangular cells without any difference between the basis and the apex, unlike in *Calendula* (and in *Arnica* [2]), in which such cells are present only at the bract basis. At the apex, through transitional stages, aspects quite similar to those observed on the abaxial page, are present.

Corolla of ray-florets

On the adaxial ligule of *Anthemis* an epidermis of the helianthoid type is present which is very similar to the corresponding one of *Arnica montana* [2], apart from the greater number and prominence of the downwards decurring striations in *Anthemis*.

A very different array of microcharacters occurs in the adaxial ligule of *Calendula*: subrectangular, very elongated cells with convex outer walls and cuticle striations decurring longitudinally only on the middle of the cells (Pl. I, Fig. 5).

The abaxial ligules show strong differences in their epidermal features: in *Anthemis* strongly striated cells (Pl. I, Fig. 6) which, very elongated at the ligule basis, become shorter and more and more sinuous in outlines towards the apex, and biseriate glandular trichomes (Pl. II, Fig. 7); in *Calendula* lenticular cells with convex outer walls and longitudinally running cuticle striations, restricted to the middle of the cells at the ligule basis (Pl. II, Fig. 8), evenly distributed over the whole cell surface at the apex (Pl. II, Fig. 9).

In the lower, tubular zone of the corolla of *Anthemis* ray-florets, cells showing the same characters as those seen on the abaxial ligule (Pl. I, Fig. 6) are present, whereas in *Calendula* the lenticular shapes of the epidermal cells disappear, rectangular striated cells being present in the upper region (Pl. II, Fig. 10). In the lower one, characterized by biseriate covering trichomes (Pl. II, Fig. 11), the cell surfaces are almost smooth.

Corolla of disk-florets

The adaxial epidermises show in *Anthemis* the same morphological features found in the adaxial ligules. In *Calendula*, on the contrary, the epidermal cells – rectangular and with smooth convex outer walls - have a quite different appearance from those described for the corresponding page of the ligule.

On the abaxial epidermis of *Anthemis* rectangular cells with smooth outer walls occur. In *Calendula*, rectangular cells with quite different ornamentation features are found at the basis (Pl. II, Fig. 12), at the apex (Pl. III, Fig. 13) of the corolla tube and on the apical teeth (Pl. III, Fig. 14).

Calyx

Unlike in *Arnica* [2], the calyx is very reduced or suppressed in both ray- and disk-florets of the two adulterant species.

Ovaries

The morphological features of the ovaries of *Anthemis tinctoria* have been described by Kynčlová [8]. They lack both covering and glandular trichomes and are provided with longitudinal bars and grooves. Inspected by SEM, the ovary surfaces show faintly striated epidermal cells and rows of mucilaginous cells (Pl. III, Fig. 15 and Pl. III, Fig. 16) located on both bars and grooves. Rows formed by more numerous mucilaginous cells are often present on the ovary surface of disk-florets (Pl. III, fig. 17).

Calendula officinalis is known to be a heterocarpous species, in which three different types of fruits are developed. The heteromorphic achenes of *Calendula* have been described by Lanza [9] and by Heyn, Dagan and Nachman [6].

The abaxial surfaces of both the winged ovaries of ray-florets and those of the inner disk-florets are covered by glandular trichomes (Pl. III, Fig. 18), which on the contrary are absent in the ovaries of the outermost disk-florets. Epidermal cells with almost smooth cuticles are present in all the ovary types.

Paleae

Present on *Anthemis* receptacles only, the paleae are covered by rectangular epidermal cells with smooth outer walls on both abaxial and adaxial sides.

Discussion

Quite different surface microcharacters have been evidenced by SEM in the epidermises covering the involucral bracts, the ray- and disk- florets of these two species, which belong to different *Compositae* tribes [7].

In the involucral bracts the differences consist in both cell ornamentation and the type of trichomes occurring; in the adaxial ligules, epidermises of the helianthoid and of the mutisioid types [1] are present in *Anthemis* and *Calendula* respectively.

As regards the abaxial corollas, diagnostic value must be attached to cell shape for the ray-florets and to the presence or absence of cuticle ornamentations for the disk-florets.

The ovary surfaces have evidenced even more striking differences between *Calendula* and *Anthemis*.

The surface scanning proved to be particularly suitable for showing up the morphological details of the *Anthemis* mucilaginous cells, which seem quite different from those observed in *Leucanthemum vulgare* [3].

The taxonomic value of the achenes, which has been already discussed [2], could be considerably improved by the microcharacters evidenced by SEM. Namely, the distribution pattern and the micromorphological aspects of the *Compositae* mucilaginous cells would merit closer consideration.

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EXPLANATIONS OF PLATES I–III

PLATE I

Fig. 1. – *Calendula officinalis*. Involucral bract, abaxial surface: biseriate covering trichomes along the margin (×150).

Fig. 2. – *Calendula officinalis*. Involucral bract, abaxial surface: epidermal cells, stomata and glandular hairs (×150).

Fig. 3. – *Anthemis tinctoria*. Involucral bract, abaxial surface: striated epidermal cells and stomata (×1,000).

Fig. 4. – *Anthemis tinctoria*. Involucral bract, abaxial surface: covering trichomes and glands (×60).

Fig. 5. – *Calendula officinalis*. Ray-floret, adaxial ligule: longitudinally striated, elongated epidermal cells (×600).

Fig. 6. – *Anthemis tinctoria*. Ray-floret, abaxial ligule: strongly striated, elongated cells at the ligule basis (×600).

PLATE II

Fig. 7. – *Anthemis tinctoria*. Ray-floret, abaxial ligule: strongly striated sinuous cells and biseriate glandular trichome towards the ligule apex (×600).

Fig. 8. – *Calendula officinalis*. Ray-floret, abaxial ligule: epidermal cells towards the ligule basis (×600).

Fig. 9. – *Calendula officinalis*. Ray-floret, abaxial ligule: epidermal cells towards the ligule apex (×600).

Fig. 10. – *Calendula officinalis*. Ray-floret, abaxial surface of the tubular zone of corolla: epidermal cells in the upper region (×600).

Fig. 11. – *Calendula officinalis*. Ray-floret, abaxial surface of the tubular zone of corolla: covering trichomes in the lower region (×600).

Fig. 12. – *Calendula officinalis*. Disk-floret, abaxial corolla: epidermal cells towards the basis (×2,000).

PLATE III

Fig. 13. – *Calendula officinalis*. Disk-floret, abaxial corolla: epidermal cells towards the apex (×1,100).

Fig. 14. – *Calendula officinalis*. Disk-floret, abaxial corolla: epidermal cells of the apical teeth (×2,000).

Fig. 15. – *Anthemis tinctoria.* Disk-floret, ovary: row of mucilaginous cells (×600).

Fig. 16. – *Anthemis tinctoria.* Ray-floret, ovary: rows of mucilaginous cells (×850).

Fig. 17. – *Anthemis tinctoria.* Disk-floret, ovary: rows of mucilaginous cells (×400).

Fig. 18. – *Calendula officinalis.* Inner disk-floret, ovary: adaxial surface lacking glandular trichomes; on the left, the glandular trichomes of the abaxial surface are evident. (×80).

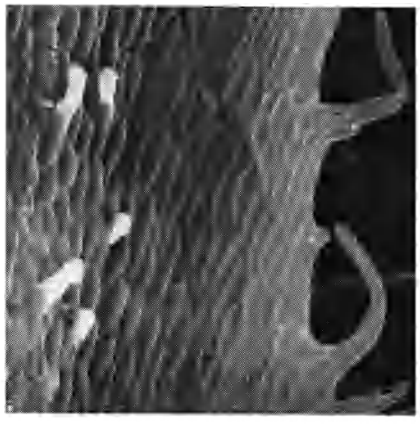
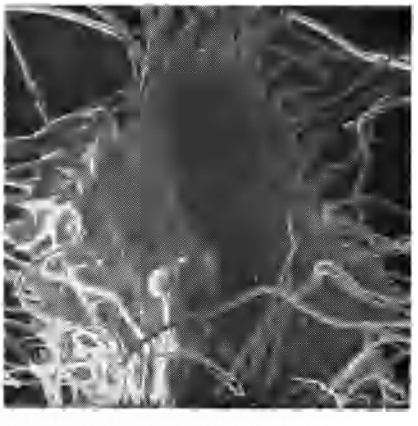


Fig. 1.

Fig. 2.

Fig. 3.



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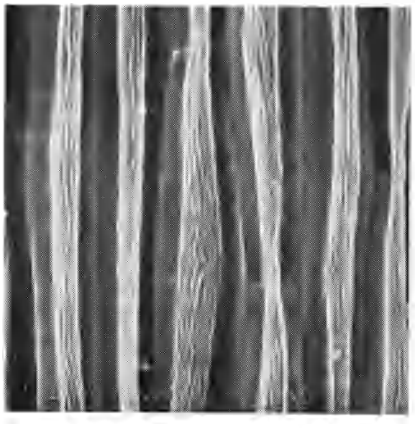

Fig. 5.

Fig. 6.

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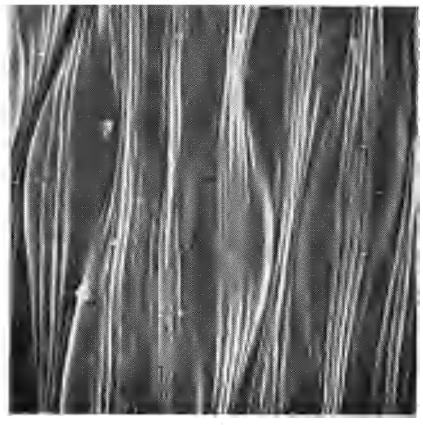

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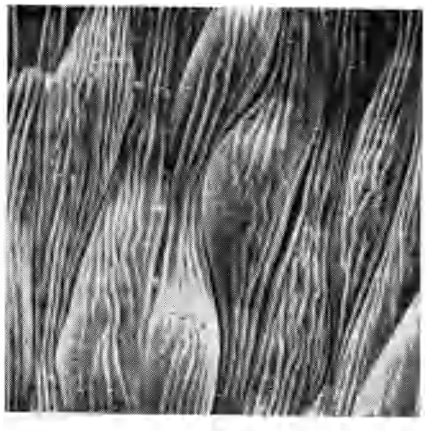

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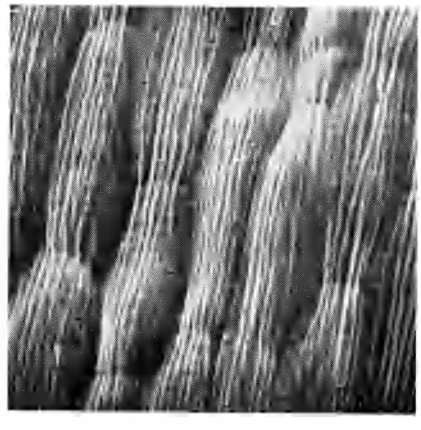


Fig. 10.

Fig. 11.

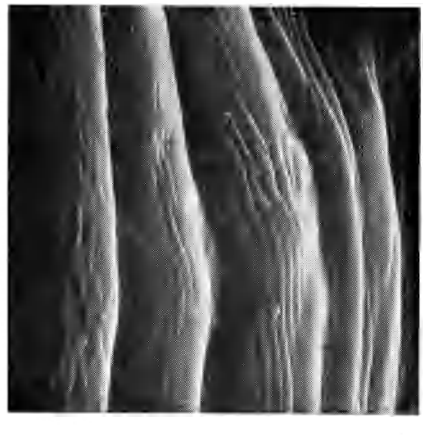


Fig. 12.

Fig. 13.

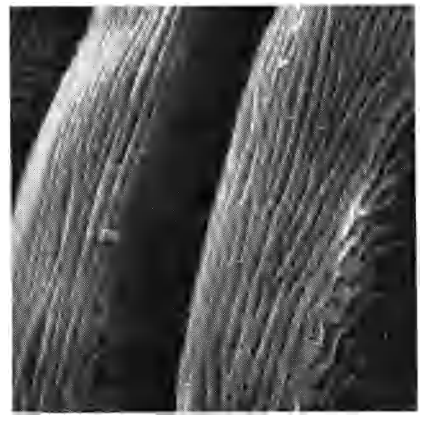


Fig. 14.

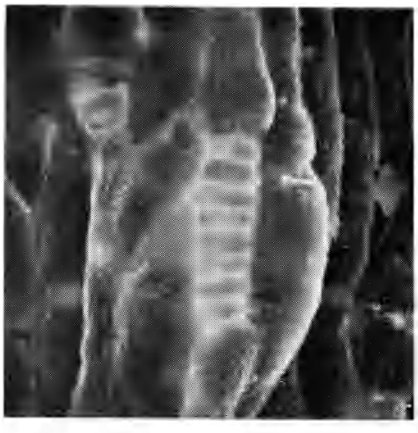


Fig. 15.

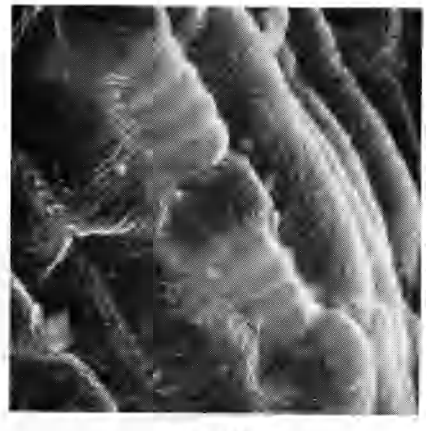
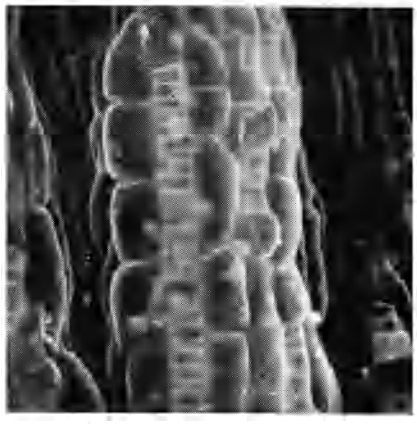


Fig. 16.



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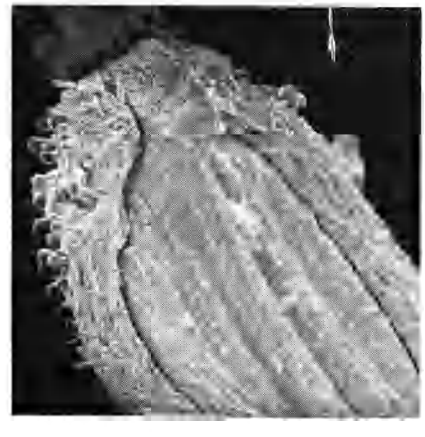

Fig. 18.