

Seed morphology and seed coat structure in Mediterranean plants: implication for evolution, taxonomy and germinative performance

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Introduction

The seed coat is the direct interface between embryo and external environment, imparts protection against pathogens and adverse conditions, contributes to embryo nutrition and growth and has key functions in seed dispersal, dormancy, and germination.

The morphology and physiological behaviour of seed coats are main features for understanding the pattern of temporal/spatial distribution of species. Seed polymorphism, in fact, can affect primary traits such as dispersal capacity, seed dormancy, predation, germinability and seedling competition, which markedly influence plant population dynamism and community structure (Debeaujon et al., 2000; Cupido et al., 2011). Seed coat traits also provide useful information for the analysis of taxonomic relations in many plant families and genera, as well as in critical *taxa*, since they usually exhibit little plasticity and almost always reflect genetic differences. In particular, scanning electron microscopy (SEM) has been employed to detect minute significant seed coat patterns, which may help to better define species characteristics (Barthlott, 1981, 1984; Fawzi, 2011).

Seed trait variations exist not only among species, but also within species and this is pivotal for seedling establishment at different habitats. Seed heteromorphism, micromorphology of epicuticular waxes, gland/trichome structure and secretion, presence of crystals in pericarp and other chemical compounds may play a role in changing seed response to selective environmental pressures across a wide geographic range, often providing a more favorable condition for plant survival (Cochrane et al., 2014).

Hence, a better understanding of morphological, ultra-structural and anatomical seed coat characteristics can contribute to explain and predict seed performance under certain

environmental conditions and select the most suitable treatments promoting seed germination.

For this reasons, the Seed Banks at the Botanic Gardens of Catania and Palermo are particularly involved in research studies aimed to detect and describe macro-morphological and micro-morphological features of seeds from different plant families and genera, using both light and scanning electron microscopy.

Main scopes of these studies are: 1) recognize and compare inter-specific and intra-specific variation of seed coat traits, with particular attention on rare and threatened species; 2) evaluate the usefulness of these traits in taxonomic and phylogenetic studies; 3) establish correlations between seed coat structures and seed germination; 4) understand the seed coat adaptive potential and sensitivity to particular ecological conditions and changing environments.

Material and methods

Exomorphic parameters and internal structures are detected from at least 100 fresh diaspores using both flatbed scanner and stereoscope (10–100×), then elaborated by image analysis systems.

Seed parts and tissues are stained with different methods, both on fresh (Phloroglucinol-HCl, Lugol's solution, TTC) and fixed material (Ruthenium Red and Light Green).

Seed coat sculpturing is analysed by SEM after gold sputter coating. The terminology from Barthlott (1981, 1984) is used to describe the seed coat considering arrangement and shape of epidermal cells, architecture of anticlinal and periclinal cell walls, primary sculpturing and secondary sculpturing (when present), occurrence, types and distribution of glands and/or trichomes.

Statistical analyses are computed on all quantitative morpho-anatomical parameters, using ANOVA after Barlett's test and post-hoc Fisher LSD test, linear regression and multivariate analyses (PCA, DFA, UPGMA).

Results and discussion

Several comparative analyses on seed morphology and seed coat microsculpturing, as well as on germination under different light and thermal conditions, were carried out to investigate variability of seed coat patterns and germinative performance traits at inter-specific and intra-specific levels.

Target groups were critical *taxa* with taxonomic and phylogenetic misclassifications or uncertain position (e.g. *Allium*, *Astragalus*, *Brassica*, *Calicotome*, *Dianthus*, *Helichrysum*), rare or phyto-geographically interesting *taxa* (*Retama*, *Sarcopoterium*), highly specialized species adapted to selective habitats such as sandy dunes, marshes, debris, high mountain belt (Salmeri et al., 2011a, b; Lantieri et al., 2011, 2012; Brullo et al., 2012; Ferrauto et al., 2015).

Our studies revealed that morpho-histological features of the seed coat, such as thick layer of epicuticular wax, tightly packed epidermal palisade cells, and various chemical substances (e.g. lignin, cutin, wax, suberin, and phenolic deposits), are particularly related to physical dormancy. Furthermore, the presence of mucilage cells or glands on exotesta was especially related with seed imbibition at early stages of germination.

Actually, no specific relation were detected between seed morphological traits (size, mass, colour, sculpturing) and germination behaviour, which mostly vary in response to different thermo- and photo-period.

On the contrary, diaspore characteristics appear to be influenced by natural selection forces

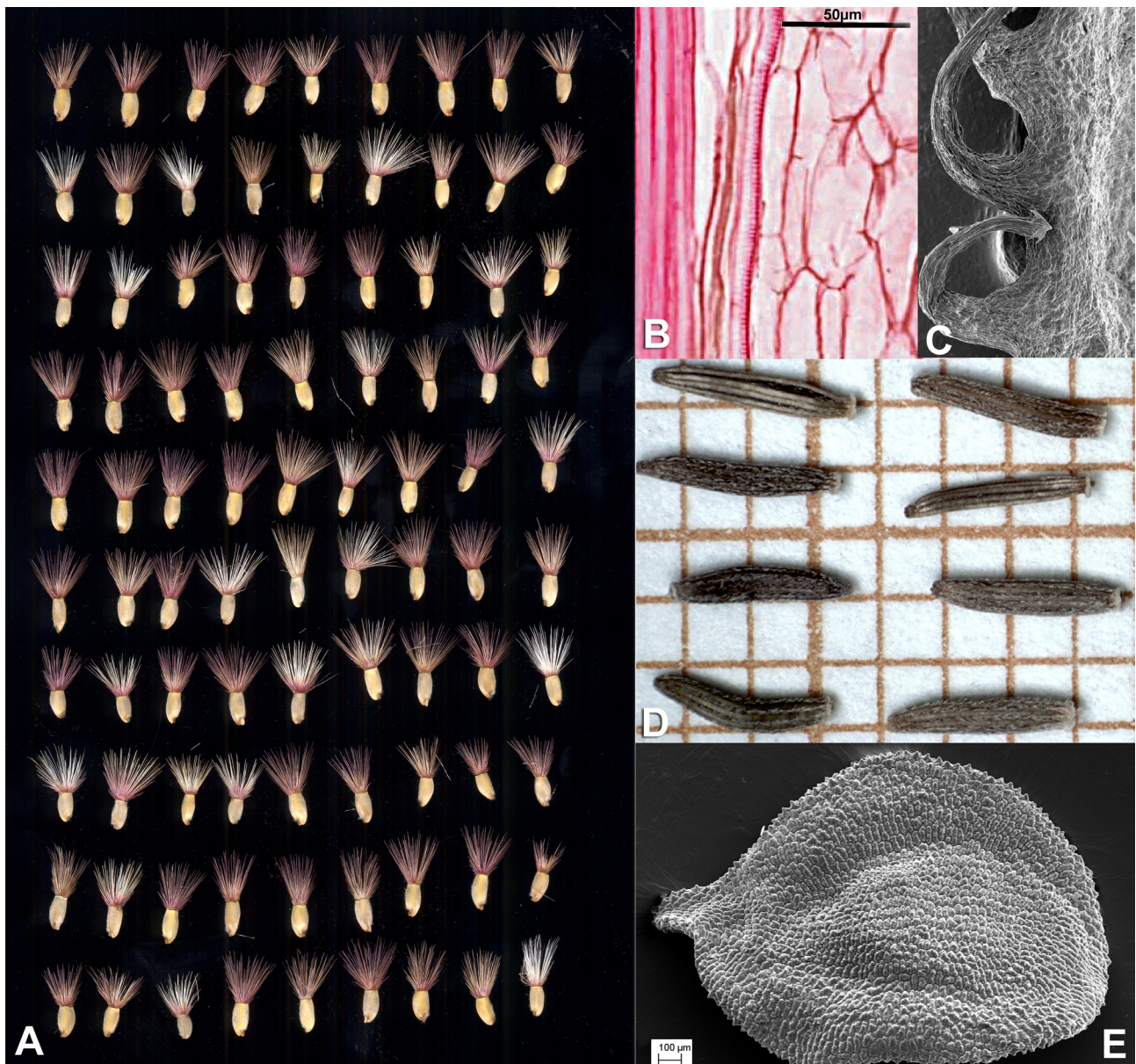


FIGURE 1. Types of macro- and microscopic seed analysis. A) Flatbed scanner image of *Centaurea tauromenitana* Guss. seeds; B) Seed coat longitudinal section of *Crepis leontodontoides* All.; C) Glandular trichomes of *Daucus carota* L. seeds (SEM); D) Seeds of *Senecio ae tnensis* Jan (light stereoscope); E) Seed coat pattern of *Dianthus rupicola* Biv. (SEM)

representing a sign of ecological adaptation, including local climate condition, or genetic drift which lead to differentiation in trait means and plasticity across different populations and taxa.

Our results also indicated the existence of very distinct ornamentation patterns which proved to be informative at sub-generic or even sub-specific level and should be included as valuable discriminating traits in taxonomic and phylogenetic studies as well as in the management procedures of seed bank collections.

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