Seed Morphology Studies on some Veronica L. Species (Plantaginaceae) with Scanning Electron Microscopy

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Abstract
In this study, the seed coat surfaces of Veronica bozakmanii M. A. Fischer, V. arvensis L., V. polita Fries, V. triphyllus L., V. hederifolia and V. cymbalaria which had grown in European Turkey were scanned by scanning electron microscope for the first time. Testa surface characteristics and micromorphological properties of examined Veronica species were determined and given comparatively which was important for the identification of the species systematically.

Key words: Plantaginaceae, seed, SEM, Turkey, Veronica.

Introduction

Plantaginaceae is a cosmopolitan family of 3 related genera, i.e. Bougueria Decne., Littorella P. Bergius and Plantago L. and about 275 species are distributed in diverse habitats throughout the world [1-3]. In a classification presented by Judd et al. [4] and APG [5], the concept of the Plantaginaceae was broadened to include many genera that were formerly assigned to the Scrophulariaceae including Veronica L. Veronica is a genus of the Plantaginaceae [6] formerly placed in Scrophulariaceae with about 500 species [7]. It is distributed over most of the Northern Hemisphere and in many parts of the Southern Hemisphere, and is ecologically highly diverse with species growing in aquatic to dry steppe habitats from sea level to high alpine regions.

The taxonomic history of Veronica clearly shows the difficulties involved in defining natural group within it, in fact, regarding delimitation of the genus and infrageneric classification [8-9]. Albach et al. [7] has proposed a new infratribal and infrageneric rearrangement for the Veroniceae and for Veronica. Nine monophyletic genera are recognized in the tribe and 13, also monophyletic, subgenera within a broadly circumscribed genus Veronica, in which up to ten genera have been determined. In Turkey, Veronica represented with over 86 species belonging to four sections (Veronicastrum, Beccabunga, Pocilla and Veronica) [10]. In the last years, there were lots of systematic and phylogenetic studies on genus Veronica L. [11-13]; [7]. Both macro- and micromorphological characters of the seed were shown to be of essential systematic importance within Veronica and, therefore, have been studied by several authors [14-19]. Later, in addition to these studies, Martinez-Ortega [20-21], and Shehata and Loutfy [9] worked with the perennial species of Veronica, they demonstrated a correlation between specific patterns of seed coat ornamentation.

In this study seed morphology of six Veronica species belonging to five subgenera [V. bozakmanii (subgenus Beccabunga), V. arvensis (subgenus Chamaedrys), V. triphyllus (subgenus Pellidosperma), V. polita (subgenus Pocilla), V. hederifolia and V. cymbalaria (subgenus Cochlidiosperma)] have been studied by scanning electron microscope. The different and similar features of these species have been described. The aim of this present
study is to investigate whether the ultrastructure of the testa [as seen under a scanning electron microscope (SEM)] can be used as an additional support to extend the previous study of Martínez-Ortega and Rico [21].

Material and Methods

Veronica samples were collected from different localities from European Turkey. These localities are listed in Table 1. Mature seeds from six Veronica belonging to five subgenera in Veronica have been studied. All seeds were obtained from herbarium specimens. Specimens were deposited in the herbarium of the Faculty of Science at Trakya University named as EDTU. Seeds were dried in a critical point dryer and were mounted on aluminum stubs. They were examined under LEO Supra 35 VP FEG SEM (Scanning Electron Microscope). Because seed sizes were very different seed general views were photographed at 65×, 70×, 99×, 120×, 200× magnifications. Samples were examined at 1000× and 1500 magnification for seed surface (testa) cells. The terminology used to describe seed coat surface sculpturing follows mainly Font Quer [22], Stearn [23], Juan et al. [19] and Martínez-Ortega and Rico [21].

Table 1. Collection data of the investigated specimens were given following

<table>
<thead>
<tr>
<th>No</th>
<th>Plant species</th>
<th>Locality / Collectors / EDTU number / Date</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Veronica bozakmanii</td>
<td>Tu(E) A1(E) Edirne: Enez, Vakif village, around Orenler, 58 m, 40° 37' 0&quot;N, 26° 15' 0&quot;E / M. Kireç (EDTU 6932) / 05.04.1994.</td>
</tr>
<tr>
<td>2</td>
<td>Veronica arvensis L.</td>
<td>Tu(E) A1(E) Edirne: Center between Karaagac-Pazarkule 1 km, 23 m, 41° 39' 28&quot;N, 26° 31' 25&quot;E / N.Ç. Demirkan (EDTU 7732) / 06.05.2000.</td>
</tr>
<tr>
<td>3</td>
<td>Veronica polita Fries</td>
<td>Tu(E) A1(E) Kirklareli: between Terzidere and Topdere villages 1 km, 482 m, 41° 59' 42&quot;N, 27° 7' 39&quot;E / Edirne: Karakasim, 29 m, 41° 31' 00&quot;N, 26° 38' 00&quot;E / C. Yarci (EDTU 7107) / 08.05.1996.</td>
</tr>
<tr>
<td>4</td>
<td>Veronica triphylllos L..</td>
<td>Tu(E) A1(E) Edirne: Enez: Buyukevren village, around Kufaci, 79 m, 40° 38' 0&quot;N, 26° 13' 0&quot;E / M. Kireç (EDTU 6941) / 30.03.1994.</td>
</tr>
<tr>
<td>5</td>
<td>Veronica hederifolia L.</td>
<td>Tu(E) A1(E) Edirne: Center: Musabeyli village, 109 m, 41° 40' 0&quot;N, 26° 40' 0&quot;E / F. Dane (EDTU 3515) / 30.03.1989.</td>
</tr>
<tr>
<td>6</td>
<td>Veronica cymbalaria Bodard</td>
<td>Tu(E) A1(E) Edirne: Center: around Faculty of Medicine campus, 26 m, 41°40'28&quot;N, 26° 33' 39&quot;E / F. Dane (EDTU 3396) / 25.04.1989.</td>
</tr>
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</table>

Results

In this study, the micromorphological characters of the seed outer surface of six Veronica species were examined by SEM (Table 1). The micromorphological features of the seeds as seen in figures (Figs 1-6) including micrographs provided an additional source of characters for the separation of the examined species (V. bozakmanii, V. arvensis, V. triphylllos, V. polita, V. hederifolia and V. cymbalaria) collected from European Turkey. SEM micrographs of all examined taxa were given for the first time in this study.

*Veronica bozakmanii* has colliculate seed coat pattern. It has obscure or sometimes shallow radial walls. This seed coat pattern sometimes have corrugations (Fig 1). *V. arvensis, V. trifillos* and *V. polita* have reticulate- verrugate seed coat pattern. In this type of seed coat, there are variable sizes of polygonal cells with radial walls irregularly thickened from
medium depth to shallow, defining a clear basal reticulum. Polygonal cells have tangential walls with a central or sometimes excentric amorphous warts (Fig 2-4). *V. hederifolia* and *V. cymbalaria* have reticulate-corrugate seed coat pattern. In this type of seed coats have shallow to medium depth radial walls, not uniformly thickened, sometimes reinforced by a striation or network (rarely with irregular thickenings on cell angles). Tangential walls regular or irregularly corrugate, irregularly compressed or sometimes slightly granulated (Fig 5-6).

**Discussion**

The seeds of *Veronica* are flat or planoconvex, but some groups indicate the need for further classifications. They range from elipsoidal to suborbicular in outline. The trend seems to be the result of ellipsoidal seeds in species of the subgenera *Beccabunga* and *Pseudolysimachion* to the suborbicular seeds of subgenus *Pentasepalae* [17]; [21]. Seeds in subgenera *Beccabunga* are the flattest (<0.2 mm) [17]. The seeds are among the cochleariform type in subgenera *Pocilla*, *Pellidosperma* and *Cochlidiosperma* [according to Hufford and McMahon [24] and also in two species in *V. subgen.* Synthyris—*V. californica* and *V. regina-nivals* while the remainder species of this subgenus have seeds that are dorsiventrally flat] turn inwards to form cymbiform or cyathiform seeds [25]. Regarding the seeds, the reticulate-corrugate pattern is shared by the perennial representatives with x = 7 or 8, but the annuals whose chromosome number is based on x = 7 present different patterns of seed coat ornamentation (colliculate and reticulate-verrucate) [25].

Reticulate-verrucate seed coat pattern is present in seven subgenera and is considered to be the most common pattern in *Veronica*. *Veronica* subgen. *Pentasepalae* and *V. subgen.* *Beccabunga* mainly display a reticulate-verrucate seed coat. All representatives of *V. subgen.* *Pentasepalae* except *V. vendettadeae*, *V. bogosensis*, *V. armena*, *V. surculosa* and *V. kurdica* are observed to have this type of seed coat. Furthermore, subgenera *V. subgen.* *Stenocarpon*, *V. subgen.* *Pocilla*, *V. subgen.* *Pegmentosperma* have reticulate-verrucate seed coat pattern [25]. In this study, *V. arvensis* belonging (subgenus *Chamaedrys*), *V. triphyllos* belonging (subgenus *Pellidosperma* and *V. polita* belonging (subgenus *Pocilla*) are observed to have reticulate-verrucate seed coat. Also in the study of Muñoz-Centeno et al. [25] *V. ceratocarpa* is documented to have reticulate-verrucate seed coat too.

The existence of the reticulate-corrugate seed coat pattern is widely observed in *V. subsect.* *Serpyllifoliae* and *V. subsect.* *Beccabunga* as well as *V. gentianoides*, all of *V. subgen.* *Beccabunga*, in the greater part of *V. subgen.* *Cochlidiosperma*, in *V. cana* an Asian representative of *V. subgen.* *Veronica*, in three representatives from *V. subgen.* *Pentasepalae* (*V. vendetta-deae*, *V. bogosensis* and *V. armena*) and in two species from *V. subgen.* *Pocola*. During the research conducted for this paper, *V. hederifolia* and *V. cymbalaria* (subgenus *Cochlidiosperma*) have been observed to have reticulate-verrucate seed coat. At this point it is important to point out that according to the study conducted by Muñoz-Centeno et al. [25]. *V. gentianoides* has reticulate-corrugate seed coat too.

Kaplan et al. [26] examined four *Veronica* seed coats which were gathered from the natural habitat present in Turkey. In their study, seed coat pattern types were listed as follows: Reticulate-papillate for *V. persica*, reticulate to reticulate-corrugate for *V. beccabunga*, colliculate to corrugate for *V. officinalis* and ribbed for *V. multifida*. Colliculate seed coat pattern is also a widely observed one. It appears in the subgeneras of *V. subgen.* *Pocilla*, *V. subgen.* *Beccabunga*, plus *V. peregrina* and *V. subgen.* *Pellidosperma*, plus in two species (*V. surculosa* and *V. kurdica*) from *V. subgen.* *Pentasepalae*. 

In the study of Muñoz-Centeno et al. [25] *V. reuterana* and *V. cardiocarpa* have reticulate-corrugate seed coat. In this study *V. bozakmanii* has colliculate seed coat. The results of this paper are in agree with the results of the study of Muñoz-Centeno et al. [25].
Figure 2. *Veronica arvensis*: a. b. General view of seed; c. Seed coat surface.
Figure 3. *Veronica triphyllos*: a. b. General view of seed; c. Seed coat surface.
Figure 4. *Veronica polita*: a. General view of seed; b. Seed coat surface.
Conclusion

The research presented in this paper concludes that the micromorphological characters of seed surfaces were important to determine the species of Veronica L. genus. The present data represent the analysis of the seed coat morphology of some Veronica L. species by scanning electron microscope which were grown in European Turkey.
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Figure 6. *Veronica cymbalaria*: a.b. General view of seed; c. Seed coat surface.

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References