Nutlet micromorphology of the genus Marrubium L. and allies and its systematic implication (Lamiaceae: tribe Marrubieae)

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Abstract. Nutlets of 20 taxa of the tribe Marrubieae were examined by scanning electron microscopy (SEM) and detailed descriptions of nutlet micromorphological features for all examined taxa are provided. The nutlets exhibited variation in size, shape, color and surface sculpturing. The nutlets shape of most species studied is ovate, but rounded, broad ovate, elliptic, lanceolate, triangular and oblong can also be found in a few species. Six basic types of the sculpturing pattern of nutlet surface can be distinguished: reticulate, foveolate, scalariform, ruminate, pustilulate and colliculate-granulate. The most common type of nutlet sculpturing among the studied species is reticulate, but the variation in size and shape of their composing cells provided useful diagnostic characters. Our investigation revealed that the type of sculpturing was more useful in taxon delimitation among Marrubium species and allies at the species rank.

Keywords. diagnostic characters, Labiatae, surface sculpturing, scanning electron microscopy, taxonomy

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INTRODUCTION

Marrubiaceae Vis., with five genera and ca. 91 species, is one of the 10 Lamioidae tribes distributed mainly in southern Europe and North Africa (Siadati et al., 2018). They are mostly nonaromatic herbs or subshrubs with campylotropous to rotate calyx and often with secondary calyx lobes, zygomorphic and 2-lipped corolla and stamens included or shortly exserted from the corolla (Siadati et al., 2018; Harley et al., 2004).

According to one of the most recent phylogenetic studies, the monophyletic Marrubiaceae contains four major clades: the first one including member of Acanthoprasium (Benth.) Spach and Moluccella L., the second one comprising species of Ballota L., the third clade containing Pseudodictamnus Fabr. and related species and the fourth one including Marrubium L. species (Siadati et al., 2018). Acanthoprasium, with two species, is an European genus with a woody habit, spiny bracteoles and upper lip of corolla totally hairy. According to Bendiksby et al. (2011), the annual or short-lived perennials Moluccella, contains eight species in southwestern Asia and the Mediterranean regions, are characterized by a zygomorphic calyx more or less expanded at the mouth, internally glabrous, and usually lobed with both primary and secondary indentations. The genus Ballota are subshrubs to perennial herbs with herbaceous bracteoles and shortly exserted stamens from the corolla and extend from Macaronesia, Europe to Mediterranean and Western Asia (Siadati et al., 2018). According to Bentham (1832-1836), the genus Ballota had traditionally been divided into three sections based on two main morphological characters, woody against herbaceous habit and the type of bracteoles (spinose against herbaceous): (1) sect. Acanthoprasium Benth., which is raised to generic rank by Bendiksby et al. (2011), (2) sect. Beringeria (Neck.) Benth., which is recently raised to generic rank by Siadati et al. (2018) and (3) sect. Ballota Benth. (Bentham 1832-1836). Based on recent phylogenetic studies, the genus Ballota re-circumscribed to encompass the members of sect. Ballota with about three species. Moreover, members of sect. Beringeria recently raised to generic rank as Pseudodictamnus, which accommodates perennial herbs with herbaceous bracteoles, expanded calyx limb with 6-20 teeth and widely distributed from Eurasia to North Africa (Siadati et al., 2018). The genus Marrubium with about 50 species have usually toothed and petiolate leaves, 5-15 (-30) lobed calyx covered by dense stellate trichomes, corolla tube shorter than calyx and stamens included in the corolla tube (Harley et al., 2004).

Studies on nutlet micromorphology within Lamiaceae showed that nutlets features e.g., shape, morphology of the abscission scar and surface sculpturing, were potentially useful at different taxonomic levels (e.g. Demissew & Harley, 1992; Marin et al., 1994; Ryding, 1995; Oran, 1996; Ryding, 1998; Salmaki et al., 2008; Moon et al., 2009; Satil et al., 2012; Krawczyk & Glowacka, 2015; Eyvaz?adeh Khosroshahi & Salmaki, 2018). Among different nutlet characters, type of sculpturing has been considered to be taxonomically most important (Oran, 1996; Kahrman et al., 2011), however, color, size and shape of nutlets were considered unimportant, either because they did not vary or the variation was random or too great (Oran, 1996). Abscission scars were invariably, but the variation at higher levels may be significant (Guerin, 2005). There are no reports on the taxonomic significance of the nutlets micromorphology among different genera of Marrubiaceae, except that by Mosquero et al. (2007), who provided a description of the morphology and anatomy structure in Marrubium vulgar L. Akgül et al. (2008) illustrated the range of variability in seed characters in Marrubium species found in Turkey and Hassan & Al-Thobaiti (2015) provided a detailed description of the morphological nutlet characteristics of Marrubium vulgar in Saudi Arabia. Thus, the main goal of this study was to provide a detailed description of nutlet micromorphology of the genus Marrubium and allies.

MATERIALS AND METHODS

Nutlets of 20 species representing all five genera of tribe Marrubiaceae were selected to investigate the value of seed characters in the classification of genera and species. Nutlets were collected from herbarium specimens, deposited in the herbaria M (Botanische Staatssammlung München), MSB (Münich Systematic Botany), and TUH (Central Herbarium of Tehran University). A list of voucher specimens is presented in Table 1. A total of two species of Acanthoprasium, two species of Moluccella, four taxa representing three species of Ballota, two species of Pseudodictamnus and 10 species of Marrubium were analyzed. Nutlets were observed in advance, using a stereomicroscope to ensure that they were of normal size and maturity. Nutlets observations were made using scanning electron microscopy.

For SEM observations, dried nutlets were mounted on aluminum stubs using double-sided adhesive and sputter-coated with a thin layer (ca. 30 nm) of gold and examined by means of a Hitachi SU3500 (Japan) scanning electron microscopy at an accelerating voltage of 5-30 kV. This paper follows the terminology of Bojnanský & Fargašová (2007) and Stearn (1983) for the seed shape and surface ornamentation.
Table 1. Taxa, voucher specimens and collection data of selected species of *Marrubium* and its allied genera deposited in the herbaria M (Botanische Staatssammlung München), MSB (Münich Systematic Botany), and TUH (Central Herbarium of Tehran University).

<table>
<thead>
<tr>
<th>Species</th>
<th>Collection data</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Acanthoprasium frutescens</em> (L.) Spenn.</td>
<td>France: Schlucht von Aiglum, 30 km westlich und nördlich Nice, O. Angerer s.n. (M)</td>
</tr>
<tr>
<td><em>Acanthoprasium integrifolium</em> (Benth.) Ryding</td>
<td>Cyprus: – 1880 (M)</td>
</tr>
<tr>
<td><em>Ballota nigra subsp. anatolica</em> P. H. Davis, Cult.</td>
<td>Turkey: Nigde, near Aydinkent, Yayikli, P. Rasmont 55330 (M)</td>
</tr>
<tr>
<td><em>Ballota nigra subsp. ruderalis</em> (SW.) Briq.</td>
<td>Jugoslavien: Makedonija, 13 km ONO von Skocivir an der Straße zum Kajmakchalan (Grenzgebirge), D. Podlech &amp; Lippert 26027 (M)</td>
</tr>
<tr>
<td><em>Ballota platyloma</em> Rech.f.</td>
<td>Iran: Mazandaran, Zentral- Elburs, Im Einzugsgebiet des oberen Tedschen-Flusses, unterhalb Kom-rud bala, lochere Gebuschvegetation, Felsschutt; 60 km ostlich von Firuzkuh, E. Behboudi &amp; P. Aellen s.n. (M)</td>
</tr>
<tr>
<td><em>Ballota saxatilis</em> Sieber ex C.Presl</td>
<td>Libanon/Syrien: –, s.n. (M)</td>
</tr>
<tr>
<td><em>Pseudodictamnus hispanicus</em> (L.) Salmaki &amp; Siadati</td>
<td>Spain: Valencia Segunto, inruninis, 508 (M)</td>
</tr>
<tr>
<td><em>Pseudodictamnus aucheri</em> (Boiss.) Salmaki &amp; Siadati</td>
<td>Iran: Chahar-Mahal-e Bakhtiari, Kuh-e Rig from Dehno, Zarre 17941 (TUH)</td>
</tr>
<tr>
<td><em>Marrubium alyssoides</em> Pomel,</td>
<td>Marokko: d’ Oujda, 5 km W El Aioun; Felder, Eucalyptushainen N der Straße nach Taza (P 1), W. Lippert 21821 (M)</td>
</tr>
<tr>
<td><em>Marrubium anisodon</em> K. Koch</td>
<td>Afghanistan: Logar, Weshang des Tera-Passes bei Nayzi, D. Podlech 18498 (M)</td>
</tr>
<tr>
<td><em>Marrubium astracanicum</em> Jacq.</td>
<td>Caucasus: Krasnoselkoiwe, montes Arenguni, in vicinitate pagi Tokludza, V. Vašák 257082 (M)</td>
</tr>
<tr>
<td><em>Marrubium catarifolium</em> Desr.</td>
<td>Caucasica: M. Senser 257078 (M)</td>
</tr>
<tr>
<td><em>Marrubium cuneatum</em> Banks &amp; Sol.</td>
<td>Iran: Kurdistan, In graminosis siccis jugi prope Salavatabad 25 km E Sanandaj, K.H. Rechinger 42784 (M)</td>
</tr>
<tr>
<td><em>Marrubium leonuroides</em> Desr.</td>
<td>Caucasus: Kislovodsk, in abruptis, I. Akinfiyev 7382 (MSB)</td>
</tr>
<tr>
<td><em>Marrubium peregrinum</em> L.</td>
<td>Austria: Nordburgenland, Parndorfer Platte, trokene, grasige Stellen am E-Fuß des Heidl bei Nickelsdorf, ca. 0.45 km SSE des Bahnübergangs am NE-Fuß des Heidl (Kote 133), T. Barta 2004-351 (M)</td>
</tr>
<tr>
<td><em>Marrubium persicum</em> C.A. Mey.</td>
<td>Armenia: Ararat, Ararat valley 12 km NW of Ararat town, 7 km NW of Vedi, Erah range, Fayvush &amp; al. XI/1040 (M)</td>
</tr>
<tr>
<td><em>Marrubium propinquum</em> Fisch. &amp; C.A. Mey.</td>
<td>Caucasus: Krasnoselskoiwe, montes Arenguni, in vicinitate pagi Tokludza, V. Vašák s.n. (M)</td>
</tr>
<tr>
<td><em>Moluccella acheri</em> (Boiss.) Scheen,</td>
<td>Iran: Baluchistan, 20 km a Zahedan meridiem versus, K.H. Rechinger 14562 (M)</td>
</tr>
<tr>
<td><em>Moluccella laevis</em> L.</td>
<td>Iraq: Erbil (Kurdistan). In agris demssis inter Mirza Punstam et Pania, ca 500 m. 11230 (M)</td>
</tr>
</tbody>
</table>

**RESULTS AND DISCUSSION**

Several main features of the investigated nutlets *i.e.* shape, length, width, as well as sculpturing patterns, projections of outer periclinal walls and anticlinal walls are summarized in Table 2. Selected SEM micrographs of nutlets studied were presented in Figures 1-3. In general, the color of nutlets in all studied species was dark brown to black. The nutlets were oblong (*e.g.*, *M. catarifolium* Desr., Fig. 2G), triangular (*e.g.*, *Mo. laevis* L., Fig. 3H), broad ovate (*e.g.*, *B. nigra subsp. ruderalis* (Sw.) Briq., Fig. 1F) to rounded (*e.g.*, *P. acheri* (Boiss.) Salmaki & Siadati, Fig. 1L) and elliptic (*e.g.*, *A. frutescens* (L.) Spenn) to lanceolate (*e.g.*, *B. nigra subsp. anatolica* P.H. Davis, Fig. 1D) in shape, but the most common type was ovate (*e.g.*, *B. platyloma* Rech. f., Fig. 1H; *M. cuneatum* Banks & Sol., Fig. 2I and *M. persicum* C.A.Mey., Fig. 3B). The size of nutlet ranged from 1.34×1.08 mm (in *M. leonuroides* Desr.) to 4.04×1.45 mm (in *Mo. acheri* (Boiss.) Scheen). Nutlets of investigated taxa exhibited six types of surface sculpturing patterns. The most common type
of sculpturing pattern was reticulate (e.g., *A. frutescens*, Fig. 1A), however, other types of sculpturing patterns, like scariosum (*A. integrifolium* (Benth.) Ryding, Fig. 1C), foveolate (*M. persicum*, Fig. 3C), ruminate (*M. alyssoides* Pomel, Fig. 2B), pusticulate (*M. astracanicum* Jacq., Fig. 2F) and colliculate-granulate (*Mo. laevis*, Fig. 3I), were observed.

Our study represents the first investigation on nutlet micromorphology on the genus *Marrubium* and its allied genera. Variation in shape, size, presence or absence of trichomes at the apex of nutlets and particularly surface sculpturing appeared to have taxonomic value in some groups of Lamiaceae (Husain et al., 1990; Oran, 1996; Navarro & El-Qualidi, 2000; Moon & Hong, 2006; Moon et al., 2009; Kahraman et al., 2011). Özkan et al. (2009) found that variation of shape, size, surface sculpturing and color were useful in distinguishing groups, species and subspecies among 12 examined species of *Salvia*. In *Stachys* (Salmaki et al., 2008) nutlet micromorphology provided valuable data in separating the related species within sections, although these characters were not useful in

<table>
<thead>
<tr>
<th>Taxon</th>
<th>Nutlet Shape</th>
<th>Nutlet length (mm)</th>
<th>Nutlet width (mm)</th>
<th>Surface Sculpturing</th>
<th>Apex Bearded</th>
<th>Outer Periclinal Wall</th>
<th>Anticlinal Wall</th>
<th>Figures</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Acanthoprasium frutescens</em></td>
<td>Elliptic</td>
<td>2.75</td>
<td>1.38</td>
<td>Reticulate +</td>
<td>Deep</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Acanthoprasium integrifolium</em></td>
<td>Triangular</td>
<td>2.75</td>
<td>1.39</td>
<td>Scalariform +</td>
<td>Shallow</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Ballota nigra</em> subsp. anatolica</td>
<td>Triangular</td>
<td>2.09</td>
<td>0.9</td>
<td>Foveolate</td>
<td>Concave</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Ballota nigra</em> subsp. ruderalis</td>
<td>Broad ovate</td>
<td>1.85</td>
<td>1.16</td>
<td>Reticulate –</td>
<td>Flat</td>
<td>–</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Ballota platyloma</em></td>
<td>Ovate</td>
<td>1.85</td>
<td>1.05</td>
<td>Reticulate –</td>
<td>Flat</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Ballota saxatilis</em></td>
<td>Ovate</td>
<td>2.3</td>
<td>1.29</td>
<td>Reticulate –</td>
<td>Concave</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Pseudodictamnus aucheri</em></td>
<td>Rounded</td>
<td>1.78</td>
<td>1.32</td>
<td>Reticulate –</td>
<td>Concave</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Pseudodictamnus hispanicus</em></td>
<td>Ovate</td>
<td>2.22</td>
<td>1.27</td>
<td>Reticulate –</td>
<td>Concave</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Marrubium alyssoides</em></td>
<td>Ovate</td>
<td>1.86</td>
<td>1.01</td>
<td>Ruminate –</td>
<td>Concave</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Marrubium anisodon</em></td>
<td>Oblong</td>
<td>1.78</td>
<td>0.99</td>
<td>Reticulate –</td>
<td>Shallow</td>
<td>Concave</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Marrubium astracanicum</em></td>
<td>Ovate</td>
<td>1.79</td>
<td>1.07</td>
<td>Pusticulate –</td>
<td>Convex</td>
<td>–</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Marrubium catarifolium</em></td>
<td>Oblong</td>
<td>2.28</td>
<td>1.22</td>
<td>Reticulate –</td>
<td>Deep</td>
<td>Concave</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Marrubium cuneatum</em></td>
<td>Ovate</td>
<td>2.21</td>
<td>1.21</td>
<td>Reticulate –</td>
<td>Flat</td>
<td>Risen</td>
<td></td>
<td>Fig. 1</td>
</tr>
<tr>
<td><em>Marrubium leonuroidei</em></td>
<td>Ovate-lanceolate</td>
<td>1.34</td>
<td>1.08</td>
<td>Reticulate –</td>
<td>Concave</td>
<td>–</td>
<td></td>
<td>Fig. 2</td>
</tr>
<tr>
<td><em>Marrubium litardierei</em></td>
<td>Ovate</td>
<td>2.09</td>
<td>1.1</td>
<td>Reticulate –</td>
<td>Shallow</td>
<td>Concave</td>
<td></td>
<td>Fig. 2</td>
</tr>
<tr>
<td><em>Marrubium peregrinum</em></td>
<td>Oblong</td>
<td>1.65</td>
<td>0.96</td>
<td>Reticulate +</td>
<td>Concave</td>
<td>–</td>
<td></td>
<td>Fig. 3</td>
</tr>
<tr>
<td><em>Marrubium persicum</em></td>
<td>Ovate</td>
<td>1.79</td>
<td>1</td>
<td>Foveolate</td>
<td>Concave</td>
<td>Risen</td>
<td></td>
<td>Fig. 3</td>
</tr>
<tr>
<td><em>Marrubium propinquum</em></td>
<td>Oblong</td>
<td>1.81</td>
<td>1.03</td>
<td>Reticulate –</td>
<td>Concave</td>
<td>Risen</td>
<td></td>
<td>Fig. 3</td>
</tr>
<tr>
<td><em>Mo. laevis</em></td>
<td>Triangular</td>
<td>4.04</td>
<td>1.45</td>
<td>Reticulate –</td>
<td>Concave</td>
<td>Risen</td>
<td></td>
<td>Fig. 3</td>
</tr>
<tr>
<td><em>Mo. laevis</em></td>
<td>Triangular</td>
<td>3.04</td>
<td>2.13</td>
<td>Coliculate-Granulate –</td>
<td>Convex</td>
<td>Represented by channels</td>
<td></td>
<td>Fig. H, I</td>
</tr>
</tbody>
</table>

*Due to lack of sufficient evidence, *Ballota saxatilis* has not yet been assigned to *Pseudodictamnus*, but it shares several morphological features with the members of this genus.*

separating large natural groups. Variations in size and the type of sculpturing as well as the shape of its composing cells had been considered to provide the most valuable characters at species level (e.g., Kahraman et al., 2011; Tarimcilar et al., 2013; Salmaki et al., 2008; Eyvazadeh Khosroshahi & Salmaki, 2018). For instance, the polygonal cells in *M. peregrinum* L. (Fig. 3A) were much smaller than those in *M. leonuroides* (Fig. 2L). Moreover, *M. anisodon* K.Koch (Fig. 2D) and *M. litardierei* (Fig. 2N) were different in the depth of pits on surface. Due to the particular value of microsculpturing, a comprehensive discussion is given below, indicating the importance of sculpturing patterns and its
systematic importance in the genus *Marrubium* and its allied genera.

Fig. 2. Scanning electron micrographs of *Marrubium* and its allied genera. A, B. *Marrubium alyssoides*; C, D. *M. anisodon*; E, F. *M. astracanicum*; G, H. *M. catarifolium*; I, J. *M. cuneatum*; K, L. *M. leonuroides*; M, N. *M. litardierei*, O. *M. peregrinum*. 

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**Acanthoprasium**—This genus includes two species based on the latest phylogenetic study (Bendiksby et al., 2011), characterized by simple hairs and broadly campanulate calyx with spiny lobes. Both species of *Acanthoprasium* are well distinguished by having bearded nutlets. The present study showed that the presence of trichomes at the apex of nutlets could be used as a taxonomic marker in the delimitation of *Acanthoprasium* nutlets from the rest of Marrubieae. However, these two species were different in shape and sculpturing pattern. While, *A. frutescens* is characterized by elliptic nutlets as well as reticulate pattern. While, *A. integrifolium* is distinguished by triangular nutlets and scalariform sculpturing (Fig. 1B–C).

**Moluccella**—The genus *Moluccella*, characterized by glabrescent stems and leaves as well as an expanded calyx, is distributed from southern Europe to central Asia, Pakistan and Kashmir (Scheen et al., 2010; Bendiksby et al., 2011). In the present study, two out of eight species of this genus were examined. Although both of these species were similar in shape and possession of the largest nutlets (Fig. 3F–H), they were different in the pattern of nutlet sculpturing. *Moluccella aucheri*, distinguished by reticulate pattern of nutlet sculpturing, differs from *M. laevis* with colliculate-granulate pattern of nutlet sculpturing. In addition, the type of anticlinal walls was different between these two species. While *M. aucheri* possesses raised anticlinal walls, *M. laevis* is characterized by anticlinal walls represented by channels. Due to the limited taxon sampling on the genus *Moluccella*, a more comprehensive study needs to be done for indicating the taxonomic importance of nutlet characters.

**Ballota**—Recently, a narrower circumscription of the genus *Ballota* was proposed by Siadati et al. (2018) to encompass the members of sect. *Ballota* (sensu Patzak, 1958) with about three species. These three species are characterized by the following morphological features: five main calyx teeth, rarely with a few additional minute teeth as well as simple and glandular indumentum (Siadati et al., 2018). Although nutlet morphology was not useful in the recognition of the genus *Ballota* from the other genera.
genera of the tribe Marrubiaceae, it was helpful in the
delimitation of taxa at species level. Ballota nigra
subsp. anatolica with lanceolate nutlets was
characterized by foveolate pattern of nutlet
sculpturing (Fig. 1D–E). On the other hand, B. nigra
subsp. ruderalis (Fig. 1F–G) and B. platyloma (Fig.
1H–I) have broadly ovate and ovate nutlets,
respectively, and possess reticulate pattern of nutlet
sculpturing.

**Pseudodictamnus**– The genus *Pseudodictamnus*,
with about 28 species, comprises perennial herbs
with herbaceous bracteoles, expanded calyx limb
with 6–20 teeth and corolla tube shorter than or
equaling the calyx with branched and simple hairs
(Siadati et al., 2018). This genus, which is
phylogenetically known to be the closest relative of
*Ballota* and *Marrubium*, shares similar nutlet
features with them. For instance, the pattern of
nutlet sculpturing among all three species of the
genus *Pseudodictamnus* was reticulate, which is a
common type among the species of *Ballota* and
*Marrubium*. Moreover, these three species were
similar in the following characters: reticulate pattern
of nutlet sculpturing with concave projection in
outer pericinal wall and raised anticinal walls.
However, they were different in the shape of nutlets
(Fig. 1 J, L, N).

**Marrubium**– *Marrubium*, with ca. 50 species, is
morphologically well characterized by the following
features: usually toothed and petiolate leaves,
thyrsoïd inflorescence; calyx 5–15(-30)-lobed,
corolla tube shorter than calyx; stamens included in
corolla-tube, posterior corolla lip 2-lobed (Harley et
al., 2004). In addition, several characters, such as
rounded or subtruncate nutlets at apex and glabrous
or with sessile glands at apex, have been mentioned
in previous studies (Harley et al., 2004).

It is not surprising that *Marrubium*, the largest
genus of the tribe Garrubieae, shows considerable
diversity in nutlet characters such as size, shape, and
sculpturing pattern. *Marrubium leonuroides* and *M.
catarifolium* possessed the smallest and biggest
nutlets among the examined species, respectively.
The common nutlet shapes among the *Marrubium*
species were ovate and oblong. The common
sculpturing pattern of nutlet in *Marrubium* species
was reticulate, however, they showed some
variation in shape and size of the composing cells.

Based on morphological characters the genus
*Marrubium* has been divided into six sections by
which was represented here by *M. astracanicum*, *M.
catarifolium*, *M. leonuroides* and *M. propinquum*,
morphologically characterized by five straight or
curved calyx teeth and purple corolla. Although all
the species attributed to this section were different
in shape, they showed similar type of sculpturing,
except in *M. astracanicum*, which possessed
pustulate sculpturing. While *Marrubium*
yllosoides and *M. persicum*, belonging to
*Marrubium* sect. *Stellata*, were similar in shape of
nutlets but differed in the type of sculpturing, both
studied species of *Marrubium* sect. *Marrubium* (*M.
anisodon* and *M. cuneatum*) differed in the shape of
nutlets but possessed a similar reticulate type of
sculpturing. *Marrubium peregrinum*, belonging to
sect. *Ramosa*, possessed the smallest oblong nutlets
(1.65×0.96 mm) among the studied species and
simple hairs at apex.

This section is morphologically well characterized
by widely branched stems and few-flowered
verticillasters with white corolla (Davis, 1982).
Nutlet sculpturing feature was helpful in the
separation of species, however, it appeared
uninformative at the level of section.

**CONCLUSION**

Nutlet micromorphology provided valuable data in
the separation of the related species within genera of
the tribe. Our findings revealed that the nutlet shape,
sculpturing pattern and the absence/presence of
simple hairs at apex of nutlet were the most
significant features in the separation of taxa at
species level. However, these characters were not
useful in the separation of sections within genera.
Our results, in accordance with previous studies of
Lamiaceae (e.g., Oran, 1996), showed that the shape
of the nutlets were insignificant in assessing the
relationships among *Marrubium* species and its
relatives in the tribe Marrubiaceae. It seemed also that,
contrary to other genera of Lamiaceae (e.g., Guerin,
2005), nutlet characters were of low phylogenetic
value in Marrubiaceae.

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