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Light and scanning electron microscopic study of seeds in *Nigella* L (Ranunculaceae)

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Abstract. Seed morphology of 6 species of *Nigella* L. (Ranunculaceae) viz., *N. sativa* L., *N. hispanica* L., *N. arvensis* L., *N. orientalis* L., *N. nigellastrum* (L) Willk. and *N. integrifolia* Regel, was studied utilising the light and scanning electron microscope to determine the significance of testa features as taxonomic characters. An artificial key based on spermoderm features is proposed to delimit the species studied. The present study supports the treatment of *N. integrifolia* Regel as a monotypic genus *Komaroffia integrifolia* (Regel) Periera.

Keywords. *Nigella* L; seed morphology; taxonomic significance; light microscopy; scanning electron microscopy.

1. Introduction

The genus *Nigella* L (Ranunculaceae) consists of 20 species indigenous to Mediterranean areas of Europe and Central Asia (Willis 1973). Various aspects of the genus *Nigella* have been examined by a number of workers, embryology (Kordyum 1957, 1959; Ly Thi Ba 1962; Vijayaraghavan and Marwah 1969 a, b), cytology (Strid 1965; Gillot 1970; Bhandari *et al* 1976; Suresh and Satyesh 1980), biosystematics (Strid 1970) palynology (Skavarla and Nowicke 1979). Recently Bouman (1978) discussed the seed and seed coat features (LM and SEM) of *Nigella damascena*. To date there is no comparative study on the microcharacters of the testa surface of the *Nigella* species presently studied. The application and use of SEM besides LM has helped in examining the surface features of species with small seeds. We have, therefore, examined 6 species of *Nigella* to assess the value of seed morphology (LM and SEM) by which the species may be distinguished and identified.

2. Materials and methods

Seed material of *Nigella sativa* L, *N. hispanica* L, *N. arvensis* L, *N. orientalis* L, *N. nigellastrum* (L) Willk and *N. integrifolia* Regel was obtained from Botanischer Garten, Der Universitat, Basel (Switzerland). Exomorphic characters like size, colour and testa pattern were observed under LM. For SEM study, the seeds were affixed on aluminium stub by transparent adhesive. The seeds were uniformly coated with gold following the sputtering technique (Damblon 1975), rotating at an angle of 45° to the vaporising filament at an accelerating potential of 10–15 kV and a part of the seed coat was uniformly photographed in all the species (using a Cambridge stereoscan microscope model S4-10 (England)).

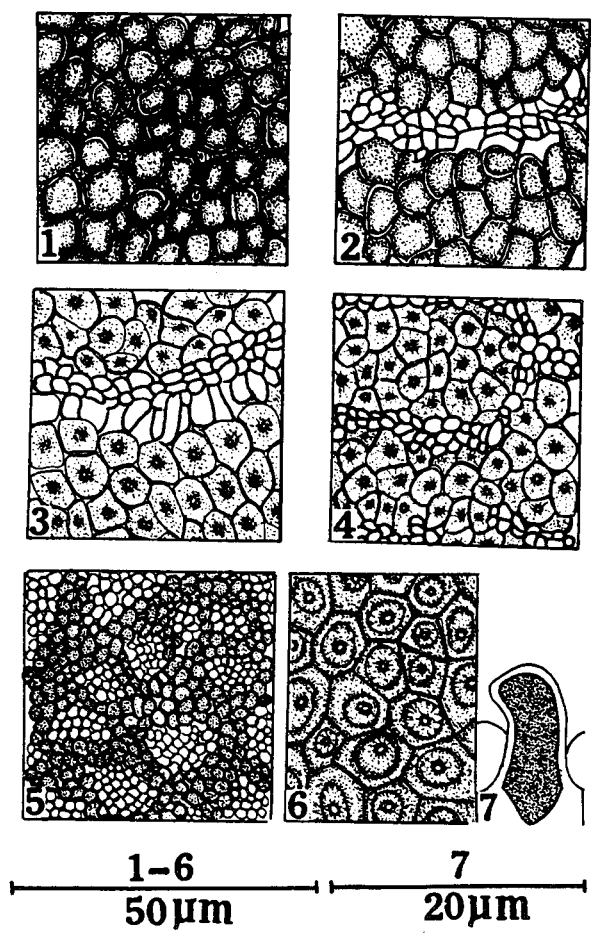
3. Observations

3.1 *Nigella sativa L.*

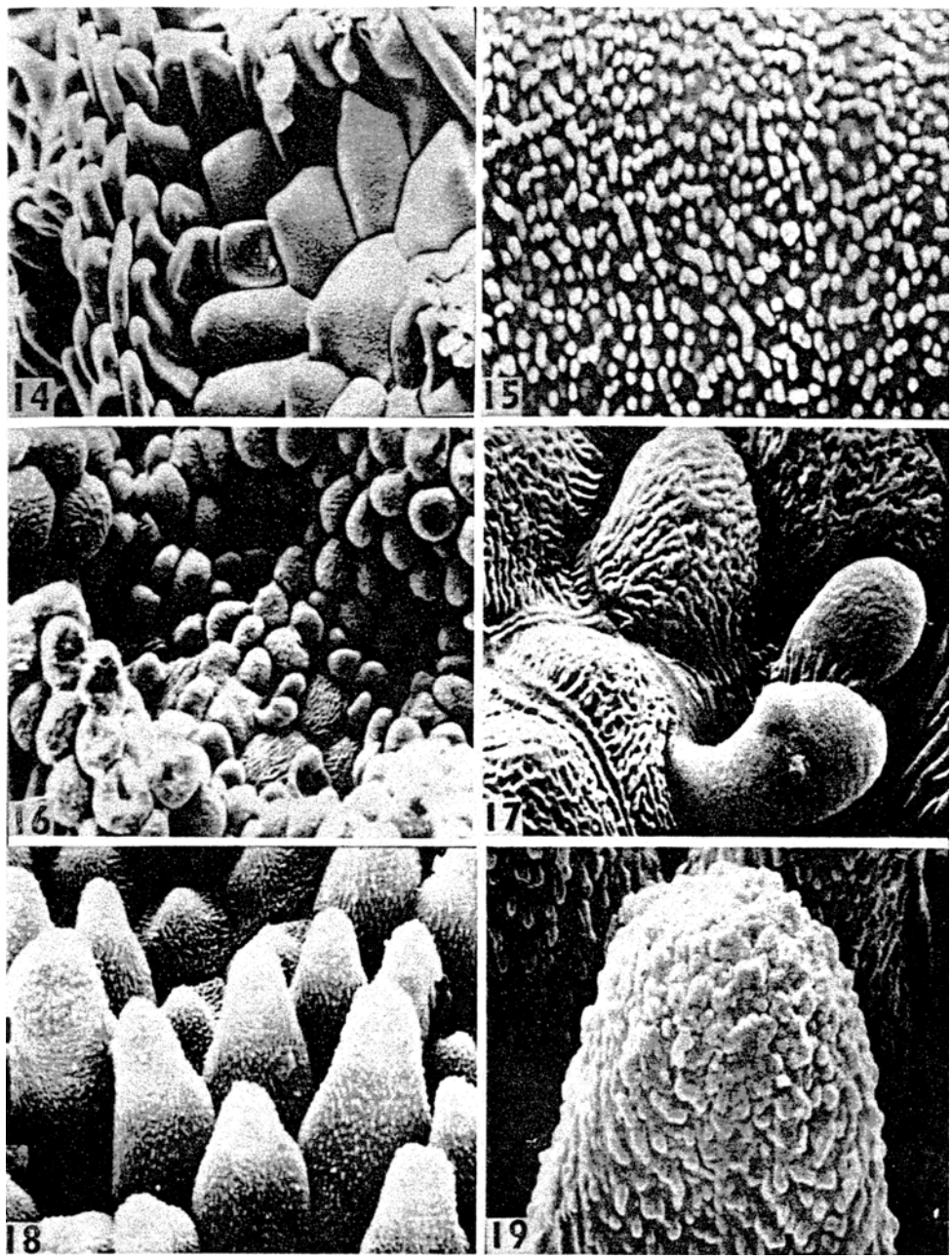
The mature seeds are 3·25 × 1·8 mm, flattened, triangular and black in colour. Testa (LM) shows a reticulate pattern of ridges. The epidermal cells between the ridges are tetragonal or circular in surface view (figure 1). Spermoderm (SEM) shows the epidermal cells with deep concavity looking like cupules and their surface is spinulose (figures 8, 9).

3.2 *Nigella hispanica L.*

The mature seeds are 2·5 × 1·6 mm, flattened, triangular and black in colour. Testa (LM) shows a reticulate pattern of ridges. The cells of elevated ridges are small, thick-walled and dark coloured. The epidermal cells between the ridges are comparatively



Figures 1–7. 1–6. Testa part of *Nigella sativa, N. hispanica, N. arvensis, N. orientalis, N. nigellastrum* and *N. integrifolia.* **7.** Papilla of *N. integrifolia.*



Figures 8–13. SEM photograph of reticulate spermoderm of **8.** *N. sativa* (× 230), **9.** spinulose surface of epidermal cell (× 2300), **10.** *N. hispanica* (× 230), **11.** undulate surface of epidermal cell (× 2300), **12.** *N. arvensis* (× 230), **13.** nipple like projections and granulate surface of epidermal cell (× 2300).

large and polygonal in surface view (figure 2). Spermoderm (SEM) shows the ridges as small, elongated cells interwoven in chains. The epidermal cells between the ridges are large, polygonal with shallow undulate surface (figures 10, 11).

3.3 *Nigella arvensis L.*

The mature seeds are 2·4 × 1·5 mm flattened, ovate and black in colour. Testa (LM) shows the prominent elevated ridges composed of short, thick-walled and dark coloured cells. The epidermal cells between the ridges are comparatively large, pentagonal to hexagonal in surface view, with nipple-like projections (figure 3). Spermoderm (SEM) shows the elevated ridges as small, fusiform to spindle-shaped cells interwoven in chains with less prominent nipple-like projections on them. The epidermal cells between the ridges are flat, penta to hexagonal with distinct nipple-like projection, and of densely granulate surface (figures 12, 13).

3.4 *Nigella orientalis L*

The mature seeds are 3·8 × 1·8 mm flattened, globoid and black in colour. Testa (LM) shows a reticulate pattern of ridges. The cells of ridges are short-elongated, thick-walled and dark-coloured. The epidermal cells between the ridges are comparatively large, flat, penta to hexagonal in surface view with nipple-like projections (figure 4). Spermoderm (SEM) shows the elevated ridges as small, clavate-shaped compactly placed cells. The epidermal cells between the ridges are flat, penta to hexagonal with distinct nipple-like projections and of less granulate surface (figures 14, 15).

3.5 *Nigella nigellastrum (L) Willk.*

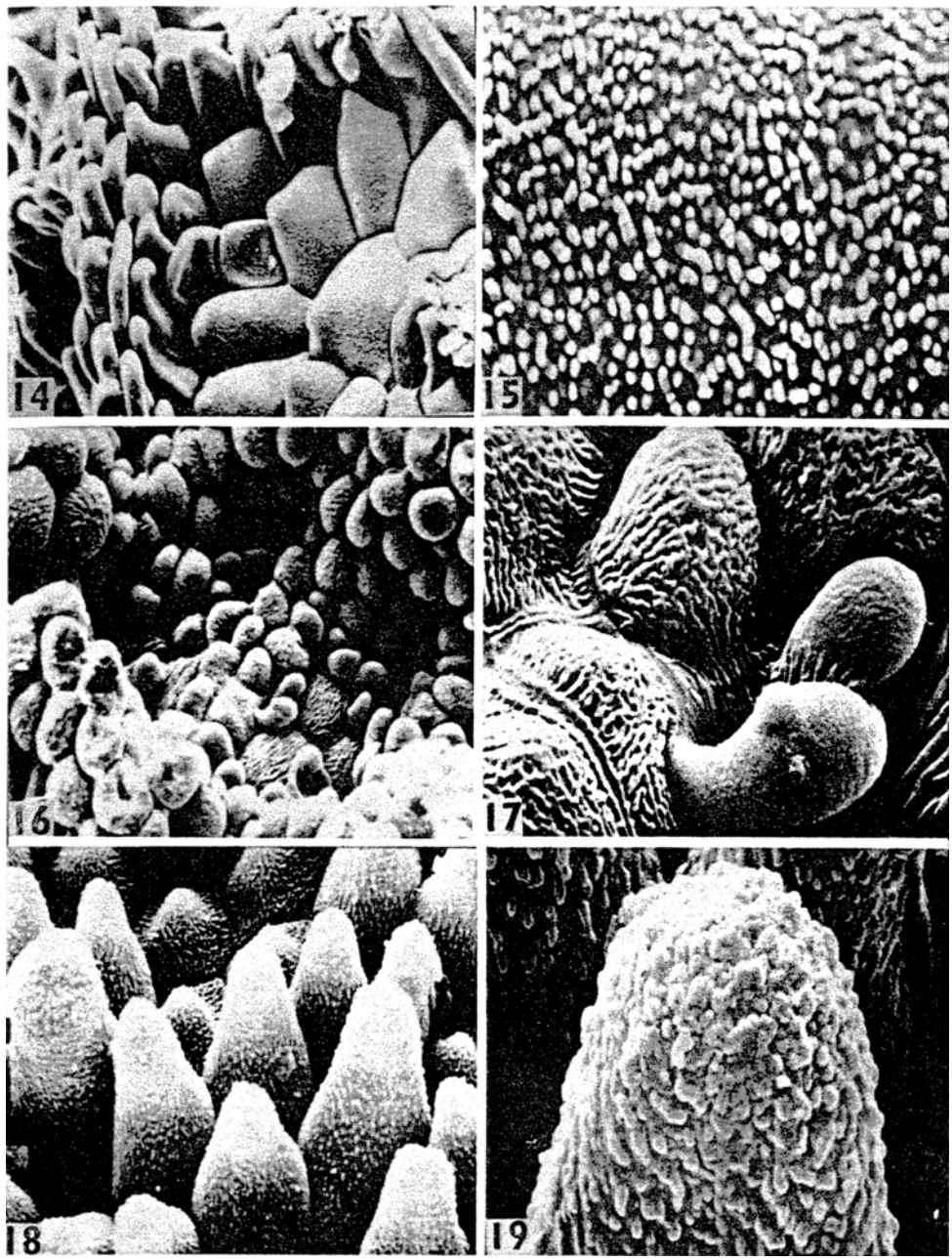
The mature seeds are 2·6 × 1·8 mm flattened, ovate and greyish black in colour. Testa (LM) shows a reticulate pattern of ridges and furrows. The cells of elevated ridges are large circular, thick-walled and dark-coloured. The epidermal cells between the ridges are short and circular in surface view (figure 5). Spermoderm (SEM) shows the elevated ridges of large, circular, reticulate cells, with few of them producing tomentose conical projections. The epidermal cells in furrows are polymorphic with secondary fine reticulations traversed with channel like net-work (figures 16, 17).

3.6 *Nigella integrifolia Regel.*

The mature seeds are 1 × 5 mm flattened, triangular and greyish in colour. Testa (LM) shows a glossy, papillate type, composed of penta to hexagonal cells projecting circular tips in surface views (figure 6). Spermoderm (SEM) shows numerous erected bottle like glossy papillae densely covered by warty structures (figures 18, 19).

4. Discussion

Mature seeds of *Nigella* species (1–3·8 mm in length) are black or greyish in colour. In general the seeds are flattened due to compact development within the capsules (Bouman 1978) and their outlines vary as ovate, globoid or triangular. The seed-coat



Figures 14–19. SEM photograph of reticulate spermoderm of **14.** *N. orientalis* (× 230), **15.** granulate surface of epidermal cell (× 2300), **16.** *N. nigellastrum* (× 230), **17.** reticulate surface of epidermal cell with conical projection (× 2300), **18.** SEM photograph of papillate spermoderm of *N. integrifolia* (× 550), **19.** a papilla with warty surface (× 2300).

surface (LM, SEM) in *N. sativa, N. hispanica, N. arvensis, N. orientalis* and *N. nigellastrum* is of more or less reticulate type, while *N. integrifolia* is of papillate type. The ridges are more prominent at the junction of flattened planes than the transversely oriented ridges of the integumentary part of the seed. The cells of ridges are small, elongated, compactly placed or in interwoven chains. The epidermal cells between the ridges are comparatively large, flat, circular to polygonal in surface view. Under the SEM, cells are cupule-like with a spinulose surface (*N. sativa*), shallow cells (*N. hispanica*), penta to hexagonal cells with prominent nipple like projections and granulate surface (*N. arvensis* and *N. orientalis*); cells polymorphic with fine secondary reticulations (*N. nigellastrum*). But the spermoderm of *N. integrifolia* is unique in possessing glossy bottle-shaped papillate outgrowths of warty surface. Skvarla and Nowicke (1979) similarly noted typical exine in pollen grains of *N. integrifolia* out of 8 species of *Nigella* studied. Further cytological studies by Strid (1970) also indicate variation in chromosomes number; *N. integrifolia* ($n = 7$) in contrast to other species ($n = 6$). Thus palynological, cytological and testa microcharacters support the treatment of *N. integrifolia* as a monotypic *Komaroffia integrifolia* (Regel) Lemos Periera. A key has been proposed on the basis of spermoderm features for the identification of various *Nigella* species studied.

Artificial key based on spermoderm features for the Nigella species studied

Seeds flattened, small, 2·4–3·8 mm, black.
Testa reticulate:
Epidermal cells cupulate, deep, spinulose surface ..... *N. sativa*
Epidermal cells polygonal, shallow, undulate surface ..... *N. hispanica*
Epidermal cells with nipple like projections, densely granulate surface, ridges cells spindle shaped interwoven in chains ..... *N. arvensis*
Epidermal cells with nipple like projections granulate surface, ridges cells clavate shaped and compactly placed ..... *N. orientalis*
Epidermal cells polymorphic, reticulate surface and with rare conical projections ..... *N. nigellastrum*
Seeds flattened, small, 1·00 mm, greyish.
Testa papillate:
Epidermal cells bottle shaped glossy papillae of warty surface... *N. integrifolia*

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