POLLEN MORPHOLOGY OF *ROSA GALLICA* L. (ROSACEAE) FROM SOUTHERN POLAND

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ABSTRACT

The morphology of pollen grains of *Rosa gallica* was studied on the basis of material from 15 natural localities in southern Poland. It was ascertained that the diagnostic features of pollen grains of *R. gallica* were: the presence of deep, often Y-shaped striae in a linear arrangement; numerous perforations of various, rather big diameters on the bottom of striae; the occurrence of operculum on the membrane of ectocolpus; the presence of costae ectocolpi and fastigium in the area of endopori. The most significant differences among the particular samples studied concern the shape of grains. The differences between pollen grains of 15 individuals of *Rosa gallica* from southern Poland are generally slight. The most discriminating measured character is length of equatorial axis of pollen grain and length of ectocolpi and thickness of exine along polar axis. The remaining differentiate the individuals with smaller significance. The differences between Silesian samples and all the other examined ones may result from origin of different Pleistocene refugia.

KEY WORDS: French rose, pollen morphology, light and scanning microscope, multivariante analyses.

INTRODUCTION

In a systematic approach, *Rosa gallica* L. (= *pumila* Jacq.) belongs to the family Rosaceae L. and is the only representative of the section *Rosa* (= Gallicanae DC.) in Poland. In the systematics proposed by Zieliński (1985), this species has no taxa of a lower rank, while the Hungarian botanist Soó (1966) distinguishes two subspecies: *R. gallica*, subsp. *gallica* and subsp. *leiostyla*. In the opinion of Popek (1996), the taxon found in Poland is the subspecies *gallica*. *R. gallica*, subsp. *gallica* has in Poland many forms: var. *austriaca*, var. *cordifolia*, var. *haplodonta*, var. *leiophyla*, var. *subglandulosa* and var. *subinermis*.

Through Polish territory runs the northern limit of *Rosa gallica* (Fig. 1). Klášterský and Browicz (1964) report that this species occurs, or occurred, in 177 localities scattered throughout southern Poland. Most of them occur in the Silesian Lowland, where the rose can be found in the Odra river valley up to Nowa Sól near Głogów and Góra. Major concentrations of species localities, can also be found in the area of Kraków and Zamość (Browicz and Gostyńska 1963; Zieliński 1987). The latest data on the distribution of this species are reported by Zając and Zając (2001). However, not all the localities they cite are reliable, some of them have not been confirmed for a long time. That is why the current localities of *R. gallica* reported in this article are a valuable addition to our knowledge on the distribution of this species in Poland. In "Polish Red Data Book of Plants", *R. gallica*, has the category vulnerable – VU (Zieliński 2001).

Palynological studies of the family Rosaceae L. and genus *Rosa* L. have been conducted long since. In particular, one should mention: Crépin 1889; Mameli Calvino 1950; Erdtman 1952; Heusser 1971; Jičínska, Konalová and Sýkorová 1976; Kuprianowa and Alyoshina 1978; Eide 1981; Katiyar 1982; Kocoń and Muszyński 1982; Fedoronchuk and Savitsky 1987; Savitsky, Dubovik and Fedoronchuk 1987; Hebda and Chinappa 1990a; Hebda, Chinappa and Smith 1988a, b, 1990b, 1991; Ueda and Tomita 1989; Ueda 1994; Jacob and Pierret 2000. Keys based on pollen-grain morphology are included in: Teppner 1965; Reitsma 1966; Eide 1981; Moore Webb and Collinson 1991.

Few researchers have dealt with pollen-grain morphology of *R. gallica*. In the key prepared by Reitsma (1966), he distinguished, apart from 13 other types of pollen grains of Rosaceae, the type – *R. gallica* characterised by wide striae, operculum and costae colpi. Also Kuprianowa and Alyoshina (1978) dealt with the sporomorphs of *R. gallica*. On the

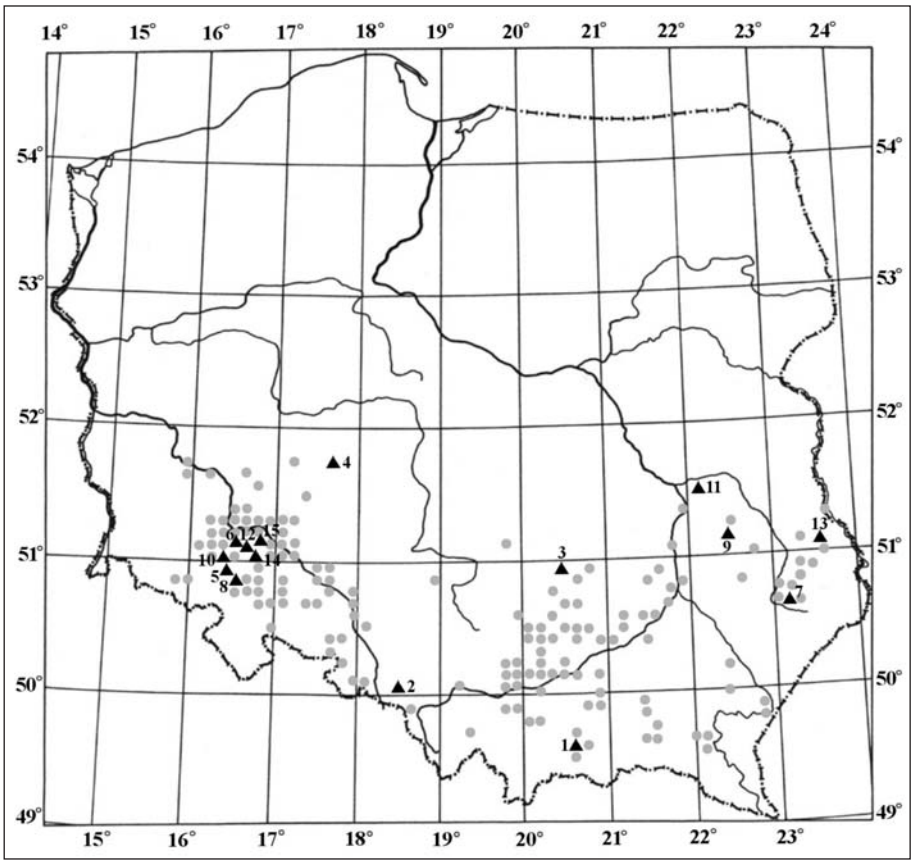

Fig. 1. Range of *Rosa gallica* (after Zając and Zając 2001) with marked positions of samples studied (1-15 as in Table 1).

basis of sculpture, Ueda and Tomita (1989) distinguished 7 types of pollen grains in roses, among them the type represented by *R. gallica* f. *versicolor.* What differentiates it from other roses are the distinct, fairly large perforations and a considerable width and spacing of muri. A description of the pollen grain sculpture of *R. gallica* from Poland, illustrated with microphotographs, is given by Popek (1996). According to him, the grains belong to the subtype *R. gallica* (type *R. canina*), with operculum typical of *R. canina.*

The principal aim of the present research was to describe the morphology of pollen grains of *Rosa gallica* on the basis of a large sample and to define the position vis a vis other authors, concerning the rank of the diagnostic features of grains of the species under study. Another major goal was to analyse the variability of *R. gallica* pollen grains as represented by 15 specimens coming from a variety of places in southern Poland.

MATERIAL AND METHODS

The pollen grains come from 15 herbaria collected by various botanists in 15 natural localities; mostly from southern Poland, one of them in the western part of Poland (Fig. 1, Table 1). Each collection was represented by 50 pollen grains; 750 pollen grains were examined in total. The analysis concerned 9 features (Table 2).

All samples were acetolysed according to Erdtman's method (1952), with insignificant modifications (Wrońska-Pilarek 1998). The pollen terminology follows Hoen (1999). The observations were carried out both with LM (types BIOLAR 2308) and SEM (ISI 60).

The pollen representing particular individuals we compared statistically using measured characters only, using multivariate analyses. The minimal and maximal values of characteristics and arithmetical means were calculated and compared. The discriminant analysis was conducted to determine the discriminant value of particular characters, described by particular Wilks' Lambda and F values. Than the discriminant analysis was used to describe the inter-individual variation and to check the differences between individuals from SW and SE part of Poland. The differences among analysed individuals were verified by the cluster analysis using the nearest neighbour method and Euclidean distances (Marek 1989; Morrison 1990; Watała 2002). The data were analysed statistically by Statistica PL for Windows software.

RESULTS
MORPHOLOGY OF POLLEN GRAINS

A description of the pollen grain morphology of *Rosa gallica* is given below and illustrated with a few SEM and LM photographs (Figs 2-8).

Pollen: tricolporate.

Tectum: perforate.

Outline: polar view; mostly circular, more rarely triangular obtuse convex, elliptic or quinquangular obtuse concave; equatorial view; mostly elliptic, more rarely circular.

Sculpture: striate. It is composed of muri and wide striae separating them. Striae are deep and form a linear pattern; they run parallel or are Y-shaped. On their bottoms are numerous perforations of various, often comparatively big diameters. The sculpture of apocolpium and mesocolpium its striate.

Size: medium; P – 35.96 (25-50) μm, E – 28.06 (20-41) μm.

TABLE 1. Location of studied samples of *Rosa gallica* (KRAM – Herbarium, Institute of Botany, Kraków, KOR – Herbarium, Institute of Dendrology, Kórnik, PZNF – Herbarium Department of Forest Botany Agricultural University, Poznań).

Number of sample	Location	Longitude E	Latitude N	Collector, date of collection, herbarium
1	Prov. Małopolska, Chełmicka Góra	20°40’	49°38’	Pawłowski B. 31.05.1920, KRAM
2	Prov. Śląsk, Gorzyce	21°50’	50°40’	Oklejewicz K. 23.06.1990, KOR
3	Prov. Świętokrzyskie, Grzęby Korzeczkowskie	20°25’	50°48’	Browicz K. 26.06.1960 KOR
4	Prov. Wielkopolska, Kobylin	17°13’	51°43’	Dembecki G. 20.06.1975, KOR
5	Prov. Śląsk, Koiszków	16°11’	51°05’	Szlachetka A., Wrońska-Pilarek D. 19.06.1999, POZNF
6	Prov. Śląsk, Koskowice	16°14’	51°11’	Szlachetka A., Wrońska-Pilarek D. 19.06.1999, POZNF
7	Prov. Lublin, Kosobudy	23°04’	50°37’	Browicz K., Gostyńska M. 17.06.1961, KOR
8	Prov. Śląsk, Krzywlina Mała	16°39’	51°21’	Bobrowicz G. 06.06.1999, POZNF
9	Prov. Lublin, Lublin	22°34’	51°14’	coll. ign. 1920, KRAM
10	Prov. Śląsk, Raczkowa	16°11’	51°08’	Szlachetka A., Wrońska-Pilarek D. 19.06.1999, POZNF
11	Prov. Lublin, between Ryki and Kurów	22°01’	51°28’	Browicz K., Gostyńska M. 11.06.1961, KOR
12	Prov. Śląsk, between Taczalin and Koskowice	16°15’	51°10’	Szlachetka A., Wrońska-Pilarek D. 19.06.1999, POZNF
13	Prov. Lublin, between Teresin and Pięciopolówka	23°33’	50°58’	Browicz K., Gostyńska M. 17.06.1961, KOR
14	Prov. Śląsk, Ogonowice	16°14’	51°06’	Szlachetka A., Wrońska-Pilarek D. 19.06.1999, POZNF
15	Prov. Śląsk, Wrocław–Osobowice	17°01’	51°06’	Zieliński J. 31.05.1971, KOR

TABLE 2. Characters of analysed pollen grains.

No.	Character
1	Length of polar axis (P)
2	Length of equatorial axis (E)
3	P/E ratio
4	Thickness of exine along polar axis (Exp)
5	Thickness of exine along equatorial axis (Exe)
6	Length of ectocolpi (Le)
7	Relative thickness of exine (P/Exp ratio)
8	Relative thickness of exine (E/Exe ratio)
9	Polar area index – PAI (d/E ratio)

P/E ratio: 1.29 (0.83-1.82). Shape of grains: suboblate (0.9%), oblate spheroidal (0.8%), spheroidal (2.0%), spheroidal prolate (16.7%), subprolate (44.3%), prolate (34.9%).

Exine: thickness of exine (Classes of exine thickness were determined on the basis of its thickness in selected species of the family Rosaceae (Eide 1981). Exine thin (0.82-1.45 µm), average (1.46-2.09 µm), thick >2.10 µm.) – average, rarely thin or thick: 1.71 (0.83-3) µm. The exine was measured along polar axis (Exp) and equatorial axis (Exe). The relative thickness of the exine (The relative thickness of the exine, its Exp/P ratio and Exe/E ratio.) measured along P averaged 0.05, and along E – 0.1. The minimum value of the coefficient for both axes is 0.03; the maximum along P is 0.08 and along E – 0.1.

Length of ectocolpi: 28.06 (18-38) µm.

Apocolpium index (PAI): 0.49 (0.31-0.75).

Apertures: 3 ectocolpi, long, deep and sharply tapering, arranged in a regular, longitudinal pattern. Width variable, usually slightly wider in the equatorial region. Outline irregular, narrowly elliptic. Edge of ectocolpi reinforced with costae colpi. The sculpturing of ectocolpus membrane much less distinct than the grain sculpturing, similar to rugulate. There is operculum on aperture membrane. It is positioned in the middle of ectocolpi and covers it partly or completely. Sculpture of operculum, usually psilate. 3 en-

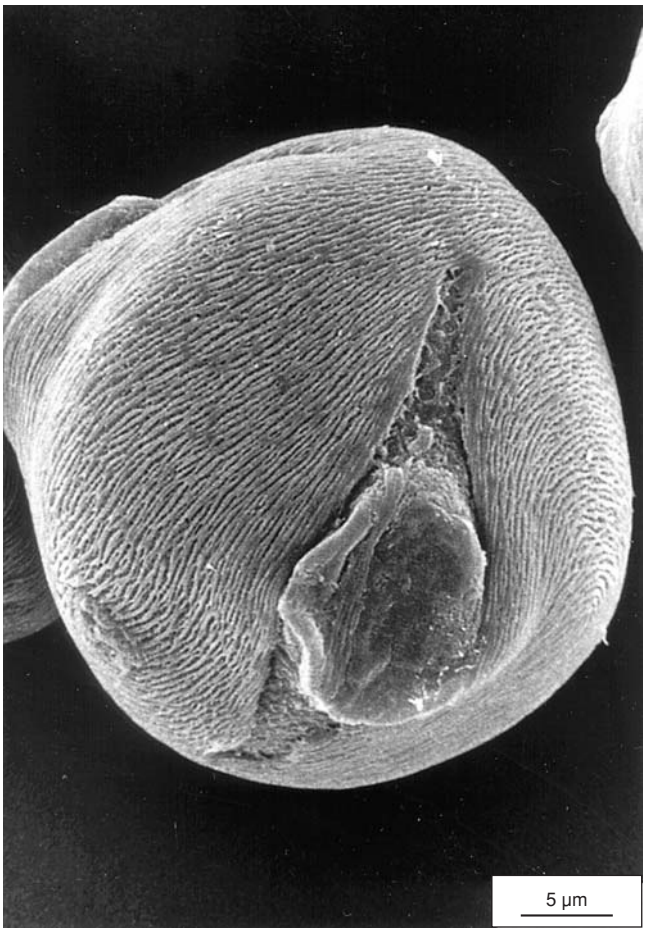

Fig. 2. (SEM). Equatorial view; sculpture of pollen grain and ectocolpi with operculum visible (×3500).

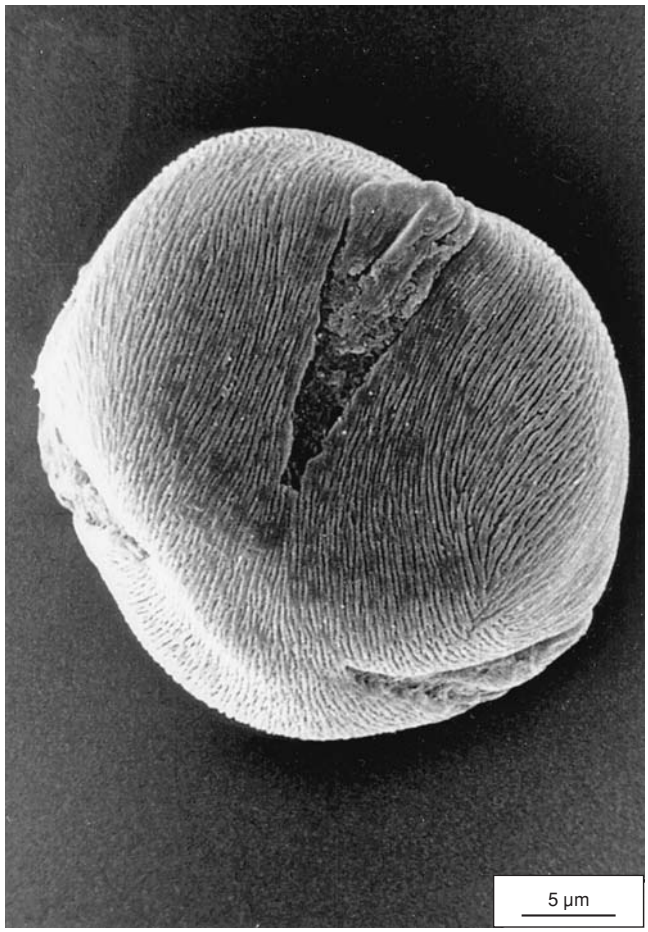

Fig. 3. (SEM). Polar view; sculpture of pollen grain and ectocolpi with operculum visible (×3500).

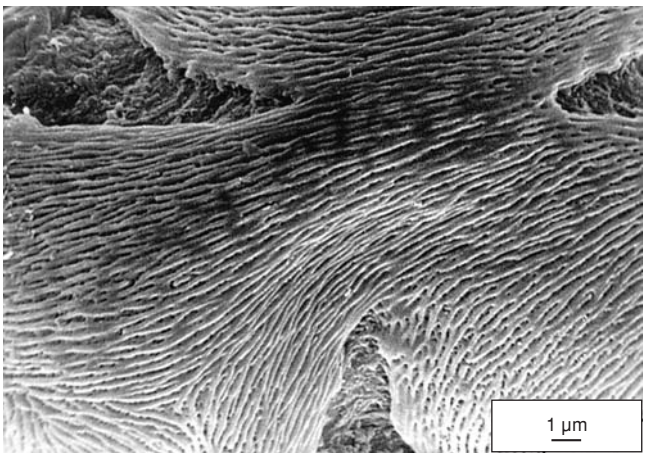

Fig. 4. (SEM). Polar area with three ectocolpi visible (×7500).

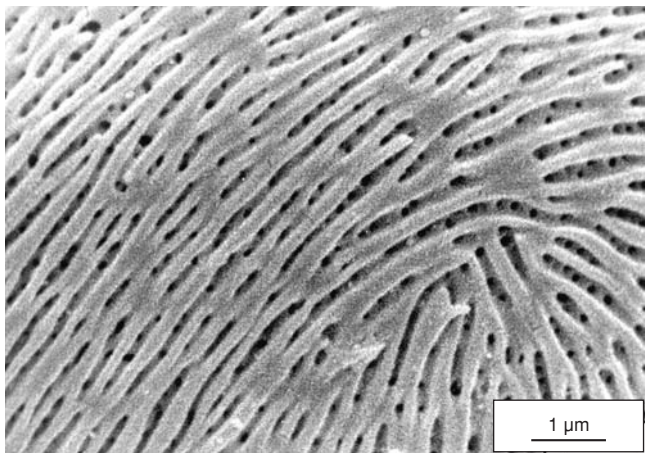

Fig. 5. (SEM). Sculpture of pollen grain; striae, muri and perforations visible (×20 000).

dopori, each placed in the middle of ectocolpi, rather poorly marked. In the endoporus area, fastigium. The edge of endopori irregular.

VARIATION BETWEEN INDIVIDUALS

All measured characters (characters 1-2 and 4-6, see Table 2) differ the individuals statistically significantly. The statistically largest differences between individuals were found in equatorial length (character 2), with partial Wilks' lambda 0.606 and F=33.94 (p=0.0000) (Table 3).

The analysis of discriminant functions of 15 specimens of *R. gallica* coming from various localities showed that the compared pollen grains differed only slightly. The three discriminant variables U_1, U_2 and U_3 account for more than 91% of information of the interindividual variation (Table 4).

The individuals which come from two centers of distribution form one dispersed group (Fig. 9), however, those from the Silesian Lowland, especially individuals 5, 6, 10, 12 and 15, are close to each other. At least a part of individuals from Lubelska Upland and other regions of Southern part of the country also form a kind of subgroup (individu-

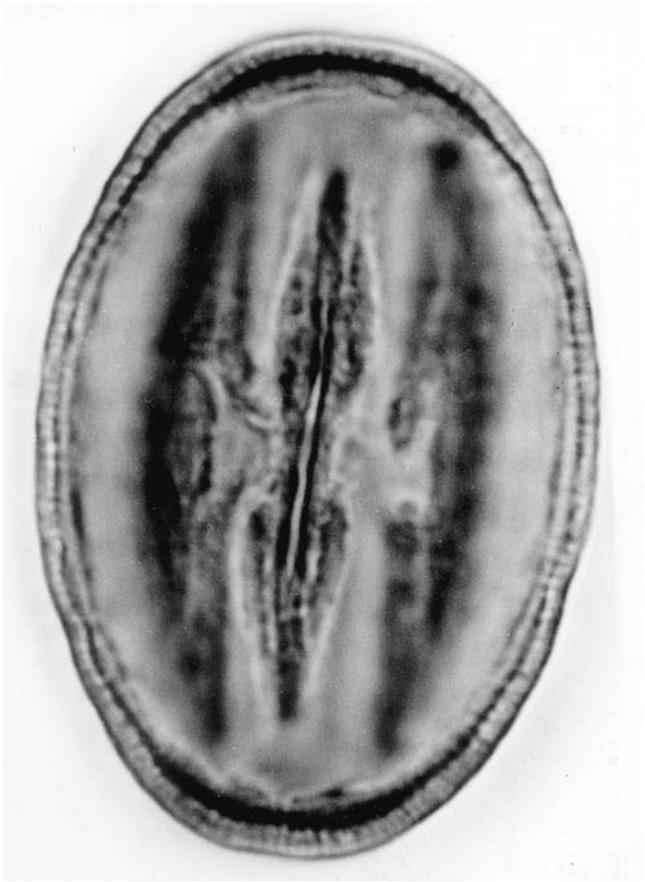

Fig. 6. (LM). Equatorial view; outline of the grain and three ectocolpi visible (×1000).

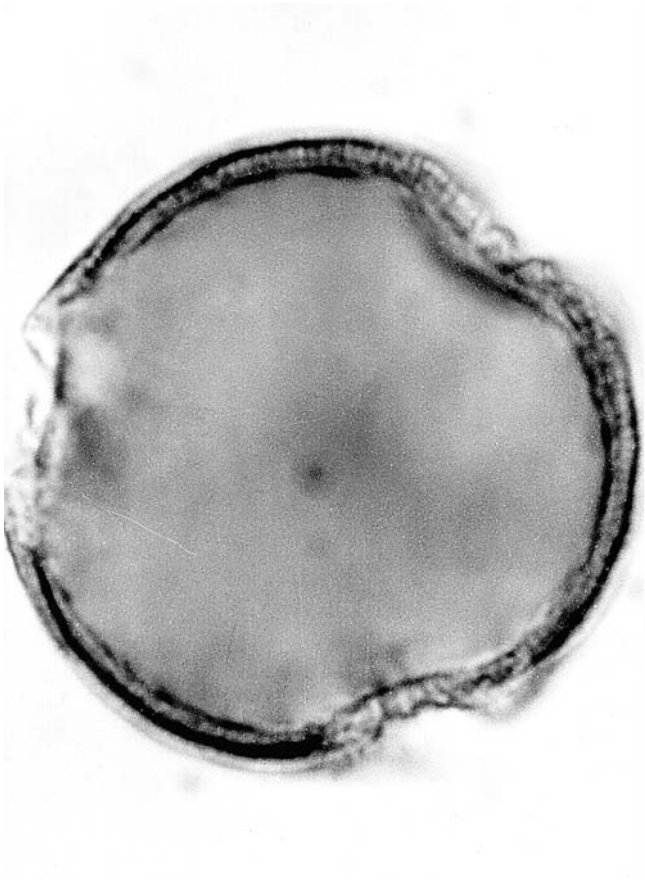

Fig. 7. (LM). Polar view; outline of the grain visible (×1000).

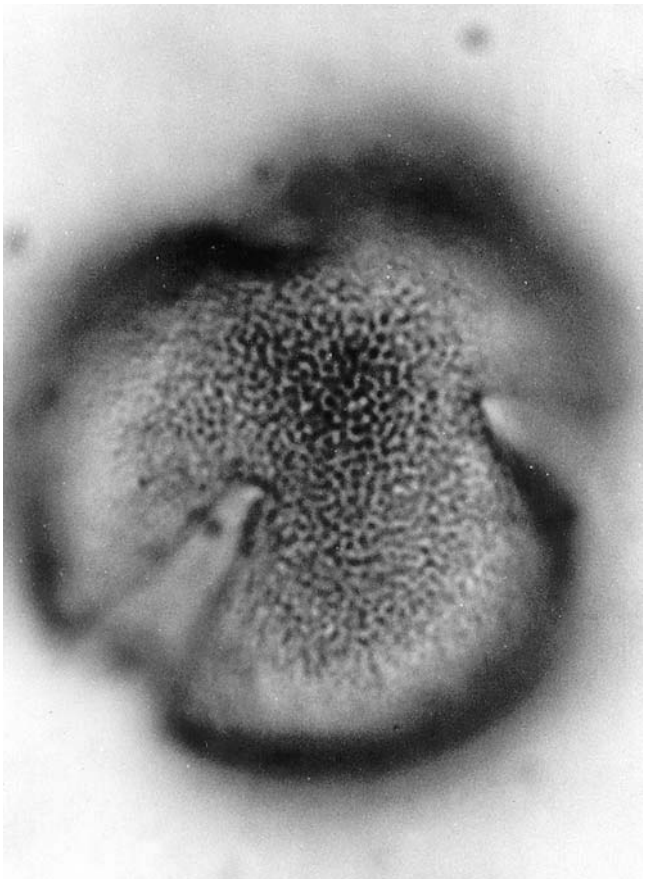

Fig. 8. (LM). Polar view; sculpture of pollen grain and ectocolpi visible (×1000).

TABLE 3. Combination of discriminant analysis for 15 samples of *Rosa gallica*.

Character	Partial Wilk's lambda	F statistic
1	0.8426	9.75
2	0.6061	33.94
4	0.8062	12.55
5	0.8989	5.87
6	0.8260	10.99

TABLE 4. The determination coefficients between discriminant variables U_1, U_2 and U_3 and 5 characters of pollen grains of *Rosa gallica* (character as in Table 2).

Character	U_1	U_2	U_3
1	8.75	16.06	1.74
2	23.42	0.81	6.76
4	13.50	12.94	1.84
5	7.34	5.61	0.69
6	29.34	0.58	4.56

al 1, 3, 4, 7, 11 and 13), but without significant distances to the subgroup from Silesia. The only exception is the individual 9 from the Lublin Upland (Fig. 9).

The dendrogram plotted on the basis of agglomerative grouping using the nearest neighbour method on Euclidean distances clearly proves the distinctness of the individual 9 from Lubelin Upland (Fig. 10). The other individuals form two different groups – the first composed by 5 individuals from the Silesian Lowland (individual 5, 6, 10, 12 and 15), the second by those coming from the South-Eastern part of the country, but also with 3 from the Silesian and Wielko-

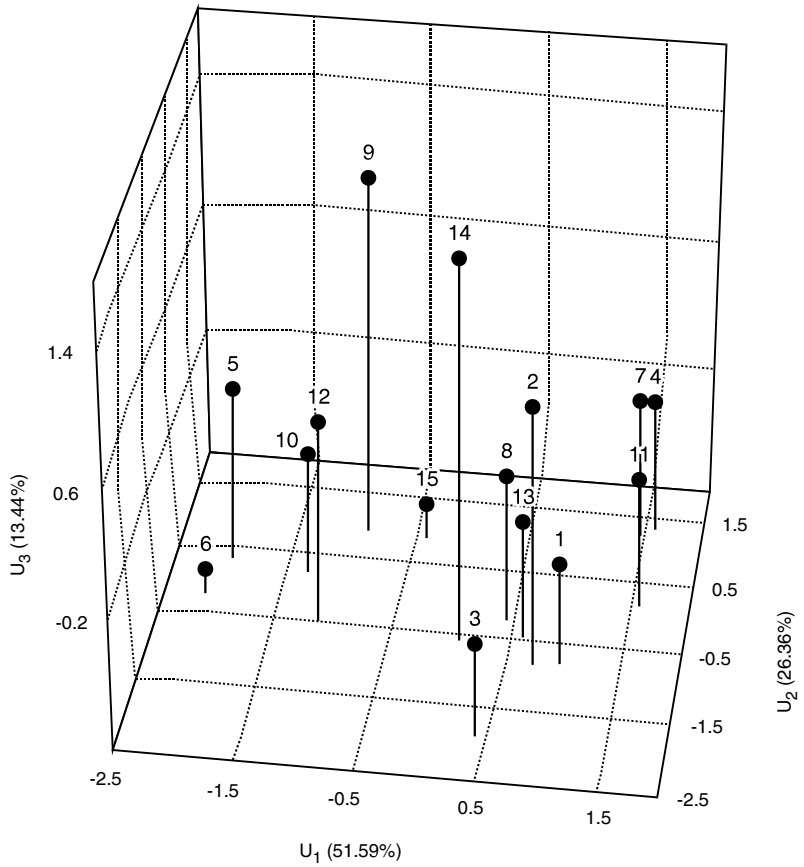

Fig. 9. Result of discriminant analysis for 15 samples of *Rosa gallica* plotted along the three first discriminant variables U_1, U_2 and U_3 (samples numbers as in Table 1).

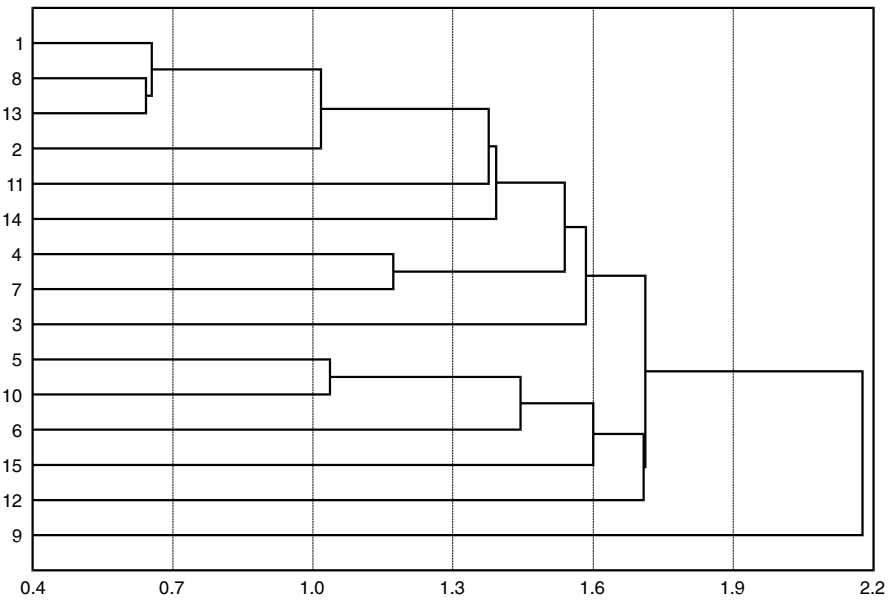

Fig. 10. Dendrogram of 15 samples of *Rosa gallica* constructed on the basis of the shortest Euclidean distances (samples number as in Table 1).

polska Lowlands (individual 1, 2, 3, 4, 7, 8, 11, 13 and 14). In generall, the material from Silesia is distant from both, that collected in the Soth-East of Poland and that from Wielkopolska. Most individuals from the Silesian Lowland have bigger pollen grains and especially longer equatorial axis (character 2). The average value of that character for the closely related individuals 5, 6, 10, 12 and 15 from Silesian Lowland is 30.7 (29.5-33.0) µm, and is significantly higher than for all other individuals.

DISCUSSION

The presented description of pollen grains of *Rosa gallica* is fully consistent with the characteristics and grain sizes given by Kuprianowa and Alyoshina (1978). It differs slightly from the characteristics of Reitsma (1966). This mainly concerns feature 1 – length of polar axis (P), according to this author, it measures a maximum of 27-34 µm, which is smaller than the value obtained in this article. The same concerns feature 9 – polar area index (PAI), which in Reitsma (l.c.) amounts to 0.25-0.35. The most significant

differences between the two studies concern grain shape. The presented results show a considerable proportion of subprolate and prolate grains, with an average frequency of spheroidal prolate grains and occasional spheroidal grains, while in the opinion of Reitsma (1966), their shapes are oblate, prolate and spheroidal, Kuprianowa and Alyoshina (1978) – spheroidal, and Eide (1981) – spheroidal prolate and subprolate. These differences may issue from decreased grain turgor produced by acetolysis, in result of which some grains could have suffered deformation.

The morphological differentiation may result from various origin of *R. gallica* population in Silesian Lowland and other regions of Poland, visible nowadays in the distribution (Browicz and Gostyńska 1963). The recognized ways of colonisation of central Europe by thermophilous plant species from S through Brama Morawska and from SE were described several times (e.g. Czubiński 1950, Tumidajowicz 1964, Kornaś and Medwecka-Kornaś 2002).

CONCLUSIONS

On the basis of the reported analyses of pollen grains of *R. gallica*, the following conclusions can be drawn:

1. The diagnostic features of pollen grains of *R. gallica* are: the presence of deep, often Y-shaped striae in linear arrangement; numerous perforations of various, rather big diameters on the bottom of striae; the occurrence of operculum on the membrane of ectocolpus; the presence of costae ectocolpi and fastigium in the area of endopori. The results obtained are more or less consistent with those of other palynologists.

2. The differences between pollen grains of 15 individuals of *Rosa gallica* from southern Poland are generally slight. The most discriminating characters are: the length of equatorial axis of pollen grain, length of ectocolpi and thickness of exine along polar axis. The remaining characters differentiate the individuals with smaller significance. The differences between Silesian samples and all other examined may result from origin of different Pleistocene refugia.

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