National Academy of Sciences of RA
Electronic Journal of
NATURAL SCIENCES

ՀՀ Գիտությունների ազգային ակադեմիա
Էլեկտրոնային ամսագիր
ԲՆԱԿԱՆ ԳԻՏՈՒԹՅՈՒՆՆԵՐ

Национальная Академия наук РА
Электронный журнал
ЕСТЕСТВЕННЫЕ НАУКИ

1(34), 2020

Botany *Բուսաբանություն* *Ботаника*

STATISTICAL ANALYSIS OF THE BASIC MORPHOLOGICAL CHARACTERISTICS OF POLLEN ON THE EXAMPLE OF SOME REPRESENTATIVES OF THE CHENOPODIACEAE VENT. FAMILY

H.H. Sonyan

Institute of Botany after A. Takhtajian of National Academy of Sciences, Republic of Armenia,
0040, Atcharjan str., 1, Avan, Yerevan
Phone.: (374 10) 61 42 41, 62 17 62
e-mail: hasmiksonyan@gmail.com

ABSTRACT

An investigation of the data of morphological characteristics of pollen of *Caroxylon* Thunb., *Kaviria* Akhani et E.H. Roalson, *Kali* Mill. genera (Chenopodiaceae fam.) was carried out using the statistical analysis. The data obtained confirm that at the level of a light microscope the diameter of pollen grains, the number and diameter of pores for all representatives these three genera can be used as the most characteristic diagnostic features.

Key words: *Chenopodiaceae, pollen–statistical analysis*

INTRODUCTION

Family Chenopodiaceae Vent. in Armenia is represented by 30-36 genera and 80-90 species of annual and perennial herbs, subshrubs, prostrate shrub and very rarely shrubs growing mainly within the lower and middle mountain zones, and the greatest taxonomic diversity is noted for Yerevan and Meghri floristic regions [1, 5].

Almost the absolutely uniformity of pollen of all Chenopodiaceae representatives, both in the form of pollen grains (spheroidal) and in the type of apertures (pantoporate), makes it necessary to search for new methods for the identification of pollen, especially at the level of light microscope (LM), which makes it possible to reveal the differences between taxa (species, genera and etc.).

Among the previously examined five morphological features of pollen, namely the diameter of pollen grains, the number of pores and diameter of pores, the exine thickness, mesoporium width the first three ones can be considered as the most constant [4].

In this work a statistical analysis of three main pollen characteristics mentioned above was carried out on the example of previously studied species (with increasing the number of specimens examined) from the general *Caroxylon* Thunb., *Kaviria* Akhani et E.H. Roalson, *Kali* Mill.

MATERIAL AND METHODS

The pollen material was obtained from the herbarium of the Institute of Botany NAS Republic of Armenia, Yerevan (ERE) as well as from living plants from the collection of the exposition plot “Flora and Vegetation of Armenia” of the Institute of Botany of the NAS of RA.

The study was carried out using a light microscope (AmScope) at magnifications of x200, x400 and x1000. The measurements were carried out on 10 pollen grains for each sample (an average of 3-8 samples). The pretreatment of pollen grains was carried out using two main methods, namely staining with basic fuchsin [3] and the simplified acetolysis method [2].

The statistical analysis was done using Microsoft Excel 2016 [9]. SD - standard deviation[1] and CV% - coefficient of variation[2] were calculated. Gomes [6] noted that CV% can be classified as low (CV%<10%), medium (CV% between 10% and 20%), high (CV% between 20% and 30%), very high (CV%>30%).

[1]The standard deviation is the degree to which the observational data or sets deviate from the mean value [7]. The formula for standard deviation (SD) is $SD = \frac{\sqrt{\sum|x-\mu|^2}}{N}$ where ∑ means "sum of", x is a value in the data set, μ is the mean of the data set, and N is the number of data points in the population [10].

[2] Coefficient of variation is the most universal indicator that reflects the degree of dispersion of values regardless of their scale and units of measurement, is equal to the ratio of standard deviation to the average value and is measured in percent [8] $CV = \frac{SD}{M} x\ 100\%$

In general, pollen characteristics of 38 speciments from 9 species of the genera *Caroxylon* Thunb., *Kaviria* Akhani et E.H. Roalson, *Kali* Mill.were analyzed.

Speciments examined: *Salsola gemmascens* Pall. (=***Caroxylon gemmascens*** (Pall.) Tzvelev): Armenia, Armavir Marz (Hoktemberyan), on the way to the Myasnikyan state farm. 27.08.1979. Leg. A. Barseghyan (ERE, 171291); Armenia, Echmiadzin district, vicinity of Yeraskhaun village, on salt marsh 18.08.2005. Leg. J. Akopian (ERE, 171293); ArmSSR, Meghri district, vicinity of Agarak. 12.05.1978. Leg. E. Gabrielyan (ERE, 171290); *Salsola nodulosa* (Mog.) Iljin (=***Caroxylon nodulosum*** Moq.): ArmSSR, Oktemberyan district, the vicinity of the village Armavir, rocky and shabby slopes (near the cemetery). 7.06.1959. Leg. M. Aslanyan, R. Karapetyan (ERE, 76102); Cuban County. Near the river Gilgilchay. 26.07.1928. Leg. I. Karjagin (ERE, 2203); *Salsola verrucosa* M.Bieb. (=***Caroxylon nodulosum*** Moq.): ArmSSR, Zovashen and Garni. Gammada on steep gypsum-bearing clay slopes. 15.07.1956. Leg. Akhverdov and Yaroshenko (ERE, 128802); Armenia, the vicinity of Yerevan, near the remote village Shorbulakh. 04.07.2007. Leg. J. Akopian (ERE, 168561); *Salsola ericoides* M. Bieb. (= ***Caroxylon ericoides*** (M. Bieb.) Akhani et E. H. Roalson): ArmSSR, Kotayk district, Garni village, Zovashen. Gammads. 12.07.1961. Leg. Ya. Mulkidzhanyan (ERE, 72306); ArmSSR, Hoktemberyan district, the vicinity of the village Shakhvarut, stony saline lands. 8.06.1959. Leg. S. Aslanyan, R. Karapetyan (ERE, 76067); Near the village Etchmiadzin 22.10.1993. Leg. A. Takhtadzhian (ERE, 7065); *Salsola nitraria* Pall. (=***Caroxylon nitrarium*** (Pall.) Akhani et E. H. Roalson): Armenia, Ararat district, environs of Ararat, in saline marshes. 4.07.1999. Leg. E. Gabrielyan (ERE, 171294); *Salsola macera* Litv. (=***Caroxylon nitrarium*** (Pall.) Akhani et E. H. Roalson): Prov. Megri, on the banks of the Araks, between the villages of Megri and Aldara. 26.07.1939. Leg. Yaroshenko (ERE, 28401); Nakhchivan ASSR, Dzhulfa, Darri-Dag, the vicinity of sulfur sources. 3.09.1967. Leg. E. Gabrielyan, P. Gambaryan (ERE, 87469); ***Caroxylon nitrarium*** (Pall.) Akhani et E. H. Roalson: Institute of Botany of NAS RA, plot of flora and vegetation of Armenia. 02.08.2017. Leg. J. Hakobyan (personal collection); *Salsola dendroides* Pall. (=***Caroxylon dendroides***) (Pall.) Tzvelev): ArmSSR, Echmiadzin district, vicinity of the lake Aigr-Lich. 28.06.1961. Leg. V. Avetisyan, E. Gabrielyan, V. Aghababyan, A. Poghosyan, V. Manukyan (ERE, 76089); Echmiadzin district, Kuru-Araks. Old bed of the Araks river. 19.07.1960. Leg. A. M. Barseghyan (ERE, 86593); ArmSSR, Meghri, Nyuvadi. 13.08.1965. Leg. Ya. Mulkidzhanyan, A. Barseghyan, P. Gandilyan (ERE, 82553); ArmSSR, Sisian district, Darabass gorge, village Urut, southwest slope. 23.07.1959. Leg. Ya. Mulkidzhanyan, S. Aslanyan (ERE, 76081); *Salsola australis* R. Br. (= ***Kali tragus*** (L.) Scop): Dzoraget and Uzunlar, the left dry bank of the Dzoraget river. 10.08.1954. Leg. Ya. Mulkidzhanyan (ERE, 76071); Prov. Megri. Meghri, along the river. 23.07.1939. Leg. Yaroshenko (ERE, 27970); Martuni district, Lichk village, lakeshore. 25.07.1949. Leg. R. Karapetyan (ERE, 47055); Vedi, sand pit. 19.06.1964. Leg. V. Manakyan (ERE, 82901); ArmSSR, Sevan lake basin, Ardanish bay near the meteorological station on travertines. 10.07.1960. Leg. S. Narinyan, R. Karapetyan (ERE, 76076); Lake Sevan. Novo-Bayazet district. Noraduz village, the western shore of the lake. 23.07.1947. Leg. R. Karapetyan (ERE, 39622); Armenia, Ararat Marz, Artashat, in the semi-desert. 20.06.1975. Leg. A. Barseghyan (ERE, 172511); ***Kali tragus*** (L.) Scop.: Institute of Botany of NAS RA, plot of flora and vegetation of Armenia. 02.08.2017. Leg. H. Sonyan (personal collection); *Salsola tamamschjanae* Iljin (= ***Kali tamamschjanae*** (Iljin) Akhani et E. H. Roalson): Vedi district, southeast of Vedi. 11.09.1935. Leg. A. Takhtadzhian (ERE, 3157); Armenia, the vicinity of Yerevan, Karmir-bloor, on red clay. 9.09.2002. Leg. J. Hakobyan (ERE, 150915); *Salsola cana* K. Koch (= ***Kaviria cana*** (K. Koch) Akhani): Dzhulfa, region semideserto 17.09.34. Leg. A. Takhtadzhian (ERE, 12181); Nakhchivan ASSR. The surroundings of the salt mine. 14.07.1972. Leg. N. Khanjyan, T. Popova (ERE, 103595); Sovetashen, Bartzrashen, gammada, on the steep clay-gravel slopes 27.07.1996. Leg. Severov and Akhverdov (ERE, 71593); SE of Vedi, gravelly slopes. 11.09.1935. Leg. Takhtadzhian (ERE, 19874); ArmSSR, Vedi district, 17 km from Arazdayan, in the direction of Gepokhchi, on the spurs of the Urts ridge, south-east slope. Friganoid vegetation. 2.09.1959. Leg. A. Takhtadzhyan, E. Gabrielyan, V. Avetisyan (ERE, 73537); Nakhichevan ASSR, neighborhood of the village Yaigi, the house on the road to the village Mastara, by the highway. 2.08.1965. Leg. Ya. Mulkidzhanyan, A. Barseghyan (ERE, 82904); Nach. ASSR. Ordubad, Nakhichevan. 29.07.1959. Leg. Y. Mulkidzhanyan S. Aslanyan (ERE, 70310); *Salsola tomentosa* (Moq.) Spach (= ***Kaviria tomentosa*** **(**Moq.) Akhani): Nakhichevan ASSR, neighborhood of the village Yaigi, the house on the road to the village Mastara, by the highway. 2.08.1965. Leg. Ya. Mulkidzhanyan, A. Barseghyan (ERE, 82903); Armenia, Yerevan Botanical Garden, 1200 above sea level. Plot of flora and vegetation of Armenia. It was grown from seeds collected from clay slopes of the vicinity of the village of Zovashen. Plants of the second year of life, second order shoot. 13.08.1992. Leg. J. Akopian (ERE, 171311); Dashlu x N. Karabakhler. on the left side of the highway; 700 m, gypsum-bearing places on very steep slopes, rocky-gravelly places. 12.07.1949. Leg. Akhverdov, Mirzoyeva (ERE, 128801) Nakhichevan ASSR, Ordubad region, the beginning of the road to Meghri, saltwort desert, on the hills, southeast slope. 20.08.1965. Leg. A. Barseghyan (ERE, 172544); ***Kaviria tomentosa*** subsp takhtadzhianii (Iljin) Akopian: Julfa, saltwort semi-desert, on the hills. 17.09.1934. Leg. A. Takhtadzhian (ERE, 12179).

RESULTS AND DISCUSSION

As was established earlier [4], the main distinguishing pollen features of representatives of the genera *Caroxylon*, *Kali*, *Kaviria* at the level of a light microscope are the diameter of pollen grains, the number and diameter of pores. Statistical analysis of these features was carried out on the basis of updated, more expanded (by the number of samples) palynomorphological data (***Table 1***).

Table 1. Data on a statistical analysis of pollen characteristics for the genera *Caroxylon, Kali, Kaviria*

Species	Pollen size (µm)		Pore number		Pore diameter (µm)	
	± SD	**CV%**	**± SD**	**CV%**	**± SD**	**CV%**
Caroxylon *gemmascens*	16.2 ±2.7	16.6%	19± 1.5	7.8%	2.6±1.1	42.3%
C. nodulosum	17.6 ±1.8	10.3%	30±2.1	7.0%	2.0±0.1	5.0%
C. ericoides	20.6 ±3.1	15.0%	32±1.1	3.4%	2.2 ±0.4	18.2 %
C. nitrarium	17.7±1.2	6.8%	30±0.8	2.6%	1.6±0.3	18.7%
C. dendroides	16.7±2.5	14.9%	28±2.2	7.8%	1.7±0.3	17.6%
Kali *tragus*	20.8±1.8	8.6%	28±0.9	3.2 %	1.8±0.3	16.6 %
K. tamamschjanae	20.6±0.1	0.5%	30±2.1	7.0%	2.2±0.3	13.6%
Kaviria *cana*	18.7±1.5	8.0%	10±0.9	9.0%	3.5±0.7	20.0%
K. tomentosa	15.7±0.7	4.4%	13±1.3	10.0%	2.6±0.3	11.5%
Intervals of variation **± SD** and **CV%**	±0.1-3.1	0.5-16.6%	±0.8-2.2	2.6-10.0%	±0.1-1.1	5.0 -42.3%

Analysis of the genus *Caroxylon* has shown that among the 5 studied species, the least variable species in point of the standard deviation and variation coefficient concerning to all three morphological characters is *C. nitrarium*. The most variable variation coefficient for all three morphological characters, as well as the maximum variable standard deviation (in the case of pore diameter) is observed on the species *C. gemmascens*. On the species *C. ericoides*, the maximum variable standard deviation was noted only for the diameter of pollen grains, and on *C. dendroides* – for the number of pores.

In general, the relatively varied variation coefficient for all five species of the genus *Caroxylon* is a sample of data on the diameter of pollen grains (6.8%–16.6%) and pore diameter (5.0%–18.7% (42.3%)), whereas the sample on the number of pores (2.6%–7.8%) seems to be the most reliable. According to the standard deviation, the data sample on the diameter of pollen grains (± 1.2−3.1) and the number of pores (± 0.8−2.2) is relatively variable and the sample on the pore diameter looks the most reliable and varies within ± 0.3−1.1.

The statistical analysis of two studied species of the genus *Kali* shows that for the species *K. tragus*, the most variable standard deviation and variation coefficient were observed relatively to the diameter of pollen grains, and the least variable – in the case of the number of pores. As for *K. tamamschjanae*, on the contrary, the most variable standard deviation and variation coefficient are noted in the case of the number of pores.

A relatively variable variation coefficient for the genus *Kali* in general is observed for the sample of data on the pore diameter (13.6%–16.6%), while sample on the diameter of pollen grains ranges between 0.5%–8.6%, and the number of pores – from 3.2% to 7.0%. A relatively variable standard deviation was observed for the sample of data on the diameter of pollen grains (± 0.1–1.8) and the number of pores (± 0.9–2.1), while the sample of pore diameter does not exceed ± 0.3 and looks most reliable.

Among the 2 species of the genus *Kaviria*, the most variable standard deviation and variation coefficient concerning to the diameter of pollen grains and the pore diameter, were observed on the species *K. cana*, the least variable – on *K. tomentosa*. At the same time, the standard deviation and the variation coefficient on the number of pores are almost the same for both species.

A relatively variable variation coefficient for the genus *Kaviria* is generally observed in the sample of data on pore diameter (11.5%–20.0%), while the sample on the diameter of pollen grains varies from 4.4% to 8.02%, and the number of pores is in the range between 9.0%–10.0 %. In general, the standard deviations for all three morphological traits varies from ± 0.3 to ±1.5.

CONCLUSION

Analysis of the coefficient of variation for all three genera shows that the sample of data on the number of pores is the low variable (CV = 2.6-10.0%). While the variation coefficient on diameter of pollen grains and pore diameter is low or medium variable (CV=0.5-16.6% and CV= 5.0-20.0% respectively).

Nevertheless, the results obtained are in the frames of data reliability (with the exception of *Caroxylon gemmascens*, where the coefficient of variation concerning to the pore diameter is very high variable (42.3%)).

Thus, the statistical analysis confirms the previously noted assumptions that, at the level of a light microscope, the diameter of pollen grains, the number and diameter of pores on the representatives of all the above three genera can be used as the most characteristic distinctive features.

ACKNOWLEDGMENTS

The studies were carried out in the Palynological laboratory of the Department of Plant Taxonomy and Geography of the Institute of Botany of the NAS RA. We express our sincere gratitude Dr. of Sci. A. M. Hayrapetyan for

consultations during the palynological investigations. We thank Associate Professor in the Department of General of the Agriculture of the ANAU S. Mamajanyan for the valuable advices in conducting statistical analysis.

REFERENCES

[1] Akopian J.A., Biomorphological peculiarities and taxonomical composition of the Chenopodiaceae Vent. family in South Transcaucasia, Abstract of the Doct. Sci. thesis... Doct. Biol. Sciences, Yerevan, 2013, 49 p., (in Russ.).
[2] Avetisyan E.M., Simplified acetolysed method of the pollen grains treatment, Bot. Zhurn., 1950, 35, 4, 385-387, (in Russ.).
[3] Smoljaninova L.A., Golubkova V.F., To the method of the pollen investigation, Dokl. AN USSR, 1950, 75, 1, pp. 125-126, (in Russ.).
[4] Sonyan H.H., New data on the pollen morphology of a number of species from the genera *Caroxylon* Thunb., *Kaviria* Akhani et E.H. Roalson and *Kali* Mill, (Family Chenopodiaceae Vent.), Biol. Journ. Arm., 2018, 4 (70), 69-74, (in Russ.).
[5] Takhtadzhyan A.L., Mulkidzhanyan Y.I., Family Chenopodiaceae. Flora of Armenia, Yerevan, 1956, 2, 519 p., (in Russ.).
[6] Vaz M.A.B., Pacheco P. S., Seidel E.J., Ansuj A.P., Classification of the coefficient of variation to variables in beef cattle experiments, Ciência Rural, 2017, v.47, N11, pp. 1-4.
[7] https://dic.academic.ru/dic.nsf/ntes/4538/%D0%A1%D0%A2%D0%90%D0%9D%D0%94%D0%90%D0%A0%D0%A2%D0%9D%D0%9E%D0%95
[8] https://studfiles.net/preview/5316293/page:3/
[9] https://microsoft-excel-2016.ru.softonic.com
[10] https://www.khanacademy.org/math/probability/data-distributions-a1/summarizing-spread-distributions/a/calculating-standard-deviation-step-by-step