RESEARCH ARTICLE

Variability of morphometric characteristics of *Dactylis glomerata* L. (Pooideae Benth.) leaf phytoliths at two stages of the growing season

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Abstract

The study is devoted to the analysis of the morphometric parameters of Crenate phytoliths of the *Dactylis glomerata* L. leaf epidermis. The use of morphometry for the nomenclature and identification of phytoliths determines the relevance of studying factors that impact on the size of silica bodies. One of the possible reasons for morphometric differences may be the phenophase in which the herbarium material was collected. We collected herbarium material of *D. glomerata* from its two habitats at the beginning and the end of the growing season. Morphometric data of phytoliths extracted from leaves were processed using descriptive statistics and analysis of variance. The Crenate study of *D. glomerata* phytoliths at different stages of the growing season shows that some parameters tend to increase, while others remain stable and may have a taxonomic potential. Finally, the increase in the size characteristics of phytoliths at the end of the growing season has been revealed. Such parameters as area, width and equivalent diameter are stable. By the end of the growing season, the phytoliths have a more elongated and irregular shape. The most stable shape parameters are roundness, compactness and aspect ratio.

Introduction

Phytoliths are a class of microfossils produced in many, but not all, families of higher plants and composed of amorphous silica or opal. They are a useful class of microfossils that are used as a proxy for reconstructing terrestrial vegetation, agricultural practices, and diets, especially in arid, semiarid, and oxidative contexts where pollen is scarce. Although phytoliths have been especially well produced and studied in grasses (Poaceae) and sedges (Cyperaceae), they are found in over 50 other families of higher plants. Given their relatively low level of taxonomic specificity, they work particularly well in multiproxy projects complementing information from other sources.

The specificity of phytoliths varies among different plant families. For temperate latitudes of Eurasia, including the south of Western Siberia, it is not possible to identify dicotyledonous plants even at the family level, but it is possible to identify such important taxa as Pinaceae, Poaceae, Cyperaceae (Gavrilov and Golyeva 2014; Lada and Gavrilov 2016; Speranskaya et al. 2018; Solomonova et al. 2016, 2018). The specificity of the structure of the grasses leaf epidermis at the level of subfamilies, phylogenetic clades and other taxonomic levels is widely known, which is reflected in the diagnostic significance of the main morphotypes of Poaceae short cells (Esau 1965; Twiss et al. 1969; Twiss 2001; Lu and Liu 2003; Bobrov et al. 2001; Rudall et al. 2014). Phytoliths of long cells, trichomes, and vesicular cells of grasses form differently than phytoliths in short cells. In the first case, phytoliths form as a cast along the cell wall due to passive evaporation during the process of transpiration, and therefore they are not much diagnostic from a taxonomy point of view. In the second case, phytoliths are under the control of enzymes, and their shape is genetically predetermined (Piperno 2006). It was the morphometric research of phytoliths that allowed taxonomic specificity of phytoliths studies to be expanded and deepened. (Hoskova et al. 2021, 2022). The morphometry of cultivated grasses has significantly advanced (Zhijun et al. 1998; Ball et al. 1996, 1999, 2017; Portillo et al. 2006; Out and Madella 2016; Wang et al. 2019; Yost et al. 2021; Chen et al. 2023). At the same time, there are data about the variability of phytoliths of short epidermal cells depending on environmental factors, for example, soil, light, humidity, temperature (Ball and Brotherson 1992; Dunn et al. 2015; Liu et al. 2016 a,b; Out and Madella 2016; Solomonova et al. 2023).

An important factor for assessing the possibility of using morphometric data is to comprehend the variability of the characteristics of the phytolith size and shape at different stages of plant maturity. The data obtained while studying the formation of phytoliths at the early stages of plant development show that the silicification of short cells begins from the moment the leaf blade of the first leaves unfolds (Black-

man 1969; Attolini et al. 2023). Mature phytolith in plants is observed even before panicle formation (Hodson et al. 1985). However, such studies cannot answer the question of whether the morphometric parameters of phytoliths will differ between phenological stages of the same plant or not. It is customary to select plants at the wilting stage for the phytolith morphometry (Ball et al. 1996, 1999, 2017; Dunn et al. 2015; Out and Madella 2016). Some scientists directly indicate that the stage of maturity should influence the phytolith size (Lisztes-Szabo et al. 2014).

The development of morphometric studies of phytoliths at different phenological stages will allow us to resolve the question of whether it is possible or not to conduct a morphometric study of phytoliths from herbarium samples collected in the summer, since this is the most frequent expedition time in temperate latitudes. According to our observations, many grasses during the wilting period, which it is recommended to collect a herbarium for phytolith research, cannot be identified as species, since they often lack caryopses and in some cases the glumes also fall off. It would be preferable to identify grasses at the flowering and fruiting stages.

The present study is devoted to Crenate (CRE) (Fig. 1), one of the most common morphotypes of C3 grasses (Solomonova et al. 2022) on the *Dactylis glomerata* L. model species. We studied two phenological stages of this plant, namely tillering and the beginning of the wilting stage. According to ISPN 2.0, crenate is classified as GSSCP (Grass silica short cell phytoliths) (Neumann et al. 2019). Two forms have been identified in earlier studies for the Crenate morphotype. For example, N.K. Kiseleva highlights wavy plastes for Agropyron, Elymus, Koeleria and double-circuit forms for Calamagrostis, Poa (Kiseleva 1989). Taking into account the number of blades, M. S. Blinnikov distinguishes two forms of phytoliths in plants and soils of the interior Pacific Northwest (USA) (Blinnikov 2005). Recent data from southern Western Siberia show that the identification of wavy and polylobed morphotypes may carry an ecological and taxonomic signal (Solomonova et al. 2019a, 2019b, 2022). A study by S. Liszte-Szabo et al. (2014) shows variability in the length and width of this morphotype in *Poa pratensis* L. (in the study, elongate crenate, but in Figure 1. C, p. 854 most forms can be compared with Crenate). The study of 6 different populations of *D. glomerata* revealed a correlation between the mean values of most morphometric characteristics of phytoliths and annual precipitation, and the phytolith width correlates with the mean January temperature (Solomonova et al. 2023). Crenate phytoliths of *D. glomerata* are larger in forest phytocenoses compared to open habitats (Solomonova et al. 2023). The influence of environmental factors on size can also be noted for other forms of phytoliths. The size of saddles in *Phragmites australis* depends directly on humidity and annual average temperature (Liu et al. 2016a). But, for example, no dependence of size on climatic factors can be traced for bilobed forms of *Pennisetum pedicellatum* phytoliths of grasses; the impact of humidity and evaporation is noted only for the morphometric characteristics of vesicular phytoliths (Issaharou-Matchi et al. 2016).
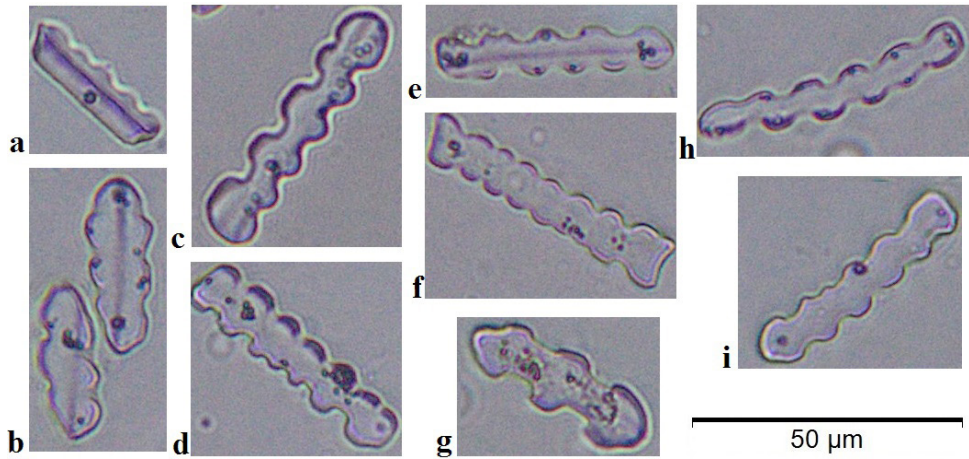
Figure 1. Diversity of CRE phytoliths in *D. glomerata* leaf: a – lateral view, b, c, e-i – measured top- view, d – half-sided phytolith, h, i – examples of "curved phytoliths".

Materials and methods

To study the seasonal variability of the morphometric characteristics of *D. glomerata* phytoliths two sites were selected in the vicinity of the city of Barnaul. Site 1, a working settlement of Yuzhny, is an apple garden with dominance of *D. glomerata* in the grass cover, while site 2 is a post-forest meadow on the edge of a pine forest on the territory of the South Siberian Botanical Garden. Five specimens of *D. glomerata*. were collected from each site twice during the growing season, namely on May 05, 2023 and on September 09, 2023. The study of phytoliths was carried out for basal and stem leaves together. Sheaths were also included in a single sample with leaf blades. Leaf sizes were measured for each sample. Fifty phytoliths were studied from each sample. Thus, the sample of phytoliths was 250 for each season at each site. In total, we measured 1000 phytoliths.

Phytolith extraction protocol according to the modified method of A.A. Golyeva (Golyeva 2001) and the phytolith research was published in Solomonova et al. 2023. 17 morphometric parameters recommended by the ICPM (International Committee on Phytolith Morphometry) were measured (Fig. 2 for size and Ball et al. 2016, Table 2 for shape).

To analyze the data obtained, descriptive statistics and analysis of variance were performed in the Past 4.03 program. Correlation analysis (linear correlation, Pearson's coefficient, significance level ≤ 0,05) was also performed between the average values of the morphometric characteristics of phytoliths and the size of basal leaves, i.e. average length and length of the largest leaf (Table 1). It was not possible to measure the leaves on the generative shoot, since they were absent in May and were in a dry state in September. The sample number of measured leaves varied from 8 leaves in May to 21 leaves in September, depending on the degree of preservation.

Table 1. Values of the mean leaf length and the length of the largest leaf (Max.) in all samples

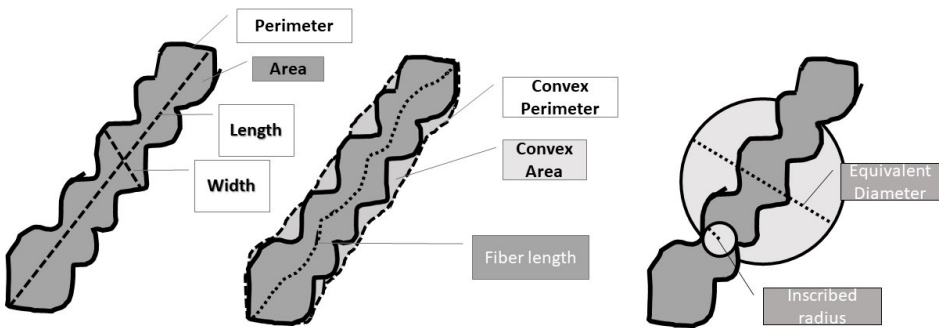|	May									
Samples №	**1.1**	**1.2**	**1.3**	**1.4**	**1.5**	**2.1**	**2.2**	**2.3**	**2.4**	**2.5**
Mean, cm ± Std. error	20± 1.8	24±2.3	36±1.7	22±1.5	27±1.8	32±2.1	41±2.4	32±1.9	33±1.7	30±1.6
Max., cm	25	30	41	25	32	42	58	40	39	36
	September									
Samples №	**1.1**	**1.2**	**1.3**	**1.4**	**1.5**	**2.1**	**2.2**	**2.3**	**2.4**	**2.5**
Mean, cm ± Std. error	56±3.1	63±4.8	54±2.8	49±4.3	58±3.2	56±3.1	43±3.4	44±2.2	67±3.1	35±2.4
Max., cm	61	73	62	60	67	61	50	54	74	42

Figure 2. Size parameters for crenate.

Result

Tables 2 and 3 present the results of descriptive statistics of morphometric data on the phytolith size dimensions of the crenate morphotype of *D. glomerata*. Table 4 shows the results of the analysis of variance of the morphotype size characteristics. Normal probability plots were created for the variables used in ANOVA analysis (For example, Fig 3). For most variables, the distribution of values closely matched the expected normal distribution (e.g., Area $p<0.020$, Convex Area $p<0.020$, Length $p<0.018$, Fiber length <0.029, Equivalent diameter $p<0.005$, Roundness $p<0.005$, Convexity $p <0.005$, Aspect Ratio $p<0.005$, Elongation $p<0.005$, and Curl <0.009). For some other variables, p values were generally above 0.05 value, but still suggested close fit to the normal distribution.
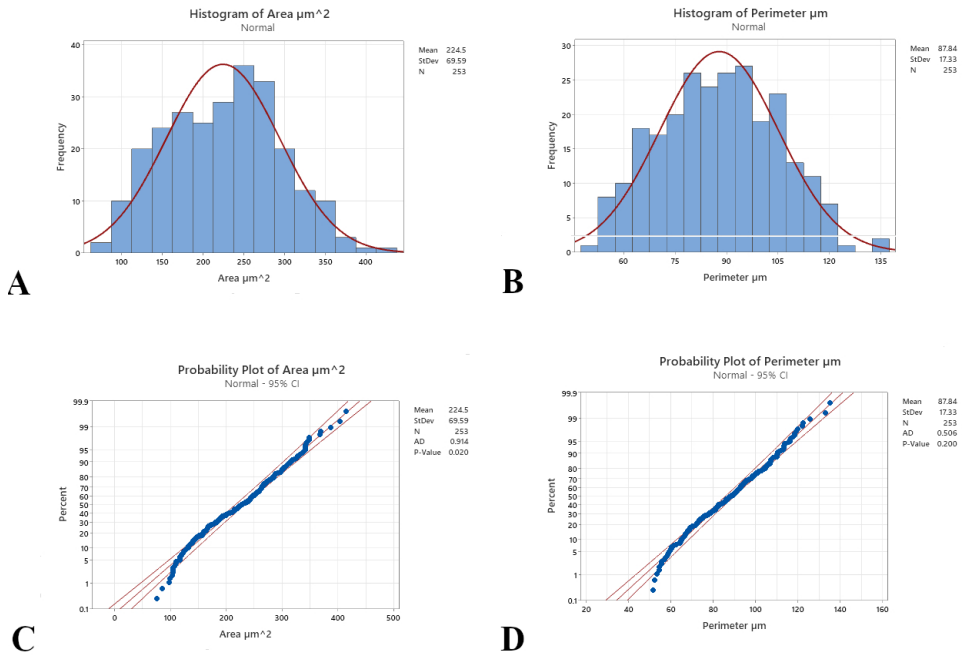
Figure 3. Normal histogram (A – area, B – perimeter) and normal probability plots (C – area, B – perimeter).

The maximum values of the crenate size parameters at site 1 show an increase in values from May to September. A similar pattern is observed at site 1, with the exception of the maximum values of the width and inscribed radius. The minimum values of the phytolith size characteristics at sample point 1 increase, with the exception of the phytolith length. At site 2, *D. glomerata* phytoliths exhibit both an increase (circumscribed perimeter, length) as well as a decrease (area, circumscribed area, width, equivalent diameter and inscribed radius) of the minimum values of a number of characteristics from May to September, and the minimum values of some indicators remain without changes (perimeter, midline length). It is also worth noting that the differences in the minimum phytolith value are not large for most characteristics between May and September, while the differences are much higher for the maximum size values. For example, the area of the smallest phytolith at site 1 differs by 9.43 μm^2 between May and September, while the area of the largest one differs by 62.37 μm^2.

Phytoliths in *D. glomerata* leaves show a slight increase in mean size values between May and September at both sample points for most characteristics. Statistically significant differences were identified in specimens of *D. glomerata* phytoliths from site 1 in the following parameters: circumscribed area, perimeter, circumscribed perimeter, length and midline length. As for site 2, the following changes in the morphometric characteristics of the studied phytoliths are significant: perimeter, length of the midline, inscribed radius. The most stable indicators at both sam-

ple points are the width, area and equivalent diameter of the studied morphotype of *D. glomerata* leaf phytoliths. The mean value of the inscribed radius decreases when comparing phytoliths of plant samples collected in May and September; these changes are statistically significant for samples of *D. glomerata* leaves from the second population.

Table 2. Characteristics of seasonal variability in the phytolith size of the crenate *D. glomerata* morphotype at site 1

	Area, μm²		Convex area, μm²		Perimeter, μm		Convex Perimeter, μm		Length, μm		Fiber length, μm		Width, μm		Equivalent Diameter, μm		Inscribed Radius, μm	
M.	5	9	5	9	5	9	5	9	5	9	5	9	5	9	5	9	5	9
Min	75.54	84.97	101.1	102.4	51.54	52.02	44.61	44.91	17.23	16.85	20.31	21.16	5.334	5.726	9.808	10.402	0.858	1.187
Max	414.7	477.0	513.3	568.9	135.2	153.2	112.5	129.7	51.05	60.02	57.76	68.26	14.72	15.24	22.98	24.65	4.716	5.523
Mn	224.8	232.8	272.2	287.4	87.95	92.54	75.17	78.85	32.96	34.61	36.94	39.05	9.646	9.607	16.71	17.01	2.715	2.595
St. er.	4.379	4.560	5.208	5.642	1.088	1.253	0.915	1.073	0.459	0.539	0.501	0.582	0.118	0.0936	0.169	0.1666	0.0492	0.0447

Note: M. – month (5 – May; 9 – September); Min – minimum parameter value; Max – maximum parameter value; Mn – average value of parameters; St. er. – standard error. All values are measured according to Ball et al. 2016.

Table 3. Characteristics of seasonal variability in the phytolith size of the crenate *D. glomerata* morphotype at site 2

	Area, μm²		Convex area, μm²		Perimeter, μm		Convex Perimeter, μm		Length, μm		Fiber length, μm		Width, μm		Equivalent Diameter, μm		Inscribed Radius, μm	
M.	5	9	5	9	5	9	5	9	5	9	5	9	5	9	5	9	5	9
Min	155.9	145.9	182.1	166.8	66.49	66.28	57.23	58.47	24.29	25.04	27.19	27.22	7.109	6.196	14.09	13.63	0.842	0.465
Max	802.4	902.6	992.4	1238	236.3	275.7	184.7	210.6	85.35	97.82	97.99	110.9	17.77	16.41	31.96	33.90	6.043	5.72
Mn	396.4	396.7	470.2	491.6	125.5	134.8	106.6	110.6	47.82	49.31	53.28	56.13	11.30	11.28	22.19	22.19	3.389	3.084
St. er.	7.883	8.253	10.01	11.55	2.111	2.396	1.698	1.829	0.851	0.900	0.952	1.041	0.126	0.133	0.2220	0.2281	0.0593	0.0544

Note: M. – month (5 – May; 9 – September); Min – minimum parameter value; Max – maximum parameter value; Mn – average value of parameters; St. er. – standard error.

Table 4. Assessment of the significance of differences between the average sizes of the studied phytoliths using the analysis of variance method

№	St	Parameters								
		Area	Convex Area	Perimeter	Convex Perimeter	Length	Fiber Length	Width	Equivalent Diameter	Inscribed Radius
1	F	1.579	3.903	7.644	6.822	5.388	7.607	0.0678	1.683	3.297
	ρ	0.2095	**0.04875**	**0.005906**	**0.009277**	**0.02067**	**0.006026**	0.7945	0.1952	0.07002
2	F	0.0007072	1.954	8.408	2.608	1.433	4.09	0.01766	0.0004232	14.45
	ρ	0.9788	0.1628	**0.003902**	0.107	0.2318	**0.04368**	0.8943	0.9836	0.0001614

Note: № – number sign, St. – statistical parameters, F – F- criterion, ρ – significance level ≤ 0,05 (significant differences are highlighted in bold).

Tables 5 and 6 present the results of descriptive statistics of morphometric data describing the shape of phytoliths of the crenate morphotype. Table 7 shows the results of the analysis of variance of the shape characteristics of the studied morphotype. The minimum values of the morphometric shape characteristics of the *D. glomerata* phytoliths from site 1 differ slightly. The phytolith shape of the site 2 samples shows the variability of the minimum values of elongation and aspect ratio. The maximum values of the phytolith shape characteristics for the site 1 samples decrease in terms of the shape factor, i.e. more irregular, namely different from a circle, particles are found among the phytoliths of plants collected in September. The maximum values of the shape factor differ slightly for the site 2 samples. The site 1 samples are characterized by the most elongated phytolith shapes of the September samples compared to the May ones. The variability of the maximum values of roundness, aspect ratio, and elongation indicators point out that the September samples contain the most elongated phytoliths. It is also worth noting a decrease in the maximum values of the convexity and solidity parameters in the samples of both sites from May to September, i.e. the autumn samples contain phytoliths with the largest recesses between the lobes.

Analysis of the mean form factor values shows a decrease in this parameter at both sites, i.e. the degree of phytolith "irregularity" relative to the circle increases from May to September. In both cases, the differences between the means are significant. The differences among the mean indicators of roundness, compactness, and aspect ratio are statistically insignificant, thus, these morphometric characteristics of the *D. glomerata* crenate form are the most stable. The mean convexity and solidity values of the studied phytoliths in the September samples are lower than in the May samples. The differences in solidity of both samples are statistically significant, as for convexity the differences in means are significant only for the site 2 samples. Both parameters decrease with the increase of recesses. In our case, this indicates the presence of more pronounced lobes in crenate phytoliths of September samples compared to plants collected in May. A statistically significant increase in elongation and curl from May to September is observed in plant samples at both sites.

Considering the stability of the phytolith aspect ratio in plants from both sites and the small differences in the phytolith length from the site 2 samples, it can be noted that elongation increases due to the greater irregularity of the phytolith shape of the September samples.

Table 5. Characteristics of seasonal variability in the phytolith shape of the crenate *D. glomerata* morphotype at site 1

	Form factor		Roundness		Convexity		Solidity		Compactness		Aspect ratio		Elongation		Curl	
M.	5	9	5	9	5	9	5	9	5	9	5	9	5	9	5	9
Min	0.193	0.197	0.117	0.123	0.742	0.739	0.726	0.672	0.341	0.351	1.73	1.735	2.088	2.124	0.767	0.786
Max	0.582	0.488	0.525	0.515	0.925	0.906	0.912	0.892	0.725	0.718	6.992	7.122	7.504	8.099	0.97	0.969
Mean	0.3671	0.3470	0.2725	0.2604	0.8558	0.8524	0.8251	0.8112	0.5173	0.5054	3.517	3.667	3.934	4.136	0.8921	0.8844
St. er.	0.00432	0.00409	0.00463	0.00462	0.00194	0.00199	0.0024	0.00234	0.004403	0.004444	0.0600	0.06408	0.06430	0.06927	0.00244	0.002164

Note: M. – month (5 – May; 9 – September); Min – minimum parameter value; Max – maximum parameter value; Mn – average value of parameters; St. er. – standard error.

Table 6. Characteristics of seasonal variability in the phytolith shape of the crenate *D. glomerata* morphotype at site 2

	Form factor		Roundness		Convexity		Solidity		Compactness		Aspect ratio		Elongation		Curl	
M.	5	9	5	9	5	9	5	9	5	9	5	9	5	9	5	9
Min	0,154	0,125	0,083	0,089	0,693	0,639	0,66	0,618	0,288	0,299	1,637	2,134	1,836	2,446	0,796	0,777
Max	0.579	0.581	0.559	0.456	0.92	0.924	0.942	0.929	0.748	0.676	9.165	9.695	9.884	10.487	0.96	0.987
Mean	0.334316	0.292156	0.2391	0.22648	0.85298	0.826364	0.849584	0.81828	0.482456	0.468948	4.354612	4.521364	4.843688	5.128656	0.898456	0.88002
St. er.	0.00583613	0.00531384	0.00495743	0.00492629	0.0029111	0.00364222	0.00306151	0.0032432	0.005047	0.00513887	0.09022538	0.09608651	0.09871323	0.1060914	0.0021674	0.00247178

Note: M. – month (5 – May; 9 – September); Min – minimum parameter value; Max – maximum parameter value; Mn – average value of parameters; St. er. – standard error.

Table 7. Assessment of the significance of differences between the average shape of the studied phytoliths using the analysis of variance method

№	St	Parameters							
		Form Factor	Roundness	Convexity	Solidity	Compactness	Aspect Ratio	Elongation	Curl
1	F	11.39	3.436	1.453	17.18	3.595	2.94	4.582	5.612
	ρ	**0.0007963**	0.06439	0.2287	**4.00E-05**	0.05852	0.08701	**0.03279**	**0.0180433**
2	F	28.53	3.261	32.59	49.26	3.517	1.601	3.867	34.45
	ρ	**1.40E-07**	0.07156	**1.96E-08**	**7.35E-12**	0.06132	0.2064	**0.0498**	**3.40E-08**

Note: № – number sign, St. – statistical parameters, F – F- criterion, ρ – significance level ≤ 0,05 (significant differences are highlighted in bold).

Discussion

The present study shows that in addition to the mean values of morphometric parameters, the minimum and maximum values are also subject to variability. At the same time, the minimum values of the phytolith size are more stable, since the three-lobed phytolith is the minimum for all samples. The number of blades for the largest phytoliths can vary.

Significant differences were noted between the means for 6 out of 9 morphometric parameters at least at one site. Some model experiments and growing plants from different populations show that environmental factors can influence the phytolith size (Ball and Brotherson 1992; Dunn et al. 2015; Out and Madella 2016). A decrease in the phytolith parameters of area, length and width with increasing light intensity was shown for the studied morphotype Crenate on the example of *Poa secunda* J Presl, (Dunn et al. 2015). In our research, the phytolith area of the May and September samples is practically the same, while at site 1, the area of the described figure (Convex area) increases, but is much less than in the experiment of R. E. Dunn et al., i.e. 15 μm² in our study and almost 200 μm² in a model experiment (Dunn et al. 2015). In our research, the phytolith width does not differ significantly between the leaf samples collected in May and September. Our study notes an increase in the mean values of morphometric indicators from May to September, and the analyzed model experiment (Dunn et al. 2015) shows a decrease in the phytolith size depending on an increase in light intensity. In the context of data comparison, we can assume that shading may also have had an effect on the phytolith size in our samples. Site 1 is located in a park, in the shade of apple trees, and an increase in crown density during the growing season could reduce insolation and affect the increase in mean size indicators (variability is observed for 5 parameters here). Site 2 is located on the edge of a pine forest and *D. glomerata* here experiences only partial shading; the morphometric parameters of phytoliths from site 2 samples are more stable (3 parameters out of 9 vary).

In addition to the factor analyzed above, the observed changes can also be explained by the degree of the plant maturity. Comparing our data with the length of the largest leaf (Table 1), we see a significant difference between the leaf sizes at site 1 and a smaller difference for site 2, in both cases the length increases, perhaps that is why the size indicators at site 1 are more variable than at site 2. At the same time, correlation analysis has not revealed a significant correlation between the phytolith size and the leaf length (Table 8).

Table 8. Correlation analysis

Size	Leaf length mean		Leaf length max		Shape	Leaf length mean		Leaf length mean	
	r	ρ	r	ρ		r	ρ	r	ρ
Area	0.049	0.837	0.099	0.678	Form Factor	-0.347	0.133	-0.354	0.126
Convex Area	0.098	0.682	0.140	0.554	Roundness	-0.165	0.486	-0.179	0.451
Perimeter	0.148	0.534	0.184	0.436	Convexity	-0.296	0.205	-0.299	0.200
Convex Perimeter	0.113	0.636	0.151	0.524	Solidity	-0.499	0.025	-0.440	0.052
Length (Feret)	0.099	0.679	0.137	0.564	Compactness	-0.161	0.498	-0.173	0.466
Fiber Length	0.131	0.579	0.170	0.473	Aspect Ratio	0.105	0.660	0.124	0.603
Width	0.002	0.994	0.044	0.855	Elongation	0.147	0.536	0.167	0.483
Equivalent Diameter	0.048	0.838	0.0969	0.684	Curl	-0.597	0.005	-0.595	0.006
Inscribed Radius	-0.248	0.291	-0.189	0.425					

Note: r – pearson's coefficient, ρ – two-tailed probabilities.

Some studies (Liszte-Szabo et al. 2014; Liu et al. 2016a; Solomonova et al. 2023) have observed variability in the morphometric parameters of CRENATE depending on the environmental conditions of the sample point. Thus, a change in environmental conditions during the growing season may be another reason for the variability in the morphometric characteristics of *D. glomerata* between the May and September samples.

A number of phytolith shape parameters also show significant variability. The phytoliths of mature plants are more irregular than those of young ones. For example, (Fig. 1h, i) curved shapes are visually marked in both planes. Some forms, for example, curved in the lateral projection of the phytolith, are technically impossible to unfold and measure in the top projection. The phytoliths become more elongated and have more pronounced lobes by September. This is also affected by an increase in size (possibly the number of blades). There is a significant inverse correlation

with the long leaf blade for the solidity and curl parameters (Table 8). As the leaf length increases, the severity of the blades increases, because the value of the solidity parameter decreases with increasing recesses. A similar dependence of solidity on the leaf length is observed in the analysis of Crenate. *D. glomerata* phytoliths in different populations (Solomonova et al. 2023). At the same time, phytoliths become more curved as the leaf enlarges, which indicates the relationship between these parameters.

The phytolith shape of the May samples corresponds to Crenate, despite the difference in some morphometric parameters. This is consistent with the studies that show the formation of a typical shape phytolith before the panicle stage (Hodson et al. 1985). A number of studies have shown that phytoliths have a species-specific shape in the early stages of leaf development (Attolini et al. 2023; Blackman 1969; Fernandes Honaine et al. 2016), and the process of silicification itself increases as the plant grows (Attolini et al. 2023; Fernandes Honaine et al. 2016) and may depend on environmental factors (Issaharou-Matchi et al. 2016). *Festuca exaltata* young leaves (3–8 cm) in a folded state have Rondels phytoliths, typical of the fescue genus (Attolini et al. 2023).

The obtained morphometric data indicate that the degree of plant maturity can influence the phytolith size and some of its form characteristics, which were used to distinguish different forms of Crenate. This should be taken into account in further research. A morphometric study of this morphotype is necessary both in various Pooideae tribes and grasses of various ecological groups, and in the soils of various phytocenoses.

Conclusion

Morphometric parameters of phytoliths are an important key to the development of paleoecological indices. A Crenate study of *D. glomerata* phytoliths at different stages of the growing season shows that some parameters tend to increase, while others remain stable and may have taxonomic potential.

Perhabs, climatic changes during the growing season, phenological changes in the studied phytocenoses, for example, increased darkening due to the formation of the crown in edificators. More pronounced lobes of Crenate phytoliths and their degree of curvature depend on the size of the leaf blade, which can be influenced by the environmental factors and the growing season.

The findings indicate the potential of morphometric studies to assess the paleoecological and taxonomic potential of phytoliths. Further direction of research may include analysis of the morphometric parameters of this morphotype in different types of grasses and in phytolith complexes of soils under different vegetation.

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