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Research Article

Comparative morphological characteristics of the chromosomes of two *Rheum* species

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Abstract. *Rheum maximowiczii* Losinsk. and *Rheum turkestanicum* Janisch. are closely related species occurring in the flora of Uzbekistan, yet their cytogenetic characteristics and karyological relationships remain insufficiently documented. In this study, chromosome number, karyotype structure, karyomorphometric characteristics, and selected morphological traits were comparatively investigated. Cytogenetic analyses were performed using metaphase chromosome spreads obtained from root apical meristem cells, and karyotype parameters and asymmetry indices were calculated. Morphological characteristics of the vegetative and reproductive organs were also compared between the two species. The results revealed that *Rheum maximowiczii* is diploid (2n = 2x = 22 = 22m), whereas *Rheum turkestanicum* is tetraploid (2n = 4x = 44 = 44m). Both species possess highly symmetrical karyotypes composed exclusively of metacentric chromosomes and were classified within Stebbins' (1971) 1A karyotype category. The total length of the monoploid chromosome set was 33.53 ± 1.82 μm in *R. maximowiczii* and 28.82 ± 0.31 μm in *R. turkestanicum*. Differences were also detected in several karyomorphometric parameters and morphological traits, particularly in stamen number, which ranged from 9 to 14 in *R. maximowiczii* but remained consistently nine in *R. turkestanicum*. These findings provide new insights into the cytotaxonomy, karyotype evolution, and systematics of *Rheum*, and establish a valuable basis for future molecular cytogenetic and phylogenetic investigations of the genus.

Key words: *Rheum maximowiczii*, *Rheum turkestanicum*, karyotype, karyomorphometry, cytogenetics, polyploidy, chromosomes.

1. Introduction

Cytogenetic studies play a pivotal role in elucidating the taxonomic classification and evolutionary relationships of plants. Since the beginning of the twentieth century, chromosome-based investigations have made substantial contributions to clarifying phylogenetic relationships among diverse taxa and to improving systematic classification (Badr & El-Shazly, 2022). Advances in karyology, systematics, and molecular genetics have demonstrated that chromosome number, chromosome morphology and DNA nucleotide sequence data constitute fundamental criteria for the identification and classification of taxa, as well as for the assessment of their phylogenetic relationships (Wolny et al. 2025; Toshtemirov et al. 2026). Furthermore, investigations of chromosome structure provide an important scientific basis for elucidating the evolutionary origin of diploid and naturally polyploid species, the processes underlying speciation and the mechanisms of plant adaptation to environmental factors (Grif, 2000; Ilnicki, 2014). Numerous studies have demonstrated that natural and artificially induced polyploid plants exhibit greater tolerance to both

abiotic and biotic stress factors than their diploid counterparts. In some cases, polyploids also display enhanced viability, adaptive capacity and reproductive success (Tossi et al., 2022).

Polyploidization is the process by which an organism undergoes a transition from the diploid state to higher ploidy levels, such as triploidy, tetraploidy, and other polyploid conditions, as a result of whole-genome chromosome set duplication (Comai, 2005). This phenomenon is recognized as one of the most significant cytogenetic mechanisms driving plant evolution, playing a central role in speciation, genome diversification, the expansion of genetic variation and the adaptation of plants to diverse environmental conditions. Polyploidization increases gene copy number, thereby facilitating the emergence of novel genetic combinations, altering patterns of gene expression and, in some cases, leading to the development of new morphological and physiological traits (Wang et al., 2005; Wan et al. 2014). Therefore, polyploidy is recognized as a widespread phenomenon in the evolutionary history of flowering plants and is considered a major evolutionary force underlying the origin, diversification, and adaptation of numerous cultivated and wild plant species (Van de Peer et al., 2021). Polyploidy represents one of the most important cytogenetic tools in the breeding of agricultural, food, and medicinal plants. Numerous economically important crops, including wheat (*Triticum aestivum*), cotton (*Gossypium hirsutum*), and cultivated strawberry (*Fragaria × ananassa*), are naturally polyploid species (Marcussen et al., 2014; Wendel & Cronn, 2003; Sattler et al., 2016; Edger et al., 2019). Furthermore, artificial polyploidization has been widely applied to increase the production of biologically active secondary metabolites, improve biomass yield, and develop novel genotypes with enhanced pharmaceutical value in medicinal plants (Dhawan & Lavania, 1996; Madani et al., 2021).

Numerous species of the genus *Rheum* are highly valued for both their medicinal and food uses (Darrudi et al., 2015; Rakhmataliev et al., 2025, 2026). The genus *Rheum* L., belonging to the family Polygonaceae, comprises approximately 60 species distributed worldwide (Wang et al., 2026). Extensive cytogenetic studies have been conducted on species of the genus *Rheum*, demonstrating the presence of both diploid and naturally polyploid species within the genus (Chin & Youngken, 1947; Wang et al., 2005; Liu et al., 2010; Wan et al., 2014). Chin & Youngken (1947) analyzed the chromosomes of 10 species from the Chinese flora and found that species of the genus had diploid (2n = 22) and tetraploid (2n = 44) chromosome numbers. The study also examined the relationship between pollen grain size and chromosome number. Liu et al. (2010) conducted a karyological study of 12 species distributed across the desert and mountainous regions of China, providing their chromosome characteristics and exact chromosome numbers. According to the authors, species of the genus possessed diploid (2n = 22) and tetraploid (2n = 44) chromosome numbers.

According to Girenko et al. (1988), species distributed in the desert and mountainous regions of Uzbekistan were reported to have diploid (2n = 22) and tetraploid (2n = 44) chromosome numbers. However, high-quality and clearly resolved chromosome images, as well as detailed statistical and karyological characteristics, are unavailable or limited in these previous studies. These diploid and naturally polyploid species are reproductively isolated and are distinguished by their ecological habitats and morphological characteristics (Vvedensky, 1971; Girenko et al., 1988). Among the species of this genus, *Rheum maximowiczii* Losinsk. and *Rheum turkestanicum* Janisch. are distributed predominantly in the mountainous and desert regions of Central Asia, respectively. These species are readily distinguished by their morphological characteristics (Korovin & Vvedensky, 1953). Previous cytogenetic studies have reported that *Rheum* species occurring in the flora of Central Asia exhibit both diploid (2n = 22) and tetraploid (2n = 44) chromosome numbers (Liu et al., 2010). However, most karyological investigations of these two species were conducted nearly four decades ago, and the available data remain limited and have not been comprehensively updated using modern cytogenetic approaches (Girenko et al., 1988). Of these species, *Rheum maximowiczii* is widely consumed by local communities in Central Asia during spring as a succulent, vitamin-rich vegetable, whereas *Rheum turkestanicum* is recognized as an important forage species for livestock (Rakhmataliev et al., 2025). However, the rhizomes of both species have been demonstrated to contain bioactive compounds with medicinal properties (Ghorbani et al., 2019; Yazdi et al., 2019; Rakhmataliev et al., 2025; Wang et al., 2026). In the present study, we aimed to determine the chromosome number, karyotype structure, and karyomorphometric characteristics of the Central Asian endemic species *Rheum maximowiczii* and *Rheum turkestanicum* and to establish the presence or absence of polyploidy in these species.

2. Materials and Methods

In this study, seeds of *Rheum maximowiczii* and *Rheum turkestanicum* were collected from natural populations in the Tashkent and Bukhara regions of Uzbekistan (Tab. 1; Fig. 1 a,b,c). For the analysis of the somatic chromosomes of *R. maximowiczii* and *R. turkestanicum*, newly developed root tips measuring 1–5 cm in length were excised from germinated seeds (Fig. 1). During the study, only root tips were used for cytogenetic analyses. Root samples were processed in 1.5 mL Eppendorf microcentrifuge tubes. Newly developed root tips

were pretreated with a colchicine solution prepared by dissolving a 1 mg colchicine tablet in 1000 mL of distilled water and incubated at room temperature (approximately 25°C) for 4 h. Following pretreatment, the root tips were rinsed three times in distilled water at room temperature for 5 min each. For fixation and maceration (tissue softening), phosphoric acid was used as a hygroscopic reagent and cell membrane hydrolyzing agent. The samples were treated with 38% H_3PO_4 solution (H_3PO_4 : H_2O = 1:2, v/v) at room temperature for 15–20 minutes (Toshtemirov et al., 2026). Subsequently, the samples were stained in 2% (w/v) orcein solution for 5 min, followed by immersion in 1% (w/v) aceto-orcein solution for an additional 5 minutes (Barykina et al., 2004). Chromosome numbers were determined by examining at least 10 well-spread metaphase plates for each species, prepared from two root tips of *Rheum maximowiczii* and seven root tips of *Rheum turkestanicum*. The stained root tips were then macerated in 45% acetic acid. For microscopic slide preparation, the stained root tip was placed on a glass microscope slide. The mitotic region was identified based on the intensity of staining, excised with a razor blade, and the remaining tissues were removed. The mitotic zone was cut into several 1–2 mm fragments, each of which was covered with a coverslip. Excess acetic acid was removed using filter paper before microscopic examination (Barykina et al., 2004; Toshtemirov et al. 2026). Metaphase images were captured at 1000× magnification using a KERN OBN 132 bright-field microscope (KERN & SOHN GmbH, Ziegelei 1, 72336 Balingen-Frommern, Germany). At least 10 metaphase plates were prepared for each species. Among these metaphase plates, the best-quality and well-spread images were selected, and three metaphase plates of *Rheum maximowiczii* (Fig. 2) and two metaphase plates of *Rheum turkestanicum* (Fig. 3) were used for morphometric analysis. Chromosome morphometric measurements and idiogram construction were performed using IdeoKar version 1.3 (Mirzaghaderi & Marzangi, 2015). KERN OPTICS S-EYE version 1.10.7 software was used for capturing metaphase images, whereas Adobe Photoshop CS6 (Adobe Systems Incorporated, 345 Park Avenue, San Jose, California, USA) was employed for chromosome pairing. During the analysis, the following standard karyotype parameters were calculated: arm ratio (AR), chromosome length (CL), relative chromosome length (RL%), centromeric index (CI), and karyotype type. For comparative karyomorphometric analysis, chromosome measurements were standardized based on the basic chromosome number (x = 11), which is common to both species. In *Rheum maximowiczii* (2n = 2x = 22), x = n = 11, whereas in *R. turkestanicum* (2n = 4x = 44), x = 11 and n = 22. Therefore, the total length of the monoploid chromosome set (TML), consisting of 11 chromosomes, was calculated for both species. TML was determined by summing the lengths of the chromosomes comprising the basic chromosome set (x = 11) in each species. This approach allows comparative assessment of chromosome dimensions between taxa with different ploidy levels but the same basic chromosome number (Peruzzi et al., 2016; Winterfeld et al., 2020 ; Mitrenina et al., 2021). In addition, the following parameters characterizing karyotype asymmetry were calculated: percentage contribution to the total chromosomal complement (F%), total length of the monoploid chromosome set (TML), total form percentage (TF%), karyotype asymmetry index (AsK%), intrachromosomal asymmetry index (A1), interchromosomal asymmetry index (A2), proportion of symmetric chromosomes (S%), mean relative length of the short arm (Xci), degree of karyotype asymmetry (A), mean relative length of the long arm (XcA), ccoefficient of variation of chromosome length (CVcl), ccoefficient of variation of the centromeric index (CVci), and the asymmetry index (AI) proposed by Romero-Zarco (1986). Detailed definitions, formulae, and measurement units for all karyotype parameters are provided in the first and second tables. Chromosomes were classified according to centromere position following the nomenclature of Levan et al. (1964) as follows: m = metacentric (AR = 1.00–1.69), sm = submetacentric (AR = 1.70–2.99), st = subtelocentric (AR = 3.00–6.99), and t = telocentric (AR ≥ 7.00). The classification was based primarily on the arm ratio (AR = L/S) and the centromeric index [CI = S/(L + S) × 100]. This nomenclature was adopted as the principal criterion for chromosome morphology. Karyotypes were classified according to their degree of asymmetry using the system of Stebbins (1971). This system evaluates karyotypes using two main parameters: the ratio of the longest to the shortest chromosome (CLmax/CLmin) and the proportion of chromosomes with an arm ratio (AR) less than 2, indicating symmetrical chromosomes. Based on four categories of length ratio (1.00; 0.99–0.51; 0.50–0.01; 0.00) and three classes of centromere position symmetry (A: mostly symmetrical; B: moderately asymmetrical; and C: highly asymmetrical), karyotypes are classified into 12 categories, ranging from 1A to 4C. This classification was used to assess the evolutionary level of karyotype structure in the studied species. All morphometric measurements were obtained using IdeoKar version 1.3 (Mirzaghaderi & Marzangi, 2015; Toshtemirov et al., 2026), whereas mean values and standard deviations were calculated in Microsoft Excel 2016 (Microsoft Corporation, One Microsoft Way, Redmond, WA). Artificial intelligence (AI) was used as a translation tool during the preparation of this manuscript and the review of the relevant literature.

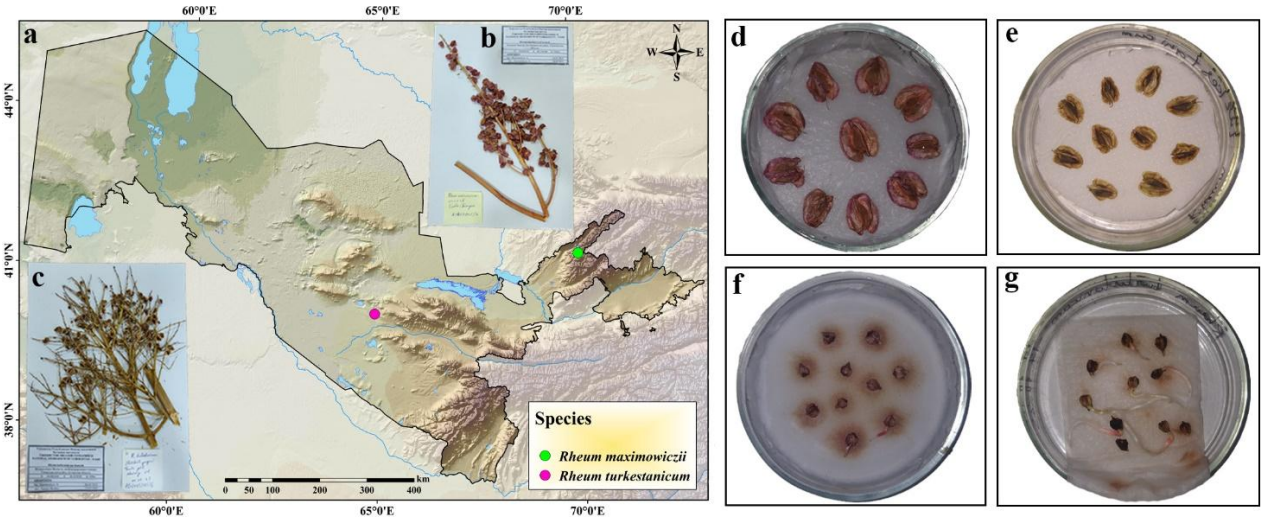

Figure 1. Study areas (a), *Rheum maximowiczii* (b, d, f) and *Rheum turkestanicum* (c, e, g).

Table 1. Collection localities and voucher numbers of the herbarium specimens.

Species	Number of specimens and populations of species	Geographic coordinates (DMS)	Altitude (m a.s.l.) according to Google Earth Pro	Vaeucher numbers
Rheum maximowiczii	1	41˚30'57.5"N, 70˚01'53.7"E	1850	A04072025/2
Rheum turkestanicum	1	40˚24'53.4"N, 64˚53'12.7"E	300	AB04052025/6

3. Results

3.1. Seed germination of the studied species.

Ten well-developed, mature, and disease-free seeds of *Rheum maximowiczii* were sown on 10 September 2025. The first seed germinated six days after sowing, followed by a second seed on the following day. The roots produced by these germinated seedlings were used for the cytogenetic analyses. The remaining eight seeds failed to germinate (Fig. 1d, f). Similarly, ten well-developed, mature, and disease-free seeds of *Rheum turkestanicum* were sown on the same date as those of *R. maximowiczii*. However, *R. turkestanicum* exhibited a considerably higher germination rate, with seven out of ten seeds successfully germinating. Root emergence began on the fifth day after sowing, and the roots grew approximately two to three times faster than those of *R. maximowiczii*. The roots obtained from the germinated *R. turkestanicum* seedlings were used for the present study (Fig. 1e, g).

3.2. Karyological characteristics of the studied species.

Cytogenetic analyses revealed the somatic chromosome number, karyotype structure, and karyomorphometric characteristics of *Rheum maximowiczii* and *Rheum turkestanicum* (Figs. 2–4). Chromosome counts from metaphase plates of *R. maximowiczii* consistently revealed 2n = 22 in three independent cells (Fig. 2a, d, g). Karyogram and idiogram analyses demonstrated that the chromosome complement consists of 11 homologous chromosome pairs, all of which are metacentric (m) (Fig. 2b, e, h; Fig. 4a–c). Accordingly, the karyotype formula was established as 2n = 2x = 22 = 22m.

Chromosome counts from metaphase plates of *R. turkestanicum* consistently revealed 2n = 44 somatic chromosomes in two independent cells (Fig. 3a, d). Karyogram and idiogram analyses revealed 22 homologous chromosome pairs, all of which were metacentric, resulting in the karyotype formula 2n = 4x = 44 = 44m. The clear separation of chromosomes in metaphase plates enabled accurate chromosome pairing and detailed karyomorphometric analyses (Fig. 3b, e; Fig. 4d, e). The karyomorphometric analysis revealed several differences between *R. maximowiczii* and *R. turkestanicum* (Table 2). The total length of the monoploid chromosome set (TML) was 33.53 ± 1.82 μm for *R. maximowiczii* and 28.82 ± 0.31 μm for *R. turkestanicum*. The total form percentage (TF%) was 44.46 ± 0.16 and 44.50 ± 0.26, respectively, whereas the karyotype asymmetry index (AsK%) was 55.54 ± 0.16 and 55.50 ± 0.26, indicating that these parameters were nearly identical between the two species. The mean centromeric index (Xci) was 0.45 ± 0.00 in *R. maximowiczii* and 0.44 ± 0.00 in *R. turkestanicum*, while the mean centromeric asymmetry (Xca) was 11.02 ± 0.28 and 11.08 ± 0.40, respectively. Among the parameters describing karyotype symmetry (Table 2), the proportion of symmetric chromosomes (S%) was higher in *R. maximowiczii* (70.41 ± 2.36) than in *R. turkestanicum* (63.76 ± 4.98). The ccoefficient of variation of chromosome length (CVcl) was greater in *R. turkestanicum* (13.31 ± 2.53) than in *R.*

maximowiczii (10.81 ± 1.39), whereas the ccoefficient of variation of the centromeric index (CVci) showed the opposite trend, being higher in *R. maximowiczii* (5.51 ± 0.25) than in *R. turkestanicum* (2.51 ± 0.12). Consequently, the asymmetry index (AI) was substantially higher in *R. turkestanicum* (522.64 ± 76.69) than in *R. maximowiczii* (196.24 ± 25.62). According to the Stebbins (1971) classification, both species were assigned to the 1A category, representing highly symmetrical karyotypes. The intrachromosomal asymmetry index (A1) was approximately 0.20 in both species, whereas the interchromosomal asymmetry index (A2) was 0.11 ± 0.01 in *R. maximowiczii* and 0.13 ± 0.03 in *R. turkestanicum* (Table 2). Morphometric analysis of individual chromosome pairs showed that, in *R. maximowiczii*, the first chromosome pair was the longest, with a total length of 3.73 ± 0.28 μm, whereas the eleventh chromosome pair was the shortest (2.80 ± 0.18 μm). Similarly, in *R. turkestanicum*, the first chromosome pair was the longest (3.30 ± 0.10 μm), and the eleventh pair was the shortest (2.09 ± 0.10 μm). The relative chromosome length (RL%) ranged from 11.06 to 8.37% in *R. maximowiczii* and from 11.44 to 7.27% in *R. turkestanicum*. In both species, the arm ratio (AR) remained within the metacentric range, varying from 1.18 to 1.37 in *R. maximowiczii* and from 1.19 to 1.33 in *R. turkestanicum*. The centromeric index (CI) ranged from 0.43 to 0.46, whereas the r-value varied from 0.74 to 0.85 in *R. maximowiczii* and from 0.75 to 0.84 in *R. turkestanicum* (Table 3). These morphometric data, together with the karyogram and idiogram analyses, confirm that all chromosomes in both species are metacentric and that their karyotypes are highly symmetrical (Fig. 4).

Table 2. Karyomorphometric parameters and karyotype asymmetry indices of *Rheum maximowiczii* and *Rheum turkestanicum*.

Parameters	*Rheum maximowiczii*	*Rheum turkestanicum*
TML	33.53 ± 1.82	28.82 ± 0.31
TF	44.46 ± 0.16	44.50 ± 0.26
AsK%	55.54 ± 0.16	55.50 ± 0.26
S%	70.41 ± 2.36	63.76 ± 4.98
Xci	0.45 ± 0.00	0.44 ± 0.00
A	0.11 ± 0.00	0.11 ± 0.00
XcA	11.02 ± 0.28	11.08 ± 0.40
CVcl	10.81 ± 1.39	13.31 ± 2.53
CVci	5.51 ± 0.25	2.51 ± 0.12
AI	196.24 ± 25.62	522.64 ± 76.69
Stebbins C	1A	1A
A1	0.20 ± 0.00	0.20 ± 0.01
A2	0.11 ± 0.01	0.13 ± 0.03

Table 3. Karyomorphometric measurements of chromosome pairs in *Rheum maximowiczii* and *Rheum turkestanicum*.

	Rheum maximowiczii								
№	*L*	*S*	*CL*	*AR*	*r-Value*	*RL%*	*F%*	*CI*	*Chr. Type*
1	2.01 ± 0.14	1.72 ± 0.14	3.73 ± 0.28	1.18 ± 0.02	0.85 ± 0.02	11.06 ± 0.26	5.09 ± 0.16	0.46 ± 0.01	m
2	1.89 ± 0.12	1.44 ± 0.10	3.32 ± 0.22	1.34 ± 0.05	0.76 ± 0.03	9.91 ± 0.43	4.26 ± 0.11	0.43 ± 0.03	m
3	1.67 ± 0.03	1.37 ± 0.07	3.04 ± 0.08	1.24 ± 0.06	0.82 ± 0.04	9.14 ± 0.27	4.10 ± 0.15	0.45 ± 0.03	m
4	1.69 ± 0.08	1.31 ± 0.08	3.00 ± 0.16	1.29 ± 0.02	0.78 ± 0.01	8.94 ± 0.05	3.90 ± 0.04	0.44 ± 0.01	m
5	1.67 ± 0.10	1.29 ± 0.05	2.95 ± 0.14	1.30 ± 0.03	0.77 ± 0.02	8.83 ± 0.05	3.85 ± 0.07	0.44 ± 0.02	m
6	1.64 ± 0.09	1.33 ± 0.05	2.96 ± 0.14	1.24 ± 0.02	0.81 ± 0.02	8.85 ± 0.06	3.98 ± 0.07	0.45 ± 0.02	m
7	1.55 ± 0.08	1.33 ± 0.10	2.88 ± 0.18	1.20 ± 0.06	0.85 ± 0.04	8.58 ± 0.21	3.93 ± 0.08	0.46 ± 0.03	m
8	1.67 ± 0.10	1.42 ± 0.12	3.09 ± 0.21	1.21 ± 0.04	0.83 ± 0.03	9.17 ± 0.13	4.19 ± 0.13	0.46 ± 0.03	m

Rheum maximowiczii									
№	***L***	***S***	***CL***	***AR***	***r-Value***	***RL%***	***F%***	***CI***	***Chr. Type***
9	1.59 ± 0.13	1.20 ± 0.08	2.79 ± 0.20	1.30 ± 0.06	0.78 ± 0.03	8.31 ± 0.39	3.58 ± 0.08	0.43 ± 0.03	m
10	1.72 ± 0.15	1.25 ± 0.05	2.97 ± 0.20	1.37 ± 0.08	0.74 ± 0.04	8.84 ± 0.25	3.76 ± 0.15	0.43 ± 0.04	m
11	1.53 ± 0.10	1.28 ± 0.08	2.80 ± 0.18	1.22 ± 0.02	0.82 ± 0.01	8.37 ± 0.35	3.81 ± 0.14	0.46 ± 0.01	m
Rheum turkestanicum									
№	*L*	*S*	*CL*	*AR*	*r-Value*	*RL%*	*F%*	*CI*	*Chr. Type*
1	1.80 ± 0.00	1.50 ± 0.10	3.30 ± 0.10	1.21 ± 0.06	0.83 ± 0.04	11.44 ± 0.22	5.20 ± 0.28	0.45 ± 0.02	m
2	1.65 ± 0.03	1.29 ± 0.06	2.93 ± 0.09	1.29 ± 0.04	0.78 ± 0.03	10.17 ± 0.21	4.46 ± 0.17	0.44 ± 0.01	m
3	1.56 ± 0.02	1.23 ± 0.05	2.79 ± 0.07	1.25 ± 0.04	0.80 ± 0.03	9.67 ± 0.14	4.27 ± 0.12	0.44 ± 0.01	m
4	1.60 ± 0.08	1.28 ± 0.06	2.88 ± 0.14	1.26 ± 0.00	0.79 ± 0.00	9.99 ± 0.37	4.44 ± 0.15	0.44 ± 0.00	m
5	1.48 ± 0.05	1.20 ± 0.03	2.68 ± 0.08	1.23 ± 0.04	0.81 ± 0.03	9.28 ± 0.18	4.16 ± 0.05	0.45 ± 0.00	m
6	1.43 ± 0.03	1.19 ± 0.05	2.61 ± 0.08	1.19 ± 0.02	0.84 ± 0.02	9.06 ± 0.18	4.12 ± 0.14	0.45 ± 0.01	m
7	1.42 ± 0.01	1.09 ± 0.00	2.51 ± 0.01	1.33 ± 0.00	0.75 ± 0.00	8.70 ± 0.14	3.77 ± 0.04	0.43 ± 0.00	m
8	1.35 ± 0.01	1.09 ± 0.03	2.44 ± 0.02	1.23 ± 0.04	0.81 ± 0.02	8.48 ± 0.17	3.80 ± 0.15	0.45 ± 0.01	m
9	1.29 ± 0.01	1.04 ± 0.02	2.33 ± 0.03	1.22 ± 0.02	0.82 ± 0.01	8.09 ± 0.19	3.62 ± 0.11	0.45 ± 0.00	m
10	1.26 ± 0.04	0.99 ± 0.04	2.26 ± 0.08	1.32 ± 0.01	0.76 ± 0.01	7.84 ± 0.37	3.45 ± 0.17	0.44 ± 0.00	m
11	1.17 ± 0.06	0.92 ± 0.04	2.09 ± 0.10	1.25 ± 0.01	0.80 ± 0.01	7.27 ± 0.43	3.21 ± 0.18	0.44 ± 0.00	m

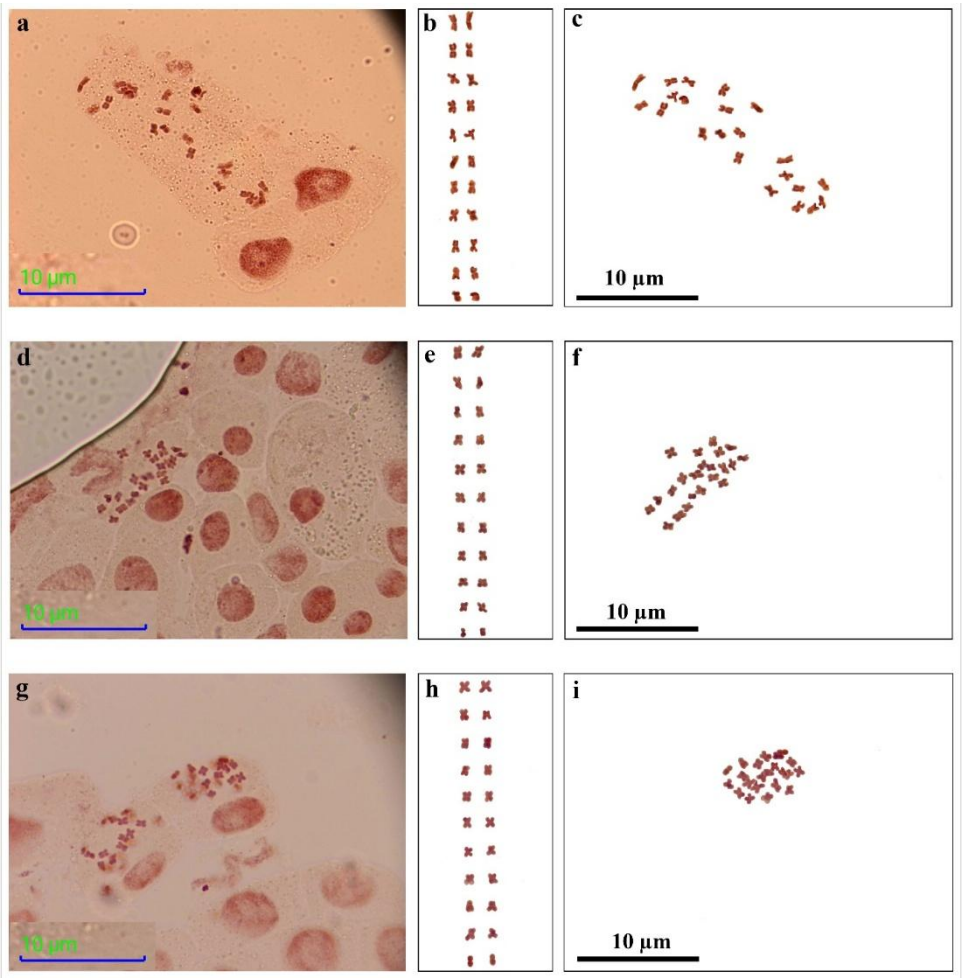

Figure 2. Somatic chromosomes and idiograms of *Rheum maximowiczii* (2n = 2x = 22): a–i, metaphase chromosomes from three root apical meristem cells obtained from germinated seeds of *R. maximowiczii*. < Scale bar = 10 µm>.

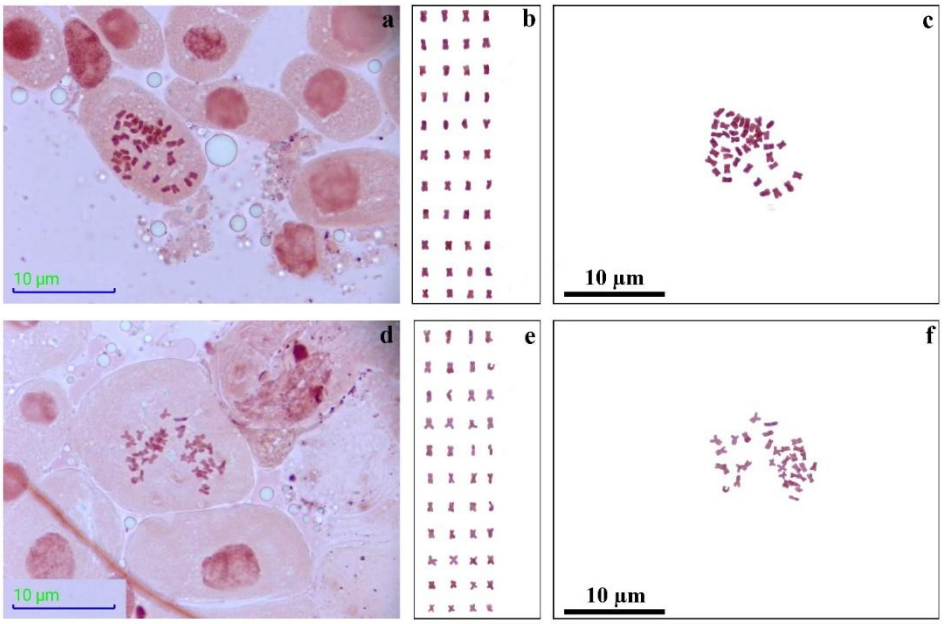

Figure 3. Somatic chromosomes and idiograms of *Rheum turkestanicum* (2n = 4x = 44): a–f, metaphase chromosomes from two root apical meristem cells obtained from germinated seeds of *R. turkestanicum*. < Scale bar = 10 µm>.

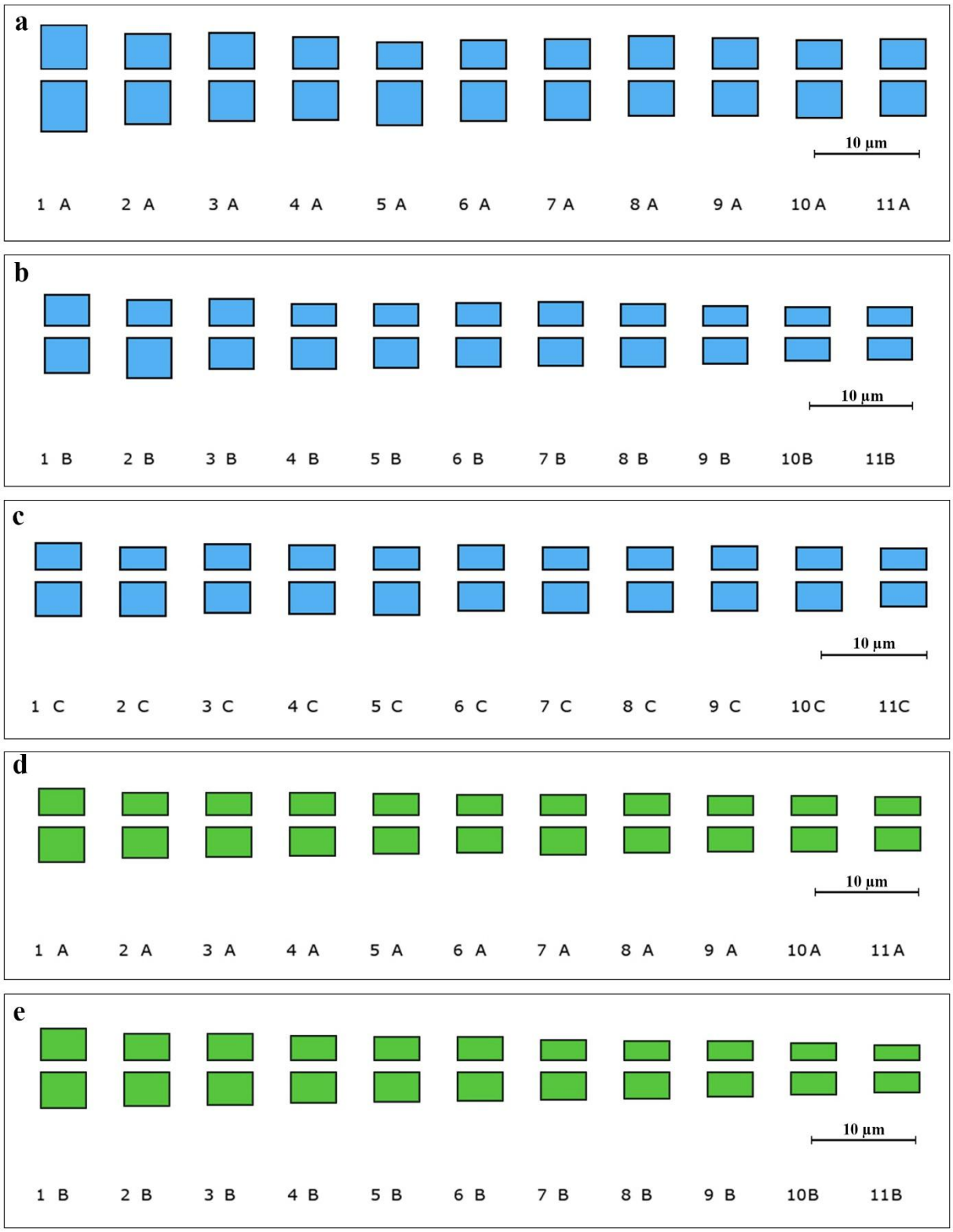

Figure 4. Idiograms of two species of the genus *Rheum* L.: a–c, *Rheum maximowiczii*; d–e, *Rheum turkestanicum*. The idiograms were constructed from chromosome images obtained from metaphase plates using IdeoKar software and Adobe Photoshop.

3.3. Morphological characteristics of the vegetative and reproductive organs.

The present study also examined differences in the morphological characteristics of *Rheum maximowiczii* and *Rheum turkestanicum* (Fig. 5). Observations showed that both species generally possess six tepals, with three outer tepals longer than the three inner ones, and a single pistil consisting of three short styles terminating in three stigmas. However, clear differences were observed in floral morphology between the two species. Examination of more than ten flowers of each species revealed that the number of stamens in *R. maximowiczii* varied from nine to fourteen, and the stamens were red, pink, or yellow in color. In all flowers, the stigmas were red to dark red.

The species was characterized by a paniculate, pyramidal inflorescence. (Fig. 5j,k,l). The leaves were reniform, with slightly undulate margins and irregular elevations (1–3 cm high), the upper leaf surface was densely covered with verrucae (wart-like protuberances). The upper surface of the petiole was reddish or green and densely, or occasionally sparsely, covered with verrucae.

R. turkestanicum consistently possessed nine stamens, which were yellow or pale yellow in color. The stigmas were white, creamy white, or pale pink. In addition, a rare morphological anomaly characterized by the fusion of floral organs (synanthy) was observed (Fig. 5q–r). The species exhibited both paniculate (Fig. 5w) and globose (Fig. 5v) inflorescences. The leaves were reniform, with irregular elevations of up to 10 cm in height located near the leaf base. The leaf margins were generally entire, and numerous verrucae were concentrated along the main veins on the abaxial (lower) leaf surface. The upper and lower surfaces of the petiole were green and usually densely covered with verrucae, although they were occasionally sparse or absent in some individuals.

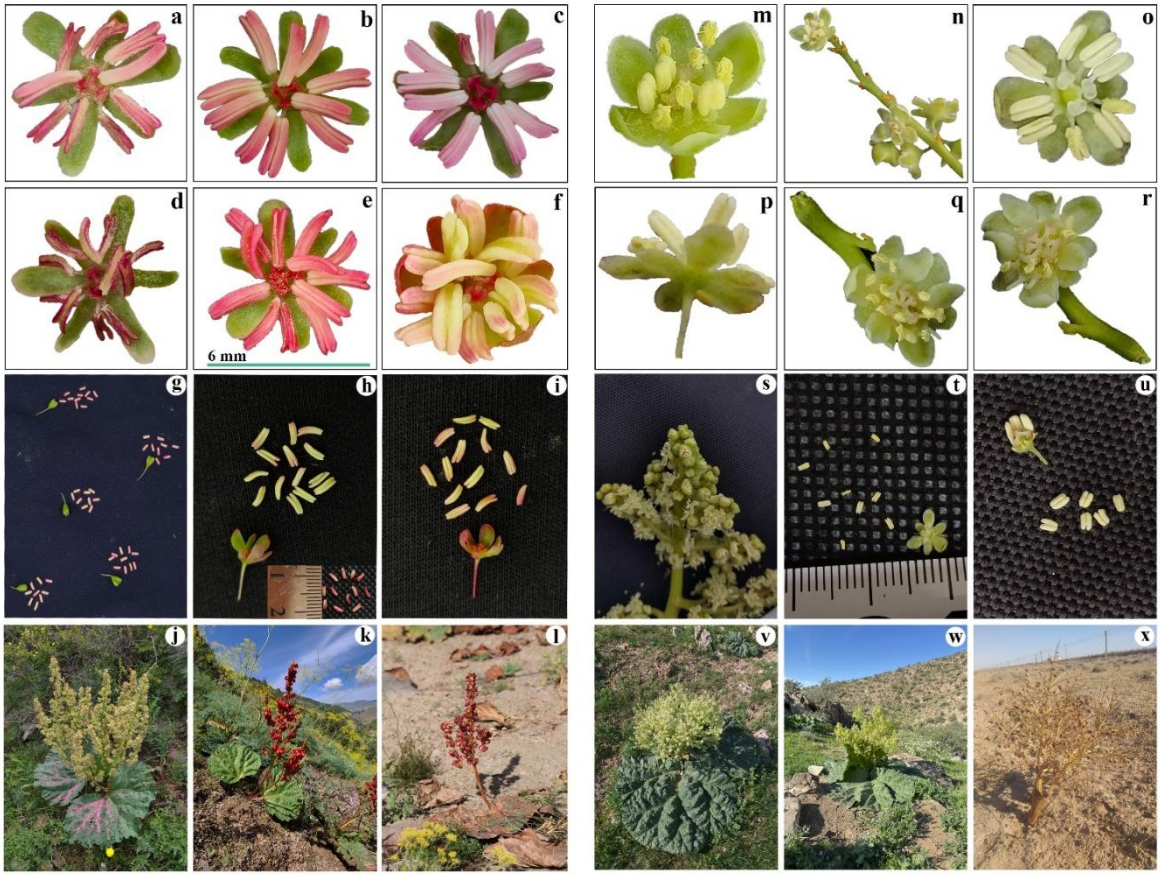

Figure 5. Morphological characteristics of two species of the genus *Rheum* L.: *Rheum maximowiczii* (a–l) and *Rheum turkestanicum* (m–x).

4. Discussion

The results of the present study provide new insights into the cytogenetic characteristics of *Rheum maximowiczii* and *Rheum turkestanicum* and allow a comparative evaluation of their karyotype structures. Previous cytological studies have shown that the basic chromosome number in the genus *Rheum* is x = 11, with diploid (2n = 22), tetraploid (2n = 44), and higher ploidy levels reported among different species (Chin & Youngken, 1947; Girenko et al., 1988; Liu et al., 2010). In the present study, *R. maximowiczii* was confirmed to be diploid (2n = 22), whereas *R. turkestanicum* was found to be tetraploid (2n = 44), corroborating the previously reported cytogenetic patterns within the genus. Furthermore, according to the chromosome size classification proposed by Lima de Faria (1980), based on the mean chromosome length (ML = TML/n), the chromosomes of *R. maximowiczii* (ML = 3.05 μm) and *R. turkestanicum* (ML = 2.62 μm) belong to the small chromosome category. These results are largely consistent with previous cytogenetic studies of *Rheum* species from China. Liu et al. (2010) reported that most species of *Rheum* possess predominantly metacentric and submetacentric chromosomes and belong to the Stebbins (1971) 2A karyotype category. They also noted that chromosomes of some diploid species are larger than those of certain tetraploid species. In contrast, our study demonstrated that all chromosomes of both *R. maximowiczii* and *R. turkestanicum* are metacentric and that both species belong to the Stebbins 1A category, representing highly symmetrical karyotypes. However, similarly, the diploid species exhibited a

greater mean chromosome length than the tetraploid species (Table 3). Polyploidy is known to induce substantial changes in the cytological, biochemical, physiological, and developmental characteristics of plants. Consequently, polyploid taxa may acquire novel adaptive traits that are absent in their diploid progenitors, enabling them to colonize new ecological niches (Levin, 2002). The diploid chromosome complement of *R. maximowiczii* and the tetraploid complement of *R. turkestanicum* suggest that polyploidization has played an important role in their evolutionary history and genome evolution. In flowering plants, polyploidization is widely recognized as one of the major evolutionary mechanisms driving speciation, ecological adaptation, and the generation of genetic diversity. Therefore, the tetraploid nature of *R. turkestanicum* may represent an adaptive advantage for survival under arid and strongly continental climatic conditions (Fedchenko, 1937; Korovin & Vvedensky, 1953; Pavlov, 1960; Stebbins, 1971; Otto & Whitton, 2000). The chromosome samples of *R. maximowiczii* were obtained from individuals growing in mountainous habitats, whereas those of *R. turkestanicum* were collected from populations inhabiting the transitional zone between the Kyzylkum Desert plains and adjacent foothills (Fig. 5 l,x). In addition, *R. maximowiczii* is distributed mainly in mountainous regions, whereas *R. turkestanicum* occurs predominantly in the Kyzylkum Desert plains and the lower foothills of desert mountains in Uzbekistan, as well as in desert habitats of Kazakhstan and Turkmenistan (Fig. 5 j,k,l,v,w,x; Fedchenko, 1937; Vvedensky, 1971; Aitzhan et al., 2024; He et al., 2024). Seed germination in *Rheum* species has been investigated extensively in previous studies (Yoo et al., 2012; Darrudi et al., 2015; Khan et al., 2022; Aldaif et al., 2025). However, during the preparation of chromosome material in the present study, differences in seed germination between the two species yielded biologically interesting observations. Seven out of ten seeds of *R. turkestanicum* germinated, and their roots developed considerably faster than those of *R. maximowiczii*, indicating a higher level of physiological activity under laboratory conditions. By comparison, only two out of ten seeds of *R. maximowiczii* germinated. This lower germination rate may be associated with physiological seed dormancy, the mechanical properties of the seed coat, the degree of seed maturity at the time of collection, or potentially with differences related to ploidy level.

Previous studies have described the general reproductive morphology of the genus *Rheum*. According to the authors of this study, the flowers of the genus *Rheum* typically possess six tepals, with the three outer tepals generally larger than the three inner ones. The androecium usually consists of nine stamens, although variation from five to ten stamens has occasionally been reported. The gynoecium consists of a single pistil with three short styles and three enlarged, recurved stigmas, while the fruit is a three-angled winged achene (Komarov, 1970; Bao & Grabovskaya-Borodina, 2003). Polyploidization may also influence floral morphology by affecting the number, relative size and spatial arrangement of floral organs (Soltis et al., 2010; Chen, 2010). More broadly, polyploidy is recognized as an important evolutionary force that influences not only plant morphology but also reproductive processes, including pollination and fertilization (Otto & Whitton, 2000). Our morphological observations demonstrated that both species retain the general floral characteristics typical of the genus *Rheum*, while exhibiting several species-specific differences. The distinguishing characteristics included variation in stamen number (9–14) and marked color polymorphism of the stamens and stigmas in *R. maximowiczii*, as well as shorter but thicker stamens and the occurrence of synanthy (fusion of floral organs), a very rare morphological anomaly, in the flowers of *R. turkestanicum* (Fig. 5). Such developmental abnormalities may arise from disturbances during plant ontogeny or may be induced by environmental factors affecting floral development (Meyerowitz et al., 1989).

5. Conclusion

This study represents one of the first comprehensive cytotaxonomic investigations integrating chromosome number, karyotype structure, karyomorphometric characteristics, and selected diagnostic morphological traits of *Rheum maximowiczii* and *Rheum turkestanicum*. Although both species possess highly symmetrical karyotypes composed entirely of metacentric chromosomes, they differ in several karyomorphometric parameters, geographical distribution, and morphological characteristics of both vegetative and reproductive organs. In particular, *R. turkestanicum* is characterized by spicate, pyramidal, and globose inflorescences and is distributed predominantly under arid and strongly continental environmental conditions, whereas *R. maximowiczii* is confined mainly to mountainous habitats. These differences reflect ecological as well as cytogenetic divergence between the two species. In addition, the number of stamens varied from 9 to 14 in *R. maximowiczii*, whereas it remained consistently nine in *R. turkestanicum*, indicating distinct patterns of morphological variation in their reproductive organs. These observations are partially consistent with previous reports describing phenotypic variation associated with polyploidy. However, establishing a direct relationship between these morphological differences and polyploidy will require more extensive molecular cytogenetic investigations involving multiple populations of both species, together with comparative analyses of additional *Rheum* species

from Central Asia. The findings of the present study provide valuable information for understanding the cytotaxonomy, evolution, and systematics of the genus *Rheum* and establish a cytogenetic framework for future molecular, evolutionary, and phylogenetic studies of these species.

References

Badr A, El-Shazly HH (2022). Chromosomes as Sources of Taxonomic Information for Plant Systematics and Evolution. *Taeckholmia*. 41(1):70–90. https://doi.org/10.21608/taec.2022.138485.1038

Wolny E, Mur LAJ, Ohmido N, Yin Z, Wang K, Hasterok R. 2025. Thriving or Withering? Plant Molecular Cytogenetics in the First Quarter of the 21st Century. *International Journal of Molecular Sciences* 26(14):7013. https://doi.org/10.3390/ijms26147013

Toshtemirov JG, Tojiboeva U, Asatulloev T, Aromov T, Boboeva N, Sharipov A, Raimov S, Toshmirov S, Yusupov Z, Tao D (2026) Revisiting chromosome diversity and karyotype evolution in five *Tulipa* species (Liliaceae) from Uzbekistan. *Journal of Asia-Pacific Biodiversity*. https://doi.org/10.1016/j.japb.2025.12.010

Grif VG (2000) Some aspects of plant karyology and karyosystematics. *International Review of Cytology* 196: 131–175. https://doi.org/10.1016/S0074-7696(00)96004-2

Ilnicki T (2014) Plant biosystematics with the help of cytology and cytogenetics. *Caryologia* 67(3): 199–208. https://doi.org/10.1080/00087114.2014.931642

Tossi VE, Martínez Tosar LJ, Laino LE, Iannicelli J, Regalado JJ, Escandón AS, Baroli I, Pitta-Álvarez SI (2022) Impact of polyploidy on plant tolerance to abiotic and biotic stresses. *Frontiers in Plant Science* 13: 869423. https://doi.org/10.3389/fpls.2022.869423

Comai L (2005) The advantages and disadvantages of being polyploid. *Nature Reviews Genetics* 6(11): 836–846. https://doi.org/10.1038/nrg1711

Wang A, Yang M, Liu J (2005) Molecular phylogeny, recent radiation and evolution of gross morphology of the rhubarb genus *Rheum* (Polygonaceae) inferred from chloroplast DNA trnL-F sequences. *Annals of Botany* 96(3): 489–498. https://doi.org/10.1093/aob/mci201

Van de Peer Y, Ashman TL, Soltis PS, Soltis DE (2021) Polyploidy: an evolutionary and ecological force in stressful times. *The Plant Cell* 33(1): 11–26. https://doi.org/10.1093/plcell/koaa015

Wendel JF, Cronn RC (2003) Polyploidy and the evolutionary history of cotton. *Advances in Agronomy* 78: 139–186. https://doi.org/10.1016/S0065-2113(02)78004-8

Marcussen T, Sandve SR, Heier L, Spannagl M, Pfeifer M, The International Wheat Genome Sequencing Consortium, Jakobsen KS, Wulff BBH, Steuernagel B, Mayer KFX, Olsen OA (2014) Ancient hybridizations among the ancestral genomes of bread wheat. *Science* 345(6194): 1250092.

Sattler MC, Carvalho CR, Clarindo WR (2016) The polyploidy and its key role in plant breeding. *Planta* 243(2): 281–296. https://doi.org/10.1007/s00425-015-2450-x

Edger PP, Poorten TJ, VanBuren R, Hardigan MA, Colle M, McKain MR, Smith RD, Teresi SJ, Nelson ADL, Wai CM, et al. (2019) Origin and evolution of the octoploid strawberry genome. *Nature Genetics* 51(3): 541–547. https://doi.org/10.1038/s41588-019-0356-4

Dhawan OP, Lavania UC (1996) Enhancing the productivity of secondary metabolites via induced polyploidy: a review. *Euphytica* 87(2): 81–89. https://doi.org/10.1007/BF00021879

Madani H, Escrich A, Hosseini B, Sanchez-Muñoz R, Khojasteh A, Palazon J (2021) Effect of polyploidy induction on natural metabolite production in medicinal plants. *Biomolecules* 11(6): 899. https://doi.org/10.3390/biom11060899

Darrudi R, Hassandokht MR, Nazeri V (2015) Effects of moist stratification, GA3 and seed age on seed germination of *Rheum khorasanicum* B. Baradaran & A. Jafari. *Journal of Applied Research on Medicinal and Aromatic Plants* 2(4): 168–173. https://doi.org/10.1016/j.jarmap.2015.07.001

Rakhmataliev A, Togaev A, Ergashov I, Yusupov Z (2025) Ethnomedicinal and nutritional applications of *Rheum maximowiczii* Losinsk. in traditional medicine in Uzbekistan. *Ethnobotany Research and Applications* 31: 1–8. https://doi.org/10.32859/era.31.44.1-8

Rakhmataliyev A, Samiyev L, Xonnazarov R, Yusupov Z (2026) The importance of medicinal rhubarb in traditional medicine. *Proceedings of the Uzbekistan–China International Symposium on Current Issues in the Cultivation and Utilization of Medicinal Plants*, Tashkent, Uzbekistan, pp. 182–192. https://doi.org/10.5281/zenodo.21356314

Wang J, Yusupov Z, Rakhmataliev A, Li L, Li T, Zeng Q, He M, Tojibaev K, Sun M (2026) Toward sustainable utilization: A multi-dimensional evaluation reveals the distinct chemical and toxicological profile of *Rheum maximowiczii*, a traditional Central Asian medicinal resource. *Industrial Crops and Products* 248: 123584. https://doi.org/10.1016/j.indcrop.2026.123584

Chin TC, Youngken HW (1947) The cytotaxonomy of *Rheum*. *American Journal of Botany* 34(10): 401–407. https://doi.org/10.2307/2437150.

Liu R, Wang A, Tian X, Wang D, Liu J (2010) Uniformity of karyotypes in *Rheum* (Polygonaceae), a species-rich genus in the Qinghai-Tibetan Plateau and adjacent regions. *Caryologia* 63(1): 82–90. https://doi.org/10.1080/00087114.2010.10589711.

Wan D, Sun Y, Zhang X, Bai X, Wang J, Wang A, Milne R (2014) Multiple ITS copies reveal extensive hybridization within *Rheum* (Polygonaceae), a genus that has undergone rapid radiation. *PLoS ONE* 9(2): e89769. https://doi.org/10.1371/journal.pone.0089769

Korovin EP, Vvedensky AI (1953) *Flora of Uzbekistan*. Vol. 2. Academy of Sciences of the Uzbek SSR, Tashkent, Uzbekistan.

Vvedensky AI (1971) *Conspectus Florae Asiae Mediae (Identification Key to the Plants of Central Asia)*. Vol. 2. Fan Publishing House of the Academy of Sciences of the Uzbek SSR, Tashkent, Uzbekistan.

Girenko MM, Ivanova KV, Komarova RA (1988) *Flora of Cultivated Plants*. Vol. 12: Leaf Vegetable Plants (Asparagus, Rhubarb, Sorrel, Spinach, Purslane, Garden Cress, Dill, Chicory, Lettuce). Agropromizdat, Leningrad, USSR, pp. 30–70.

Yazdi AJ, Javanshir S, Soukhtanloo M, Jalili-Nik M, Jafarian AH, Iranshahi M, Hasanpour M, Khatami SM, Hosseini A, Amiri MS, Ghorbani A (2019) Acute and sub-acute toxicity evaluation of the root extract of *Rheum turkestanicum* Janisch. *Drug and Chemical Toxicology* 42(6): 624–633. https://doi.org/10.1080/01480545.2018.1561713

Ghorbani A, Amiri MS, Hosseini A (2019) Pharmacological properties of *Rheum turkestanicum* Janisch. *Heliyon* 5(7): e01986. https://doi.org/10.1016/j.heliyon.2019.e01986

Barykina RP, Veselova TD, Devyatov AG, Dzhalilova KhKh, Ilyina GM, Chubatova NV (2004) *Handbook of botanical microtechnique: Fundamentals and methods* [*Spravochnik po botanicheskoy mikrotekhnike. Osnovy i metody*]. Moscow State University Publishing, Moscow.

Mirzaghaderi G, Marzangi K (2015) IdeoKar: an ideogram constructing and karyotype analyzing software. *Caryologia* 68(1): 31–35. https://doi.org/10.1080/00087114.2014.998526

Levan A, Fredga K, Sandberg AA (1964) Nomenclature for centromeric position on chromosomes. *Hereditas* 52(2): 201–220. https://doi.org/10.1111/j.1601-5223.1964.tb01953.x

Romero Zarco C (1986) A new method for estimating karyotype asymmetry. *Taxon* 35(3): 526–530. https://doi.org/10.2307/1221906

Stebbins GL (1971) Chromosomal evolution in higher plants. Edward Arnold, London.

Komarov VL (1970) *Flora of the U.S.S.R.* Vol. 5. Israel Program for Scientific Translations, Jerusalem, Israel, pp. 482–501. (Original work published 1936).

Bao, B. J., & Grabovskaya-Borodina, A. E. (2003). *Rheum*. In Flora of China, Vol. 5, pp. 341–350. Science Press & Missouri Botanical Garden Press.

Otto SP, Whitton J (2000) Polyploid incidence and evolution. *Annual Review of Genetics* 34: 401–437. https://doi.org/10.1146/annurev.genet.34.1.401

Stebbins GL (1980) Polyploidy in plants: Unsolved problems and prospects. In: Lewis WH (ed.) *Polyploidy: Biological Relevance*. Springer, Boston, MA, pp. 495–520. https://doi.org/10.1007/978-1-4613-3069-1_26

Fedchenko BA (1937) *Flora of Turkmenistan*. Vol. 2, Pt. 1. Turkmen State Publishing House, Ashgabat, Turkmenistan.

Pavlov NV (1960) *Flora of Kazakhstan*. Vol. 3. Academy of Sciences of the Kazakh SSR, Alma-Ata, Kazakhstan.

Aitzhan M, Aimenova Z, Sumbembayev A (2024) Studying the species distribution of rhubarbs (*Rheum* L.) in Kazakhstan. *BIO Web of Conferences* 100: 04034. https://doi.org/10.1051/bioconf/202410004034

He M, Asatulloev T, Yusupov Z, Li L, Meng X, Li Y, Tojibaev K, Sun M. 2024. Rhubarb: A potent medicinal herb linking traditional medicine research between Uzbekistan and China. Plant Diversity of Central Asia 3(2):1–18. https://doi.org/10.54981/PCDA/vol3_iss2/art1

Lima-de-Faria A (1980) Classification of genes, rearrangements and chromosomes according to the chromosome field. *Hereditas* 93(1): 1–46. https://doi.org/10.1111/j.1601-5223.1980.tb01043.x

Brochmann C (1993) Reproductive strategies of diploid and polyploid populations of arctic *Draba* (Brassicaceae). *Plant Systematics and Evolution* 185(1–2): 55–83. https://doi.org/10.1007/BF00937720

Meyerowitz EM, Smyth DR, Bowman JL (1989) Abnormal flowers and pattern formation in floral development. *Development* 106(2): 209–217. https://doi.org/10.1242/dev.106.2.209

Yoo JH, Hwang IS, Seong ES, Lee JG, Kim NJ, Kim MJ, Lim JD, Ham JK, Ahn YS, An TJ, Yu CY

(2012) Establishing optimal germination for stored *Rheum palmatum* L. seeds. Korean Journal of Medicinal Crop Science 20(2): 85–88. https://doi.org/10.7783/KJMCS.2012.20.2.085

Khan MI, Shahzad A, Ganie IB, Pandith SA (2022) Organogenesis, direct somatic embryogenesis, and shoot proliferation of *Rheum spiciforme* Royle: An endemic and vulnerable medicinal herb from Indian Trans Himalayas. *In Vitro Cellular & Developmental Biology – Plant* 58(1): 35–50. https://doi.org/10.1007/s11627-021-10211-2

Aldaif M, Ucar S, Yigider E, Aydin M, Kasapoglu AG, Ilhan E (2025) Micropropagation and genetic fidelity of *Rheum ribes* L.: A threatened and endemic medicinal herb from the Turkish Eastern Anatolia region. *Plant Cell, Tissue and Organ Culture* 162: 1. https://doi.org/10.1007/s11240-025-03111-5

Chen ZJ (2010) Molecular mechanisms of polyploidy and hybrid vigor. *Trends in Plant Science* 15(2): 57–71. https://doi.org/10.1016/j.tplants.2009.12.003

Soltis DE, Buggs RJA, Doyle JJ, Soltis PS (2010) What we still don't know about polyploidy. *Taxon* 59(5): 1387–1403. https://doi.org/10.1002/tax.595006

Levin DA (2002) Phenotypic consequences of chromosome doubling. In: *The Role of Chromosomal Change in Plant Evolution*. Oxford University Press, New York, NY, pp. 134–149. https://doi.org/10.1093/oso/9780195138597.003.007

Peruzzi, L., Carta, A. & Altinordu, F. 2017. Chromosome diversity and evolution in *Allium* (Allioideae, Amaryllidaceae). Plant Biosystems 151: 212–220. https://doi.org/10.1080/11263504.2016.1149123

Mitrenina, E.Y., Erst, A.S., Peruzzi, L., Skaptsov, M.V., Ikeda, H., Nikulin, V.Y. & Wang, W. 2021. Karyotype and genome size variation in white-flowered *Eranthis* sect. *Shibateranthis* (Ranunculaceae). PhytoKeys 187: 207. https://doi.org/10.3897/phytokeys.187.75715

Winterfeld, G., Ley, A., Hoffmann, M.H., Paule, J. & Röser, M. 2020. Dysploidy and polyploidy trigger strong variation of chromosome numbers in the prayer-plant family (Marantaceae). Plant Systematics and Evolution 306: 36. https://doi.org/10.1007/s00606-020-01663-x