

Karyotypic variability in some species of the genus *Chondrilla* (Asteraceae)

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Abstract. Karyotypic variability of plants was evaluated in 17 populations of six species of the genus *Chondrilla* (*C. ambigua* Fisch., *C. breviostris* Fisch, et Mey., *C. laticoronata* Leonova, *C. canescens* Kar., Kir., *C. juncea*, *C. pauciflora* Ledeb.) in the Astrakhan, Voronezh and Saratov regions, the Republic of Kalmykia, and Western Kazakhstan. It is maintained that *C. ambigua* is a strict diploid ($2n = 2x = 10$) species, while its close relative - *C. pauciflora* - is a strict triploid ($2n = 3x = 15$) taxon. The research demonstrates that the plants of the apomictic taxa *C. breviostris*, *C. canescens*, *C. juncea* and *C. laticoronata* are characterized by the genomic instability in the form of chromosome instability which results in these species' mixed-ploidy populations.

1 Introduction

The research on genetic polymorphism and morphological variability of a number of *Chondrilla* species in European Russia [1] requires a thorough examination of the species' karyotypic variability.

2 Materials and Methods

2.1 Object of study

In order to evaluate the karyotypic variability of plants, 17 populations of six *Chondrilla* species were subject to the research. Below are the names of the species, the numbers of the populations (in brackets), and the populations' locations:

C. ambigua (1030) – Astrakhan region, Krasnoyarsky district, near the village of Dosang; (1031) – Republic of Kalmykia, Yashkulsky district, near the village of Khulhuta.

C. breviostris (1029) – Astrakhan region, Akhtubinsky district, near the village Bolhuny; (1046) – Astrakhan region, Kharabalinsky district, near the village of Volnoe; (1055) – Astrakhan region, Kharabalinsky district, near the station Verblyuzhya; (1058) – Astrakhan region, Krasnoyarsky district, near the village of Dosang; (1059) – Republic of Kalmykia,

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Yashkulsky district, near the village of Khulhuta; (1077) – Kazakhstan, West Kazakhstan region, Bokeyorda district, near the village of Giekkum.

C. canescens (1040) – Saratov Region, Khvalynsky district, near the town of Khvalynsk, Belenkaya Mountain; (1054) – Astrakhan region, Akhtubinsky district, near the station Kapustin Yar; (1074) – Voronezh region, Bogucharovsky district, near the village Tereshkovo.

C. juncea (1027) – Saratov region, Krasnokutsky district, near the village of Dyakavka.

C. laticoronata (1056) – Astrakhan region, Akhtubinsky district, near the village of Verblyuzhye; (1070) – Astrakhan region, Kharabalinsky district, near the village of Sasykoli; (1071) – Astrakhan region, Kharabalinsky district, near the village of Tambovka; (1076) – Kazakhstan, West Kazakhstan region, Bokeyorda district, near the village of Giekkum.

C. pauciflora (1078) – Kazakhstan, West Kazakhstan region, Bokeyorda district, near the village of Urda.

The achenes that served as the material were collected from the natural populations in the years 2015-2017. The research started with examining the populations of those taxa that had been proved independent species by the earlier research on their morphological variability; and of those two taxa (*C. canescens* and *C. juncea*) the independence of which has not yet been established [1].

2.2 Methods

In order to evaluate the karyotypic variability of plants in the populations, the squash technique was employed to measure the chromosome number in the cells of seedlings' root apical meristems [2]. The chromosome number was calculated in root apical meristems of germinated seeds that had been formed in two flowering regimes (free flowering and dust-free flowering).

The seeds were germinated until the length of roots reached 10-12 mm. The root tips were pre-treated with an aqueous solution of bromonaphthaline for 3.5 hours at 40C. Roots were then fixed in a solution of 3 part 95% ethanol and 1 part 45% acetic acid. The fixed material was stained in aceto-haematoxylin; and the slides were prepared using the squash method in an 80% cloral hydrate solution.

The slides were analysed using an Axio Scope A1 microscope (Carl Zeiss) with immersion oil under 100×16 magnification. The microimages were taken with a high-definition digital camera AxioCam MRc 5 (D) and an adapter 60N-C 1" 1.0x. Chromosomes were counted in no less than 10 cells of each slide [3].

Whenever a plant was diagnosed with aneu- or mixoploidy, its basic ploidy level was considered the one that was characteristic of the majority of apical meristem cells.

3 Results

The basic chromosome number $x = 5$ was used to determine the ploidy level [4, 5]. It is revealed that only two of the studied taxa are constituted of plants of the same ploidy level. Specifically, all *C. ambigua* plants are diploid ($2n = 2x = 10$), while all plants of its close relative – *C. pauciflora* – are triploid ($2n = 3x = 15$). These results are in line with the data recorded in the literature [6].

C. juncea plants from the Krasnokutsky district in the Saratov region were either triploid or tetraploid. The majority of *C. canescens* plants were also triploid, with about 10% of diploid plants collected in the population in the Khvalynsky district of the Saratov region. Regardless of the location, *C. laticoronata* plants were triploid, with only a small portion of pentaploid plants in one of the populations in the adjacencies of the Tambovka village in the Astrakhan region.

Table 1. Ploidy levels in the root apical meristem cells of *Chondrilla* under different flowering regimes in 2015-2017.

Species name	№ pop.	Year of study	Flowering regime	Analyzed seedlings					
				total, units.	of them %				
					x	2x	3x	4x	5x
<i>C. ambigua</i>	1030	2015	f/f	3	0	100	0	0	0
		2016	f/f	3	0	100	0	0	0
		2017	f/f	36	0	100	0	0	0
	1031	2015	f/f	4	0	100	0	0	0
<i>C. brevirostris</i>	1029	2015	f/f	8	0	0	12.5	87.5	0
		2016	f/f	9	0	0	100	0	0
			d/f	3	0	0	100	0	0
	2017	f/f	47	0	0	100	0	0	
	1046	2016	f/f	62	0	0	100	0	0
	1055	2016	f/f	24	0	8.3	87.5	4.2	0
		2017	f/f	19	0	0	100	0	0
	1058	2016	f/f	10	0	0	90.0	10.0	0
			d/f	2	0	0	100	0	0
	1059	2016	f/f	61	1.6	98.4	0	0	0
1077	2017	f/f	71	0	0	100	0	0	
<i>C. canescens</i>	1040	2016	f/f	54	0	9.3	90.7	0	0
	1054	2016	f/f	12	0	0	100	0	0
	1074	2017	f/f	5	0	0	100	0	0
<i>C. juncea</i>	1027	2015	f/f	11	0	0	72.7	27.3	0
			d/f	2	0	0	50.0	50.0	0
	2016	f/f	19	0	0	52.6	47.4	0	
<i>C. laticoronata</i>	1056	2016	f/f	39	0	0	100	0	0
	1070	2016	f/f	10	0	0	100	0	0
	1071	2016	f/f	2	0	0	100	0	0
		2017	f/f	35	0	0	94.3	0	5.7
	1076	2017	f/f	53	0	0	100	0	0
<i>C. pauciflora</i>	1078	2017	f/f	19	0	0	100	0	0

Note. x=5. Flowering regime: f/f – free flowering, d/f – dust-free flowering.

Of all the studied populations, *C. brevirostris* had the highest degree of ploidy variation. In two out of six populations of this species (populations no. 1046 and 1077 of the Astrakhan region and Western Kazakhstan respectively) plants had stable ploidy 3x=15. In other populations ploidy ranged from x=5 (one plant of population no. 1059) to 4x=20 (7 plants of population no. 1029 of the year 2015 and rare plants of populations no. 1055 and 1058 of the year 2016).

Furthermore, a detailed analysis revealed a significant proportion of aneu- and mixoploid plants among *C. brevirostris*, *C. canescens*, *C. juncea*, and *C. laticoronata*. Also, for these species one and the same apical meristem may contain cells with different ploidy levels and diverse variants of aneuploidy. It is only in *C. ambigua* and *C. pauciflora* plants where aneuploidy and mixoploidy never occurs.

4 Discussion

The research revealed the genomic instability in the form of chromosome instability in *C. brevirostris*, *C. canescens*, *C. juncea*, and *C. laticoronata* plants. Earlier these species were

found to have notable variability in the frequencies of gametophytic apomixis in the populations [7]. The data suggest that in these species the variability of apomixis frequency results from the genome instability in apical meristem cells, while the genome instability itself comes from the hybridisation of the studied taxa or their interbreeding with other species of the genus. The strict genome diploidy of *C. ambigua* asserts that the species is obligate apomictic. Although the *C. pauciflora*'s capacity for apomictic seed formation has not been investigated, the strict genome triploidy of its plants provides evidence that it is an apomictic taxon and, most likely, a cytotype deriving from *C. ambigua* which it has very similar morphological [8] and molecular and genetic [9] features with. Apparently, this cytotype is not linked to the parental species by gene flow and is obligate apomictic.

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