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Karyotype variation within some native populations of oriental spruce (*Picea orientalis*) in Turkey

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Abstract: Karyological studies have been investigated within 8 native Anatolian populations of oriental spruce (*Picea orientalis* (L.) Link) in Turkey. The somatic chromosome number of $2n = 2x = 24$ has been observed in all accessions. The karyotypes are generally asymmetrical with most of the chromosomes having median to median-submedian centromeres. Inter-population variability of the karyotype was summarized with cluster analysis. We found that the karyotypes have positively correlated with the altitudes of the natural habitats. The high value of karyotype asymmetry may be attributed to both microenvironment and natural regeneration methods used in oriental spruce.

Key words: chromosomes; cluster analysis; karyotype asymmetry; oriental spruce

Introduction

A wide array of techniques has been used in the studies of forest tree relationships and variation (Nkongolo 1999). Initially, descriptive morphology was widely used and is still useful. This was followed by studies of growth, physiological and ecological attributes. In the past ten years, isoenzyme chemistry and DNA technologies have been employed to analyze the genetic structure of the populations of several forest trees and to delineate species (Copes & Beckwith 1977; Devey et al. 1991; Besse et al. 1993; Yeh et al. 1995; Campbell et al. 2005).

In the genus *Picea* Dietr., which comprises some 40 to 60 species distributed over the cooler areas of the Northern Hemisphere of both Old and New Worlds, genetic relationships among species have been mainly determined by crossability and morphological characteristics (Nkongolo 1999). Due to a high level of variation, classification of such a large genus has proved difficult and many ambiguities still remain (Gordon 1996).

Oriental spruce is present in most botanical gardens and some nurseries. Several cultivated forms exist. For ornamental purposes, the dwarf ones, especially 'Aurea', are beautiful additions to a rock garden (Warren 1982).

Oriental spruce is widespread in the northeast Anatolia, known as the centre of its diversity. The species covers about 350000 ha in this region. The mixed and pure oriental spruce stands begin just west of the Melet River of Ordu province in Turkey and extend along to the southern section of the Caucasian Mountains in Georgia (Turna 2004).

In many plant groups, including gymnosperms, karyotypic data have been useful in understanding phylogenetic relationships and variations. However, to our knowledge, karyological studies on oriental spruce are restricted to only chromosome numbers. No detailed karyotype analysis among oriental spruce populations using cluster analysis is available in the literature. The aim of this study is to investigate the karyotype variation within 8 native Anatolian populations of oriental spruce in Turkey.

Material and methods

Plant material

The study was carried out in 8 native populations of oriental spruce in north-east Anatolia, Turkey. Seeds were taken from 20 randomly selected individuals per population. Altitudes and coordinates of the natural habitats are given in Table 1.

Chromosome analysis

Mitotic chromosome preparations were obtained from root tips of germinating seeds. The seeds germinated in Petri dishes. The tips of roots were cut off and pretreated with 0.05% colchicine for 2–5 h (Inceer et al. 2002). They were then fixed in ethanol-acetic acid (3:1, v/v) for at least 24 h at 4 °C, hydrolysed in 1 N HCl at 60 °C for 15 min and then rinsed in tap water. Staining was carried out in 1%

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Table 1. Localities of oriental spruce populations studied.

No	Population	Altitude (m a.s.l.)	Latitude (North)	Longitude (East)
1	Örümcek (Gümüşhane)	1500	40° 39′ 40″	39° 01′ 10″
2	Kürtün (Gümüşhane)	1500	40° 43′ 30″	39° 13′ 15″
3	Ordu	1800	40° 38′ 58″	37° 56′ 16″
4	İkizdere (Rize)	1800	40° 38′ 50″	40° 32′ 12″
5	Çaykara (Trabzon)	1800	40° 34′ 30″	40° 23′ 10″
6	Çamlıhemşin (Rize)	1800	40° 52′ 20″	40° 56′ 40″
7	Dereli (Giresun)	1200	40° 35′ 50″	38° 27′ 15″
8	Artvin	1200	41° 07′ 23″	41° 37′ 22″

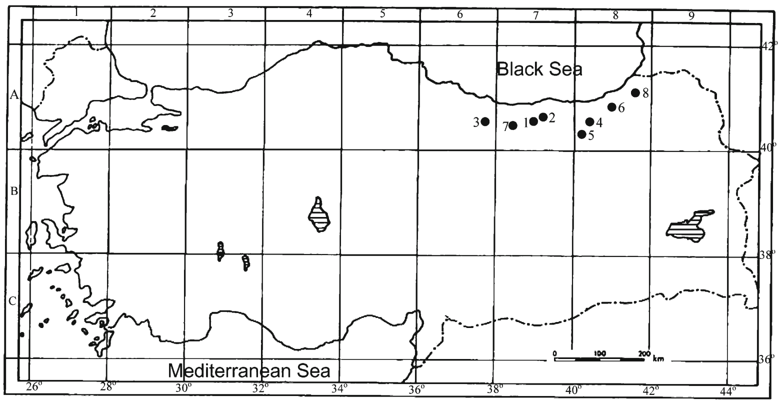


Fig. 1. Locations of 8 native populations of oriental spruce in Turkey. The populations are numbered as in Table 1.

aqueous aceto-orcein for 12–18 h at room temperature, and squashes were made in 45% acetic acid. The best well-spread metaphase plates were photographed with Zeiss Axiovert microscope or Leica DM 4000 microscope (Germany).

The best metaphase plates of each population, normally at least five, were photographed and idiograms were prepared from enlarged prints by measuring the total length of the chromosomes arms and secondary constriction. Chromosomes were classified according to Levan et al. (1964). The karyotype asymmetry index (AI) was calculated according to the formula proposed by Paszko (2006). In the haploid idiograms, the chromosomes were arranged according to their average lengths.

Cluster analysis

Several quantitative values were obtained from chromosome character measurements: total length (L+S), long arm length (L), short arm length (S), arm ratio (L/S), centromeric index (CI) and karyotype asymmetry index (AI). The grouping of the populations of the species was performed using the cluster analysis (CA) method including single linkage, unweighted paired group with arithmetic mean (UPGMA), carried out by means of Minitab (Version 13) computer program. A standardized data matrix was used for cluster analysis.

Results

Karyology

The results of karyotype analysis of the investigated populations are given in Tables 2–9. Detailed karyotypes and idiograms of oriental spruce are presented here for the first time. All the mitotic cells exhibited normal prophase, metaphase, anaphase and telophase. Chromosome counts at metaphase revealed no aneuploidy which confirms the genetic stability of the materials studied (Fig. 2). Representative somatic metaphases and haploid idiograms of each population are shown in Fig. 2 and Figs 3–10, respectively. The total karyotype length, roughly indicative of the DNA content, ranges from 109.2 to 154.2 µm among the populations. Karyotype asymmetry index corresponds to Pearson's coefficient of dispersion and gives an idea of the asymmetry caused by different lengths of the chromosomes.

Örümcek (Gümüşhane) Population: Chromosome 3 is of M-type (strictly median centromere). Chromosomes 1, 2, 4, 5, 6, 7 and 8 have median centromeres (m-type), chromosomes 9, 10, 11 and 12 have submedian (sm) centromeres (Figs 2A, 3, Table 2). Chromosomes 2 and 3 have secondary constructions. The total karyotype length is 117.1 µm and the chromosome length ranges from 6.45 to 12.93 µm.

Kürtün (*Gümüşhane*) *population*: Chromosome 3 is of M-type. Chromosomes 1, 2, 4, 5, 6, 7 and 8 have median centromeres (m-type), chromosomes 9, 10, 11 and 12 have submedian (sm) centromeres (Figs 2B, 4, Table 3). Chromosomes 2 and 3 have secondary constructions. The total karyotype length is 109.2 µm and the chromosome length ranges from 6.29 to 11.51 µm.

Ordu population: Chromosome 4 is of M-type. Chromosomes 1, 2, 3, 5, 6, 7 and 8 have median centromeres (m-type), chromosomes 9, 10, 11 and 12 have submedian

Table 2. Karyotype data for oriental spruce from Örümcek (Gümüşhane) population.

Pair	L	S	L+S	L/S	SC	CI	RL	Type
I	6.88±0.25	6.05±0.10	12.93±0.30	1.1	–	46.8	11.0	m
II	6.60±0.27	5.58±0.43	12.18±0.24	1.2	+	45.8	10.4	m
III	5.79±0.17	5.79±0.17	11.58±0.33	1.0	+	50.0	9.9	M
IV	5.76±0.34	5.01±0.26	10.77±0.59	1.1	–	46.5	9.2	m
V	5.60±0.61	4.51±0.24	10.11±0.84	1.2	–	44.6	8.6	m
VI	5.45±0.69	4.31±0.59	9.76±0.87	1.3	–	44.2	8.3	m
VII	5.28±0.30	4.23±0.76	9.51±0.76	1.2	–	44.5	8.1	m
VIII	4.75±0.31	4.31±0.25	9.06±0.44	1.1	–	47.6	7.7	m
IX	5.90±0.20	2.88±0.30	8.78±0.48	2.0	–	32.8	7.5	sm
X	5.45±0.42	2.88±0.30	8.33±0.59	1.9	–	34.6	7.1	sm
XI	4.93±0.30	2.73±0.15	7.66±0.44	1.8	–	35.6	6.5	sm
XII	4.36±0.42	2.09±0.10	6.45±0.53	2.1	–	32.4	5.5	sm

Note: Pairs of homologous chromosomes are arranged in order of decreasing length. Chromosomal type is according to Levan et al. (1964). L, mean length of the long arm of the chromosome ± SD (μm); S, mean length of the short arm of the chromosome ± SD (μm); L+S, mean total length of the chromosome ± SD (μm); L/S, length ratio of long and short arms of the chromosome (R, ratio in Levan et al. 1964); CI, centromeric index; SC, secondary constriction; RL, relative length of the chromosome; Karyotype formula: $2n = 2x = 24 = 2\mathrm{M} + 14\mathrm{m} + 8\mathrm{sm}$. Karyotype asymmetry index = 0.03.

Table 3. Karyotype data for oriental spruce from Kürtün (Gümüşhane) population.

Pair	L	S	L+S	L/S	SC	CI	RL	Type
I	6.25±0.82	5.26±0.46	11.51±1.25	1.2	–	45.7	10.5	m
II	6.10±0.73	4.88±0.48	10.98±1.19	1.3	+	44.4	10.1	m
III	5.19±0.38	5.19±0.38	10.38±0.75	1.0	+	50.0	9.5	M
IV	5.41±0.61	4.91±0.38	10.32±0.98	1.1	–	47.6	9.5	m
V	5.45±0.66	4.51±0.31	9.96±0.94	1.2	–	45.3	9.1	m
VI	5.34±0.43	4.23±0.76	9.57±0.92	1.3	–	44.2	8.8	m
VII	5.14±0.42	3.95±0.52	9.09±0.91	1.3	–	43.5	8.3	m
VIII	4.88±0.62	3.69±0.54	8.56±0.98	1.3	–	43.1	7.8	m
IX	5.25±0.65	2.75±0.33	8.00±0.96	1.9	–	34.4	7.3	sm
X	5.13±0.78	2.53±0.38	7.65±1.08	2.0	–	33.1	7.0	sm
XI	4.56±0.48	2.31±0.59	6.87±1.04	2.0	–	33.6	6.3	sm
XII	4.19±0.78	2.10±1.39	6.29±1.12	2.0	–	33.4	5.8	sm

Note: See Table 2 for abbreviations. Karyotype formula: $2n = 2x = 24 = 2\mathrm{M} + 14\mathrm{m} + 8\mathrm{sm}$. Karyotype asymmetry index = 0.07.

Table 4. Karyotype data for oriental spruce from Ordu population.

Pair	L	S	L+S	L/S	SC	CI	RL	Type
I	8.16±0.91	6.61±0.68	14.77±1.47	1.23	+	44.8	10.5	m
II	7.45±0.46	6.49±0.72	13.94±1.16	1.15	–	46.6	9.9	m
III	7.19±0.48	6.11±0.64	13.30±1.11	1.18	–	45.9	9.5	m
IV	6.51±0.53	6.51±0.53	13.02±1.05	1.00	–	50.0	9.3	M
V	6.98±0.94	5.66±0.56	12.64±1.05	1.23	–	44.8	9.0	m
VI	6.70±1.01	5.55±0.27	12.25±1.21	1.12	–	45.3	8.7	m
VII	6.56±0.72	5.45±0.77	12.01±1.25	1.20	–	45.4	8.6	m
VIII	6.39±0.67	5.09±0.62	11.48±1.15	1.26	–	44.3	8.2	m
IX	6.83±0.77	3.88±0.49	10.71±1.25	1.76	–	36.2	7.6	sm
X	6.45±0.45	2.84±0.47	9.29±0.84	2.27	–	30.6	6.6	sm
XI	5.75±0.29	2.95±0.39	8.70±0.58	1.95	–	33.9	6.2	sm
XII	5.44±0.55	2.70±0.37	8.14±0.57	2.01	–	33.2	5.8	sm

Note: See Table 2 for abbreviations. Karyotype formula: $2n = 2x = 24 = 2\mathrm{M} + 14\mathrm{m} + 8\mathrm{sm}$. Karyotype asymmetry index = 0.05.

(sm) centromeres (Figs 2C, 5, Table 4). Chromosome 1 has a secondary construction. The total karyotype length is 140.3 μm and the chromosome length ranges from 8.14 to 14.77 μm.

İkizdere (Rize) population: Chromosome 5 is of M-type. Chromosomes 1, 2, 3, 4, 6, 7 and 8 have median centromeres (m-type), chromosomes 9, 10, 11 and 12 have submedian (sm) centromeres (Figs 2D, 6, Table 5). Chromosomes 1 and 2 have secondary constructions. The total karyotype length is 154.2 μm and the chromosome length ranges from 8.31 to 16.87 μm.

Çaykara (Trabzon) population: Chromosome 5 is of M-type. Chromosomes 1, 2, 3, 4, 6, 7 and 8 have median centromeres (m-type), chromosomes 9, 10, 11 and 12 have submedian (sm) centromeres (Figs 2E, 7, Table 6). Chromosomes 2 and 5 have secondary constructions. The total karyotype length is 147 μm and the chromosome length ranges from 8.33 to 15.83 μm.

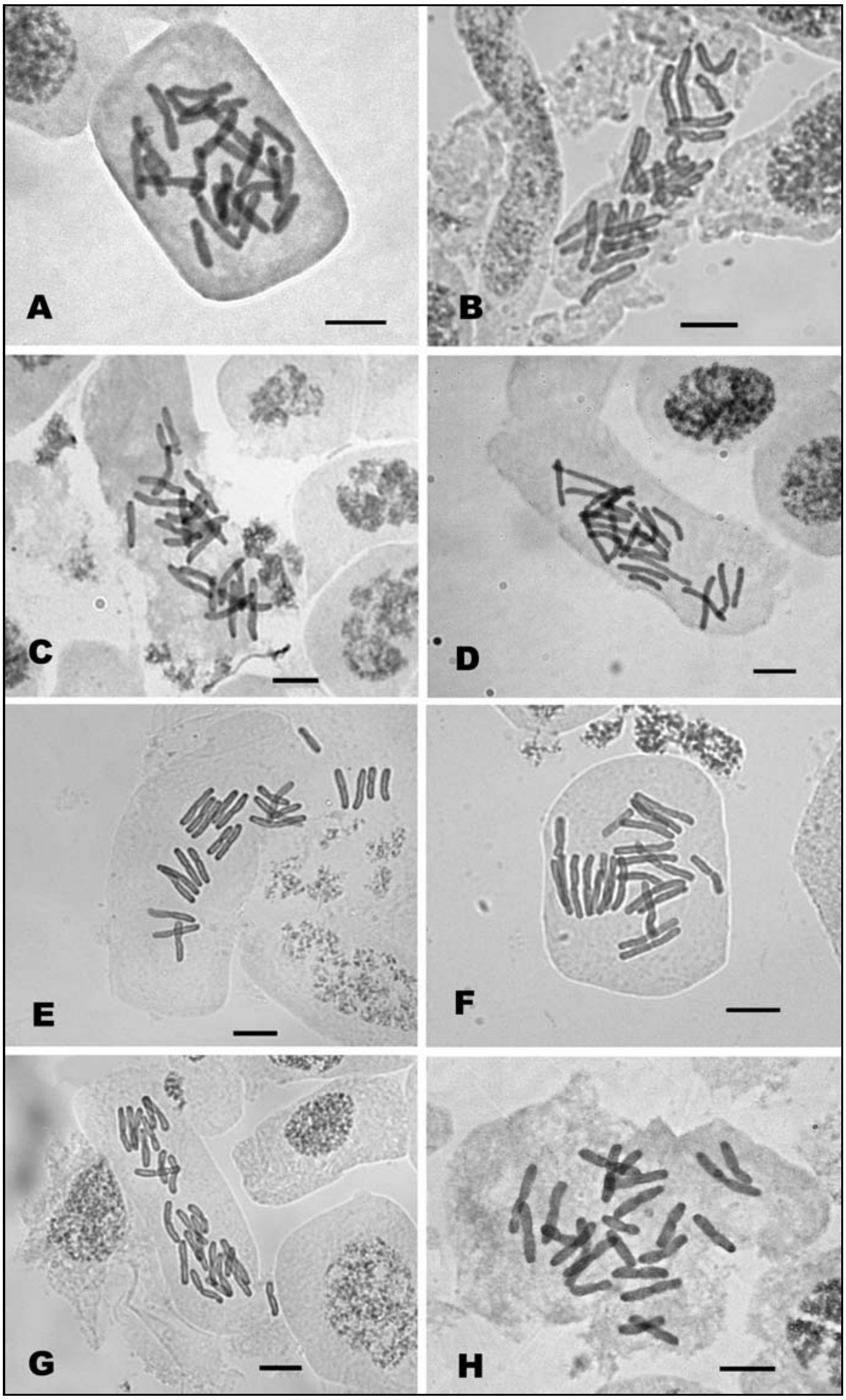


Fig. 2. Somatic metaphases of the populations studied. A– Örümcek (Gümüşhane); B – Kürtün (Gümüşhane); C – Ordu; D – İkizdere (Rize); E – Çaykara (Trabzon); F – Çamlıhemşin (Rize); G – Dereli (Giresun); H – Artvin. Scale bars 10 μm.

Çamlıhemşin (Rize) population: Chromosome 5 is of M-type. Chromosomes 1, 2, 3, 4, 6, 7 and 8 have median centromeres (m-type), chromosomes 9, 10, 11 and 12 have submedian (sm) centromeres (Figs 2F, 8, Table 7). The short arm of chromosome 1 has a secondary construction. The total karyotype length is 141.5 μm and the chromosome length ranges from 8.05 to 15.50 μm.
Dereli (Giresun) population: Chromosome 4 is of M-type. Chromosomes 1, 2, 3, 5, 6, 7 and 8 have median centromeres (m-type), chromosomes 9, 10, 11 and 12 have submedian (sm) centromeres (Figs 2G, 9, Table 8). Chromosomes 4 and 5 have secondary constructions. The total karyotype length is 131.1 μm and the chromosome length ranges from 7.14 to 13.69 μm.
Artvin population: Chromosome 5 is of M-type. Chromosomes 1, 2, 3, 4, 6, 7 and 8 have median centromeres (m-type), chromosomes 9, 10, 11 and 12 have submedian (sm) centromeres (Figs 2H, 10, Table 9). Chromo-

Table 5. Karyotype data for oriental spruce from İkizdere (Rize) population.

Pair	L	S	L+S	L/S	SC	CI	RL	Type
I	8.86±0.95	8.01±0.88	16.87±1.74	1.1	+	47.5	10.9	m
II	8.25±0.97	7.26±0.60	15.51±1.50	1.1	+	46.8	10.1	m
III	7.94±1.07	6.84±0.63	14.78±1.53	1.2	–	46.3	9.6	m
IV	7.61±0.89	6.63±0.38	14.24±1.24	1.1	–	46.6	9.2	m
V	6.96±0.60	6.96±0.60	13.92±1.19	1.0	–	50.0	9.0	M
VI	7.36±0.11	6.16±0.88	13.52±0.94	1.2	–	45.6	8.8	m
VII	7.01±0.45	6.04±0.43	13.05±0.66	1.2	–	46.3	8.5	m
VIII	7.09±0.75	5.49±0.49	12.58±0.48	1.3	–	43.6	8.2	m
IX	7.76±0.92	3.70±0.14	11.46±0.94	2.1	–	32.3	7.4	sm
X	7.15±0.18	3.60±0.45	10.75±0.51	2.0	–	33.5	7.0	sm
XI	6.20±0.34	2.98±0.29	9.18±0.37	2.1	–	32.5	6.0	sm
XII	5.36±0.44	2.95±0.10	8.31±0.50	1.8	–	35.5	5.4	sm

Note: See Table 2 for abbreviations. Karyotype formula: $2n = 2x = 24$ = 2M + 14m + 8sm. Karyotype asymmetry index = 0.09.

Table 6. Karyotype data for oriental spruce from Çaykara (Trabzon) population.

Pair	L	S	L+S	L/S	SC	CI	RL	Type
I	8.25±0.29	7.58±0.30	15.83±0.47	1.1	–	47.9	10.8	m
II	8.11±0.55	6.70±0.70	14.81±1.21	1.2	+	45.2	10.1	m
III	7.61±0.61	6.59±0.83	14.20±1.34	1.2	–	46.4	9.7	m
IV	7.50±0.60	6.00±0.18	13.50±0.51	1.3	–	44.4	9.2	m
V	6.55±0.20	6.55±0.20	13.10±0.39	1.0	+	50.0	8.9	M
VI	7.03±0.32	5.86±0.36	12.89±0.44	1.2	–	45.5	8.8	m
VII	6.71±0.45	5.75±0.41	12.46±0.45	1.2	–	46.1	8.5	m
VIII	6.90±0.18	4.80±0.22	11.70±0.32	1.4	–	41.0	8.0	m
IX	7.17±0.24	3.81±0.24	10.98±0.23	1.9	–	34.7	7.5	sm
X	6.69±0.22	3.43±0.30	10.12±0.17	2.0	–	33.9	6.9	sm
XI	6.00±0.00	3.04±0.08	9.04±0.08	2.0	–	33.6	6.2	sm
XII	5.76±0.43	2.56±0.23	8.33±0.52	2.3	–	30.7	5.7	sm

Note: See Table 2 for abbreviations. Karyotype formula: $2n = 2x = 24$ = 2M + 14m + 8sm. Karyotype asymmetry index = 0.04.

Table 7. Karyotype data for oriental spruce from Çamlıhemşin (Rize) population.

Pair	L	S	L+S	L/S	SC	CI	RL	Type
I	8.00±0.71	7.50±0.71	15.50±1.41	1.1	+	48.4	11.0	m
II	7.60±0.82	6.48±1.05	14.08±1.84	1.2	–	46.0	10.0	m
III	7.41±0.94	6.36±0.62	13.77±1.54	1.2	–	46.2	9.7	m
IV	7.06±1.05	6.08±0.62	13.14±1.65	1.2	–	46.3	9.3	m
V	6.44±0.76	6.44±0.76	12.88±1.52	1.0	–	50.0	9.1	M
VI	6.79±0.98	5.48±0.33	12.27±1.16	1.2	–	44.7	8.7	m
VII	6.26±0.48	5.80±0.59	12.06±1.06	1.1	–	48.1	8.5	m
VIII	6.70±1.00	4.54±0.35	11.24±1.27	1.5	–	40.4	7.9	m
IX	6.40±0.41	3.61±0.27	10.01±0.66	1.8	–	36.1	7.1	sm
X	6.44±0.59	2.98±0.21	9.42±0.51	2.2	–	31.6	6.7	sm
XI	5.79±0.67	3.28±0.34	9.07±0.62	1.8	–	36.2	6.4	sm
XII	5.46±0.63	2.59±0.18	8.05±0.78	2.1	–	32.2	5.7	sm

Note: See Table 2 for abbreviations. Karyotype formula: $2n = 2x = 24$ = 2M + 14m + 8sm. Karyotype asymmetry index = 0.23.

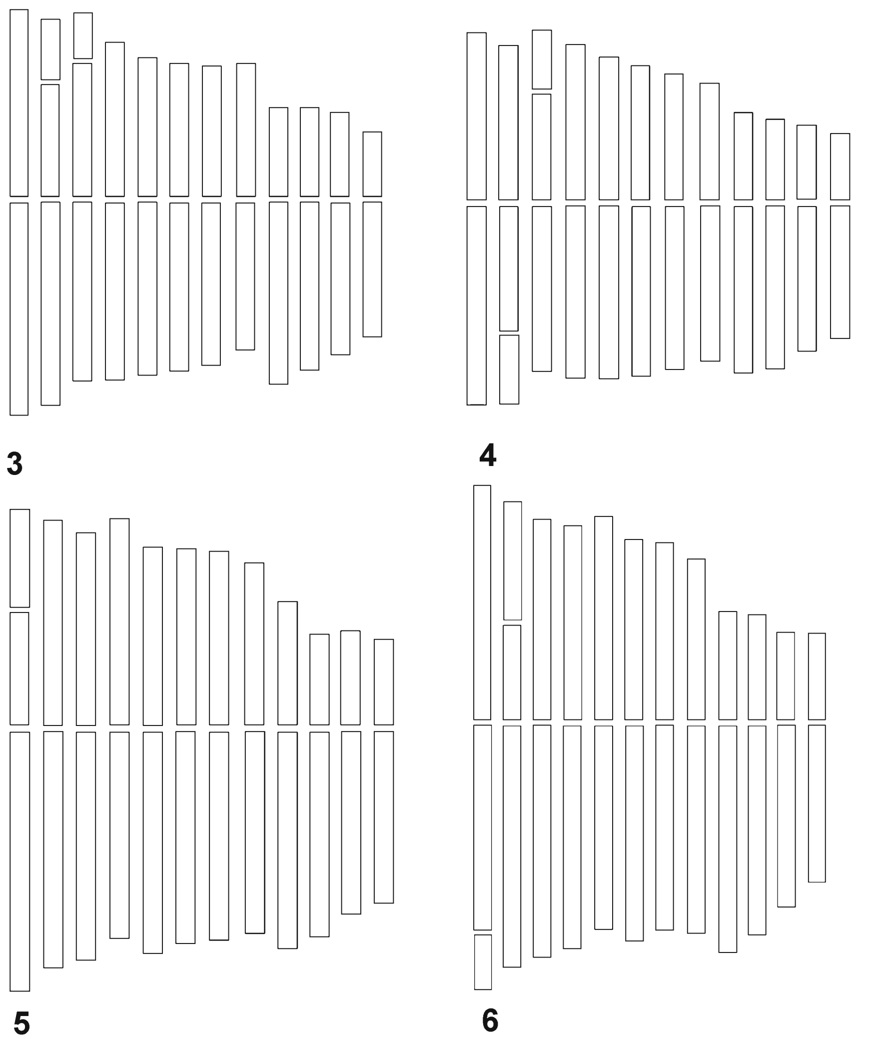
somes 1 and 2 have secondary constructions. The total karyotype length is 142.8 µm and the chromosome length ranges from 7.79 to 15.39 µm.

Cluster analysis

The dendrogram resulting from UPGMA is represented in Fig. 11. In the dendrogram, the populations of oriental spruce are connected with each other at several levels, depending on their chromosomal characteristics. The cluster analysis shows that there are three main groups. Both the first and the second group include 2 populations although the third group consists of 4 populations. Karyologic distances are low in Örümcek to Kürtün populations, Dereli to Artvin populations and Ordu to Çaykara populations, whereas they are high in Çamlıhemşin population.

Discussion

The results listed above and in Tables 2–9 confirm that the chromosome number is $2n = 2x = 24$ in *Picea orientalis*. Our diploid count is the first cytological report of this species from Turkish populations. In the literature, its chromosome number



Figs 3–6. Haploid idiograms of the populations studied. 3 – Örümcek (Gümüşhane); 4 – Kürtün (Gümüşhane); 5 – Ordu; 6 – İkizdere (Rize).

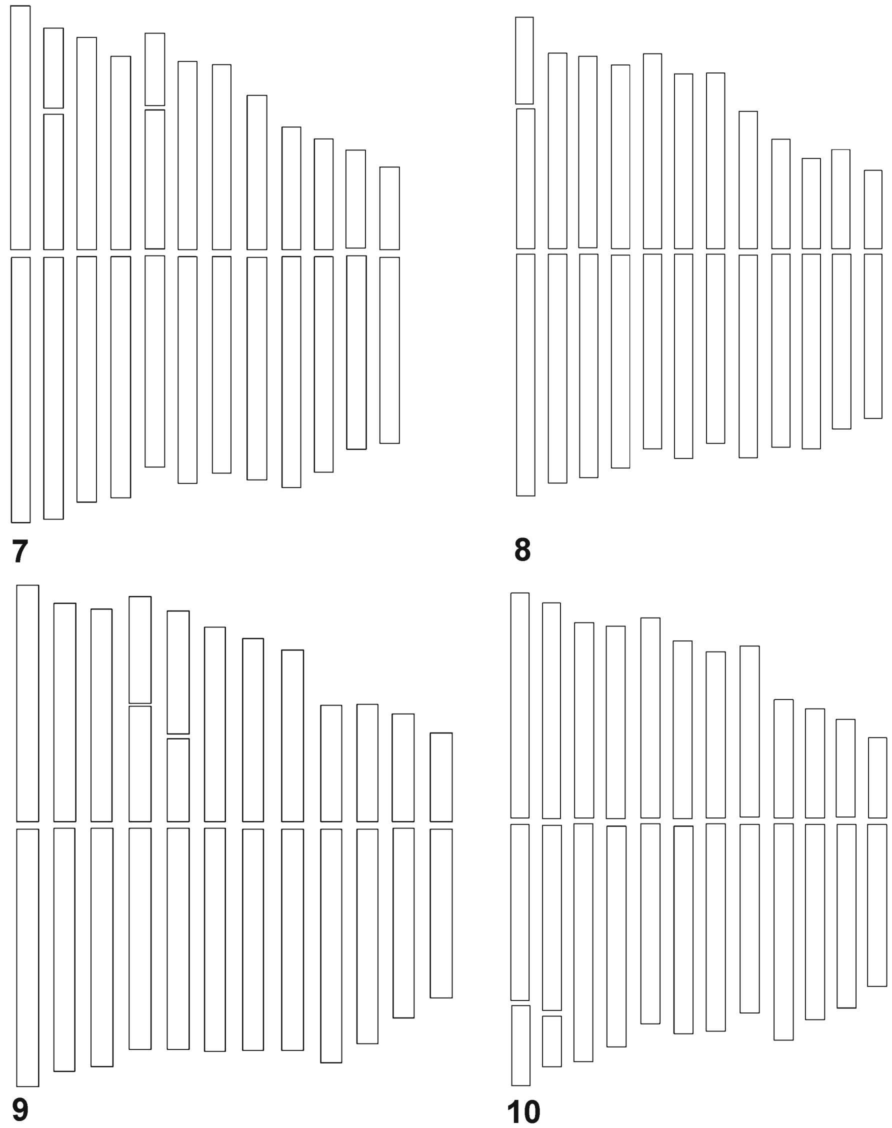
Table 8. Karyotype data for oriental spruce from Dereli (Giresun) population.

Pair	L	S	L+S	L/S	SC	CI	RL	Type
I	7.13±0.46	6.56±0.64	13.69±0.94	1.09	–	47.9	10.4	m
II	6.75±0.62	6.05±0.43	12.80±0.85	1.12	–	47.3	9.8	m
III	6.63±0.48	5.90±0.39	12.53±0.85	1.12	–	47.1	9.6	m
IV	6.13±0.41	6.13±0.41	12.26±0.82	1.00	+	50.0	9.4	M
V	6.14±0.46	5.71±0.30	11.85±0.75	1.08	+	48.2	9.0	m
VI	6.21±0.31	5.38±0.43	11.59±0.65	1.15	–	46.4	8.8	m
VII	6.13±0.16	5.06±0.80	11.19±0.72	1.21	–	45.2	8.5	m
VIII	6.14±0.57	4.74±0.40	10.88±0.59	1.30	–	43.6	8.3	m
IX	6.49±0.73	3.23±0.46	9.76±1.06	2.00	–	33.1	7.4	sm
X	5.94±0.52	3.25±0.37	9.19±0.82	1.83	–	35.4	7.0	sm
XI	5.25±0.50	2.99±0.25	8.24±0.74	1.76	–	36.3	6.3	sm
XII	4.70±0.65	2.44±0.28	7.14±0.92	1.93	–	34.2	5.4	sm

Note: See Table 2 for abbreviations. Karyotype formula: $2n = 2x = 24 = 2M + 14m + 8sm$. Karyotype asymmetry index = 0.08.

is given as $2n = 2x = 24$ (Hizume 1988). It is also the most common chromosome number in the family Pinaceae (Ohri & Khoshoo 1986; Gajdošová 1988; Hizume & Kondo 1992; Muratova 1995). No chromosomal aberrations were observed in oriental spruce.

The cytogenetic characteristics of *Picea* species include shared basic and diploid chromosome numbers ($2n = 2x = 24$), little karyotype differentiation between species, and the absence of polyploidy. With respect to these characteristics, *Picea* is representative of other conifer genera, which has led Khoshoo (1961) to hy-



Figs 7–10. Haploid idiograms of the populations studied. 7 – Çaykara (Trabzon); 8 – Çamlıhemşin (Rize); 9 – Dereli (Giresun); 10 – Artvin.

Table 9. Karyotype data for oriental spruce from Artvin population.

Pair	L	S	L+S	L/S	SC	CI	RL	Type
I	8.18±0.61	7.21±0.99	15.39±1.53	1.1	+	46.8	10.8	m
II	7.54±0.56	6.91±0.38	14.45±0.73	1.1	+	47.8	10.1	m
III	7.63±0.48	6.25±0.50	13.88±0.63	1.2	–	45.0	9.7	m
IV	7.05±0.66	6.16±0.27	13.21±0.82	1.1	–	46.6	9.2	m
V	6.41±0.36	6.41±0.36	12.83±0.72	1.0	–	50.0	9.0	M
VI	6.66±0.85	5.69±0.48	12.35±0.62	1.2	–	46.1	8.6	m
VII	6.64±0.45	5.34±0.57	11.98±0.53	1.2	–	44.6	8.4	m
VIII	6.04±0.34	5.46±0.35	11.50±0.38	1.1	–	47.5	8.1	m
IX	6.93±0.15	3.80±0.34	10.73±0.49	1.8	–	35.4	7.5	sm
X	6.23±0.21	3.50±0.08	9.73±0.25	1.8	–	36.0	6.8	sm
XI	5.88±0.28	3.13±0.15	9.00±0.16	1.9	–	34.8	6.3	sm
XII	5.20±0.49	2.59±0.14	7.79±0.47	2.0	–	33.2	5.5	sm

Note: See Table 2 for abbreviations. Karyotype formula: $2n = 2x = 24 = 2M + 14m + 8sm$. Karyotype asymmetry index = 0.11.

pothesise that the evolution of conifer chromosomes has proceeded slowly compared with angiosperms (Brown et al. 1998).

The karyotypes of the investigated populations are generally different in their chromosomal characteristics although the chromosome types are generally similar to each other (Fig. 11). Most of the chromosomes are of median (58.33%) and submedian (33.33%) centromeres.

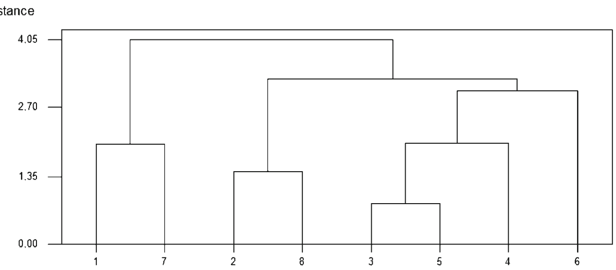


Fig. 11. UPGMA cluster analysis of the populations studied. 1 – Örümcek (Gümüşhane); 2 – Dereli (Giresun); 3 – Ordu; 4 – İkizdere (Rize); 5 – Çaykara (Trabzon); 6 – Çamlıhemşin (Rize); 7 – Kürtün (Gümüşhane); 8 – Artvin.

The karyotypes are generally asymmetrical with the smaller chromosomes being more submedian than the larger ones. These karyotypes are a common feature in the genus *Picea* and the family Pinaceae (Nkongolo 1999). Our results confirm that *Picea* is less advanced than *Larix* Link and *Abies* Miller and more advanced than *Pinus* L. The karyotype in the genus *Pinus* is primitive, the chromosomes having median centromeres and small differences in relative size (Saylor 1983) while in *Larix*, the chromosomes points are divided into two groups. One group of points indicating large, metacentric chromosomes and the other group indicating smaller, submetacentric chromosomes (Nkongolo & Klimaszewska 1995).

Karyologically most advanced karyotypes were observed in the Çamlıhemşin (Rize) population with marked chromosomal differences as to length and morphological features. The karyotype asymmetry index also reveals the higher asymmetry of the karyotype of the Çamlıhemşin population compared to other populations. There is a general assumption that asymmetrical karyotypes are derived from symmetrical ones in an evolutionary process (Stebbins 1974; Gorenflot & Raicu 1980; Inceer & Beyazoglu 2004). The high value of karyotype asymmetry may be attributed to both the microenvironment and the natural regeneration methods used over years in oriental spruce.

Our results from the karyological investigation of 8 native populations of oriental spruce show that a good correlation exists between the chromosomal characteristics and the altitude of the natural habitats despite being biogeographically distant (Fig. 11). It was determined that the populations, karyologically similar and in the same altitude, have been found to have the same clusters in the dendrogram although some populations such as Ordu and Çaykara (Trabzon) are biogeographically distant from each other. The similar karyotype in the same altitude may reflect the ecological adaptation of oriental spruce.

As seen in the dendrogram (Fig. 11), distances of the karyotypes among the populations confirm that structural chromosomal rearrangements as translocation, inversion and duplication or deficiency play an important role in the evolution of oriental spruce. According to the dendrogram, oriental spruce in Turkey can be divided into three basic groups: (i) Marginal Mountain Oriental Spruce Formation, (ii) Central Mountain Oriental Spruce Formation and (iii) High Marginal Mountain Oriental Spruce Formation. This pattern of interpopulation differentiation suggests that after the warming of the climate, the migration of oriental spruce to higher altitudes and the recolonisation of sites also with other tree species would have occurred at different speeds with varying success. This has probably led to the disruption of the range of oriental spruce, and the creation of isolated, small groups of the populations. Previous studies on oriental spruce have revealed the existence of considerable variation in morphologic and genetic characters (Turna 1996, 2004). Similar findings were also found in other forest species (Calamassi et al. 1988; Conkle et al. 1988; Turna et al. 2006).

In conclusion, these results indicate that the environmental conditions such as altitude may have an effect on karyotypes within different populations of oriental spruce. More investigations including RAPD markers would help to a better understanding of the mechanism of evolution of these populations within the species.

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