Karyological studies on four Quercus L. species in Turkey

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Abstract — In this study, the karyotype analyses of the four species of *Quercus* L. were examined. The chromosome numbers in *Q. libani* Olivier, *Q. petraea* (Mattuschka) Lieb. subsp. *iberica* (Steven ex Bieb.) Krassiln., *Q. coccifera* L. and *Q. infectoria* Olivier subsp. *infectoria* Olivier were determined as 2n=24. The karyotypes of all species consist of only metacentric chromosome pairs. Chromosome lenghts change between 0.81-2.18 in *Q. li-bani*, 0.86-1.66 in *Q. petraea* subsp. *iberica*, 0.93-1.98 in *Q.coccifera* and 0.91-1.96 in *Q. infectoria* subsp. *infectoria*, respectively. The karyotype analysis of these species were first time counted in Turkey.

Key words: Fagaceae, karyotype analysis, Quercus, Turkey.

INTRODUCTION

The genus *Quercus* is a member of Fagaceae family. This family comprises trees, small trees, and shrubs that are important sources of hard wood for industrial products, foods for animals, tannins mainly as an antiseptic, and edible nuts for humans. It includes nine currently recognized genera: *Castanea L., Castanopsis Spach., Chrysolepsis* Hjelmquist, *Colombobalanus* Nixon & Crepet, *Fagus L., Formanodendron* Nixon & Crepet, *Lithocarpus* Blume, *Trigonobalanus* Forman, and *Quercus L.* (DOĞAN *et al.* 2000; BORGARDT and KATHLEEN 1999).

Also this family includes approximately 1000 species. The biggest and the best-known group of this family is the *Quercus* with about 600 species in the world.

Turkey is one of the important diversity center of *Quercus* genus according to species number and geographical distrubition of the genus. The species of *Quercus* were represented by 18 species and 4 infraspecific taxa in three different sections in Turkey (MANOS *et al.* 2001). So far, no study has been done on their chromosomes. The taxonomic value of chromosome has not been used in order to characterize the *Quercus* species directly, but they are quite helpful to delimit the bounderies of some other species during systematical studies. The species that were analysed in this study belonged to different *Quercus* sections, such as *Quercus libani*: section *Cerris*, *Q. petraea* subsp. *iberica* and *Q. infectoria* subsp. *infectoria*: section *Quercus*, and *Quercus coccifera*: section *Ilex* (MANOS *et al.* 2001).

In the higher plants, vegetative characters are often considered as risky evidence because there are many cases where superficially similar morphological features are found in quite unrelated plants. Especially, because of wide spread introgressive hybridization (BORAZAN and BABAÇ 2003) the genus *Ouercus* is one of the most problematic groups in Turkey as problematical as do in the world. For this reason, generally cytotaxonomic characters such as chromosome number, morphology and chromosomal behaviours of the genus are preferred in the systematic studies. In addition to karyological studies, recently cpDNA analyses on the genus Quercus are also carried out for phenetic and phylogenetic classifications (BORDACS et al. 2002; DUMOLIN et al. 1995; DUMOLIN-LAPEGUE et al. 1999; FINESCHI et al. 2002; SOLTIS et al. 1992; PETIT et al. 1997).

In this study, the chromosome numbers and mophometric parameters of *Q. libani*, *Q. petraea* subsp. *iberica*, *Q. coccifera* and *Q. infectoria* subsp. *infectoria* were determined.

MATERIALS AND METHODS

The plant samples were collected during 2007 in different areas of Turkey. The localities, specimen numbers and species are shown in Table 1 and all specimens are being kept in the AIBU Herbarium.

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For karyotype studies of *Quercus* species, acorns of these samples were put into plastic cups filled with water and kept in room temperature. Root tips of germinated acorns in plastic cups were collected. As a first treatment, the root tips were pretreated in α -monobromonaphthalene for 16-17 hours at 4°C. Afterwards, roots were fixed in 3:1 absolute alcohol-glacial acetic acid mixture for over night. They were stored in 70 % alcohol at refrigerator untill analyses. Prior to staining, hydrolysis was done in 1 N HCl at 60°C for 30 minutes. Then, root tips were stained with 2 % aceto-orcein

for 2 hours and squashes were made with 45 % acetic acid. The preparations were frozen in liquid nitrogen and made permanent with Entellan.

The photographs enlarged on 10x100 were taken using OLYMPUS BX51 microscope with camera DP71 attachment. Chromosomes were classified according to the nomenclature of LEVAN *et al.* (1964). Additionally, the karyotype asymmetry parameters: Centromeric index (I^c), Intrachromosomic asymmetric index (A_1) and Interchromosomic asymmetric index (A_2) are followed ROMERO ZARCO (1986).

Table 1 — Localities of *Quercus* species used for caryological studies.

Species name	Localities	Specimen number
Quercus libani	Between Erzincan-Tercan: 35-36.km. after Tercan, 1270 m.	2007-62
Quercus petraea subsp. iberica	Samsun: 2-3.km. on the Ladik road after Havza, 780 m.	2007-49
Quercus coccifera	Edirne: 30.km. on Suluca village after Keşan, 120 m.	2007-106
Quercus infectoria subsp. infectoria	Sakarya: Bilecik road to Taraklı from Geyve, 2-3. km after Geyve, 380 m.	2007-85

RESULTS

The chromosome numbers and morphometric parameters of *Q. libani*, *Q. petraea* subsp. *iberica*, *Q. coccifera* and *Q. infectoria* subsp. *infectoria* were determined but only detailed karyotype analyses were made on *Q. libani* and *Q. petraea* subsp. *iberica*. *Q. coccifera* and *Q. infectoria* subsp. *iberica*. *Q. coccifera* and *Q. infectoria* subsp. *infectoria* were only observed according to chromosome number, length range, L/S (Largest / Shortest) and A₂ values.

The somatic chromosome numbers of all investigated species are diploid with 2n=24 and the averages of chromosomal lengths ranged from 0.81 to 2.18 µm. Among these, *Q. petraea* subsp. *iberica* has the smallest chromosomes but their observed chromosomal morphology (Fig. 1-b) was more obvious than other studied species (Figs. 1-a,c,d). Karyotypic analysis of these species were found to be very similar having all metasentric type chromosomes (Table 2, Fig. 2).

The comparison of morphometric parameters of investigated species are given in Table 3. *Q. petraea* subsp. *iberica* has the smallest value, according to parameter of haploid complement. However, the other three species have very close haploid complement values. *Q. libani* and *Q. petraea* ssp. *iberica* showed very similar intrachromosomal asymmetry (A_1) (Table 3). On the contrary to intrachromosomal asymmetry, interchromosomal asymmetry (A_2) was found to be a little distinct between these two species. But fundamentally, all of the studied species showed very similar interchromosomal asymmetry.

DISCUSSION

This work represents the first chromosomal study on some Turkish Quercus species. When the results obtained from this study are compared with previous studies on the chromosome numbers of Quercus taxa do not show much variations except Q. petraea. Chromosome number of Q. petraea may be 2n=24+1,2,3 (ZOLDO et al. 1998). Previous reports related to karyotype on *Quer*cus show that all species of Quercus investigated have diploid chromosome with 2n=24 (ZOLDO et al. 1998; Duffield 1940; Stairs 1964; Kurokawa and YONEZAWA 2004; D'EMERICO et al. 1995, 2000; OHRI and AHUJA 1990). Our studied Quercus chromosome number findings verify 2n=24, accessory chromosomes or ploidy situations are not observed in this study.

According to MANOS *et al.* (2001), the studied species are grouped in three sections. *Q. libani* is represented in *Cerris*, *Q. petraea* subsp. *iberica* and *Q. infectoria* subsp. *infectoria* in *Quercus* and



Fig. 1 — Somatic chromosomes in *Quercus* a) *Q. libani* b) *Q. petraea* subsp. *iberica* c) *Q. coccifera* d) *Q. infectoria* subsp. *infectoria*. Bar = 10 μm.

Table 2 — Chromosome numbers and karyotypic description of studied oaks.

Species	Somatic chromosome number	Karyotypic description	Length range (µm)
Q. libani	2n=24	24m	(0.81-2.18)
Q. petraea ssp. iberica	2n=24	24m	(0.86-1.66)
Q. infectoria ssp. infectoria	2n=24	24m	(0.91-1.96)
Q. coccifera	2n=24	24m	(0.93-1.98)

Q. coccifera in *Ilex* section. But SCHWARZ (1964) evaluate the same species in three subgenus as *Cerris*, *Quercus* and *Sclerophyllodrys* in respective order of sections.

When results provided from our study are compared with other studied species from the point of section (D'EMERICO *et al.* 1995), they are not entirely mathced. Nevertheless, they are not completely very distant from those values either. But when studied species are evaluated with each other in this study, morphometric parameters show less distinction.

In general, our results showed very least parametric values from *Quercus* species studied by D'EMERICO *et al.*(1995; 2000). And although the species belong to different sections, they all show the same chromosome numbers and similar chromosome parameters, except *Q. libani*. This species can be differentiated by having highest values of haploid complement, A_2 (interchromosomal index) and L/S.

This situation can be reason for *Quercus* living in different geographical regions. Because of existence of hybridization and self-incompatibility system in *Quercus* species, can make the variations increased. Beside that, our studied species have less haploid complement amount and foundamentally, this could be the reason for dissimilarity between the parametric values of two group species. Furthermore a comprehensive study in-

Table 3 — Morphometric parameters of investigated species of *Quercus*. L/S = largest/shortest chromosome; I^{C} = centromeric index; A_{1} = intrachromosomal asymmetry; A_{2} = interchromosomal asymmetry. In parameters = standart error.

Species	Haploid complement (µm)	L/S	I^{C}	A ₁	A ₂
Q. libani	16.53 (±0.11)	2.69 (±0.22)	44.60 (±0.37)	0.19 (±0.01)	0.29 (±0.11)
Q. petraea ssp. iberica	14.33 (±0.06)	1.93 (±0.16)	45.87 (±0.38)	0.15 (±0.02)	0.19 (±0.06)
Q. infectoria ssp. infectoria	16.17 (±0.08)	$2.15(\pm 0.18)$			0.22 (±0.09)
Q. coccifera	16.46 (±0.08)	$2.12(\pm 0.18)$			0.23 (±0.09)



Fig. 2 — Idiograms of two taxa of Quercus (Lenght: µm). a) Q. libani b) Q. petraea subsp. İberica.

volving more represented species from the different section in Turkey is necessery to understand *Quercus* taxonomy.

In this study, karyotypes of two taxa, *Q. libani* and *Q. petraea* subsp. *iberica* have been done for the first time. Chromosome numbers and lengths of *Q. coccifera*, *Q. infectoria* subsp. *infectoria* were also determined for the first time in Turkey. All species studied have diploid chromosome with

2n=24. At the same time, it has also noticed that *Q. libani* has the highest haploid complement and *Q. petraea* subsp. *iberica* has the least haploid complement values. This make them slightly different from the other studied species.

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