crabgrass but was identified with more than 65% genetic similarity with isolates from rice. The standard mating type tester was as well branded to 4 separate lineages, H to L. The correlation between VCGs and clonal lineages revealed low genetic diversity among all the isolates and far relationship between isolates from rice and crabgrass, whereas the possibility of speciation.

#### P0406 – ePoster

#### Studies on B-chromosomes in conifers

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B-chromosomes are found in animals, musci. gymnosperm and angiosperm plants. At present about 30 species with B-chromosomes are known among conifers. The paper deals with the results of B-chromosome studies in representatives of Pinaceae family. Among investigated by author species B-chromosomes are found in two Larix (larch) species and nine Picea (spruce) species. For karyological investigations seeds of different coniferous species (Pinaceae family) from many provenances were used. The materials were prepared and analyzed according to generally accepted techniques for coniferous plants with some modifications. The germinating seeds were pretreated in 0.5% colchicine solution for 6-8 hours, fixed in 3:1 ethanol : acetic acid mixture and stained with acetohematoxylin. Root tip meristem cells were used for study, and slides were prepared using the improved squash technique. Species from the genera Picea and Larix are the stable diploids and contain 24 somatic or A-chromosomes (2n=24). On morphological type in Picea 8 pairs of A-chromosomes are long metacentrics and 4 pairs of A-chromosomes are short meta- or submetacentrics. Karyotypes of some species include B-chromosomes. In P. schrenkiana, P. jezoensis, P. pungens, P. x fennica and P. breweriana one B-chromosome occurs (2n=24+1B), in P. koyamae and P. engelmannii 1-2 (2n=24+1-2B), in P. ajanensis and P. meyeri 1-3 (2n=24+1-3B), in P. obovata 1-4 (2n=24+1-4B), in P. glehnii 1-5 B-chromosomes (2n=24+1-5B). It was the first study case occurrence of B-chromosomes in four species - P. breweriana, P. pungens, P. koyamae and P. schrenkiana. At present Bchromosomes are found in 19 Picea species. Larix karyotype includes 6 pairs of long metacentric chromosomes and 6 pairs of short submetacentric ones. B-chromosomes are found in two Larix species: L. gmelinii and L. sibirica. Occurrence of B-chromosome in L. gmelinii was the first study case for the genus Larix. Sizes of A-chromosomes Picea and Larix are from 9 to 15 µm. Length of B-chromosomes of these species composes 25-30 % from the A-chromosomes one (4-6 µm). On their morphology B-chromosomes of Picea species can be metacentric (B1 type) and submetacentric B2 type. In P. schrenkiana, P. koyamae and P. breweriana the first type of B-chromosomes occurs only. In *P. pungens* the second type of B-chromosomes occurs only. P. obovata, P. ajanensis, Larix gmelinii, L. sibirica have the both type of B-chromosomes. It is supposed that submetacentric B-chromosomes originated in the result of pericentric inversion of metacentric ones. Other variants of B-chromosomes were found in P. glehnii. In this species five morphological types of B-chromosomes were found: large metacentric, two short meta- and submetacentric, one small metacentric, and very small submetacentric. In the opinion of some authors presence of B-chromosomes can be connected with unfavourable ecological factors (Moir, Fox 1977; Teoh, Rees 1977; Jones, Rees 1982; Broka 1990; Muratova et al. 2001, 2002 and others). Effects of B-chromosomes can have adaptive character. The results obtained allow us to consider that this system is a general phenomenon and Bchromosomes are of importance for populations and species and possibly play a role for their adaptation.

#### P0407 – ePoster

### Development and characterization of microsatellite markers on Pistachio (*Pistacia vera*)

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Pistacia vera L. (Anacardiaceae) is an economically important species as the main source of pistachio nuts. The application of genetic tools for the improvement of its commercial culture is crucial. In this sense, a linkage map based on molecular markers of this species will facilitate future breeding programs. The linkage maps are useful in important aspects such as sex-determination or marker-assisted selection for desirable traits issues (disease-resistance, growth rate, productivity...). The characterization of DNA markers is the first step towards the development of these maps. In this sense, we have constructed microsatellite-enriched libraries in pistachio by using the hybrid capture method. The libraries were made from genomic DNA samples and were enriched in di- and trinucleotides. Different libraries for males -Peter cultivar - and females - Kerman cultivar - were constructed to explore the possible association of these markers with the sex-determining loci. We also examined the utility of conserved microsatellite markers in other important Pistacia species (i.e. P. lentiscus or P. terebinthus). We believe that these data will contribute to a better knowledge of this group of species.

#### P0408 – ePoster

### Characterization of repetitive sequences on Pistachio (*Pistacia vera*)

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The genus *Pistacia* L. (Anacardiaceae) includes around a dozen of species. Some of them have economical and ecological importance, highlighting the species *Pistacia vera* – the source of pistachio nuts – or *Pistacia* 

terebinthus and Pistacia lentiscus - important elements of Mediterranean flora. Moreover, all Pistacia species are dioecious. Dioecy is a rare condition that occurs in ~7% of flowering plants genera. For all these reasons, this group of plants represents an interesting model for biological analyses being good candidates to shed light on the way gender is determined in plants. Repetitive DNA has been proved a useful tool for several evolutionary studies. Thus, it is commonly used as a molecular marker in taxonomic and phylogenetic analyses. Furthermore, its implication on sexchromosome origin and evolution has been reported. In the present work, we aim to isolate different repetitive DNA sequences from pistachio genome, focusing on satellite-DNA, a significant fraction of the eukaryotic genome. These repeats are tandemly-arrayed sequences. They are the main constituents of heterochromatin and show a pattern of evolution known as concerted evolution. The characterization of repetitive DNA from Pistacia species genome will allow us to perform different evolutionary analysis. Their organization, chromosomal location and possible differential accumulation between males and females individuals and their implication on pistachio tree sex-determination will be also discussed. Additionally, these repetitive sequences will be useful DNA markers for early identification of gender in the pistachio cultivars.

#### P0409 - ePoster

# Molecular analysis of adaptive traits associated with flooding stress tolerance in the genus *Oryza*

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Crop tolerance to flooding is an important agronomic trait. Although rice (Oryza sativa) is considered a floodtolerant crop, only some cultivars display a real tolerance to prolonged submergence, which is largely attributed to the presence of the Sub1A gene. Secondary Oryza gene pools have the potential to unveil new adaptive mechanisms and shed light on the evolution of submergence tolerance traits. In this study, we screened 109 Oryza genotypes for flooding tolerance. O. nivara and O. rufipogon accessions showed a wide range of submergence responses. The tolerance-related Sub1A-1 and the intolerance-related Sub1A-2 alleles were found in tolerant and sensitive accessions respectively. This suggests that the presence of the Sub1A gene is the result of evolutionary pressure on the Oryza species in flooding-prone areas. Flooding tolerant O. rhizomatis (IRGC-103421) and O. eichingeri (IRGC-101429) genotypes were also identified. Sub1A-1 was absent in these two species, which possess Sub1 orthologues with a high degree of similarity to O. sativa Sub1C. The coding sequence of these Sub1C-like genes revealed the presence of truncated versions of the transcript in correspondence to the ERF domain. Although Sub1A-1 was absent from these species, the expression patterns of submergence-induced genes were similar to tolerant O. sativa genotypes containing Sub1A-1. The results indicated that Sub1A-1 is not essential to confer submergence tolerance in rice relatives with the C genome that were analyzed.

#### P0411 – ePoster

# Relationships between three flowering cherry cultivars bearing green flowers

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Flowering cherry is a well-known flower in Japan. It is also very popular as an ornamental plant all over the world. We have investigated three flowering cherry cultivars bearing green flowers: Prunus 'Grandiflora' bearing yellow-green flowers, Prunus 'Gioiko' bearing variegated flowers with light green sectors and Prunus 'Shin-nishiki' bearing variegated flowers with deep green sectors. One 'Shin-nishiki' tree has been conserved in the Botanical Garden of Osaka City University. There are very few records about this cultivar, but it would appear that no other trees with the name of 'Shin-nishiki' have survived. Recently there has been confusion concerning nursery trees of 'Gioiko'. Two different types have been called 'Gioiko'. One is the original 'Gioiko', and another is 'Shin-nishiki like Gioiko'. The latter is believed to have arisen during proliferation. In this study we tried to clarify the characteristics of the three original cultivars and reveal the relationships among cultivars.

All three cultivars are double-flowered. The diameter of 'Shin-nishiki' flowers was on average 2.8 cm. This was smaller than that of 'Grandiflora' and 'Gioiko' which both had flowers bigger than 4.0 cm in diameter. The length and width of the petals were also smaller in 'Shinnishiki' compared to the other two cultivars. 'Shinnishiki' had intermediate petal-stamen floral organs. The shape of the calyx tube is one of the important characteristics distinguishing different cultivars of flowering cherry. 'Shin-nishiki' had a funnel-shaped calyx tube, whereas 'Grandiflora' and 'Gioiko' had bellshaped ones. Flowers of 'Grandiflora' and 'Gioiko' possessed similar morphological traits, whereas the flower colour of these cultivars differed as described above. All three cultivars have stomata on their petal surfaces as shown by using a KEYENCE digital microscope (KEYENCE, Japan) and SEM Miniscope (HITACHI, Japan). More stomata were observed on the abaxial side of petals, and the number of stomata was higher in the green sectors than in the white background area. 'Shin-nishiki' had high stomatal density of ca 20/mm2, whereas 'Grandiflora' and 'Gioiko' had only 7-8 stomata /mm2. In contrast, almost no stomata were observed in other flowering cherry cultivars bearing pink flowers used as a control. The total chlorophyll content of petals of 'Shin-nishiki' was 0.32mg/gFW, which was fourfold higher than in 'Grandiflora' or 'Gioiko'. This result agreed well with the differences in flower colour appearance between the three cultivars.

Based on the morphological characteristics of the flowers, 'Shin-nishiki' could be distinguished easily and clearly from the other two green cherry cultivars. Flower characteristics of 'Shin-nishiki like Gioiko' were almost same as those of 'Shin-nishiki'. Molecular analysis using SSR markers is now ongoing so that genetic relationships among three cultivars (which include four types) can be defined. Stomatal distribution and total chlorophyll