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SEQUENCE DIVERSITY AND LINKAGE DISEQUILIBRIUM IN BLACK AND WHITE POPLAR

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Association studies for trait mapping depend critically on the extent of linkage disequilibrium between functional alleles and surrounding single nucleotide polymorphism (SNPs) markers. The strategy would result very effective in natural populations of outcrossing long-lived plants thanks to: i) no need to create a mapping population and ii) higher resolution than traditional map-based strategies due to the larger number of meiotic recombination events occurred over many generations.

The availability of the *Populus trichocarpa* genomic sequence makes poplar an attractive model system to test association mapping for quantitative/adaptive traits in tree species. Recent data on natural populations of *Populus tremula* sampled across Europe pointed to a high level of nucleotide polymorphisms (π = 0.011) and a low linkage disequilibrium (LD), on the contrary in the North-American species, *Populus trichocarpa*, the nucleotide diversity is low (π = 0.0019) whereas the LD is generally high (R^2 = 0.37 over 600 bp). Considering this contrasting findings and to broaden the knowledge of the genus *Populus*, we have analyzed the sequence diversity and the LD structure in two European poplar species, *Populus nigra* and *Populus alba* (black and white poplar respectively), and compared the results on the basis of the history and distribution of populations.

Through direct sequencing of PCR amplicons, sequence diversity was surveyed at 25 loci (11 gene fragments and 14 single-copy anonymous genomic fragments) from 12 genotypes of *P. nigra* and other 12 loci (all gene fragments) from 12 genotypes of P. alba. Furthermore, for a direct comparison between the two species, sequence diversity analysis was carried out at 8 different loci from 24 genotypes of P. nigra and P. alba (collected across Europe). We found a SNP frequency of 1.1 per 100 bp in the loci analysed in white poplar with no significant difference between coding and non coding regions; in black poplar SNPs were more frequent in non coding regions (1 per 97 bp) than in the coding ones (1 per 207 bp). The overall nucleotide diversity was 0.003 and 0.0027 in white and black poplar, respectively, and sequence diversity was on average organised in 4 haplotypes per locus in both species. To evaluate the allelic distribution at the loci analysed and its deviation from neutral expectations, Tajima's D statistics and the Hudson, Kreitman and Aguadé's Test (HKA test) were computed, taking into account only SNPs. On the basis of the tests, none of the loci considered deviated significantly from the expectations based on a neutral evolution model. Overall LD estimated within the loci analysed (involving distances up to 5 kb) was found to be low but statistically significant if compared with the levels observed between physically unlinked loci. The average R^2 at 200 bp was approximately 0.25 and 0.40 in P. alba and nigra, respectively.

On the basis of this data candidate gene association mapping can be a valid approach to map interesting traits in the poplar genome.