TILLING: ALLELE MINING FOR AGRONOMICAL MAJOR TRAITS IN AN ITALIAN RICE VARIETY

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TILLING (Targeting Induced Local Lesion IN Genomes) is a reverse-genetics approach combining chemical mutagenesis with a sensitive DNA screening-technique to identify point mutations in target genes. A TILLING rice population (RICETILL) was developed from an Ethyl Methan Sulphonate (EMS) seed treatment of the rice variety Volano. The Volano cultivar was chosen as being representative of the traditional rice quality and for its relevance in ongoing breeding programs in Italy. This genetic resource was created from a starting population of 20,000 EMS-mutagenized seeds from which 1862 M2 fertile lines were obtained. From each line leaves were sampled and DNA extracted with an automated method. Five candidate genes relevant for responses to grain quality, abiotic stress, plant architecture and flowering time were chosen for investigation: Sd-1( Semidwarfing Gene), PCS -phytochelatin synthase (targeted to heavy metals incorporation into the grain), Waxy (GBSS Granule-Bound Starch Synthase for the grain amylose content), SNAC1 (related to the plant responses to drought tolerance), Hd-1 (control of heading date).

The molecular screening performed on the population for mutations in the M2 DNA samples, was performed at the PTP Genomics Platform with two methods: FLUOTILL analysis of 8- to 12-fold DNA pools and MULTISEQ with 2x DNA pools. The RICETILL population, although developed for reverse-genetics purposes, is also suitable for forward-genetics analysis and is being tested to identify variants in specific traits important for rice breeding as plant height, resistance to blast, flowering time, amylose content, panicle size. This latter part is described in detail in the poster Cavigiolo S. et al., Obtaining new genetic resources for rice breeding from an EMS-mutagenized population.

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