Determination of genes involved in the microRNA turnover in Arabidopsis thaliana

Firas LOUIS, PhD

Laboratory of Cell Cycles of Algae, Centre Algatech, Institute of Microbiology,

Czech Academy of Sciences, Trebon

Abstract

microRNAs are short 21-24nt small RNAs responsible of the regulation of proteins level in both plants and animals and as crucial regulators, they need to be tightly regulated. In plants, miRNAs are transcribed from a MIR gene and form a long transcript with a stem-loop structure necessary for processing into a smaller dsRNA by the Dicing body. The duplex is then methylated by HEN1 that confers protection against 3' nucleases. One the two strands is loaded into its partner, an ARGONAUTE protein to form the RISC complex that degrades complementary mRNAs. miRNAs can then be truncated by SDN1 or uridylated by HESO1 and/or URT1 to promote their degradation. However, SDN1 cannot degrade uridylated miRNAs and the protein responsible is still unknow. During my PhD, I tried to uncover this protein using a reverse genetic approach by rescuing the *hen1* background with different candidate genes predicted to interact with HESO1. This led us to a small phenotypic rescue of the *hen1* background by the splicing factor sua, despite not having a miRNA level increase in the vegetative stage. The alternative splicing analysis between *sua* and the wild type (Col-0) highlighted an unknown nuclease of interest (namely RNase X). The *rnase x* mutant shows a higher level of a subset of miRNAs, an increased tailing and have an improved fitness compared to Col-0, indicative of the importance of this gene. While more needs to be uncovered on this RNase X, the preliminary results suggest that it regulates a subset of miRNAs and that it may be involved in the degradation of a subset of U-tailed miRNAs.