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Avoiding positional effects in transgene expression by the use of genetic insulators in Arabidopsis

Ana Perez-González and **Elena Caro**
Centre for Plant Biotechnology and Genomics (UPM-INIA)
Campus Montegancedo UPM
28223-Pozuelo de Alarcón (Madrid), Spain

Stable and high expression of transgenes is essential for the development of genetically engineered crops. The high variability amongst transgenic lines due to gene silencing, joined to the low expression of transgenes, are nowadays a big challenge for plant synthetic biology and plant molecular farming. Positional effects due to the integration site of T-DNA are an important cause of variability, since the host genomic context where the transgene has been inserted can influence its expression due to the regulatory elements present in adjacent loci. Matrix Attachment Regions (MARs) elements are the best characterized genetic insulators and function as boundaries of DNA loops that insulate the genes they flank, thus defining a regulatory domain of gene expression1. We have performed the first parallel and systematic study of the use of four different genetic insulators flanking two transcriptional units (one with luciferase as a reporter gene, and one with the selection marker) integrated randomly within the Arabidopsis genome. Eight lines for each insulator plus a control were monitored for luciferase activity to determine the level and stability of transgene expression. Also, intergeneration, interline and interindividual differences were measured. Whole Genome Re-sequencing (WGR) was performed to map T-DNAs integration sites, and results show that some of the insulators tested allow transgene expression even when they are inserted in heterochromatic regions of the genome.

Biography

Ana Pérez-González is a PhD student at Center for Plant Biotechnology and Genomics (UPM-INIA) in Madrid. She studied Biology in Universidad Complutense in Madrid, and has been working on plant biotechnology field since then. Currently, she is finishing her PhD in the group of Elena Caro, focused on transgene silencing in plant biotechnological applications.

ana.perez@upm.es

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