ABSTRACT

In this study , we have developed activities that are part of a research project that was previously in operation regarding tomato plant. The relationship between male sterility and parthenocarpy in tomato is one of the research topics of the laboratory where I developed my project (Medina et al 2013; Rojas-Gracia et al 2017). The aim of this study was to obtain male sterile tomato plants using CRISPR / Cas9 technology. The objective of the present work is to identify those plants from this population that contain mutations in the target gene, but not the editing construct (no longer needed) and to obtain stable mutants for further genetic studies. During the course of the project, a plant mutant plant was generated, this plant was edited in a gene that is required for the development of pollen in the tomato plant. The original plant carried the CRISPR / Cas9 construct and is kanamycin resistant, and its flowers showed male sterility. The target gene was sequenced, finding changes in the sequence of the gene itself, specifically deletions of -5 and -2 nucleotides in variable proportions. The obtained plant was crossed with pollen of the wild-type genotype obtaining a first generation F1 that has been used in the present project. The results obtained were satisfactory obtaining 7out of 14 plants heterozygous and fertile. Besides, we were able to demonstrate the importance of discarding plants containing the Cas9 construct, as shows the analysis of plant 6(Sp6), which still contains the Cas9 protein, shows the presence of the -5 nucleotides and a new deletion of -17 nucleotides. The presence of mutations in the two copies of the gene (-5 and -17) indicates that the plant is a biallelic mutant; thus demonstrating that the presence of the Cas9 protein can result in additional mutations.

Key words: Pollen, Androsterilty, CRISPR/Cas9, Tomato.