

**Keywords:** Angiosperm; Pectin polysaccharides; GC content; Gene duplication; Cell adhesion

## Introduction

Pectin polysaccharides, which make up a large fraction of the immediate contact between cells, are necessary for cell adhesion in plants. The main cell wall is produced through a combination of cellulose secretion at the plasma membrane and secretion through the endomembrane system of pectin, hemicellulose, and other polysaccharides, which is where the cell wall is initially deposited during cell division [2]. The rigidity and elasticity of the cell wall can be significantly affected by enzymatic activity that further modifies the polymers. For instance, pectin methyl esterase (PME) mediates the de-esterification of pectin, which is inhibited by PME inhibitors (PMEI), which controls the charge and calcium dependent crosslinking and is related to changes in wall extensibility and adhesion. The degree of pectin esterification can affect the activity of polygalacturonases

WAKs differentiate these OGs from native pectin, it has been proposed that long polymers and fragmented pectin compete for WAK activation to activate alternative pathways [6].

**M** *Microarray*, **M** *Microarray*  
**P** *Phylogenetic tree*, **G** *Genomic*, **E** *Electrophoresis*, **A** *Agarose gel*, **I** *Immunoblotting*

In this investigation, transformation was carried out on the P. sojae-resistant soybean cultivar "Williams 82," which carries the Rps1k resistance gene. In a growth environment with a 14-h photoperiod, a relative humidity of 70–100%, day/night temperatures of 22°C/18°C, and a light intensity of 350 mol m<sup>-2</sup>s<sup>-1</sup>, seeds obtained from the T0 generation were planted in pots filled with sterile vermiculite [7].

The dominating race in Jilin Province, P. sojae race 1, PSR01, was graciously donated by Professor Shuzhen Zhang and her team. In Heilongjiang, it was isolated from diseased soybean plants.

**D** *Database*, **N** *Nucleotide*, **C** *CRISPR/Cas9*

The Phytozome database was used to download the sequence for the soybean endogenous gene GmTCP19L (Glyma.05G050400.1). The CRISPR-P web tool was used to find potential siRNA target locations within the GmTCP19L gene. Using Primer Premier 5.0, the primer binding sites for the amplification of particular siRNA target sites were created. CDSearch was used to forecast the functional domain [8]. The sequence of Cas9 was assembled downstream of the CaMV 35S promoter along with the siRNA driven by the Glycine max U6 promoter (GmU6) within its T-DNA region to create the GmTCP19L-CRISPR/Cas9 vector, carrying both GmTCP19L targeted siRNA and Cas9 cassettes. The bar gene driven by a CaMV 35S promoter was used as a screening marker.

In the T0, T1 and T2 generations, genomic DNA was isolated from each plant's leaves using the modified cetyltrimethylammonium bromide (CTAB) procedure. Next, the 621 bp GmTCP19L target area was amplified by PCR using Phanta Super Fidelity DNA Polymerase. A 1% agarose gel electrophoresis was used to find the PCR products and the GmTCP19L-F and GmTCP19L-R primers were used to sequence the results. Sequence peaks can distinguish between three different kinds of gene editing. While the wild-type (WT) and homozygous mutations had distinct peaks at the target location, the heterozygous mutations displayed overlapping peaks [9]. Sequence alignment with the WT sequence allowed for the detection of the homozygous mutations, and CRISPR/Cas9-induced frameshift mutations can result from small base insertions or deletions. We simultaneously screened T1 and T2 progenies for the tcp19l mutants devoid of the BAR and Cas9 sequences of the CRISPR/Cas9 vector. Following the manufacturer's instructions, the PAT/Bar test strip was used to identify the BAR protein, and the primers Cas9-F/R were used to amplify the fragment (349 bp) of the Cas9 gene [10].

**E** *Ensembl Plants database*

We used the online website tool CRISPR-P to analyse the potential off-target sites in order to determine whether either of the target sites might exhibit off-target activity. The sequences for the two GmTCP19L-SP1 and GmTCP19L-SP2 regions with the highest potential for off-target activity were downloaded from the Ensembl Plants database. Each off-target site's primers were created to amplify

300–500 bp regions. Through the use of the PCR technique, the regions spanning the target sites were amplified. Sequencing analysis was then used to determine the various kinds of potential off-target site editing. Detection of enzyme activities.

To test whether GmTCP19L could affect superoxide dismutase (SOD) activity and peroxidase (POD) activity, the activities of SOD and POD were measured in tcp19l mutants one gram of fresh roots were harvested at 3 days after inoculation with zoospores of P. sojae.

The SOD and POD activities were measured as described previously by Li et al. (2015) [11]. Non-transformed seedlings were used as controls.

**D** *Diagram*, **B** *Bar chart*

The relationship between GC3-50 and the synteny status of plant genes, particularly in grasses, points to a possible mechanism or pathways connecting these two traits. Individual genes can undergo non-tandem duplication, in which case the new copy is neither syntenic nor redundant, and the original (syntenic) copy can be lost by mutation with little to no effect on fitness. The likelihood that members of a gene family are not syntenic increases with repeated gene duplications and loss in either lineage. A subset of the gene space that is less duplication prone, or situations in which the duplicate copies were chosen against and swiftly lost, presumably due to gene dosage balance, is represented by genes that are still syntenic with distantly related genomes [13].

Therefore, compared to genes that have stayed single copy and syntenic, plant genes that have undergone more non-tandem gene duplications have encountered a distinct mutational environment.

Since most alterations to the third base of a codon do not affect the encoded protein, the bimodal distribution of GC content in grasses is unlikely to be the result of selection. While there is a substantial association between the GC content of grass genes and gene expression levels [8] and/or tissue specificity, the effect seems to be quite small. While selection is probably only going to have a little impact, GC content is the main determinant of codon bias in a variety of species [14].

The majority of observed SNPs in plant genomes are often explained by C–T transitions. For instance, 52.1% of SNPs in Arabidopsis and 70% of SNPs in rice are C/T polymorphisms. In the absence of selection, the C/T mutation rates are the main determinant of C/G frequency since, in rice and Arabidopsis, respectively, 15% and 25.2% of other SNPs contain A/T or C/G polymorphisms. Since gene conversion and higher mutation brought on by methylation cytosine both increase C–T polymorphisms, one or both of these mechanisms—or both—could be responsible for the observed regional difference in GC content [15].

**C** *Chart*

In the field of animal research, the regulation of RHO family proteins by PTMs has long been known. But the study of these PTM regulation mechanisms in ROP signalling in plants is still in its infancy. Here, we report information on barley ROP's in vivo ubiquitination.

The discovered ubiquitination site has previously been characterized for mammalian RACB-homologs and is conserved in all ROPs from barley, rice, and Arabidopsis. It is shown that ubiquitination affects protein abundance and that the lysine residue corresponding to RACB K167 is a common target across kingdoms.

**A** *Figure*  
 None

**C** **None**

None

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