

Two novel EMS-induced mutant alleles of *lazy plant 1 (la1)* gene in the Mo17 background

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The classical *lazy plant 1 (la1)* mutation in maize confers a striking phenotype of prostrate growth with reduced shoot gravitropism and defective inflorescence development [1]. Recently, map-based cloning of the *la1* mutant resulted in identifying mutations in *ZmLal* gene that cause *la1* phenotype [2]. This gene corresponds to gene model GRMZM2G135019 and is involved in regulating polar auxin transport and auxin signaling in maize [2]. Five *la1* alleles have been sequenced [2, 3].

We discovered two novel maize alleles of *la1* gene in Mo17 genetic background while screening M2 EMS-treated populations. Plants in two M2 families expressed a lazy plant phenotype in the field and as early as 15 days after planting in greenhouse conditions (Figure 1). Both mutant events segregate for the mutant phenotype in a 3:1 fashion consistent with a single-gene recessive mutation. Genetic complementation tests conducted by crossing heterozygous plants from both families with reference *la1-ref* heterozygous mutants resulted offspring that segregate for the lazy plant phenotype, providing strong evidence that the newly identified mutations are allelic to *la1*, located in the *ZmLal* gene and are referred to as *la1-MM115* and *la1-MM1901*.

We sequenced *ZmLal* gene from mutant plants exhibiting *la1* phenotype. *la1-MM115* is caused by 222 nucleotide deletion in the third exon (nucleotides 164 to 384 of *ZmLal* cDNA, RefGen_v2 coordinates chr4: 17,981,796..17,982,017) causing the deletion of 74 amino acids from the corresponding protein (Figure 2). This deletion was present in both genomic DNA and cDNA and no evidence for alternative splicing was observed using RT-PCR. *la1-MM1901* is caused by a nonsense substitution of C to T in the third intron (position 677 in cDNA, RefGen_v2 coordinate 17,981,504). The resulting stop codon (CAG -> TAG) leads to synthesis of a truncated protein of 225 amino acids instead of 413 amino acids (Figure 2). Seeds carrying the *la1-MM115* and *la1-MM1901* alleles have been deposited at the Maize Genetics Cooperation stock center.

Figure 1. Novel alleles of *la1* show classical prostrate growth phenotype with reduced shoot gravitropism when grown in the field (A) and in the greenhouse (B).

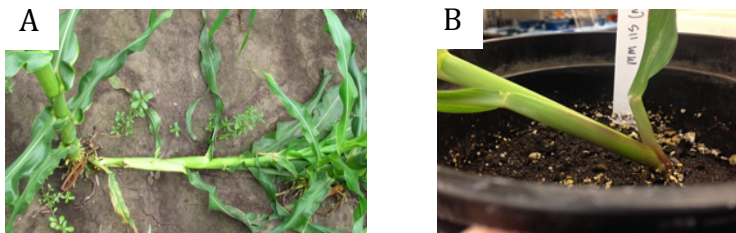
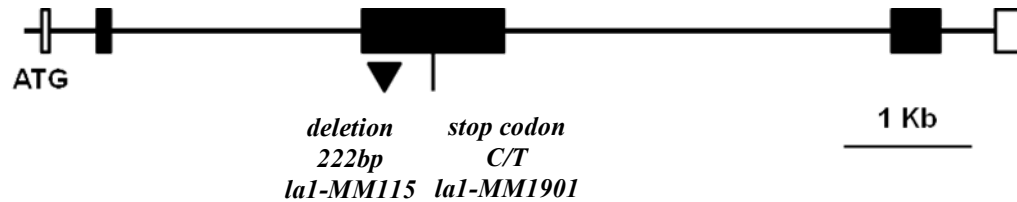


Figure 2. Position of mutations in *la1-MM115* and *la1-MM1901*. Black boxes indicate the exons, and lines between black boxes represent introns. White boxes indicate untranslated regions. The positions of the two mutant alleles are marked and described.



References:

- [1] Van Overbeek J (1936) "Lazy," an A-geotropic form of maize "gravitational indifference" rather than structural weakness accounts for prostrate growth-habit of this form. *J Hered* 27:93–96
- [2] Dong Z, Jiang C, Chen X, Zhang T, Ding L, Song W, Luo H, Lai J, Chen H, Liu R, Chen H, Zhang X, and Jin W (2013). Maize LAZY1 Mediates Shoot Gravitropism and Inflorescence Development through Regulating Auxin Transport, Auxin Signaling, and Light Response. *Plant Physiology* 163:1306-1322.
- [3] Howard TP III, Hayward AP, Tordillos A, Fragoso C, Moreno MA, Tohme J, Kausch AP, Mottinger JP, Dellaporta SL (2014) Identification of the maize gravitropism gene *lazy plant1* by a transposon-tagging genome resequencing strategy. *PLoS One* 9(1):e87053