

Does Alternative Transposition, a potential genome-restructuring activity, occur in Petunia?

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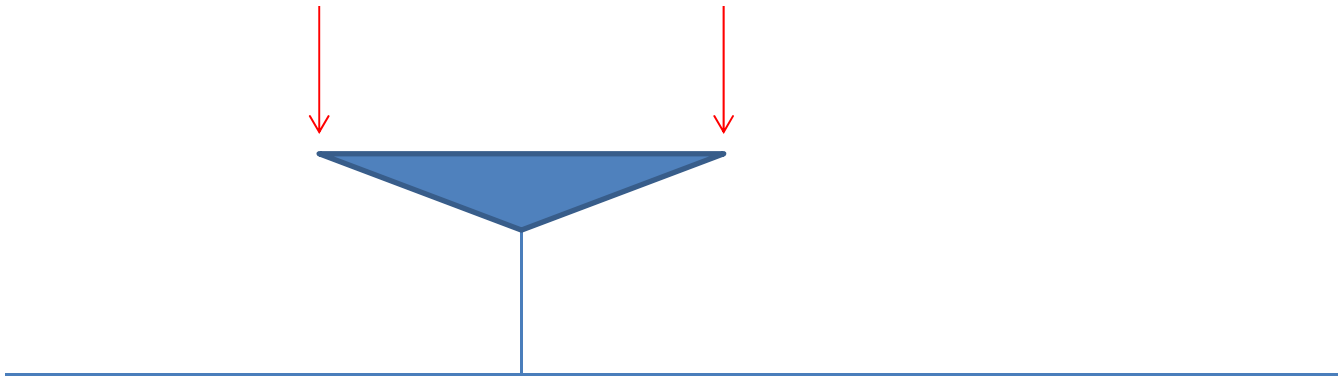


What are Transposons?

- Transposable elements (TEs) are genetic material that can move throughout the genome
- Transposase is the enzyme responsible for excising and reinserting the transposons

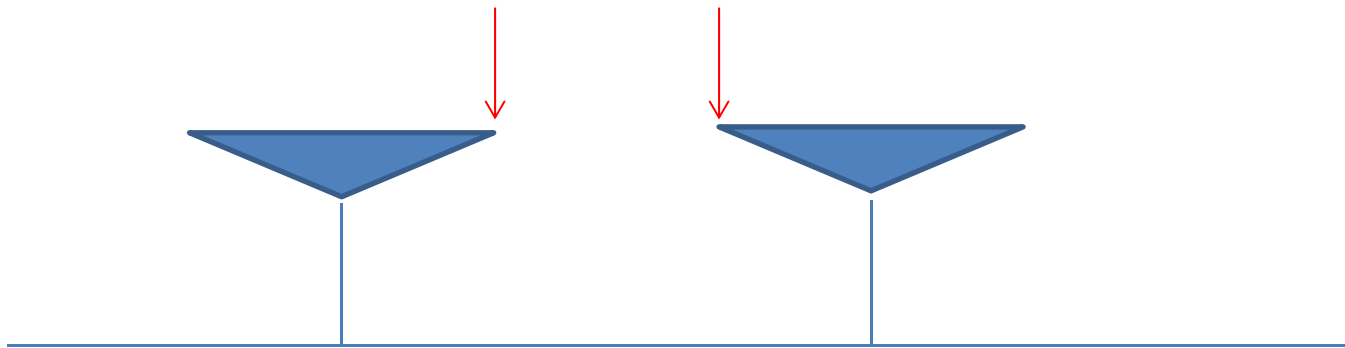
Standard Transposition

- Standard Transposition occurs when the transposase enzyme acts on the termini of the same transposable element



Alternative Transposition

- Alternative Transposition occurs when the transposase enzyme acts on the termini of two different transposable elements
- Can result in chromosomal rearrangements



Why use Petunias as a model?

- Petunias have small, non-autonomous and highly active transposons called *dTph1* elements
- Part of the hAT transposon superfamily
- S857 allele has two *dTph1* elements located approximately 30 bp apart and facing in opposite orientations

Methods

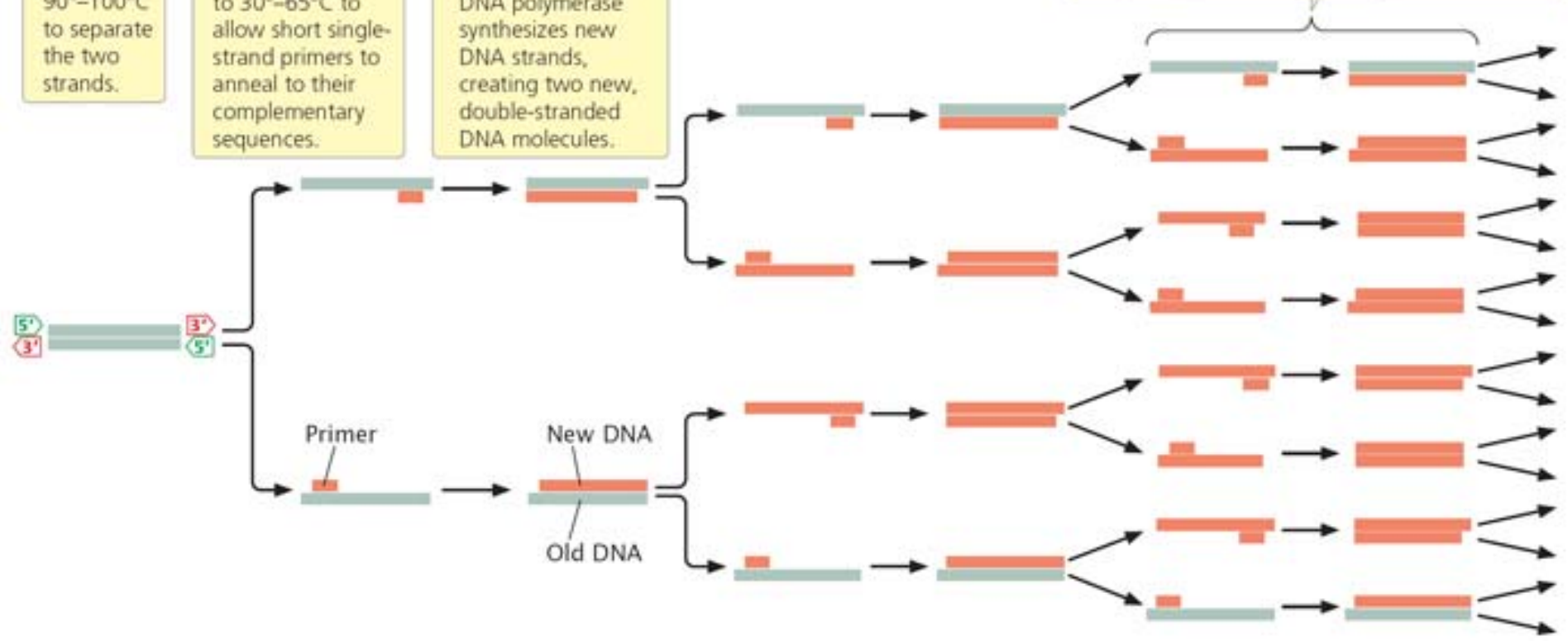
- Polymerase Chain Reaction (PCR)
- Amplifies a specific region of DNA using primers
- Amplified DNA will be run on an agarose gel
- Significant bands can be excised from gel and sequenced

1 DNA is heated to 90°–100°C to separate the two strands.

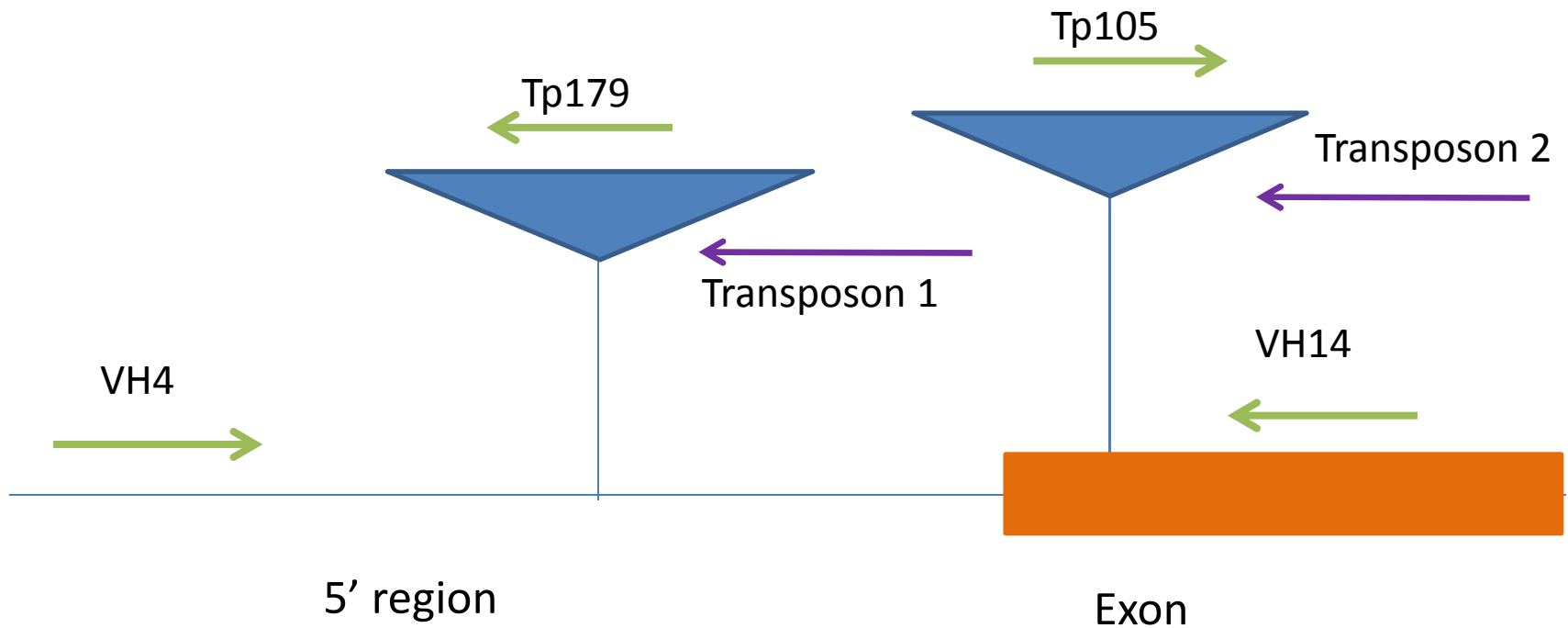
2 The DNA is quickly cooled to 30°–65°C to allow short single-strand primers to anneal to their complementary sequences.

3 The solution is heated to 60°–70°C; DNA polymerase synthesizes new DNA strands, creating two new, double-stranded DNA molecules.

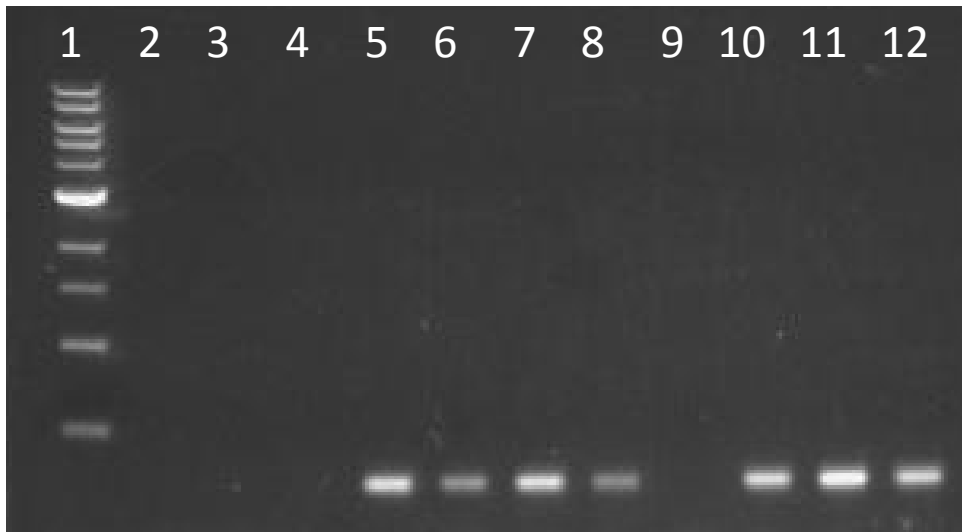
The entire cycle is repeated. Each time the cycle is repeated, the amount of target DNA doubles.



S857 Allele in Petunias

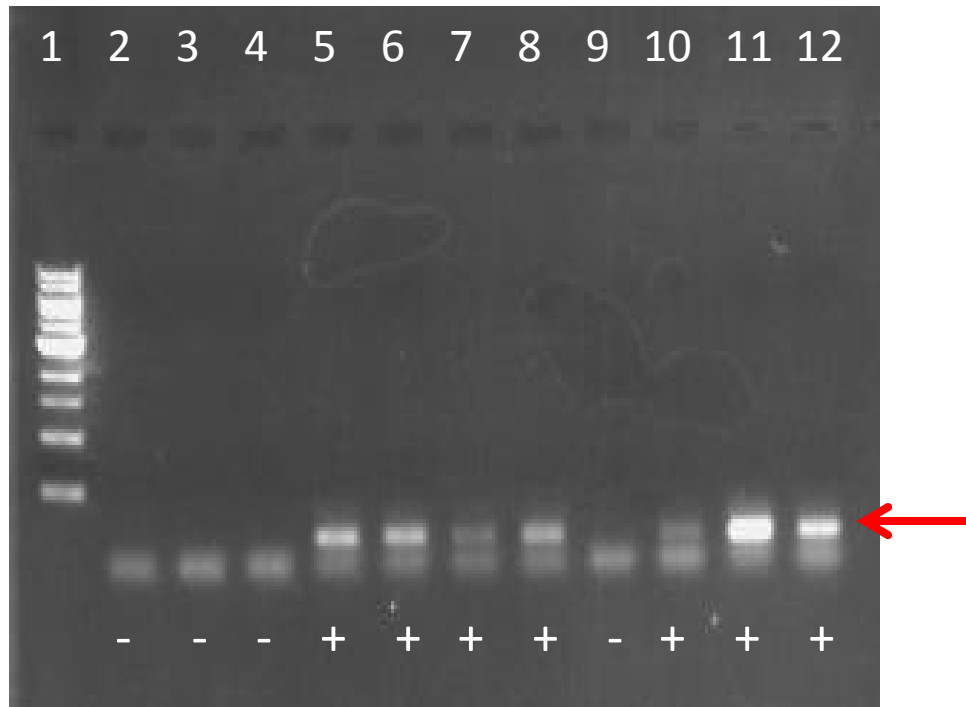


Testing for Transposon 1



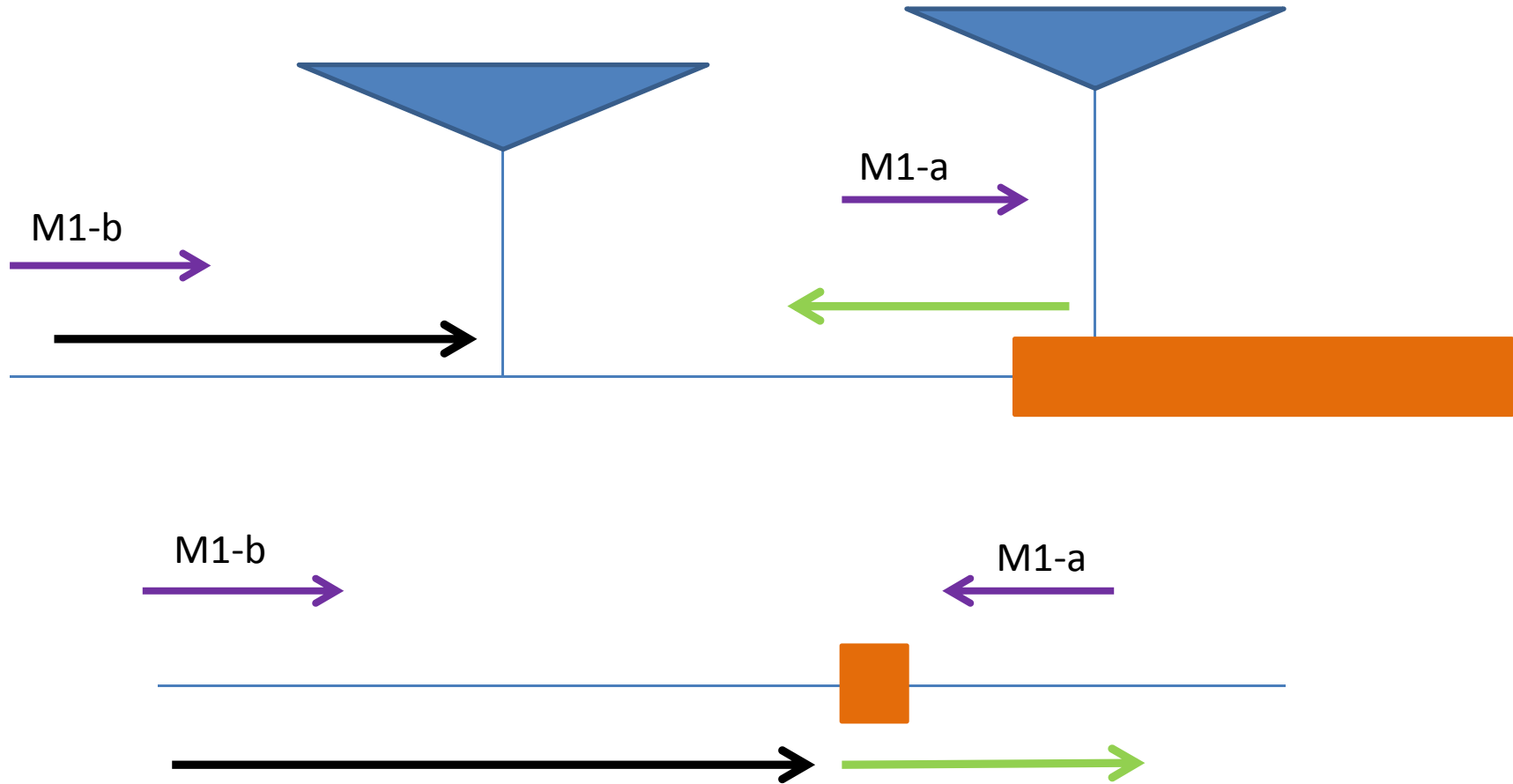
Lane	Sample
1	DNA Ladder
2	H2O
3	2:12-7
4	2:12-8
5	2:12-9
6	2:12-10
7	2:12-11
8	2:12-12
9	2:12-13
10	2:12-15
11	2:18-1
12	2:13-4

Testing for Transposon 2

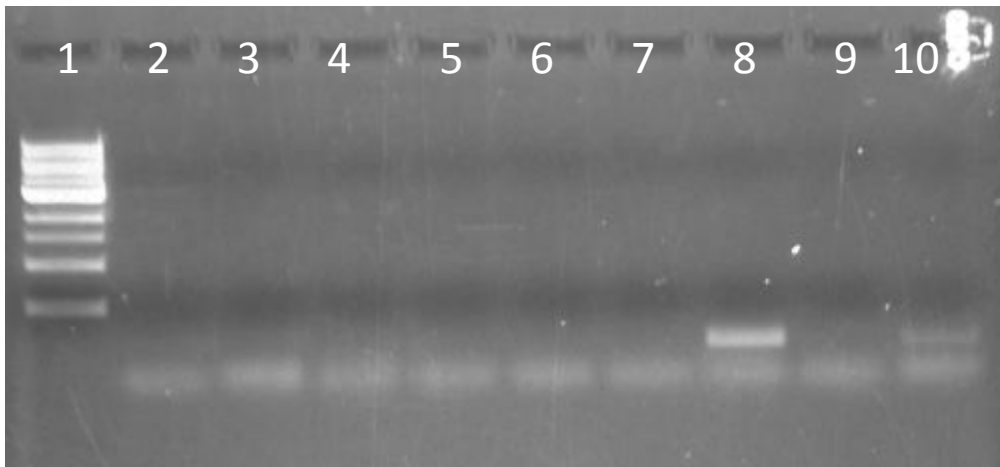


Lane	Sample
1	DNA Ladder
2	H2O
3	2:12-7
4	2:12-8
5	2:12-9
6	2:12-10
7	2:12-11
8	2:12-12
9	2:12-13
10	2:12-15
11	2:18-1
12	2:13-4

Model 1 for Alternative Transposition

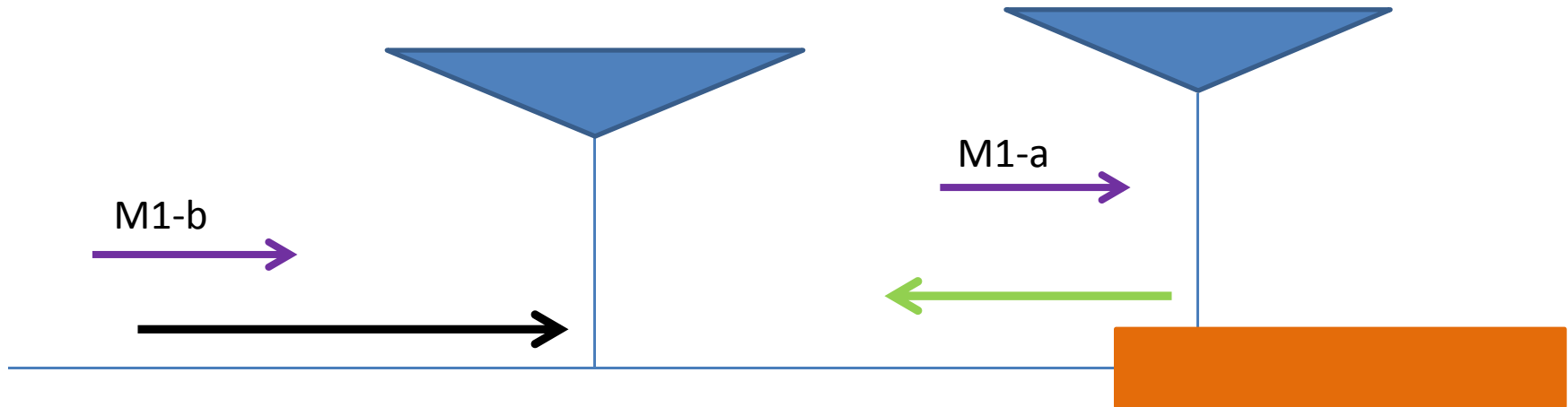


Testing for Alternative Transposition Model 1



Lane	Sample
1	DNA Ladder
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3	2:12-9
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5	2:12-11
6	2:12-12
7	2:12-15
8	2:18-1
9	2:13-4
10	2:14-2

Sequencing



Conclusion and Significance

- Evidence supports the occurrence of Alternative Transposition in Petunia
- The occurrence of Alternative Transposition in different species suggests that it may have had a significant impact on the evolution of plant genomes

Future Testing

- Continue to test plants with S857 allele for model 1 of Alternative Transposition
- Hypothesize other models for Alternative Transposition