

Development of simple sequence repeat (SSR) markers in *Medicago ruthenica* and their application for evaluating outcrossing fertility under open-pollination conditions

Jun Li^a, Hongyan Li¹, Zhiyong Li^{1*}

^a Institute of Grassland Research, Chinese Academy of Agricultural Sciences

1. Introduction

Medicago ruthenica, a diploid ($2n=2x=16$) perennial species, is a relatively unique species of *Medicago* that is adapted to dry, stony locations with extremely low snowfall and very cold winters in Siberia, Mongolia and China. *M. ruthenica* has been widely distributed in northern China as an important forage grass in cultivated and natural grasslands. However, genomic and sexual reproductive information is very limited, hindering its use in genetic and traditional breeding research.

2. Results

Using transcriptome data, we developed 213 polymorphic SSR markers. Additionally, we used these markers to quantify selfing and outcrossing rates of *M. ruthenica* plants under field conditions. The outcrossing rate was calculated to be 75.9%.

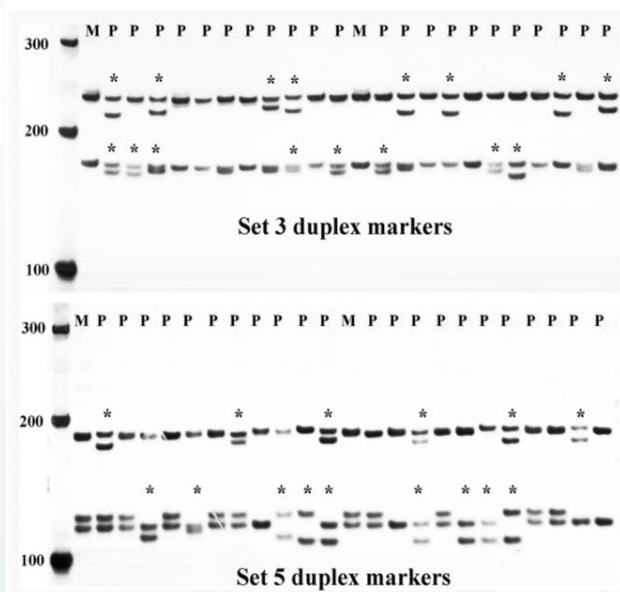


Fig. 1 DNA profile of maternal (M) parent 56-3 and progeny (P) genotypes amplified with SSR markers. The asterisk indicates a crossed offspring genotype that differed from its maternal parent. The numbers indicate the sizes on the molecular weight ladder (bp).

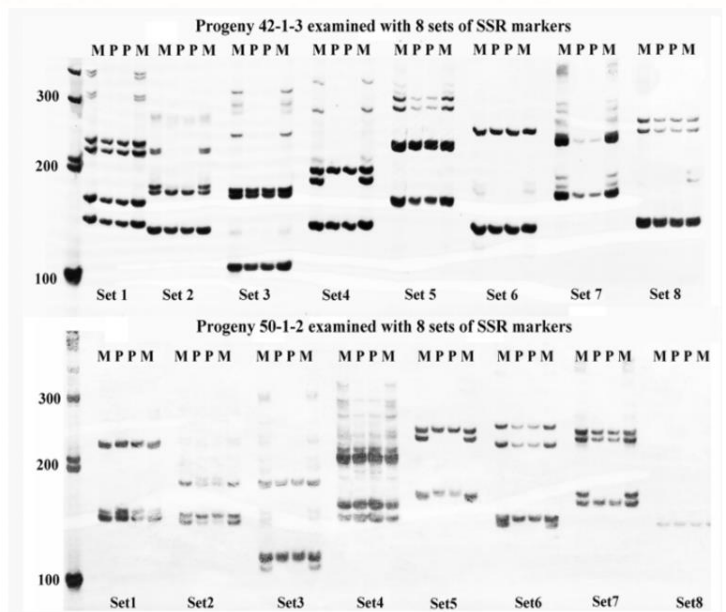


Fig. 2 Gel image of progeny (P) 42-1-3 and 50-1-2 with their respective maternal (M) parents, genotyped by 8 sets of duplex markers. SSR marker information is listed at the bottom in each group. The numbers indicate the sizes on the molecular weight ladder (bp).

3. Conclusion

Here, we first report the development of a large number of novel and efficient SSR molecular markers and outcrossing rates in *M. ruthenica*. These developed SSR markers will provide a useful tool for marker-trait association, QTL mapping, and genetic diversity analysis in *M. ruthenica*. In addition, the accurate fertilization mode under open pollination in the field has increased our knowledge of the sexual reproductive biology of *M. ruthenica*. The results should be highly valuable for molecular and traditional biological research.

