CALIFORNIA STATE SCIENCE FAIR
2004 PROJECT SUMMARY

Name(s)

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Project Number

S1699

Project Title

Transgene Expression in Wheat Engineered through Particle Bombardment

Abstract

Objectives/Goals

My objective is to reduce the amount of material necessary to make Andros spring wheat herbicide resistant by showing that the use of the GFP gene could simplify the process of homozygous plant selection at an early stage of embryo formation.

Methods/Materials

5 wheat lines were transformed using the plasmid psGFP-BAR and 5 lines were transformed using the plasmid psGFP-BAR and the plasmid pAct1-F. These lines were grown to maturity and were tested for GFP and GUS expression, as was their progeny. All transgenic wheat plants were tested for resistance to the herbicide BASTA.

Results

All analyzed transgenic lines generated after co-bombardment with psGFP-BAR and pAct1-F showed 3:1 ratio of inheritance in T1 progeny for both GFP and GUS genes. The segregation ratio in the progeny of self-pollinated T1 plants vary at a high degree. Chi square analysis revealed that some of the ratios otained are very close to the predicted 2:1 heterozygous: homozygous pattern which is characterized for the single locus insertion (3:1 Mendelian segregation). However, there was significant variation in expression levels of reporter genes among independent transformants. All tested homozygous progeny were resistant to 1% BASTA.

Conclusions/Discussion

Primary transformants with high GFP and/or GUS activities produced progeny plants with the same characteristics. Inheritance of both reporter genes was stable, and the transmission of the transgenes and the inheritance of their expression followed Mendelian ratios in the majority of the analyzed lines. A gradual reduction in gus expression was observed over two generations, which was not accompanied by a similar reduction in gfp expression. No embryos were found which showed GUS expression without GFP activity. This suggests that the reporter genes are linked together in the genome. Lack of GUS gene expression may reflect some inherent instability of GUS gene in the population examined. The results show that activity of GUS gene is more difficult to determine in wheat transgenic tissues, probably due to a lower sensitivity of histochemical methods of GUS expression compared to the highly sensitive fluorometric method of GFP expression analysis. This suggests that not only genetic and physiological factors may potentially contribute to distorted segregation.

Summary Statement

This study surveys the inheritance and expression of the gfp gene in comparison with gusA gene in T1 and T2 transgenic progeny in order to produce homozygous wheat population resistant to the herbicide BASTA.

Help Received

Used lab equipment at the Station Biotron in Pushchino, Russia under the supervision of Dr. Dmitry Miroshnichenko