Soybean Transformation with Cysteine Protease Inhibitor (CPI1) Gene for Resistance and Tolerance (In Progress…)

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Background

Soybeans are one main source of edible oils and vegetable proteins and are used to produce industrial raw materials such as biofuels. Soybeans also produce isoflavones that have a positive effect on plant life and human health. They are an important crop and are found useful in daily life, however, soybean plants are susceptible to pests and drought. Scientists are committed to genetic modification to increase soybean production and tolerance. *Agrobacterium*-mediated transformation is widely used today in molecular biology. Successful soybean transformation using *Agrobacterium* will allow for soybean seeds that are not as resistant to unfavorable conditions. The goal of this research is to insert the Cysteine Protease Inhibitor gene (CPI1) into the soybean. The CPI1 gene will control protease levels that are released during times of stress, which can lead to death of the plant. Overcoming programmed cell death will create resistant and tolerant soybeans. [1]

Research Methods

Sterile *Glycine max* seeds are germinated for five days and then excised from the medium and vertically bisected through the hypocotyl region removing the epicotyl region. Seeds are then sliced vertically along the axis about the junction between the cotyledon and hypocotyl region. Explants are co-cultivated with *Agrobacterium* containing the pHL665 construct for 5 days. Explants are washed and placed in shoot induction (SI) for 2 weeks, then seeds are transferred to new SI medium for another 2 weeks.

Abstract

Soybeans (*Glycine max*) are one of the major agricultural important crops in the United States. These legumes have a variety of uses, including food and industrial applications. Soybean growth is susceptible to drought, pest infection, and bacterial disease. These conditions strain soybean growth worldwide, dampening the effect that soybeans have on the economic network. It has been reported that the introduction of an active Cysteine Protease Inhibitor gene (CPI1) originally identified in *Arabidopsis thaliana*, will enhance soybean disease resistance and drought tolerance. The CPI1 gene helps the soybean combat programed cell death, which occurs when soybeans are exposed to unfavorable conditions and stress. Protease enzymes, released during cell death, break down the proteins and peptides necessary for the cell to live. Therefore, with the introduction of the CPI1 gene into soybean DNA protease levels can be controlled to help overcome programmed cell death. This will produce a heartier soybean that will more readily survive in a variety of adverse environmental conditions. The transgenic soybeans overexpressing CPI1 genes will grow transgenic soybeans that will promote soybean growth and its role in the economy. [2]

Seed Germination

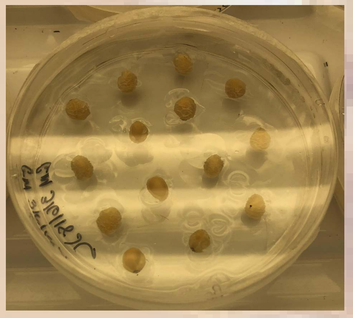


Figure 1: Sterilized seeds placed in germination medium

Figure 2: Germinated seeds after 5 days

Infection/Co-cultivation

Figure 3: Agrobacterium plated on YEP medium

Figure 4: Cut seeds on cocultivation medium

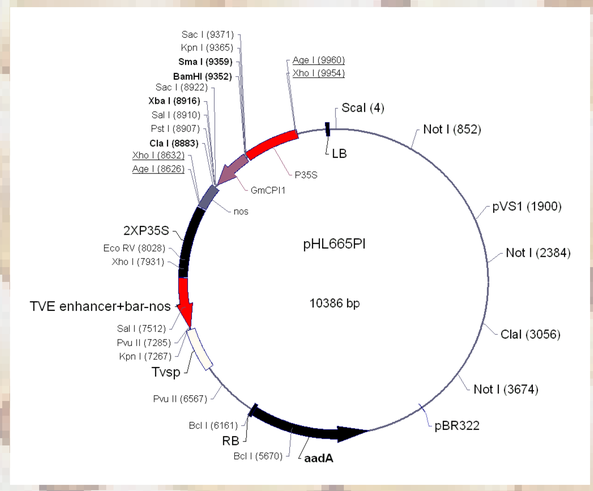
Shoot Induction

Figure 5: After 5 days of cocultivation, seeds are placed into shoot induction medium

Results and Future Work

Transgenic plants have not yet been obtained, this project will continue until molecular analysis confirms that the seeds are transformed with the CPI1 gene. Herbicide resitance will also be a form of analysis.

Genetic Engineered Binary Vector



References

[1] Erickson, D. R. (1995). Practical handbook of soybean processing and utilization. Champaign, IL: AOCS. Chapter 21- Industrial Uses of Soybeans pg 380-427

[2] Mazal Solomon, a., Beatrice Belenghi, a., Massimo Delledonne, a., Ester Menachem, a., & Alex Levine, a. (1999). The Involvement of Cysteine Proteases and Protease Inhibitor Genes in the Regulation of Programmed Cell Death in Plants. The Plant Cell, (3), 431. doi:10.2307/3870871

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