CALIFORNIA STATE SCIENCE FAIR
2014 PROJECT SUMMARY

Name(s)

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Project Number

S0526

Project Title

DNA Sequence Analysis Reveals Differences between Epazote Downy Mildew and Other Downy Mildews

Abstract

Objectives/Goals

Downy mildew caused by Peronospora effusa is a serious disease of spinach in coastal California and worldwide. I hypothesized that downy mildew from Epazote (Dysphania ambrosioides), a popular crop in Mexican cuisine and grown extensively in California, is genetically identical to the pathogen that causes disease on spinach. Both of these crops belong to the Chenopodiaceae family. My goal was to employ the ITS (internal transcribed sequence) region from the rDNA to determine the genetic similarity between these pathogens. This analysis will help determine the validity of a DNA-based diagnostic assay already developed for specific detection of the spinach downy mildew pathogen.

Methods/Materials

DNA from infected Epazote leaves was extracted using a commercial DNA extraction kit and the ITS region was amplified by Polymerase Chain Reaction (PCR). The PCR product was visualized on an agarose gel and transferred into a TOPO vector. The vector containing the cloned insert was sequenced and returned in the form of a chromatogram, which was analyzed and cloned sequences were obtained. The National Center for Biotechnology Information (NCBI) database was searched by the Basic Local Alignment Search Tool (BLAST) using the cloned sequence. Newly cloned sequences were then aligned with closely matched sequences from NCBI and phylogenetic relationships were determined.

Results

Of the first hits, P. effusa, P. farinosa, and P. corydalis were the closest matches, having a sequence identity of 93%. P. effusa infects spinach, P. farinosa infects beet, and P. corydalis infects squirrel corn. From the sequence analysis, it was determined that the species of Peronospora that affects Epazote is distinct from P. effusa and that the former most likely represents a novel species of Peronospora.

Conclusions/Discussion

My studies indicated that Peronospora effusa (the pathogen that infects spinach) is genetically distinct from the downy mildew species that infects Epazote. These results rejected my initial hypothesis that the two pathogens are genetically similar. Epazote downy mildew has not been previously reported in California to our knowledge. The species of downy mildew that infects Epazote is still undetermined. Future studies will be focused on describing this new species, and also its pathogenicity on other selected crops or weeds in California.

Summary Statement

Analyses of the ITS rDNA sequences determined that spinach and Epazote downy mildew are genetically distinct.

Help Received

Dr. Steven Klosterman and Mrs. Amy Anchieta accommodated me in their laboratory and guided me through all phases of my study.