



CALIFORNIA STATE SCIENCE FAIR  
2017 PROJECT SUMMARY

<b>Name(s)</b> Vivian Hoang; Trevina Tan	<b>Project Number</b> <b>S0512</b>
<b>Project Title</b> <b>Plant Defense Transcription: Who Is in Control, the Plant or Pathogen?</b>	
<b>Abstract</b> <b>Objectives/Goals</b> The Mudgett laboratory hypothesized that WRKY transcription factors bind to the tomato DH1 promoter to regulate DH1 gene expression during pathogen infection and that XopD may interact with the WRKY transcription factors to interfere with DH1-regulated defense responses. The goal of our project was to test these hypotheses by identifying WRKY factors that bind to the DH1 promoter. In tomato, there are 81 annotated WRKY transcription factors. To identify WRKYs that regulate DH1 gene expression, we had two specific aims: Aim 1: We determined the subcellular localization pattern of 81 tomato WRKY transcription factors by transiently expressing GFP-WRKY fusion proteins in plants cells using the Agrobacteria-mediated transient expression assay and identified WRKY factors that display similar subnuclear localization patterns to that of XopD. Aim 2: We identified WRKY factors that altered DH1 gene transcription by expressing the GFP-WRKY factors in transgenic tomato leaves containing the DH1 promoter fused to the $\beta$ -glucuronidase (GUS) gene.	
<b>Methods/Materials</b> We used transformed Agrobacterium expressing the GFP-WRKYs from our mentors and created inoculums that were injected into N. Benthamiana. After taking samples, we examined them under the microscope to determine localization. Secondly, we looked at WRKY expression by inoculating agrobacterium in transgenic tomatoes and determined their biological roles in DH1 expression using gus assays and a fluometer. We determined the significance of the data using a t-test using SPSS statistics software.	
<b>Results</b> Of the 81 WRKY transcription factors tested, 40 localized to the nucleus, 11 in the nucleolus, 10 in both the nucleus and cytoplasm, 7 in the cytoplasm, and 13 in nuclear foci, which is consistent with XopD localization. WRKY6 and WRKY40 activated the pDH1::GUS reporter gene, suggesting that these two proteins may directly regulate DH1 transcription and form a complex with XopD.	
<b>Conclusions/Discussion</b> We determined that tomato WRKY transcription factors (WRKY6 and WRKY40) regulate DH1 gene expression and are potential targets of the Xanthomonas XopD virulence factor. WRKY transcription factors are DNA-binding proteins that regulate many immunity processes in plants, especially tomatoes. Researching WRKY activity in DH1 transcription can lead to solutions that increase tomato immunity.	
<b>Summary Statement</b> Through our project, we determined two WRKY transcription factors (WRKY6 and WRKY40) bind to the tomato DH1 promoter to regulate DH1 gene expression during pathogen infection.	
<b>Help Received</b> We worked on the experiment using equipment at Mudgett laboratory in the Biology Department at Stanford University and were inspired by their prior work. We got help in understanding and received advice from Prof. Mary Beth Mudgett. Dr. Jung-Gun Kim monitored our project and gave us the	