Reading Guide for Hajeri *et al.*, 2014
A Supplement to "Combatting the Southern Pine Beetle"

by

Ann T.S. Taylor, Department of Chemistry, Wabash College, Crawfordsville, IN

Rebecca L. Leuschen-Kohl, Quantitative Plant Pathology Program, Purdue University, West Lafayette, IN

In order to prepare for the case study "Combatting the Southern Pine Beetle," read the following article and answer the questions below:

- Hajeri, S., N. Killiny, C. El-Mohtar, W.O. Dawson, & S. Gowda. (2014). *Citrus tristeza* virus-based RNAi in citrus plants induces gene silencing in *Diaphorina citri*, a phloem-sap sucking insect vector of citrus greening disease (Huanglongbing). *Journal of Biotechnology* 176: 42–9. <https://doi.org/10.1016/j.jbiotec.2014.02.010>

Questions

1. Name the following:
   a. The two names for the disease of interest:

   b. The bacterium of interest:

   c. The bacterium-carrying vehicle of interest:

   d. The viral DNA vector of interest:

2. What is Awd? What is PDS? What previously published findings are important to the current study?

3. What hypothesis is the current paper testing?

4. In Figure 1, we see a schematic representation of the CTV genome in a binary vector.
   a. What is the point of the 35S promoter?

   b. The schematic shows the creation of four separate constructs (shown after the p23 ORFs). What does each construct represent?

5. Describe what is happening in the images of Figure 2a. Include the following terms in your description: *Nicotiana benthamiana, GFP-tagged line 16c, CTV vector,* and *tGFP.*

6. In Figure 2c, we see a northern blot showing the sgRNA expression of CTV-wt versus CTV-tGFP.
   a. What does a northern blot identify?

   b. Compare and contrast the bands of the northern blot for CTV-wt and CTV-tGFP.

   c. Why did the researchers test GFP silencing before the tPDS or tAwd constructs?

7. In Figure 3, what phenotype did we see in *C. macrophylla* plants treated with *Agrobacterium* carrying CTV-tPDS?

8. Explain why the researchers tested whether or not the CTV-tAwd construct could be grafted on to other species of citrus.

9. In Figure 5E, what is the purpose of TubA? What is the purpose of the data shown in Figure 5E?

10. How did the researchers measure the efficacy of the CTV-tAwd vector in managing the *D. citri* population? What were the results?