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Nebraska Soybean Board
Year-End Research Findings Report

Please use this form to summarize the practical benefits of your research project and what has been accomplished. Your answers need to convey why the project is important and how the results impact soybean production.

Project Title: Improve soybean resistance to Nematodes with synthetic RNA interference

Contractor & Principal Investigator: Bin Yu

Please check/fill in appropriate ☐ ***box: Continuation research project***
☒ ***Year 1 of 3 year research project***

1. ***What was the focus of the research project or educational activity?***

As the most destructive pest disease for soybean, The soybean cyst nematode (SCN) costs huge losses every year. This project is expected to develop novel technology to generate soybean varieties with high resistance to SCN. We will use four different Pol III promoters to drive the expression of four short hairpin RNAs at the same time in soybean, respectively. Each short hairpin RNAs (shRNAs) will repress the expression of an essential gene of SCN when consumed by SCN, resulting in death of SCN. In addition, the adaption of SCN to this variety will be also examined. Furthermore, we will examine the combination of natural SCN resistant trait with shRNAs will reduce the SCN adaptation. This aspect is less explored in the field.

2. ***What are the major findings of the research or impacts of the educational activity?***

Vectors harboring four siRNAs targeting four different genes of SCN have been transformed in Dr. Clemente's Lab. We are in the process to evaluate the expression of transgenes and the generation of siRNAs. We are also in the process to design additional vectors used for transformation. In this period, we also developed deep-seq based method to identify the insertion position of transgene in soybean. This is important since knowing the transgene insertion position is fundamental to the application of crop engineering and the method is accurate and cost-efficient. This method is based on the newly developed nanopore technology that sequence long DNA fragments.

3. ***Briefly summarize, in lay terms, the impact your findings have had, or will have, on improving the productivity of soybeans in Nebraska and the U.S.***

As the most destructive pest disease for soybean, SCN cost huge losses every year. However, the limited number of resistant soybean varieties has challenged the management of SCN. This project will generate a SCN resistant soybean variety, and therefore has the potential to help the management of SCN and to benefit soybean production in US including Nebraska. In addition, this project applies the next generation RNAi technology to improve soybean traits. If succeed, it will open doors to apply this technology to improve other important soybean traits, such as fungi disease resistance. Thus, this project should have the potential to benefit soybean production more broadly. Moreover, this study will study the adaption of SCN to this strategy and combined

this strategy with natural SCN resistance. We expect this combination will reduce the adaption and help soybean breeding in long run in an environmental friendly and cost efficiently way

4. Describe how your findings have been (or soon will be) distributed to (a) farmers and (b) public researchers. List specific publications, websites, press releases. etc.

Dou, Y., Li, S., Yang, W., Liu, K., Du, Q., Ren, G., Yu, B., and Zhang, C. (2017). Genome-wide Discovery of Circular RNAs in the Leaf and Seedling Tissues of Arabidopsis Thaliana. Current Genomics **18,** 360-365.

Li, S., Castillo-Gonzalez, C., Yu, B., and Zhang, X. (2017a). The functions of plant small RNAs in development and in stress responses. Plant J **90,** 654-670.

Li, S., Liu, K., Zhang, S., Wang, X., Rogers, K., Ren, G., Zhang, C., and Yu, B. (2017b). STV1, a ribosomal protein, binds primary microRNA transcripts to promote their interaction with the processing complex in Arabidopsis. Proc Natl Acad Sci U S A **114,** 1424-1429.

Ren, G., Wang, X., and Yu, B. (2017). Analysis of the Uridylation of Both ARGONAUTE-Bound MiRNAs and 5′ Cleavage Products of Their Target RNAs in Plants. Methods in molecular biology **1640,** 23-27.

Wang, H., Zhang, C., Dou, Y., Yu, B., Liu, Y., Heng-Moss, T.M., Lu, G., Wachholtz, M., Bradshaw, J.D., Twigg, P., Scully, E., Palmer, N., and Sarath, G. (2017). Insect and plant-derived miRNAs in greenbug (Schizaphis graminum) and yellow sugarcane aphid (Sipha flava) revealed by deep sequencing. Gene **599,** 68-77.

Yu, Q., Liu, Y., Li, M., and Yu, B. (2017). Small RNA Biogenesis and Degradation in Plants. Plant Epigenetics, 107-127.

5. Did the NE soybean checkoff funding support for your project leverage any additional state or Federal funding support? (Please list sources and dollars approved.)

We submitted a grant to NSF and NIH based on the data generated through this project this March but not funded. We are going to continue this effort and will submit a grant to USDA