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| Please use this form to clearly and concisely report on project progress. The information included should reflect quantifiable results that can be used to evaluate and measure project success. Comments should be limited to the designated boxes. Technical reports, no longer than 4 pages, may be attached to this summary report. |
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| Project Title:  | Southern Root-Knot Nematode in Maturity Group 4 Soybean: Characterization of Resistance Mechanisms and Breeding for Resistance |
| Organization:  | University of Arkansas System, Div of Ag |
| Principal Investigator Name: | Travis Faske |
| Report Period: |  6/15/2024 to 9/15/2024 |
| Project Status: |
| **Objective 1. Characterization of the mechanism of resistance to SRKN. (Faske and Watson; Univ of Arkansas and LSU, respectively)**A greenhouse experiment was designed to evaluate the reproduction vs galling in the PI lines that are being evaluated for the MOR to SRKN in this study at the Lonoke Extension Center in Arkansas. Based on the percentage of root system galled, all resistant lines (Forrest, PI 438489B, PI 567305, and PI 567516C) had lower (P = 0.05) than the susceptible control, Magellan. However, no single resistant cultivar had lower SRKN reproduction than Magellan. These data support that genes that control nematode reproduction are not necessary the same that control galling. Furthermore these data support the direction of this project to evaluate resistant germplasm that has both low reproduction and galling. This experiment was only conducted once and is currently in the final stages of completion at the Lonoke Extension Center. **Objective 2: Genetic characterization and development of functional markers for new sources of resistance to SRKN. (Nguyen, Univ. of Missouri)**Marker-Assisted Selection: Received 80 lines from Dr. Feng Lin's group (Fisher Delta Research Center, Portageville, MO) for screening for SRKN resistance. DNAs were extracted from the leaf samples of these 80 lines and were screened to identify major SRKN-resistant loci on Chromosome 10 using KASP PCR. Among all the soybean breeding lines, an elite line S21-22067 was found to be resistant to SRKN. This elite line is a promising resource that can be integrated into the breeding program for SRKN resistance.Identification of RILs for Chr. 10 & Chr. 13 QTLs – Continuing the previous year's study, we identified about 32 RILs developed from a cross of Mag × PI 438489B based on major resistance on Chro. 10 and minor resistant on Chr. 13. These contrasting RILs were selected with a combination of QTL on Chr.10 + Chr. 13, only Chr.13 QTL, only Chr. 10 QTL, and further these RIL lines along with the parents were planted in the BREC field of the University of Missouri.Seed increase: We planted seeds of the major RKN lines such PI 438489B, PI 567305, Forrest, PI 567516C, Magellan, and NILs at the University of Missouri & Arkansas for seed increase.**Objective 3. Development of breeding populations and MG IV soybean varieties with resistance to SRKN.**

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| **University of Arkansas, Caio Vieira**: Yield trials at various testing locations are progressing smoothly, with harvesting starting in mid-September. We initiated breeder selection and maturity note-taking. Additional morphological traits, including pod color, pubescence, and lodging will also be recorded. Harvest preparation is in full swing, with the organization of harvest bags, tag preparation, site selection, and calibration of field equipment well underway. Crossing block activities concluded by the first week of September. Once the maturity crosses are harvested, they will be sorted, counted, and shipped to off-season nurseries for generation advancement. Additionally, leaf tissues from all preliminary entries have been collected and DNA isolated for Soy3KSNP genotyping. Seed samples have also been sent out for proprietary disease and abiotic stressors panel screening.**2025 Potential Release:** R19-45980 is a tentative SRKN-resistant release in 2025. It achieved great results in the ARVT with 100.5% of the test mean and placed 3rd out of 37 lines in the 2023 USDA Uniform Trials. Pre-foundation seed is being grown in Stuttgart, AR, and conversion to Enlist-E3® and XtendFlex® backgrounds are ongoing in off-season nurseries.**2024 Pre-commercial Stage**: Four high-yielding SRKN-resistant lines were advanced to pre-commercial stages. These lines continue to develop in our 2024 Pre-commercial Yield Trials, the 2024 USDA Southern Uniform Yield Trials, and the 2024 Arkansas Variety Testing, covering a total of 30 environments (Table 1). Additionally, the introgression of herbicide technology traits Enlist-E3® and XtendFlex® continues in our winter nursery. These advancements will offer farmers SRKN-resistant lines with enhanced yield potential and flexibility in herbicide technologies. **Table 1.** SRKN-resistant lines advanced to pre-commercial stages

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| Name | Pedigree | MG | 23 Yield | 23 % Xtend | 23 % E3 | 24 Season |
| R21KB-06852 | R15-7016/TN11-5140 | 5E | 67.1 | 93.1 | 94.4 | PCM / UP5 / VT / TI |
| R21KB-03657 | R13-13997/NCC09-200719-1-37 | 5E | 67.7 | 94.0 | 95.4 | PCM / UP5 / VT / TI |
| R21KB-05522 | R16-141/R13-13997 | 5E | 66.2 | 101.4 | 101.7 | PCM / UP5 / VT / TI |
| R19-45980 | Ellis/R04-357 | 5E | 67.9 | 95.7 | 91.1 | PCM / UT5 / VT / Pre-F / TI |

**2024 Finals Stage:** Lines R22KB-16609 and R22KB-02812, which possess the SRKN-resistant trait, are progressing through our 2024 final yield trials at Marianna, Pine Tree, Rohwer, Stuttgart, and Fisk, MO locations. These lines have also undergone evaluation in our internal flood tests during the V1 and R2 stages. Furthermore, we continue working on the integration of herbicide resistance traits (Enlist-E3® and XtendFlex®) into R22KB-16609.**2024 Preliminary Stage**: Evaluation of 1,176 lines is ongoing at Marianna, Pine Tree, and Rohwer replicated preliminary trials. Rogueing to remove off-types based on flower color, pod characteristics, and pubescence is in progress and will continue through the harvest period. Additionally, tissue samples from all preliminary lines have been collected for genotyping using the Soy3KSNP chip. These samples are also being evaluated with a proprietary disease and abiotic stressors marker panel that assesses responses to 25 different stressors.**2024 Progeny rows and Generation Advancement**: Approximately 16,000 F4:5 progeny rows are continuing their growth development in Kibler, AR. Breeder selections will begin by the end of September, focusing on overall plant architecture, maturity, and pod load. Additionally, 90 SRKN populations with at least one SRKN-resistant parent are progressing through generation advancement in the winter nursery.**2024 Crossing Block:** Sixteen SRKN-resistant parents were included in the 2024 crossing block, resulting in 87 new bi-parental populations with SRKN resistance. In addition, these populations carry desirable alleles for multiple biotic and abiotic stressors traits, including flooding and drought tolerance, cyst nematode resistance, stem canker resistance, among others.**2024 Seed Purity and Increase:** All lines evaluated at various stages of the breeding pipeline are grown in seed increase and purity plots in Fayetteville, AR. These plots undergo a rigorous rogueing process to remove off-types based on morphological traits such as flower color, growth habit, pod color, pubescence, and maturity. Rogueing will continue throughout the season, with maturity notes being recorded biweekly. |

**University of Missouri, Feng Lin:**  Currently, or projects listed below are in the field and awaiting harvest. Promising lines in the regional test: In total, we sent for RKN phenotype screening 25 lines present in the USDA Southern Regional Testes. From these, 3 promising lines, maturity group 4, carry the RKN gene of Resistance to Root-Knot nematode. These lines are listed in the table below. **Table 1:** Advanced lines in the Southern Regional Testes – USDA for yield test.

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| Line | MG |
| S21-11972HP | 4-L |
| S21-17588LL55 | 4-M |
| S21-22067 | 4-E |

AYT Lines: In 2024, a total of 100 lines are in Advanced yield trials (AYT). These lines were submitted for phenotypic screening, to be performed during the growing season. These lines were also analyzed using the CORTEVA panel and University of Missouri molecular markers. Out of the 100 lines, 34 showed the presence of the RKN resistance gene on chromosome 10 (more than 30%).Progeny Rows: We have about 100 bi-parental populations derived from nematode-resistant pedigrees. These lines will be planted in the next weeks, according to the weather conditions, in Portageville and compared to widely grown commercial XtendFlex checks. These lines will be evaluated based on yield potential and other agronomic traits. Breeding population advancement: In 2023, Two crosses were made with G11-7013 (elite line with a gene from PI96354) x University of Missouri RKN resistant lines. These crosses are listed in the table below. In addition, another 80 crosses were made using at least one source of Resistance to Nematode. These crosses will return in 2025 as progeny rows. |