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| Project Number:  |  |
| 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 |
| Project Lead Name: | Travis Faske |
| Reporting Period:*Please select the appropriate reporting period for this report.* |  [x]  December [ ]  March [ ]  June [ ]  September [ ]  Final |
| The information included in this detailed report should reflect quantifiable results that can be used to evaluate and measure project success.If Progress Report – What key activities were undertaken and what were the key accomplishments during this reporting period? List each key deliverable from the proposal and describe progress made (or not made) toward achieving it, including metrics were appropriate.If Final Report – What were the key accomplishments during the life of the project? List each deliverable from the proposal and describe progress made (or not made) toward achieving it, including metrics where appropriate. |
| Objective 1. Characterization of the mechanism of resistance to SRKN. (Faske and Watson; Univ of Arkansas and LSU, respectively)Two greenhouse experiments were conducted at the Lonoke Extension Center to evaluate the reproduction and galling in the PI lines that are being evaluated in this study. A lower percentage of root system galled was observed with Forrest, PI 56751C and PI 567305 than PI 438489, NIL of PI 438489B and Magellan. In contrast, reproduction was lower on PI 438489, NIL of PI 438489B, and Forrest than Magellan and PI 567305. These data indicate that although PI 567305 has low galling it also has a lot of reproduction, which would not be a good selection as a breeding line to reduce nematode reproduction. These data emphasize the importance of evaluating lines for both galling and reproduction before using them in a breeding program. Objective 2: Genetic characterization and development of functional markers for new sources of resistance to SRKN. (Nguyen, Univ. of Missouri)Seed increase: we harvested the seeds of major RKN lines such PI 438489B, PI 567305, Forrest, PI 567516C, Magellan, and Recombinant inbred lines for Chr. 10 and 13 at the University of Missouri & Arkansas to share with collaborators for genotyping and SRKN phenotyping.Marker assisted selection: Developed six markers from Chr. 13 QTL (from PI 567516C) that can distinguish between both resistant (PI 567516C) and susceptible (Magellan) parents and currently these markers are being tested for other important RKN lines and different sources of RKN resistance. ​Obj. 3: Development of breeding populations and MG4 soybean varieties with resistance to SRKN. Caio Vieira, Univ of ArkansasDecember 15, 2024 – Yield trials were completed across five locations, with data on maturity, yield, and breeder’s notes analyzed. Molecular marker analysis identified SRKN-resistant lines with strong yield performance for advancement. Genotyping data is being processed for genomic prediction. Hybrid F1 seeds from the 2025 crosses were sent to the winter nursery for generation advancement. The December selections meeting finalized lines for the 2025 trials and testing locations and the SRKN-MSSB Meeting in Fayetteville, AR, reviewed progress and set objectives for 2025.2025 Potential Release: R19-45980 is a tentative SRKN-resistant release in 2025. It performed consistently well and achieved great results in the 2023 and 2024 ARVT, with 100.5% and 97% of the test mean, respectively. Additionally, it placed 3rd out of 37 lines in the 2023 USDA Uniform Trials. Pre-foundation was harvested in Stuttgart, AR, and conversion to Enlist-E3® and XtendFlex® backgrounds is ongoing in off-season nurseries.2025 Pre-commercial Stage: Line R21KB-05522 is a promising MG 5E with the SRKN-resistant trait. In our internal tests, yield performance was 102% and 99% of the check mean during the 2023 and 2024 seasons, respectively. Additionally, R21KB-05522 yielded 88% of the adjusted means in the OVT, while yielding 90.3% of the check mean in the USDA uniform preliminary trials. In 2025, this line will be evaluated in our internal pre-commercial, OVT, and USDA uniform trials. In addition, line R22KB-16609, possessing the SRKN-resistant trait, was advanced from the final to the pre-commercial stage to be evaluated in the 2025 growing season. Finally, integration of the herbicide-resistance traits (Enlist-E3® and XtendFlex®) into R22KB-16609, continues in winter nursery.2025 Final Stage: Ten lines posing the SRKN-resistant trait with promising yield performance in the 2024 preliminary yield trials were advanced to be tested during the 2025 final (AF) yield trials. It is noteworthy that line R24PR-00010E has both the SRKN-resistant trait as well as the herbicide-resistant trait Enlist-E3® (Table 1).Table 1. Yield performance of AR elite lines possessing the SRKN-resistant trait selected for 2025 advancement.

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| Genotype | MG | % Check 2024 | 2025 Plan | Traits |
| R23KB-03866 | 3L | 118 | AF | SRKN |
| R23KB-03901 | 3L | 11 | AF | SRKN |
| R23KB-03934 | 3L | 109 | AF | SRKN |
| R23KB-03985 | 3L | 94 | AF | SRKN |
| R23KB-07416 | 3L | 95 | AF | SRKN |
| R23KB-10870 | 5E | 103 | AF | SRKN |
| R23KB-10229 | 5E | 100 | AF | SRKN |
| R23KB-10441 | 5E | 101 | AF | SRKN |
| R24PR-00010E | 4L | 91 | AF | SRKN / Enlist-E3® |
| R23KB-09628 | 5E | 95 | AF | SRKN |

2025 Generation Advancement: Seventy-nine populations, with at least one parent having the SRKN-resistant trait, were obtained from the 2024 crossing block. In total, 1,006 F1 seeds were planted on Nov 15th in the Puerto Rico off-season nursery and will be advanced until reaching the F4 stage. Additionally, these populations carry desirable alleles for multiple biotic and abiotic stressor traits, including flooding and drought tolerance, cyst nematode resistance, and stem canker resistance. Simultaneously, materials in the F2 and F3 stages continue development in the off-season nursery. 2025 Progeny rows: Around 124 different populations will be entered into the 2025 progeny rows; of these, 47 populations have at least one SRKN-resistant parent and will be evaluated in Stuttgart, Arkansas, during the upcoming season.2025 Crossing Block: Marker data and transfer agreements are underway to obtain new genetic material with the SRKN trait. Similarly, SoySNP3K data is being processed, and crossing simulations are underway to obtain better yielding and adapted lines using historical data. A final number of combinations will be presented in the next quarterly report. 2025 Introducing New Genetic Diversity: Ten lines predicted to exhibit low galling scores, despite lacking the resistance allele on chromosome 10, were previously selected using genomic models. Seed increases for these lines were successfully grown and harvested in Stuttgart, AR. Concurrently, Dr. Faske evaluated these lines for low galling during the 2024 season. Preliminary results identify four promising lines with low galling scores, consistent with genomic predictions regarding the absence of the chromosome 10 resistance allele. Additionally, a second year of SRKN screening will be conducted under greenhouse and field conditions by Dr. Faske at Hope, AR, to validate these results. Also, a new SRKN screening protocol is being developed at the Fayetteville campus to accelerate the identification of resistant materials and increase genetic diversity. Report December 15 – Feng Lin1. Screening for Resistance: A total of 96 AYT lines were sent to the University of Georgia for galling score analysis. Out of these, 26 lines displayed a low galling rate (1 to 2) on a 1-5 scale compared to the checks. Notably, among these 26 lines, 23 exhibited the presence of a marker on chromosome 10 associated with resistance to SRKN. We also collected soil samples in Clarkton from an area with highly infected sandy soil for screening trials in 2025. The density of SRKN nematodes ranged from 315 to 4,140 nematodes per 100 cc of soil. Soil samples with more than 300 nematodes per 100 cc are considered highly infected and suitable for analysis.
2. 2024 Crosses: We attempt 88 crosses carrying at least one parent with RKN resistance genes in the 2024 season. The crosses were planned for planting on three dates (approximately every 15 days) to increase the opportunities and likelihood of success. Our field was unfortunately affected by a flood, resulting in some unsuccessful crosses. For these, the parents were sent for another attempt of cross in the winter nursery.
3. Breeding population advancement: The successful crosses were sent to the winter nursery to advance generation. We expect these crosses to return in April 2026 to be planted as progeny rows and evaluated for agronomic traits and yield potential.
4. Progeny Rows: We will receive in 2025 more than 160 populations crossed in 2023. At least 30% of these populations have at least one parent with resistance to SRKN.
5. Preliminary yield trials: A total of 1500 progeny rows were selected to be tested in the preliminary yield trials in 2025 based on the agronomic performance and yield potential. From these, most have an SRKN-resistant parent.
6. Advanced yield trials: We are currently analyzing the data from the PYT to advance lines to AYT 2025 trials. After selecting the lines, we will send them for molecular marker screening to confirm resistance to SRKN (~100 lines).
7. USDA Trials: To compound the Preliminary Uniform trial for USDA (UP) in 2025, we selected from AYT 2024, 8 lines that have resistance to SRKN from maturity group 3L to 4L. These lines were tested for yield in 6 outside locations (IL, AR, MO, and TN) and 2 locations in the University farm (Clay and Loam soil). The data are shown below. For the UT data, we do not receive all the information needed to analyze and decide which lines will be included in the UT 2025 USDA trials.

Table 1: Yield in Bushel/acre and percentage (%) compared to the checks for the SRKN-resistant lines selected.

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|  |  |  |  | **Portageville, MO** | **Outside locations** |
| **Name** | **RM** | **MG** | **RKN** | **Bu/ac** | **% XT** | **% E** | **% C** | **% CK** | **Bu/ac** | **% XT** | **% E** | **% C** | **% CK** |
| S22PR-260E3 | 3.6 | 3L | R | 60.8 | 98 | 96 | 92 | 95 | 61.1 | 84 | 86 | 92 | 87 |
| S22PR-205E3 | 3.9 | 3L | R | 78.8 | 127 | 100 | 91 | 106 | 52.5 | 76 | 71 | 80 | 75 |
| S22-24366 | 4.3 | 4E | R | 71.6 | 115 | 91 | 83 | 96 | 69.3 | 100 | 93 | 105 | 99 |
| S22-16004 | 4.4 | 4E | R | 64.2 | 104 | 81 | 75 | 86 | 70.6 | 102 | 95 | 107 | 101 |
| S22-24401 | 4.4 | 4E | R | 59.7 | 96 | 75 | 69 | 80 | 70.5 | 101 | 95 | 107 | 101 |
| S22-24339 | 4.6 | 4L | R | 77.3 | 107 | 105 | 107 | 106 | 66.1 | 98 | 97 | 105 | 100 |
| S22-8718HP | 4.8 | 4L | R | 71.8 | 99 | 97 | 99 | 99 | 66.0 | 98 | 97 | 104 | 100 |
| S22-15639 | 4.6 | 4L | R | 63.2 | 87 | 86 | 87 | 87 | 53.6 | 80 | 79 | 85 | 81 |

\*Light green indicates a check yield of 95% or more, while dark green signifies a yield greater than 100%.Checks: XT: Xtend Flex; E: Enlist; C: Conventional.1. Releases: We are currently analyzing the data across the locations to decide the best lines to release in 2025. We have identified at least four promising lines with SRKN resistance in the Uniform trials that should be considered for release.
2. Lines in Winter Nursery for conversion: In addition to our conventional releases, we have several lines in Winter Nursery for conversion from conventional to the XtendFlex technology (Table 2). From these lines, four have RKN resistance confirmed by molecular markers. These lines will return next year, and their performance will be tested in yield trials.

Table 2: RKN lines in winter nursery for conversion to XtendFlex technology.

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| **Line** | **RKN** | **XtendFlex conversion** |
| S20-7901 | R | Yes |
| S21-2267 | R | Yes |
| S21-11972 | R | Yes |
| S19-10701 | R | Yes |

1. Publications

Argenta, J. *et al.* ‘S19-10701’: A semi-determinate maturity group IV conventional soybean cultivar with high yield and broad disease resistance. Under internal revision |