

NIFA SCRI Project 2023-05687
Annual Progress Report: September 2025–August 2026

Project Title: Development of Germplasm Resources and Molecular Breeding Tools to Combat Endemic and Emerging Diseases in U.S. Spinach Production

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Non-Technical Summary

Spinach (*Spinacia oleracea* L.) production is threatened by several diseases that can severely reduce crop yield, quality, and marketability. Developing disease-resistant cultivars is the most effective and sustainable approach to managing these diseases. However, conventional spinach breeding relies heavily on time-consuming phenotypic selection, which can delay cultivar development and release. Molecular breeding can accelerate this process by enabling breeders to identify and select plants carrying genes associated with disease resistance. This project builds upon the accomplishments of a previous NIFA-SCRI project (2017-51181-26830), which investigated resistance to three major spinach diseases: downy mildew (DM), white rust (WR), and Fusarium wilt (FW). That project established a foundation of genomic resources and molecular breeding tools, including single-nucleotide polymorphism (SNP) markers and genetic maps. The current project focuses on quantitative trait locus (QTL) mapping, molecular marker validation, and the integration of these tools into spinach breeding programs. It seeks to accelerate the development of cultivars resistant to DM, WR, FW, and two emerging diseases: Stemphylium leaf spot (SLS) and anthracnose leaf spot (ALS). Improved genome-enabled resources will be developed and used to introduce disease-resistance traits into cultivar-development pipelines. The project's long-term goal is to develop disease-resistant spinach cultivars using advanced genetic tools and selection methods. These resources will benefit public and private spinach breeders, growers, and the broader spinach industry. By integrating genetics, genomics, disease phenotyping, and innovative breeding methods, the project will help address current and emerging disease threats and support more sustainable spinach production. This project addresses two priority areas of the USDA Specialty Crop Research Initiative: (1) research in plant breeding, genetics, genomics, and other methods to improve crop characteristics; and (2) efforts to identify and address threats from pests and diseases, including emerging and invasive species.

Goals and Objectives

Spinach (*Spinacia oleracea* L.) is an economically important vegetable crop worldwide, with an estimated annual value of \$11.8 billion. The United States is the world's second-largest spinach producer and a leader in fresh-market spinach production, with an annual production value of approximately \$500 million. Increasing interest in healthy diets has contributed to sustained demand for spinach during the past three decades. Despite this growing demand, commercial spinach production is threatened by diseases that can substantially reduce yield and quality. Five particularly important diseases are downy mildew (DM), white rust (WR), Fusarium wilt (FW), Stemphylium leaf spot (SLS), and anthracnose leaf spot (ALS). The spinach industry requires the continued development of improved, locally adapted germplasm and cultivars with resistance to these diseases. The overarching goals of this project are to develop spinach germplasm resistant to these five diseases and to develop molecular tools for efficiently introducing disease-resistance traits into cultivar-development pipelines. The project's long-term goal is to accelerate the release of resistant spinach cultivars suitable for conventional and organic production while reducing reliance on fungicides.

The specific objectives are:

1. **Characterize and validate molecular markers for resistance to endemic and emerging spinach diseases.**

Hypothesis: Spinach germplasm collections contain genetically diverse sources of resistance to the five targeted diseases that can be used for cultivar development.

2. **Introgress disease resistance into spinach breeding lines for cultivar development.**

Hypothesis: Integrating molecular selection with disease phenotyping will accelerate spinach cultivar development.

3. **Develop a grower-oriented outreach program based on experiential learning and economic decision-support tools for spinach disease management.**

Hypothesis: Interactive and demonstrative educational approaches will increase the adoption of disease-management practices, reduce production risks, and improve profitability.

The project will identify genomic regions associated with disease resistance, develop and optimize practical molecular tools for selection, and generate resistant spinach breeding materials. Overall, these efforts will advance spinach breeding, improve genomic and phenotyping resources, generate new scientific knowledge, expand disease-resistant germplasm, and support sustainable and economically viable spinach production in the United States.

* **Target Audience** (8000 character limit)

Spinach Producers, Growers, and Seed Companies: Spinach producers, growers, and seed companies were engaged through field days, project meetings, presentations, publications, and other communication channels. Seed companies have used molecular markers associated with disease resistance to support their spinach breeding programs.

Scientists: Project outputs—including QTL mapping and association studies, SNP marker discoveries, peer-reviewed publications, abstracts, presentations, and reports—provide valuable resources for scientists and plant breeders. These contributions support continued advances in spinach genetics, genomics, disease resistance, and breeding technologies.

Students and Early-Career Researchers: Graduate students, postdoctoral researchers, research associates, and other trainees received instruction and practical experience in classical and molecular plant breeding. Training included crossing, population advancement, disease phenotyping, and cultivar development, as well as QTL mapping, genome-wide association studies (GWAS), genomic prediction, SNP discovery and genotyping, whole-genome sequencing (WGS), whole-genome resequencing (WGR), genotyping-by-sequencing (GBS), bioinformatics, marker-assisted selection (MAS), and genomic selection (GS).

Products (8000 character limit)

Peer-Reviewed Publications

2026

1. Alkabkabi, H., K. Chiwina, Y. Qu, R. Du, R. Khanal, Q. Luo, M.M. Ullah, H. Xiong, P. Rosas-Anderson, B. Mou, and A. Shi. 2026. Genome-wide association study and genomic prediction of bolting trait in spinach (*Spinacia oleracea* L.). *Frontiers in Plant Science* 17:1795277. <https://doi.org/10.3389/fpls.2026.1795277>
2. Feng, C., P.F. Fichoux, M.I. Villarroel-Zeballos, V.L. Castroagudin, H.V. Zima, K. Lamour, K.J. Clark, S.J. Klosterman, and J.C. Correll. 2026. Phenotypic and genotypic characterization of novel strains of the spinach downy mildew pathogen. *Plant Disease* 110(3):754–761. <https://doi.org/10.1094/PDIS-01-25-0064-RE>
3. Khuimphukhieo, I., J.C. Chavez, C.A. Avila, A. Shi, W. Ojeda, and J. Enciso. 2026. Integrating vegetation indices and texture features from UAS imagery to improve yield prediction and growth analysis of spinach (*Spinacia oleracea* L.). *International Journal of Agronomy* 2026:1411368. <https://doi.org/10.1155/ioa/1411368>
4. Liu, S.S., Clark, K. Anchiet, A., Ayala, H. Correll, J. C., Montazar, A., Putman, A. I., and Steven J. Klosterman. 2026. Estimation of airborne sporangia numbers of lettuce and spinach downy mildew pathogens in three California Valleys. 2026. *PhytoFrontiers* <https://doi.org/10.1094/PHYTOFR-04-25-0045-R>
5. Qu, Y., R. Du, K. Chiwina, H. Alkabkabi, M.M. Ullah, R. Khanal, Q. Luo, P. Rosas-Anderson, H. Xiong, C.A. Avila, B. Mou, and A. Shi. 2026. Genome-wide association study and genomic prediction of fruit weight in spinach. *Scientific Reports* 16:19430. <https://doi.org/10.1038/s41598-026-46903-1>
6. Srungarapu, R.S., A. Shi, and C.A. Avila. 2026. Genome-wide association and genomic prediction of anthracnose (*Colletotrichum dematium*) resistance in spinach. *The Plant Genome* 19(2):e70254. <https://doi.org/10.1002/tpg2.70254>

2025

7. Bhattarai, G., B. Liu, J. Correll, and A. Shi. 2025. Genome-wide association study and genomic prediction of leaf spot (*Stemphylium vesicarium*) resistance in a spinach diversity panel. *Frontiers in Plant Science* 16:1663650. <https://doi.org/10.3389/fpls.2025.1663650>

Manuscripts Submitted

8. Alkabkabi, H., I. Alatawi, H. Xiong, K. Chiwina, Q. Luo, Y. Qu, R. Du, R. Khanal, M.M. Ullah, P. Rosas-Anderson, B. Mou, K.-S. Ling, and A. Shi. 2026. Evaluation of seedling-stage salt tolerance in USDA spinach (*Spinacia oleracea* L.) accessions. Submitted to *HortScience*.
9. Gyawali, S., A. Shi, J.C. Correll, and L.J. du Toit. 2026. Genome-wide association study and genomic prediction of Fusarium wilt resistance in USDA spinach germplasm. Submitted to *Plant Pathology*.
10. Olaoye, D., G. Bhattarai, J. Correll, and A. Shi. 2026. Identification of novel genomic regions for downy mildew resistance and validation of predictive breeding in spinach (*Spinacia oleracea* L.). Submitted to *BMC Plant Biology*.

Manuscripts in Internal Review

11. Daba, S.D., S. Gyawali, A. Shi, and L.J. du Toit. 2026a. Screening cultivated (*Spinacia oleracea*) and wild (*Spinacia turkestanica*) spinach accessions for resistance to two races of *Fusarium oxysporum* f. sp. *spinaciae*. Intended for submission to *Plant Disease*; currently under internal review.
12. Daba, S.D., S. Gyawali, A. Shi, and L.J. du Toit. 2026b. Genome-wide association study and candidate gene identification for Fusarium wilt resistance in a panel of cultivated spinach (*Spinacia oleracea*) accessions. Intended for submission to *Molecular Breeding*; currently under internal review.

Manuscript in Preparation

13. Daba, S.D., M.I. Gordon, A. Shi, and L.J. du Toit. 2026c. Comparative transcriptome analysis of resistance in spinach (*Spinacia oleracea*) to race 1 of *Fusarium oxysporum* f. sp. *spinaciae*. Manuscript in preparation; journal to be determined.

Book

Correll, J.C., L.J. du Toit, M.I. Villarroel-Zeballos, B.S. Gerdeman, and T.W. Miller, eds. 2026. *Compendium of Spinach Diseases, Pests, and Disorders*. APS Press, St. Paul, Minnesota, USA. Online ISBN: 978-0-89054-710-6. [APS Press book page](#)

Book Chapter

Kik, C., B. Mou, G. Bhattarai, A. Shi, J.C. Correll, and L.J. du Toit. 2026. F₁ hybrid breeding and the genetics of disease and pest resistance. In J.C. Correll, L.J. du Toit, M.I. Villarroel-Zeballos, B.S. Gerdeman, and T.W. Miller, eds., *Compendium of Spinach Diseases, Pests, and Disorders*. APS Press, St. Paul, Minnesota, USA. <https://doi.org/10.1094/9780890547106.01.03.1>

Abstracts and Meeting Presentations

2026

1. Alkabkabi, H., I. Alatawi, H. Xiong, K. Chiwina, Y. Qu, R. Du, R. Khanal, M.M. Ullah, B. Mou, and A. Shi. 2026. Genome-wide study of salt tolerance in USDA spinach germplasm at the seedling stage. ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
2. Avila, C.A., R.S. Srungarapu, and A. Shi. 2026. Breeding for resistance to anthracnose (*Colletotrichum dematium*) in spinach. National Association of Plant Breeders Annual Meeting, College Station, Texas, USA, May 2026.
3. Chiwina, K., Y. Qu, H. Alkabkabi, M.M. Ullah, R. Khanal, B. Mou, R. Du, Q. Luo, and A. Shi. 2026. Genome-wide association study and genomic prediction of monoecy in spinach. ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
4. Daba, S.D., M.I. Gordon, A. Shi, and L.J. du Toit. 2026d. Searching for sources of resistance to two races of *Fusarium oxysporum* f. sp. *spinaciae*. Western Washington Seed Workshop, Mount Vernon, Washington, USA, January 9, 2026.
5. Daba, S.D., M.I. Gordon, S. Gyawali, A. Shi, and L.J. du Toit. 2026e. Putative candidate genes in *Spinacia oleracea* for resistance to races 1 and 2 of *Fusarium oxysporum* f. sp. *spinaciae*. Poster presented at Plant Health 2026, Providence, Rhode Island, USA, August 1–4, 2026.
6. Du, R., Y. Qu, H. Alkabkabi, K. Chiwina, R. Khanal, Q. Luo, M.M. Ullah, E. Wijnker, L. Vorstenbosch, R. van Treuren, C. Kik, L.J. du Toit, J. Correll, B. Mou, and A. Shi. 2026. Genome-wide association study of resistance to multiple races of the spinach downy mildew pathogen (*Peronospora effusa*). ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
7. Higgins, G. 2026. Winter spinach production and pests. Grow Food Boston. February 3, 2026.
8. Khanal, R., H. Alkabkabi, K. Chiwina, Y. Qu, R. Du, M.M. Ullah, M. Grusak, B. Mou, and A. Shi. 2026. Genome-wide association study and candidate gene identification for mineral accumulation in USDA spinach germplasm. ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.

9. Qu, Y., R. Du, K. Chiwina, H. Alkabkabi, M.M. Ullah, R. Khanal, Q. Luo, P. Rosas-Anderson, H. Xiong, C.A. Avila, B. Mou, and A. Shi. 2026. Genome-wide association study and candidate gene identification for fruit weight in USDA spinach germplasm. ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
10. Samy, B.A.A., D. Thompson, A. Shi, and V. Joshi. 2026. Genetic and physiological markers of heat resilience in spinach cultivated under organic field conditions. ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
11. Shakeel, M., Fallahi, M., Villarroel Zeballos, M. I., Zima, H. V., Remon, Rylie, and James C. Correll. 2026. Examination of Pythium species affecting commercial hydroponic spinach production. APS Meeting 2026.
12. Shi, A., J. Correll, G. Bhattarai, B. Liu, H. Xiong, D. Olaoye, Y. Qu, M.M. Ullah, R. Du, K. Chiwina, H. Alkabkabi, R. Khanal, L.J. du Toit, S. Gyawali, S.D. Daba, C.A. Avila, J.J. Rameneni, R.S. Srungarapu, J. Enciso, S. Zapata, L. Stein, V. Joshi, B. Analin, B. Mou, B. Poudel-Ward, C. Detranaltes, and S. Scheufele. 2026. Development of germplasm resources and molecular breeding tools to combat endemic and emerging diseases in U.S. spinach production. ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
13. Srungarapu, R.S., A. Shi, and C.A. Avila. 2026. Dissecting the genetic architecture of anthracnose (*Colletotrichum dematium*) resistance in spinach through genome-wide association studies and genomic prediction. SAES Annual Meeting, Mission, Texas, USA, February 2026.
14. Srungarapu, R.S., A. Shi, and C.A. Avila. 2026. Genome-wide association and genomic prediction of anthracnose (*Colletotrichum dematium*) resistance in spinach. Society for Experimental Biology Annual Conference, Florence, Italy, July 2026.
15. Srungarapu, R.S., K. Mandadi, and C.A. Avila. 2026. High vitamin C enhances salinity tolerance in spinach (*Spinacia oleracea* L.) through improved photosynthetic performance and stomatal regulation. ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
16. Srungarapu, R.S., A. Shi, and C.A. Avila. 2026. Integrating genome-wide association and genomic prediction to enhance anthracnose resistance (*Colletotrichum dematium*) in spinach (*Spinacia oleracea* L.). ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
17. Ullah, M.M., G. Bhattarai, M. Fallahi, A. Riaz, H. Alkabkabi, K. Chiwina, R. Khanal, Y. Qu, R. Du, P. Rosas-Anderson, B. Mou, J. Correll, and A. Shi. 2026. Temporal transcriptomic profiling reveals early defense mechanisms in spinach against downy mildew (*Peronospora effusa*). ASHS 2026 Annual Conference, Dallas, Texas, USA, August 3–7, 2026.
18. Xicay A. E., S.D. Zapata, M. Garcia, P. Omara. “*Simulation-Based Optimization for Decision Making Under Uncertain Production Environments*” Selected Paper. 2026 Agricultural and Applied Economics Association Annual Meeting. Kansas City, MO, July 27, 2026.
19. Xicay A. E., S.D. Zapata, M. Garcia, P. Omara. “*Simulation-Based Optimization for Decision Making Under Uncertain Production Environments*” Selected Paper. 2026 International Food and Agribusiness Management Association. Academic Symposium, Cork, Ireland, June 15, 2026.
20. Xicay A. E., S.D. Zapata. “*Simulation-Based Optimization for Decision Making Under Uncertain Production Environments*” Selected Paper. 2026 Symposium for Agricultural & Applied Economics Research. College Station, TX, March 20, 2026.
21. Xicay A. E., S.D. Zapata, M. Garcia, P. Omara. “*Simulation-Based Optimization Application to Farm Management Under Uncertainty*” Selected Paper. 2026 Southern Agricultural Economics Association Annual Meeting. Louisville, KY, February 03, 2026.

2025

22. Avila, C.A., R.S. Srungarapu, and A. Shi. 2025. Genome-wide association mapping and genomic prediction of anthracnose (*Colletotrichum dematium*) resistance in spinach (*Spinacia oleracea* L.). 11th International Spinach Conference, Monterey, California, USA, October 21–22, 2025.
23. Correll, J.C., C. Feng, M. Fallahi, M.I. Villarroel-Zeballos, B. Dhillon, A. Shi, H.V. Zima, K. Clark, S.J. Klosterman, and B. Irish. 2025. A 35-year perspective on downy mildew of spinach

caused by *Peronospora effusa*. 11th International Spinach Conference, Monterey, California, USA, October 21–22, 2025.

24. Daba, S.D., M.I. Gordon, S. Gyawali, A. Shi, and L.J. du Toit. 2025. Genome-wide association study of spinach accessions for resistance to two races of *Fusarium oxysporum* f. sp. *spinaciae*. 11th International Spinach Conference, Monterey, California, USA, October 21–22, 2025.
25. Fallahi, M., M.I. Villarroel-Zeballos, H.V. Zima, S.J. Klosterman, A. Shi, and J.C. Correll. 2025. Race diversity in the spinach downy mildew pathogen *Peronospora effusa*. 11th International Spinach Conference, Monterey, California, USA, October 21–22, 2025.
26. Mou, B., and A. Shi. 2025. Leafminer resistance in spinach. 11th International Spinach Conference, Monterey, California, USA, October 21–22, 2025.
27. Scheufele, S.B., G. Higgins, and A. Shi. 2025. Winter spinach production in New England. 11th International Spinach Conference, Monterey, California, USA, October 21–22, 2025.
28. Shi, A., J. Correll, G. Bhattarai, B. Liu, H. Xiong, Y. Qu, D. Olaoye, L.J. du Toit, S. Gyawali, S.D. Daba, C.A. Avila, J.J. Rameneni, R.S. Srungarapu, L. Stein, V. Joshi, B. Analin, and B. Mou. 2025. Genome-wide analysis of disease resistance, nutrients, and health-promoting compounds in spinach. 11th International Spinach Conference, Monterey, California, USA, October 21–22, 2025.

Other Products

No additional products were reported during this reporting period.

*Accomplishments (8000 character limit)

What was accomplished under these goals?

1. Characterize and Validate Molecular Markers for Resistance to Spinach Diseases

1.1 Downy Mildew

- Seventy spinach genotypes were evaluated in Arizona during the 2025–2026 winter season, and more than 20 genotypes exhibited resistance to downy mildew.
- A genome-wide association study (GWAS) of 243 spinach accessions evaluated for resistance to *Peronospora effusa* (*Pe*) race 5 identified 93 resistance-associated SNPs distributed across all six chromosomes. Candidate genes included *Spo12821* on chromosome 3 of the Sp75 reference genome and its ortholog, *SOV3g001250*, in the Monoe-Viroflay reference genome. Genomic prediction achieved predictive abilities of up to $r = 0.90$ across validation scenarios (Olaoye et al., 2026; submitted to *BMC Plant Biology*).
- A GWAS of 378 diverse spinach accessions from the Centre for Genetic Resources, the Netherlands (CGN), evaluated for resistance to 15 *P. effusa* races, identified a major resistance region on chromosome 3 and several race-specific loci on chromosomes 2, 4, and 6. Candidate resistance genes included *SOV3g001250*, *SOV3g000990*, *SOV3g000230*, and *SOV3g000290* (Du et al., 2026; ASHS Annual Conference).

1.2 White Rust

- Segregating populations derived from crosses between white rust-resistant and susceptible parental lines were evaluated for resistance to *Albugo occidentalis*. QTL mapping of white rust resistance is underway.

1.3 Fusarium Wilt

- A total of 379 spinach accessions were evaluated in 2025 for resistance to *Fusarium oxysporum* f. sp. *spinaciae* (*Fos*) races 1 and 2. Among these accessions, 52 were resistant to *Fos* race 1 and five were resistant to *Fos* race 2 (Daba et al., 2026a,d). GWAS identified genomic regions associated with resistance to both races, including loci near genes encoding protein kinases,

transcription factors, transporters, cell-wall biosynthesis proteins, and antioxidant-related proteins (Daba et al., 2026b,e).

- RNA sequencing of spinach accessions with contrasting responses to *Fos* races 1 and 2 identified candidate genes and molecular pathways associated with Fusarium wilt resistance (Daba et al., 2026c).

1.4 *Stemphylium* Leaf Spot

- A GWAS of 311 spinach genotypes identified four key SNP markers associated with resistance to *Stemphylium* leaf spot, caused by *Stemphylium vesicarium*. Genomic prediction achieved an accuracy of $r = 0.51$ (Bhattarai et al., 2025).

1.5 *Anthraco* Leaf Spot

- GWAS identified 20 SNPs associated with resistance to anthracnose leaf spot, caused by *Colletotrichum spinaciae*, including a major QTL explaining up to 66.8% of the phenotypic variance. Genomic prediction achieved an accuracy of up to $r = 0.92$, while prediction based on the 20 GWAS-derived SNPs achieved an accuracy of $r = 0.51$ (Srungarapu et al., 2026).

2. Introgress Disease Resistance into Spinach Breeding Lines

- Recurrent selection was used to combine multiple downy mildew resistance genes. Progeny from 34 crosses, together with resistant and susceptible checks, were evaluated in replicated field trials at the USDA-ARS Sam Farr United States Crop Improvement and Protection Research Center in Salinas, California.
- Approximately 650 spinach lines were screened at the Texas A&M AgriLife Research and Extension Center in Weslaco for disease resistance and desirable leaf characteristics.
- Approximately 200 new breeding lines were evaluated during the 2023–2026 winter seasons and advanced for further testing at the Alma research site in Arkansas.

3. Develop Outreach and Economic Tools for Growers

3.1 *Commercial Showcase Trials in Crystal City, Texas*

- Fifty-one commercial spinach varieties from global seed companies were evaluated for resistance to *Stemphylium* leaf spot from December 2025 through February 2026. Disease ratings and trial results were shared with growers and seed-industry representatives at the Texas Spinach Field Day in Crystal City on March 3, 2026.
- Thirty commercial spinach varieties were displayed and discussed during the March 3, 2026, field day.
- In a separate trial conducted from December 2024 through February 2025, 30 leading commercial varieties were evaluated for resistance to four diseases. ‘Baboon’ displayed no disease symptoms but had poor stand establishment. ‘Shearwater’, ‘PV-1569’, ‘Bonnethead’, and ‘Gerenuk’ were among the most promising varieties for *Stemphylium* leaf spot resistance.

3.2 *Industry and Stakeholder Engagement*

- The USDA-ARS leafy greens breeding program was presented at a joint Seed Central and California Leafy Greens Research Board event held at Hartnell College in Salinas, California, on June 25, 2026. The event was attended by more than 100 professionals, primarily representing seed companies.

3.3 *Economic Impact in Texas*

- Drought, disease pressure, and the closure of a processing facility created substantial challenges for the Texas spinach industry. Screening trials at Tiro Tres Farms identified varieties with improved disease tolerance and adaptation to local production conditions.
- Adoption of LifeGard reduced fungicide use by approximately 40%, lowering production costs and supporting the long-term sustainability of the Texas spinach industry.

3.4 Economic Analysis of White Rust Resistance

- Four white rust-management scenarios were evaluated using historical yield data, market information, and estimated production costs:
 1. **Baseline production:** 730.1 cartons per acre, with an estimated net return of \$1,738.60 per acre.
 2. **No disease treatment:** 291.6 cartons per acre, with an estimated net return of -\$1,597.34 per acre.
 3. **GH-107 resistant lines:** 687.6 cartons per acre, with an estimated net return of \$3,285.91 per acre.
 4. **GH-108 resistant lines:** 710.6 cartons per acre, with an estimated net return of \$3,517.50 per acre.
- Monte Carlo simulations using 1,000 iterations indicated that resistant lines significantly improved expected profitability, with the GH-108 lines providing the strongest economic performance.

4. Other Significant Research Findings

- Fruit size: GWAS identified six SNPs significantly associated with fruit weight, and genomic prediction (GP) achieved prediction accuracy of up to $r = 0.51$ using the six GWAS-derived SNPs (Qu et al., 2026).
- Salt tolerance: Ten accessions were identified as highly salt-tolerant. Three SNPs significantly associated with relative salt tolerance for chlorophyll (RST_C) were identified on chromosomes 2 and 6. GP achieved a prediction accuracy of $r = 0.55$ for salt tolerance.
- Bolting: GWAS identified seven significant loci distributed across chromosomes 2, 4, and 6, including a major-effect locus on chromosome 6 (~13.54 Mb). GP achieved a prediction accuracy of approximately $r = 0.39$ for bolting in spinach.
- *P. effusa* strains: Three new races and 18 novel strains of *P. effusa* were identified through pathogenicity evaluations. In addition, 70 commercial spinach cultivars were evaluated for their reactions to selected races and novel strains, providing valuable information for cultivar selection and resistance breeding (Feng et al., 2026).
- Breeding: Approximately 100 spinach breeding lines were evaluated and harvested during the 2025–2026 winter season at the Alma Research Site in Arkansas to support continued germplasm improvement.

What opportunities for training and professional development has the project provided? (8000 character limit)

1. Personnel Training and Professional Development

- One master's level graduate student assisted Dr. Zapata in developing a simulation-based optimization framework for identifying optimal spinach management strategies under uncertainty.
- Two postdoctoral research associates received project-related training. One joined Dr. Carlos Avila's program at Texas A&M AgriLife in January 2025, and Dr. Sintayehu Daba has worked in Dr. Lindsey du Toit's program since 2024.
- A postdoctoral research associate and a research scientist in Dr. Avila's program received training in molecular breeding, spinach disease phenotyping, data analysis, scientific presentations, and manuscript preparation.
- Dr. Modan Das, a postdoctoral research associate, and an undergraduate student joined Dr. Beiquan Mou's USDA-ARS program in Salinas, California, in 2025 and received training in spinach breeding and research methods.

- A research associate who joined Dr. Ainong Shi's laboratory in June 2024 and a Ph.D. student who joined the project in January 2025 received training in spinach breeding, disease evaluation, molecular-marker analysis, data analysis, and dissemination of research results.

2. Undergraduate Training

- Three undergraduate students completed 15-week professional internships at Texas A&M AgriLife in Weslaco:
 - One student, supervised by Dr. Samuel Zapata, developed a stochastic framework for evaluating the economic value of new spinach cultivars.
 - Two students, supervised by Dr. Avila, received training in DNA isolation, polymerase chain reaction (PCR), marker-assisted selection, and conventional plant breeding.
- An undergraduate student from the University of Texas Rio Grande Valley completed a 10-week summer internship in Dr. Avila's laboratory through the USDA-funded PANACEAS Research and Extension Experiences for Undergraduates program. The student received training in produce-quality evaluation, seed collection, conventional and molecular disease identification, PCR-based diagnostics, and marker-assisted selection.
- Two undergraduate students completed summer internships in Dr. Avila's program at Texas A&M AgriLife in 2026. They received hands-on training in crop maintenance, harvesting, phenotypic evaluation, and field-data collection.
- An undergraduate student in Dr. Mou's USDA-ARS program received training in spinach research and field and laboratory methods.

3. Graduate Education and Course Instruction

- A special-topics course in Molecular Plant Breeding was offered in Fall 2026 (<http://spinachdb.com/MB/>). Twelve students received training in molecular-marker development, genetic analysis, and the application of molecular tools in plant breeding.
- The project provided hands-on training to graduate students in experimental design, spinach disease phenotyping, molecular breeding, data analysis, and scientific communication.

4. Extension and Broader Professional Development

- Genevieve Higgins, working with Susan Scheufele's program, delivered one professional-development presentation to agricultural service providers.
- Postdoctoral researchers, research staff, graduate students, and undergraduate interns gained experience in presenting research findings, preparing manuscripts, collaborating across institutions, and communicating project results to growers and seed-industry stakeholders.

How have the results been disseminated to communities of interest? (8000 character limit)

During the reporting period of September 15, 2025–August 31, 2026, project results were disseminated through field days, demonstration trials, diagnostic services, scientific and industry meetings, Extension programs, publications, online resources, and direct communication with growers and seed companies.

1. Field Days and Demonstration Trials

- **The 11th International Spinach Conference** was held in Monterey, California, on October 21, 2025. Presentations covered spinach production, breeding, molecular biology, disease and pest management, food safety, marketing, and future industry challenges. Dr. Jim Correll led and chaired the conference, while Dr. Ainong Shi, Dr. BeiQuan Mou, Dr. Larry Stein, Dr. Lindsey du Toit, and Susan Scheufele contributed as authors and presenters. Additional information is available through the conference meeting portal. <https://spinach.uark.edu/salinas-ca-international-spinach-conference-meeting-portal/>

- The associated **International Spinach Conference Field Day** was held in the Salinas Valley on October 22, 2025. Participants toured research and commercial fields and reviewed cultivar performance, disease resistance, and spinach production practices.
<https://ucanr.edu/blog/salinas-valley-agriculture/article/registration-open-11th-international-spinach-conference>
- The **Yuma Spinach Field Day** was held at the Yuma Agricultural Center on February 17, 2026. Dr. Correll and collaborators presented results from commercial cultivar trials and downy mildew resistance evaluations. Participants received updates on disease monitoring, cultivar performance, and the use of genetic resistance to manage *Peronospora effusa*.
<https://ucanr.edu/blog/salinas-valley-agriculture/article/yuma-spinach-field-day-will-take-place-february-17-2026>
- The **Texas Spinach Industry Field Day** was held at Tiro Tres Farms in Crystal City on March 3, 2026. Approximately 70–75 growers, researchers, Extension personnel, seed-company representatives, and other stakeholders attended. Presentations and field tours addressed spinach breeding, fungicide trials, seedling diseases, white rust, *Stemphylium* leaf spot, commercial cultivar evaluations, and SCRI project activities.
- Fifty-two commercial spinach cultivars were evaluated at the Texas site for resistance to *Stemphylium vesicarium*. ‘Diamondback’ had the lowest disease severity, followed by ‘LSPH-0299’, ‘Boomslang’, ‘Bonnethead’, ‘Gerenuk’, ‘KSP0141’, ‘Arakas’, ‘Ovenbird’, and ‘NUN 07575 SPS’. These findings were shared with growers, seed companies, and other stakeholders.
- A **Downy Mildew Field Day** was held at the Yuma Agricultural Center on March 12, 2026. Field demonstrations addressed fungicide efficacy, resistance management, disease diagnostics, and genetic resistance to downy mildew in spinach and other leafy vegetables. Dr. Correll’s adjacent spinach cultivar trials were also available for observation.
<https://acis.cals.arizona.edu/agricultural-ipm/vegetables/vipm-archive/vipm-plant-view/march-2026-downy-mildew-field-day>

2. Plant Diagnostic and Disease-Monitoring Services

The Yuma County Cooperative Extension Plant Diagnostic Clinic provided direct diagnostic assistance to spinach growers. During the reporting period, 37 independent spinach cases were diagnosed using samples submitted to the clinic or collected from symptomatic fields.

Major problems included *Stemphylium* and *Cladosporium* leaf spots, *Pythium* and *Rhizoctonia* root rots, stubby-root nematodes, spinach mites, and abiotic disorders such as herbicide injury, sandblasting, and nutrient deficiencies. Diagnostic findings and management recommendations were communicated directly to growers and pest control advisors.

The project also continued to monitor spinach downy mildew populations in Arizona, California, Texas, and other production regions. Low downy mildew pressure was reported in southwestern and coastal California, and this information was shared through field days, diagnostic activities, and direct communication with growers and industry representatives.

3. Cultivar Evaluation and Fungicide Trials

Nine fungicide treatments were evaluated for control of spinach downy mildew on the F₁ hybrid ‘Banjo’ at the Yuma Agricultural Center from January through March 2026. However, downy mildew did not develop in the treatment plots or adjacent cultivar trials, preventing comparisons of treatment efficacy. Sentinel field trials were conducted in Yuma, Arizona, and the Salinas Valley, California, to evaluate commercial spinach cultivars under naturally occurring downy mildew pressure. These trials provided growers and seed companies with current information about cultivar performance and disease resistance.

4. Scientific and Industry Presentations

- Dr. Sintayehu Daba presented GWAS results for resistance to two races of *Fusarium oxysporum* f. sp. *spinaciae* at the 11th International Spinach Conference in October 2025.

- Dr. Daba presented an update on Fusarium wilt research at the Western Washington Seed Workshop in Mount Vernon, Washington, on January 9, 2026.
- Dr. Daba presented a poster on candidate genes associated with resistance to races 1 and 2 of *F. oxysporum* f. sp. *spinaciae* at Plant Health 2026 in Providence, Rhode Island, on August 1–4, 2026.
- Project scientists delivered multiple presentations at the ASHS 2026 Annual Conference in Dallas, Texas, on August 3–7, 2026. Topics included downy mildew resistance, disease-related transcriptomics, disease-resistance breeding, salt tolerance, mineral accumulation, monoecy, and fruit weight.
- The USDA-ARS leafy greens breeding program was presented at a joint Seed Central and California Leafy Greens Research Board event held at Hartnell College in Salinas, California, in June 2026. The event reached more than 100 seed-industry professionals and other stakeholders.

5. Extension and Industry Collaboration

The UMass Extension team disseminated spinach production and disease-management recommendations through *Vegetable Notes*, grower meetings, and online and in-person educational programs. These activities reached 439 individuals during the reporting period.

Project scientists collaborated with the International Working Group on *Peronospora* in Spinach, seed companies, Naktuinbouw, Plantum, universities, and Extension specialists to characterize novel *P. effusa* isolates, maintain standard spinach race differentials, and coordinate downy mildew monitoring.

<https://worldseed.org/our-work/disease-resistance/other-initiatives/>

6. Publications and Other Scientific Communication

During the reporting period, project investigators published six peer-reviewed journal articles and submitted three additional manuscripts. These publications communicated advances in spinach disease resistance, molecular-marker development, genomic prediction, high-throughput phenotyping, and cultivar improvement.

Project investigators also delivered abstracts, posters, and oral presentations at international, national, and regional meetings, including the 11th International Spinach Conference, Plant Health 2026, the ASHS 2026 Annual Conference, and the Western Washington Seed Workshop. Major topics included:

- Resistance to downy mildew, Fusarium wilt, Stemphylium leaf spot, and anthracnose;
- Genomic prediction and marker-assisted breeding;
- Transcriptomic responses to disease infection;
- Salt tolerance and mineral accumulation;
- Spinach monoecy and fruit weight; and
- Development of improved spinach germplasm and molecular breeding tools.

Project results were further distributed through Extension publications, project reports, conference abstracts, posters, oral presentations, online resources, grower meetings, field tours, and direct communication with seed companies and other industry partners.

What do you plan to do during the next reporting period to accomplish the goals? (8000 character limit)

During the next reporting period, the project team will continue field and controlled-environment disease evaluations, genetic and genomic analyses, development of disease-resistant spinach lines, stakeholder engagement, and dissemination of research results.

1. Field Evaluations

Spinach germplasm, commercial cultivars, breeding populations, and advanced breeding lines will be evaluated under field conditions for:

- Downy mildew resistance in Yuma, Arizona, and Salinas, California; and

- White rust, Stemphylium leaf spot, and anthracnose leaf spot resistance in Crystal City, Texas. These trials will identify resistant materials and validate their disease-resistance performance across environments.

2. Greenhouse and Growth-Chamber Evaluations

Controlled-environment disease screening will include:

- Downy mildew resistance at the University of Arkansas;
- Anthracnose leaf spot resistance at Texas A&M AgriLife; and
- Fusarium wilt resistance to races 1 and 2 at Washington State University.

Fusarium wilt transcriptomic studies will continue to identify genes, regulatory pathways, and biological processes associated with resistance to the two pathogen races.

3. Genetics of Disease Resistance

QTL mapping, GWAS, and genomic prediction will be conducted to investigate resistance to five major spinach diseases:

- Anthracnose leaf spot;
- Downy mildew;
- Fusarium wilt;
- Stemphylium leaf spot; and
- White rust.

The project will identify and validate SNP markers, candidate genes, and genomic regions associated with disease resistance. Promising markers will be evaluated for use in marker-assisted selection, while genomic prediction models will be assessed for their application in genomic selection.

4. Development of Disease-Resistant Spinach Lines

Breeding, selection, seed increase, and advancement of disease-resistant spinach lines will continue at participating locations:

- **Downy mildew resistance:** USDA-ARS in Salinas, California, and the University of Arkansas in Fayetteville;
- **Anthracnose leaf spot, Stemphylium leaf spot, and white rust resistance:** University of Arkansas and Texas A&M AgriLife Research and Extension Center in Weslaco; and
- **Fusarium wilt resistance:** Washington State University and the University of Arkansas.

Promising plants and breeding lines will be selected for additional disease screening, seed production, marker validation, and advancement into subsequent breeding cycles.

5. Outreach and Stakeholder Engagement

Spinach field days will be organized in Crystal City, Texas; Salinas, California; and Yuma, Arizona. These events will provide growers, seed companies, pest control advisors, Extension personnel, and researchers with opportunities to:

- Observe commercial cultivars and experimental breeding lines;
- Review disease-resistance and fungicide trials;
- Learn about newly developed molecular markers and breeding tools; and
- Provide feedback on industry needs and project priorities.

The project will continue collaborating with seed companies, Extension programs, and the International Working Group on *Peronospora* in Spinach to monitor emerging downy mildew races and communicate findings to stakeholders.

6. Publications and Conferences

The project team plans to submit at least five manuscripts to peer-reviewed journals and deliver more than 10 presentations at international, national, and regional conferences, workshops, stakeholder meetings, and field days.

Research findings will also be disseminated through Extension publications, project reports, online resources, grower meetings, field tours, and direct communication with seed-industry partners.

Changes/Problems

Describe major changes/problems in approach and reason(s) for these major changes. If applicable, provide special and/or additional reporting requirements specified in the award Terms and Conditions.

None.

Estimated Project FTEs for The Project Duration

Role	Non-Students or Faculty	Students with Staffing Roles			Computed Total by Role
		Undergraduate	Graduate	Post-Doctorate	
Scientist	6	3	2	2	13
Professional	2				2
Technical	2				2
Administrative					
Other					
Computed Total					
	10	3	2	2	17

Activities

Research	80 %
Extension	10 %
Education	10 %

Research Effort Categories

Basic	20 %
Applied	60 %
Developmental	20 %

Classification

Knowledge Area (KA)	Subject of Investigation (SOI)	Field of Science (FOS)	Percent
201	1430	1080	25
202	1430	1080	25

Knowledge Area (KA)	Subject of Investigation (SOI)	Field of Science (FOS)	Percent
201	1430	1081	25
202	1430	1081	25

Knowledge Area

201 - Plant Genome, Genetics, and Genetic Mechanisms; 202 - Plant Genetic Resources

Subject Of Investigation

1430 - Greens and leafy vegetables (includes endive, lettuce, spinach, turnip-greens, celery, rhubarb, parsley, asparagus)

Field Of Science

1080 - Genetics; 1081 – Breeding