

**U. S. DEPARTMENT OF ENERGY, OFFICE OF SCIENCE
INTEGRATED SUPPORT CENTER—CHICAGO OFFICE**

**NATIONAL ENVIRONMENTAL POLICY ACT (NEPA)
ENVIRONMENTAL EVALUATION NOTIFICATION FORM**

To be completed by "Applicant," i.e., organization with responsibilities for a "Federal action" involving application to DOE for a permit, license, exemption or allocation, or other similar actions. For assistance with this Form, refer to "Instructions for Preparing ISC-CH F-560, Environmental Evaluation Notification Form."

Solicitation/Award No. (if applicable): DE-SC0023305

Organization Name: Donald Danforth Plant Science Center

Proposed Action Title: Harnessing regulatory variation to elucidate drought resilience mechanisms in sorghum

Total DOE Funding/Total Funding: \$2,699,194.00

I. Project Description: (Use explanation pages if additional space is required)

A. Proposed Project/Action (if applicable, delineate Federally funded/Non-Federally funded portions)

This project investigates the innate drought resilience of sorghum [*Sorghum bicolor* (L.) Moench], a bioenergy feedstock and cereal crop. The overall project objective is to define and functionally characterize genes and pathways related to drought stress tolerance in sorghum and the molecular mechanisms by which these factors drive phenotypic diversity. This will accelerate the development of improved varieties that are high-yielding with limited water resources. Specific objectives include: 1) Leveraging a regulatory genomics framework to pin-point genetic variation that contributes to drought response across sorghum diversity, 2) Enhancing understanding of how genotype drives phenotype and environmental adaptation using high-resolution, field-based phenotyping of sorghum diversity and mutant collections, and 3) Experimentally validating predictions of gene function using molecular and genetic assays and targeted gene editing.

Please see continuation in explanation pages.

B. Would the project proceed without Federal funding? Yes No

If "yes," use explanation page.

II. Description of Affected Environment: (Use explanation pages if additional space is required)

There will be no affected environment in this project.

III. Preliminary Questions:

- | | Yes | No |
|---|--------------------------|-------------------------------------|
| A. <u>Is the DOE-funded work routinely administrative or <i>entirely</i> advisory or a “paper study?”</u> | <input type="checkbox"/> | <input checked="" type="checkbox"/> |

If “Yes”, ensure that the description in Section I reflects this and go directly to Section V.

- B. Is there any potential whatsoever for: (Provide an explanation for each “Yes” response)

- | | | |
|--|-------------------------------------|-------------------------------------|
| 1. Work to be performed outdoors? | <input checked="" type="checkbox"/> | <input type="checkbox"/> |
| 2. Major modification of a building interior? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 3. Threat of violation of applicable statutory, regulatory, or permit requirements for environment, safety, and health? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 4. Siting, construction or major expansion of waste treatment, storage, or disposal facilities? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 5. Disturbance to hazardous substances, pollutants, or contaminants preexisting in the environment? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 6. The presence of any environmentally-sensitive resources? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 7. Any potential whatsoever for high consequence impacts to human health or the environment? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 8. The work being connected to another existing/proposed activity that could potentially create a significant impact? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 9. Nearby past, present, and/or reasonably foreseeable future actions such that collectively significant impacts could result? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 10. Scientific or public controversy, uncertainty over potential impacts, or conflicts regarding resource usage? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |

If “No” to ALL Section III.B. questions, go directly to Section V.

IV. Potential Environmental Effects: (Provide an explanation for each “Yes” response)

- A. Environmentally Sensitive Resources: Could the proposed action potentially result in changes and/or disturbances to any of the following resources?

- | | Yes | No |
|--|--------------------------|-------------------------------------|
| 1. Threatened/Endangered Species and/or Critical Habitats | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 2. Other Protected Species (e.g., Burros, Migratory Birds, Pollinators) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 3. Sensitive Environments (e.g., Tundra/Coral Reefs/Rain Forests) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 4. Cultural or Historic Resources | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 5. Important Farmland | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 6. Non-Attainment Areas for Ambient Air Quality Standards | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 7. Class I Air Quality Control Region | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 8. Special Sources of Groundwater (e.g. Sole Source Aquifer) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 9. Navigable Air Space | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 10. Coastal Zones | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 11. Areas with Special National Designation (e.g. National Forests, Parks, Trails) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 12. Floodplains and/or Wetlands | <input type="checkbox"/> | <input checked="" type="checkbox"/> |

- B. Regulated Substances/Activities: Would the proposed action involve any of the following regulated Items or activities?

- | | | |
|--|--------------------------|-------------------------------------|
| 13. Natural Resource Damage Assessments | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 14. Invasive Species or Exotic Organisms | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 15. Noxious Weeds | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 16. Clearing or Excavation greater than one acre or Removal of Trees Governed by Local Requirement | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 17. Dredge or Fill (under Clean Water Act, Section 404, greater than one acre) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |

III. Preliminary Questions:

- | | Yes | No |
|---|--------------------------|-------------------------------------|
| A. <u>Is the DOE-funded work routinely administrative or <i>entirely</i> advisory or a “paper study?”</u> | <input type="checkbox"/> | <input checked="" type="checkbox"/> |

If “Yes”, ensure that the description in Section I reflects this and go directly to Section V.

- B. Is there any potential whatsoever for: (*Provide an explanation for each “Yes” response*)

- | | | |
|--|-------------------------------------|-------------------------------------|
| 1. Work to be performed outdoors? | <input checked="" type="checkbox"/> | <input type="checkbox"/> |
| 2. Major modification of a building interior? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 3. Threat of violation of applicable statutory, regulatory, or permit requirements for environment, safety, and health? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 4. Siting, construction or major expansion of waste treatment, storage, or disposal facilities? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 5. Disturbance to hazardous substances, pollutants, or contaminants preexisting in the environment? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 6. The presence of any environmentally-sensitive resources? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 7. Any potential whatsoever for high consequence impacts to human health or the environment? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 8. The work being connected to another existing/proposed activity that could potentially create a significant impact? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 9. Nearby past, present, and/or reasonably foreseeable future actions such that collectively significant impacts could result? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 10. Scientific or public controversy, uncertainty over potential impacts, or conflicts regarding resource usage? | <input type="checkbox"/> | <input checked="" type="checkbox"/> |

If “No” to ALL Section III.B. questions, go directly to Section V.

IV. Potential Environmental Effects: (*Provide an explanation for each “Yes” response*)

- A. Environmentally Sensitive Resources: Could the proposed action potentially result in changes and/or disturbances to any of the following resources?

- | | Yes | No |
|--|--------------------------|-------------------------------------|
| 1. Threatened/Endangered Species and/or Critical Habitats | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 2. Other Protected Species (e.g., Burros, Migratory Birds, Pollinators) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 3. Sensitive Environments (e.g., Tundra/Coral Reefs/Rain Forests) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 4. Cultural or Historic Resources | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 5. Important Farmland | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 6. Non-Attainment Areas for Ambient Air Quality Standards | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 7. Class I Air Quality Control Region | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 8. Special Sources of Groundwater (e.g. Sole Source Aquifer) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 9. Navigable Air Space | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 10. Coastal Zones | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 11. Areas with Special National Designation (e.g. National Forests, Parks, Trails) | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 12. Floodplains and/or Wetlands | <input type="checkbox"/> | <input checked="" type="checkbox"/> |

- B. Regulated Substances/Activities: Would the proposed action involve any of the following regulated items or activities?

- | | | |
|--|--------------------------|-------------------------------------|
| 13. Natural Resource Damage Assessments | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 14. Invasive Species or Exotic Organisms | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 15. Noxious Weeds | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 16. Clearing or Excavation greater than one acre or Removal of Trees Governed by Local Requirement | <input type="checkbox"/> | <input checked="" type="checkbox"/> |
| 17. Dredge or Fill (under Clean Water Act, Section 404, greater than one acre) | <input type="checkbox"/> | <input type="checkbox"/> |

B. Regulated Substances/Activities: Would the proposed action involve any of the following regulated Items or activities? (continued)

	Yes	No
18. Noise (in excess of regulations)	<input type="checkbox"/>	<input checked="" type="checkbox"/>
19. Asbestos Removal	<input type="checkbox"/>	<input checked="" type="checkbox"/>
20. Polychlorinated biphenyls (PCBs)	<input type="checkbox"/>	<input checked="" type="checkbox"/>
21. Import, Manufacture, or Processing of Toxic Substances	<input type="checkbox"/>	<input checked="" type="checkbox"/>
22. Chemical Storage/Use	<input type="checkbox"/>	<input checked="" type="checkbox"/>
23. Pesticide Use	<input type="checkbox"/>	<input checked="" type="checkbox"/>
24. Hazardous, Toxic, or Criteria Pollutant Air Emissions	<input type="checkbox"/>	<input checked="" type="checkbox"/>
25. Liquid Effluents	<input type="checkbox"/>	<input checked="" type="checkbox"/>
26. Spill Prevention/Surface Water Protection	<input type="checkbox"/>	<input checked="" type="checkbox"/>
27. Underground Injection	<input type="checkbox"/>	<input checked="" type="checkbox"/>
28. Hazardous Waste	<input type="checkbox"/>	<input checked="" type="checkbox"/>
29. Underground Storage Tanks	<input type="checkbox"/>	<input checked="" type="checkbox"/>
30. Radioactive or Radioactive Mixed Waste	<input type="checkbox"/>	<input checked="" type="checkbox"/>
31. Radiation Exposure	<input type="checkbox"/>	<input checked="" type="checkbox"/>
32. Nanoscale Materials	<input type="checkbox"/>	<input checked="" type="checkbox"/>
33. Genetically Engineered Microorganisms/Plants or Synthetic Biology	<input checked="" type="checkbox"/>	<input type="checkbox"/>
34. Ozone Depleting Substances	<input type="checkbox"/>	<input checked="" type="checkbox"/>
35. Greenhouse Gas Generation/Sustainability	<input type="checkbox"/>	<input checked="" type="checkbox"/>
36. Off-Road Vehicles	<input type="checkbox"/>	<input checked="" type="checkbox"/>
37. Biosafety Level 3-4 Laboratory	<input type="checkbox"/>	<input checked="" type="checkbox"/>
38. Research on Human Subjects or other Vertebrate Animals	<input type="checkbox"/>	<input checked="" type="checkbox"/>
39. Facility footprint exceeds 5,000 Square Feet	<input type="checkbox"/>	<input checked="" type="checkbox"/>

C. Other Relevant Information: Would the proposed action involve the following?

	Yes	No
40. Disproportionate Nearby Presence of Minority and/or Low Income Populations	<input type="checkbox"/>	<input checked="" type="checkbox"/>
41. Existing, Modified, or New Federal/State Permits	<input type="checkbox"/>	<input checked="" type="checkbox"/>
42. Involvement of Another Federal Agency (e.g. license/permit, funding, approval)	<input type="checkbox"/>	<input checked="" type="checkbox"/>
43. Action in a State with NEPA-type law	<input type="checkbox"/>	<input checked="" type="checkbox"/>
44. Expansion of Public Utilities/Services	<input type="checkbox"/>	<input checked="" type="checkbox"/>
45. Depletion of a Non-Renewable Resources	<input type="checkbox"/>	<input checked="" type="checkbox"/>
46. Subject to an Existing Institutional Work Planning and Control Process	<input type="checkbox"/>	<input checked="" type="checkbox"/>
47. Other Pertinent Information Which Could Impact Human Health or the Environment	<input type="checkbox"/>	<input checked="" type="checkbox"/>

V. Applicant certification that to the best of their knowledge all information provided on this form is accurate:

Does this disclosure contain: classified, sensitive business, or other exempt information that DOE would not be obligated to disclose pursuant to the Freedom of Information Act. Yes No

A. Organization Official (Name and Title): Melissa A. Kerckhoff, Sponsored Research Compliance Manager

Signature: Melissa A. Kerckhoff Digitally signed by Melissa A. Kerckhoff
Date: 2022.09.22 01:53:44 -05'00' Date: September 22, 2022

e-mail: grantsadmin@danforthcenter.org Phone: 314-587-1285

B. Optional Secondary Approval (Name and Title): _____

Signature: _____ Date: _____

e-mail: _____ Phone: _____

Remainder to be completed by DOE

VI. DOE Concurrence/Recommendation/Determination:

A. DOE Project Director/Program Manager or Contract/Grant Management Specialist:

	Yes	No
Has the Applicant completed this Form correctly?	<input checked="" type="checkbox"/>	<input type="checkbox"/>
Does an existing generic categorical exclusion apply?	<input type="checkbox"/>	<input checked="" type="checkbox"/>

If yes, indicate: _____

Name and Title: Daniella Duverne, Contract Specialist

Signature: DANIELLA DUVERNE Digitally signed by DANIELLA DUVERNE
Date: 2022.09.22 13:21:53 -05'00' Date: _____

B. DOE NEPA Team Review (if requested):

	Yes	No
Is the class of action identified in the DOE NEPA Regulations (Appendices A-D to Subpart D (10 CFR § 1021))?	<input checked="" type="checkbox"/>	<input type="checkbox"/>

If yes, specify the class(es) of action: B3.6, B3.8

Name and Title: _____

Signature: _____ Date: _____

C. DOE Counsel (if requested):

Name and Title: _____

Signature: _____ Date: _____

D. DOE NEPA Compliance Officer:

The preceding pages are a record of documentation required under DOE Final NEPA Regulation, 10 CFR § 1021.410.

- Action may be categorically excluded from further NEPA review. I have determined that the proposed action meets the requirements for Categorical Exclusion referenced above.
- Action requires approval by Head of the Field Organization. Recommend preparation of an Environmental Assessment.
- Action requires approval by Head of the Field Organization or a Secretarial Officer. Recommend preparation of an Environmental Impact Statement.

Comments/limitations if any:

NEPA Compliance Officer:

Name: PETER SIEBACH Digitally signed by PETER SIEBACH
Date: 2022.09.22 13:49:41 -05'00'

Signature: _____ Date: _____

Optional Additional Narrative: (add additional detail to description to Sections I and II or explanations to responses in Sections 3 and 4.

Additional narrative from 1.A:

The proposed work leverages a field-based phenotyping infrastructure at Maricopa, AZ, which provides an exceptional capability for managed stress trials in a hot and arid environment through controlled irrigation. An automated field scanner system collects high-resolution phenotyping data using a variety of sensors throughout the growth season, from seedling establishment to harvest. A customized sorghum diversity panel will be phenotyped under the field scanner to compare drought-stressed and well-watered plants. This will provide abundant opportunities for GWAS analyses, which will be integrated with information collected from sequence-indexed sorghum mutant populations that have also been phenotyped at UA-MAC. This will facilitate associations between genotype and drought-responsive phenotype and ultimately gene discovery. The same diversity panel will be phenotyped on the Danforth Center's Lemnatec Scanalyzer in a controlled-environment drought stress experiment where RNA-seq performed on each line will allow for interrogation of regulatory variation across sorghum diversity. The project will also leverage an extensive regulatory genomics framework that was established with prior funding to identify genes and regulatory elements that control variation in drought-responsive phenotypes. Candidate loci prioritized through the integration of these resources will be functionally validated in planta using gene editing.

Explanation to "yes" in B.1:

Fieldwork will be performed at the University of Arizona field research site at Maricopa, AZ (UA-MAC). Sorghum plants will be grown in the field for phenotyping over the course of a growing season. Fieldwork will also be performed at research field sites at Purdue University and Donald Danforth Plant Science Center. At these two sites, sorghum plants will be grown for phenotyping and also crosses and seed propagation.

Explanation to "yes" in B.33:

The molecular component of this project is to functionally test hypotheses made based on genomic predictions in planta, and this typically involves editing the locus of interest using CRISPR/Cas9-based gene editing. Here, Cas9 and a series of synthetic guide (g)RNAs designed to target a genetic region of interest, are expressed in plants. Other times, a gene product such as an enzyme or transcription factor may be overexpressed behind a constitutive promoter or specifically expressed using its native promoter or tissue-specific promoter. This work will be done in Sorghum bicolor. Transgenic plants are monitored closely and propagated in controlled growth environments at the Danforth Center Integrated Growth Facility. For edited lines, the Cas9 transgene can be crossed out in later generations rendering the plants non-transgenic. Transgenic or edited plants will never be grown out in the field in this project. All constructs are transformed into plants through Agrobacterium after being assembled in E. coli.

DOE NEPA Tracking Number