Using Phenotyping Technology to Diagnose Chemical Stress in Plants

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Diagnosing Herbicide Stress in Plants

- Herbicide injury in plants is diagnosed through visual evaluation or through tissue chemical analysis
- Drawbacks
    - Subjectivity of manual evaluation
    - High cost and destructive nature of tissue analysis
    - Biochemical changes occur prior to symptom development

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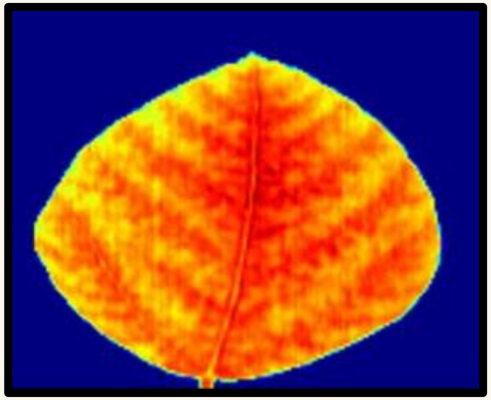
Diagnosing Plant Stress Through Imaging

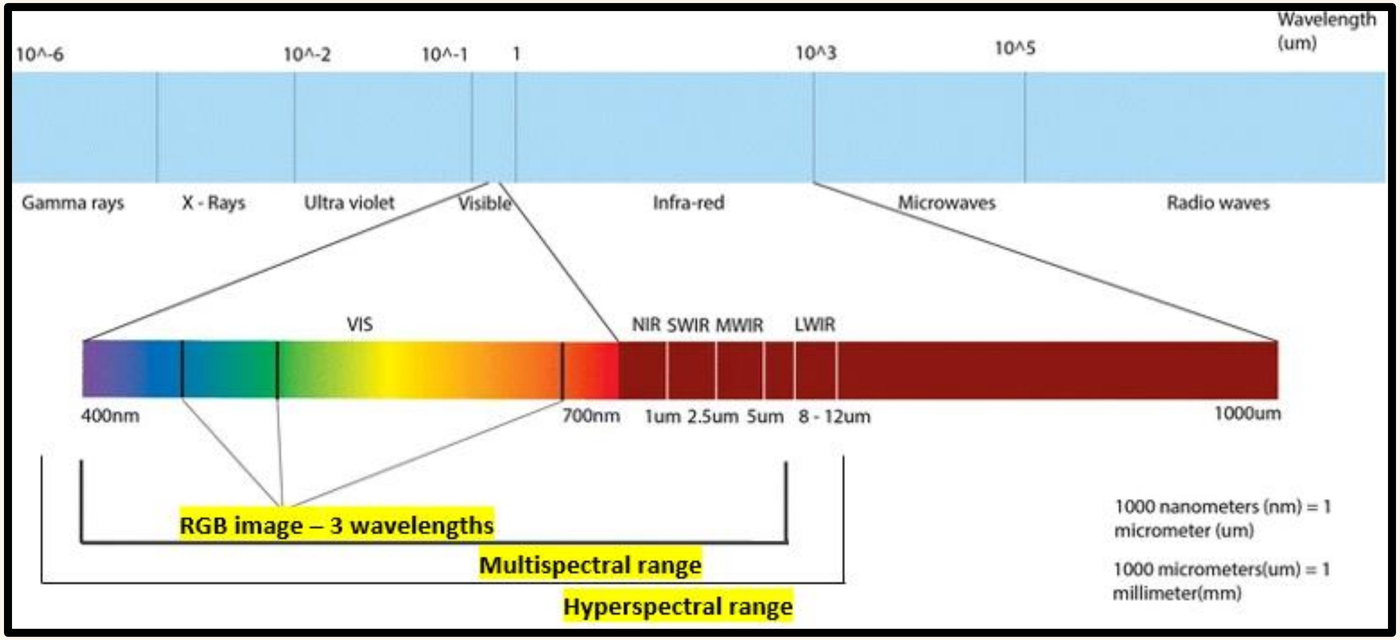
- The light spectral profile of plant tissue is altered by biochemical changes that result from abiotic and biotic stressors[1]
    - Reflected in changes of leaf shape, leaf color, and morphology of the canopy
- Spectral profile differences can be detected through imaging systems[1]



[1] Cheshkova (2022)

Diagnosing Plant Stress Through Imaging

- ❖Hyperspectral imaging evaluates the physiological and morphological parameters of plant tissues simultaneously[1]
- ❖Currently used for measuring leaf moisture, chlorophyll content, nutrient content, and disease presence[1]



[1]Cheshkova (2022)

Photo Credit: Lowe et al (2017)

Imaging Use in Herbicide Research

- ❖Spectral differences in soybeans treated with a dose range of dicamba can be observed 1 week after treatment using an overhead hyperspectral imager.[1]
- ❖Hyperspectral imaging can predict the ability of corn to recover from varying levels of glyphosate injury with high accuracy.[2]
- ❖High throughput imaging using a UAV sensor is more precise in evaluating the severity of crop injury from herbicide stress than visual evaluation.[3]

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[1] Zhang et al. (2019)
[2] Zhang et al. (2021)
[3] Duddu et al. (2019)

Current Hyperspectral Imaging Models

- ❖ Capture single angle
    - o Overhead fixed imagers
    - o UAV imagery
- ❖ Many sources of noise
    - o Ambient lighting
    - o Shadows
    - o Soil backgrounds
    - o Varied leaf angles
    - o Specular lighting

Sources of Imaging Variability

Shadows

Sources of Imaging Variability

Shadows

Soil background

Sources of Imaging Variability

Shadows

Soil background

Varied Leaf Angles

Sources of Imaging Variability

Shadows

Soil background

Specular lighting

Varied Leaf Angles

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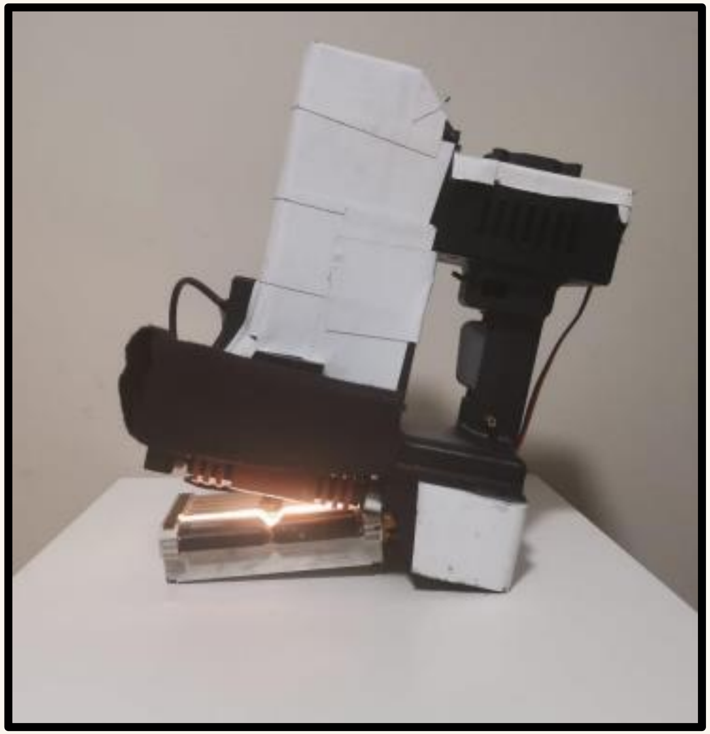
LeafSpec Imager



Early LeafSpec Prototype

LEAFSPEC

- ❖Developed by Dr. Jian Jin’s lab at Purdue University
- ❖Handheld, high resolution hyperspectral imager
- ❖Geo-referenced imaging data provided in real-time
- ❖Clamps to top and bottom leaf surfaces for set imaging lighting, distance, and angles

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Hypothesis

The LeafSpec imager can be used to accurately classify plant injury from foliar herbicide applications prior to visual symptom development.

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Objectives

- Quantify the accuracy of LeafSpec to detect exposure of soybeans (*Glycine max*) to low doses of dicamba and 2,4-D choline.

- Determine the accuracy of LeafSpec classification for herbicides in different site of action groups on waterhemp (*Amaranthus tuberculatus*).

Materials and Methods
Soybean Trial

- Application
  - Soybeans were treated at the V2 growth stage
  - Moving track sprayer applied 140 L ha^{-1} solution
  - 14 replicates per treatment
- Data Collection
  - Visual estimates of injury (0-100%) at 7 and 14 DAT (data not shown)
  - Hyperspectral images collected with LeafSpec at 1, 4, 7 and 14 DAT

Auxin Treatments	Rate
2,4-D choline	1/25th [1]
2,4-D choline	1/75th [1]
Dicamba	1/4000th [2]
Dicamba	1/1000th [2]
Nontreated	-

1. Fraction of 1065 g ae ha^{-1} labeled rate
2. Fraction of 560 g ae ha^{-1} labeled rate

Materials and Methods
Waterhemp Trial

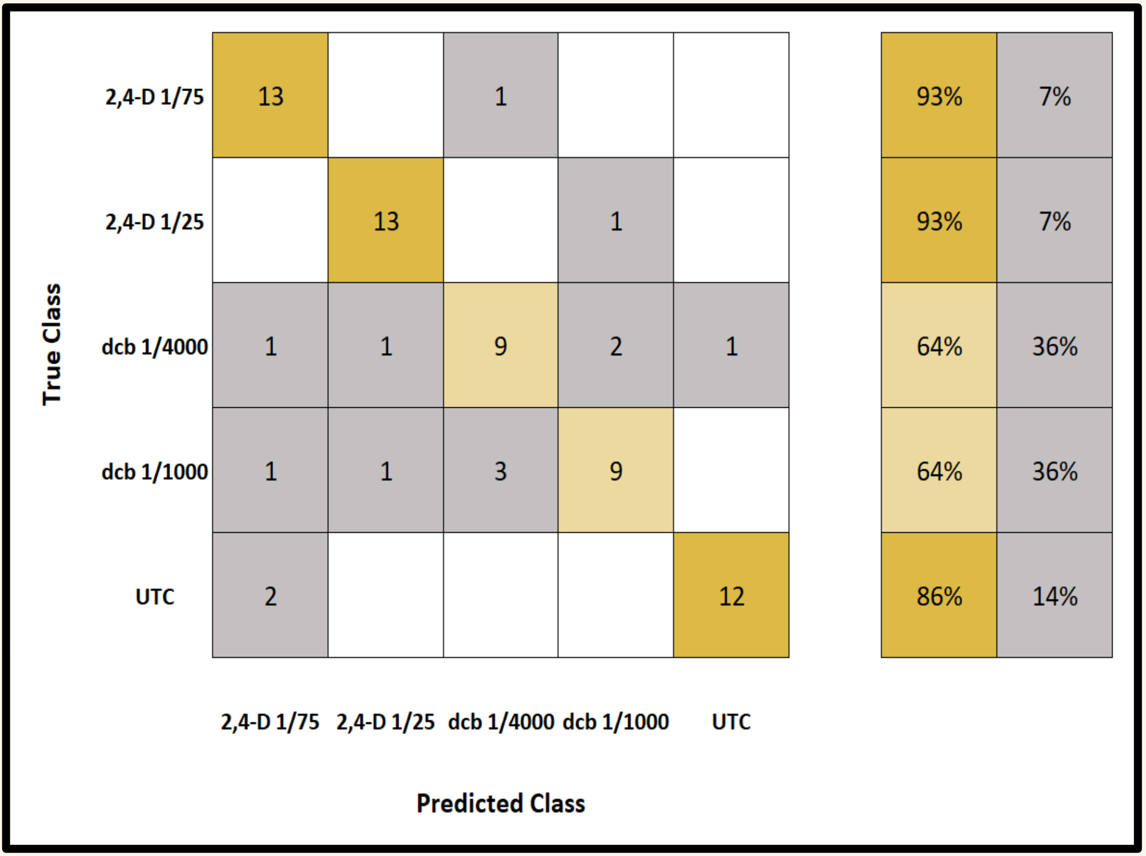
SOA Treatments	Rate
Dicamba	17.5 g ae ha^{-1}
2,4-D choline	17.5 g ae ha^{-1}
Atrazine	375 g ai ha^{-1}
Flumioxazin	1.17 g ai ha^{-1}
Mesotrione	18.75 g ai ha^{-1}
Norflurazon	270 g ai ha^{-1}
Nontreated	-

- Application
  - Waterhemp plants had at least 12 leaves at application. Leaves measured 5 cm in length
  - Atomizer applied herbicide spray to wet
  - 16 replicates per treatment
- Data Collection
  - Visual estimates of injury (0-100%) at 4 and 7 DAT (data not shown)
  - Hyperspectral images collected with LeafSpec at 1, 4, 7, and 14 DAT

Materials and Methods
Data Processing

- Various algorithms were used for noise reduction and compared for injury classifications

- Model classification accuracy results were represented as **confusion matrices**



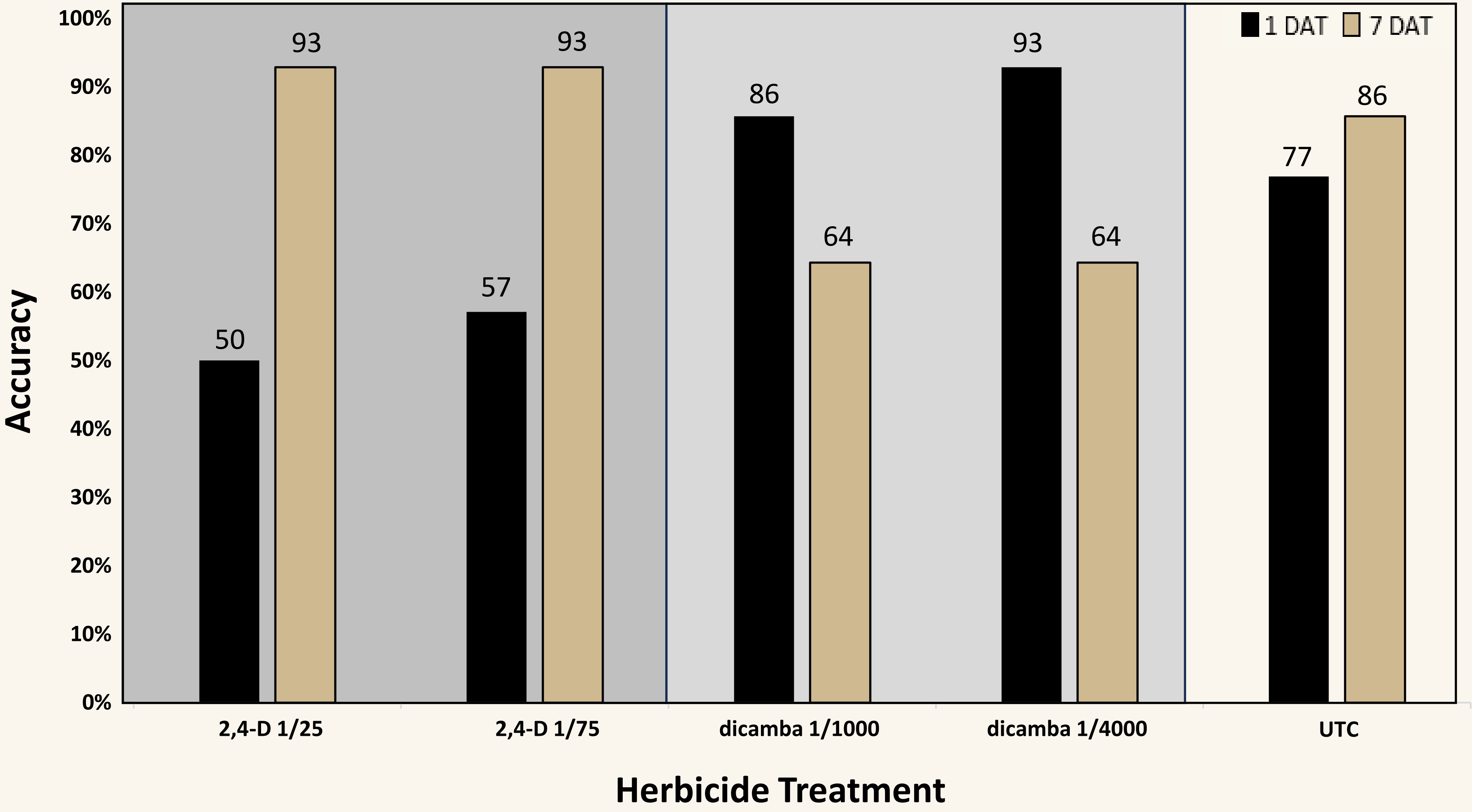
True Class

	2,4-D 1/75	2,4-D 1/25	dcb 1/4000	dcb 1/1000	UTC		
2,4-D 1/75	13		1			93%	7%
2,4-D 1/25		13		1		93%	7%
dcb 1/4000	1	1	9	2	1	64%	36%
dcb 1/1000	1	1	3	9		64%	36%
UTC	2				12	86%	14%

Predicted Class

Results

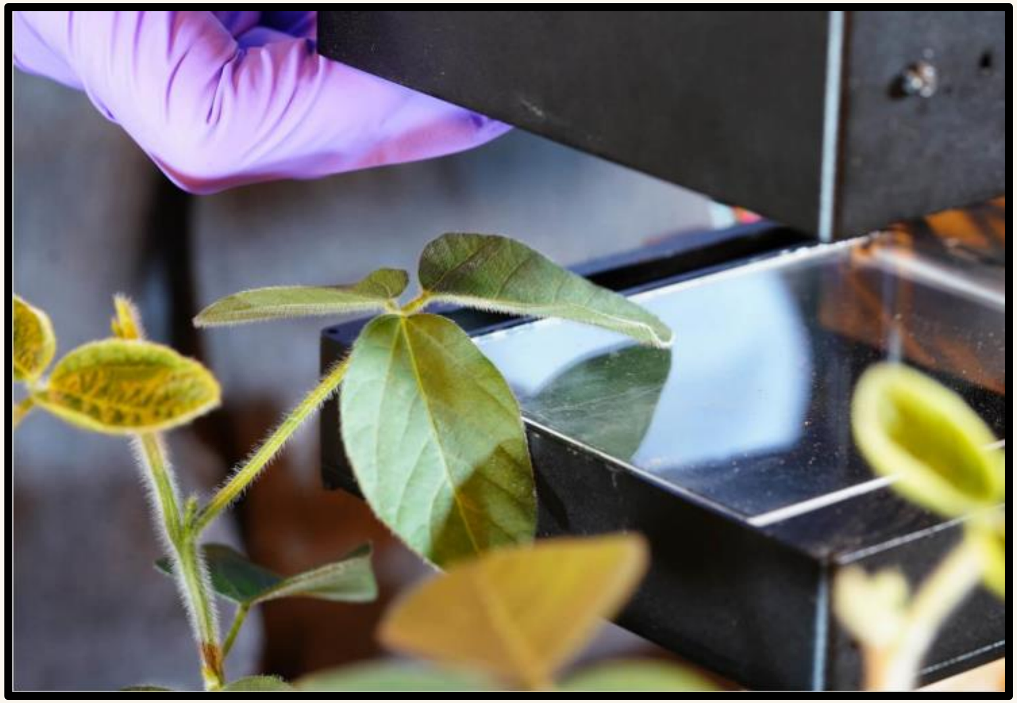
Soybean Injury Classification Accuracy
1 DAT vs 7 DAT



Soybean Injury Classification Accuracy

- ❖Dicamba exposure was classified with LeafSpec with greater than 85% accuracy at 1 DAT before visual injury symptoms were present.

- ❖2,4-D classification accuracy increased with time after application to 93% accuracy at 7 DAT.



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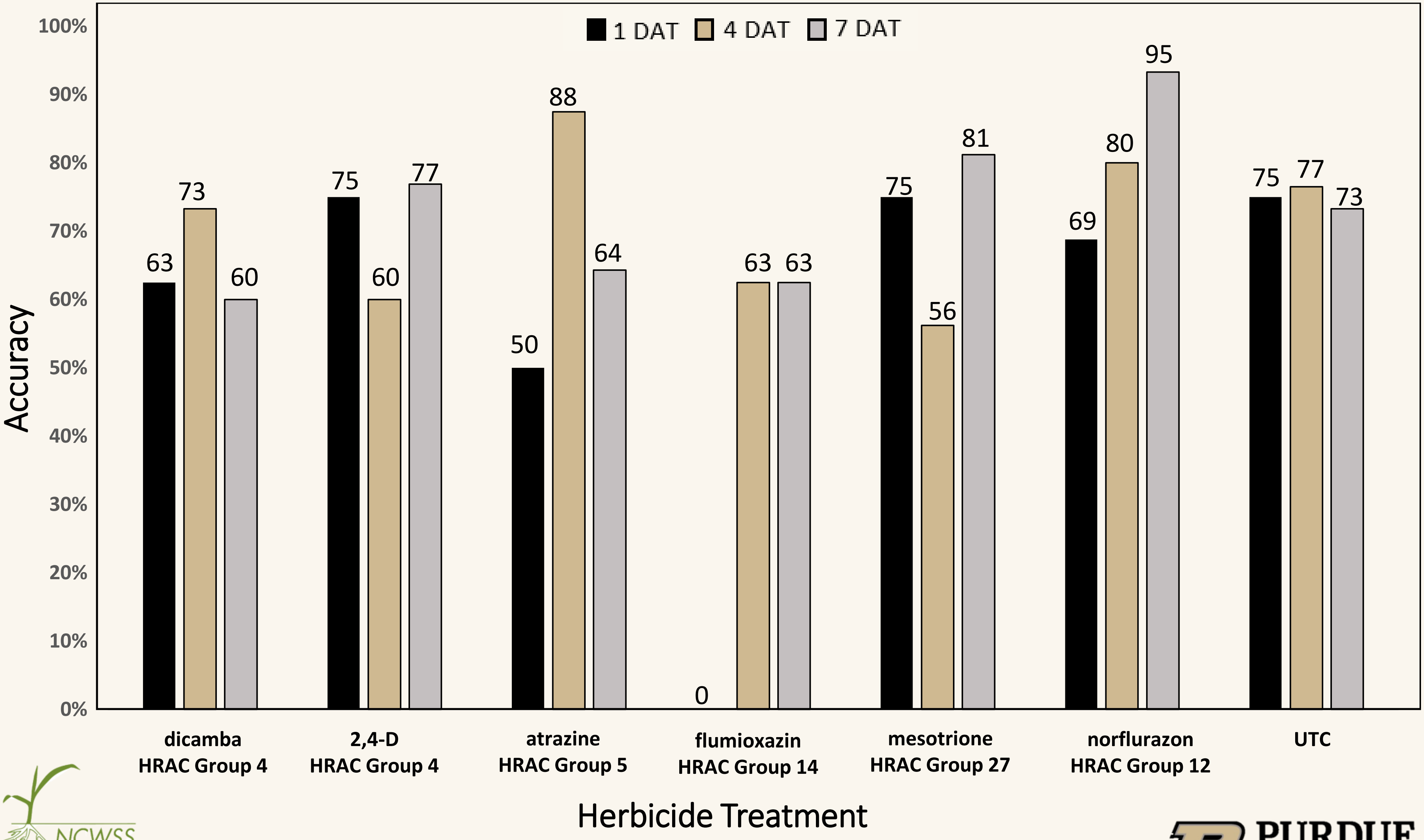
Waterhemp Classification Accuracy
1, 4, 7 DAT



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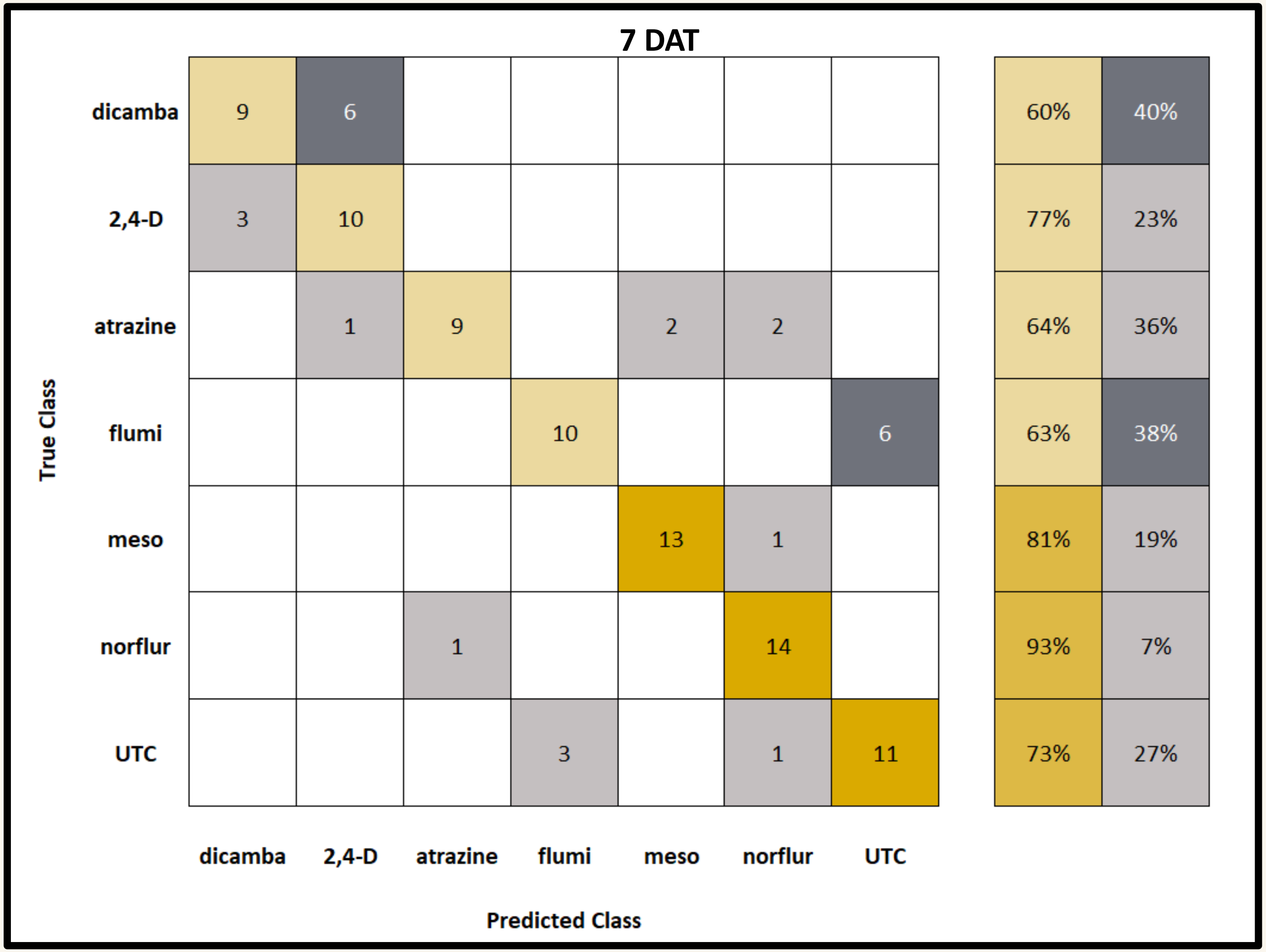
Waterhemp Confusion Matrix



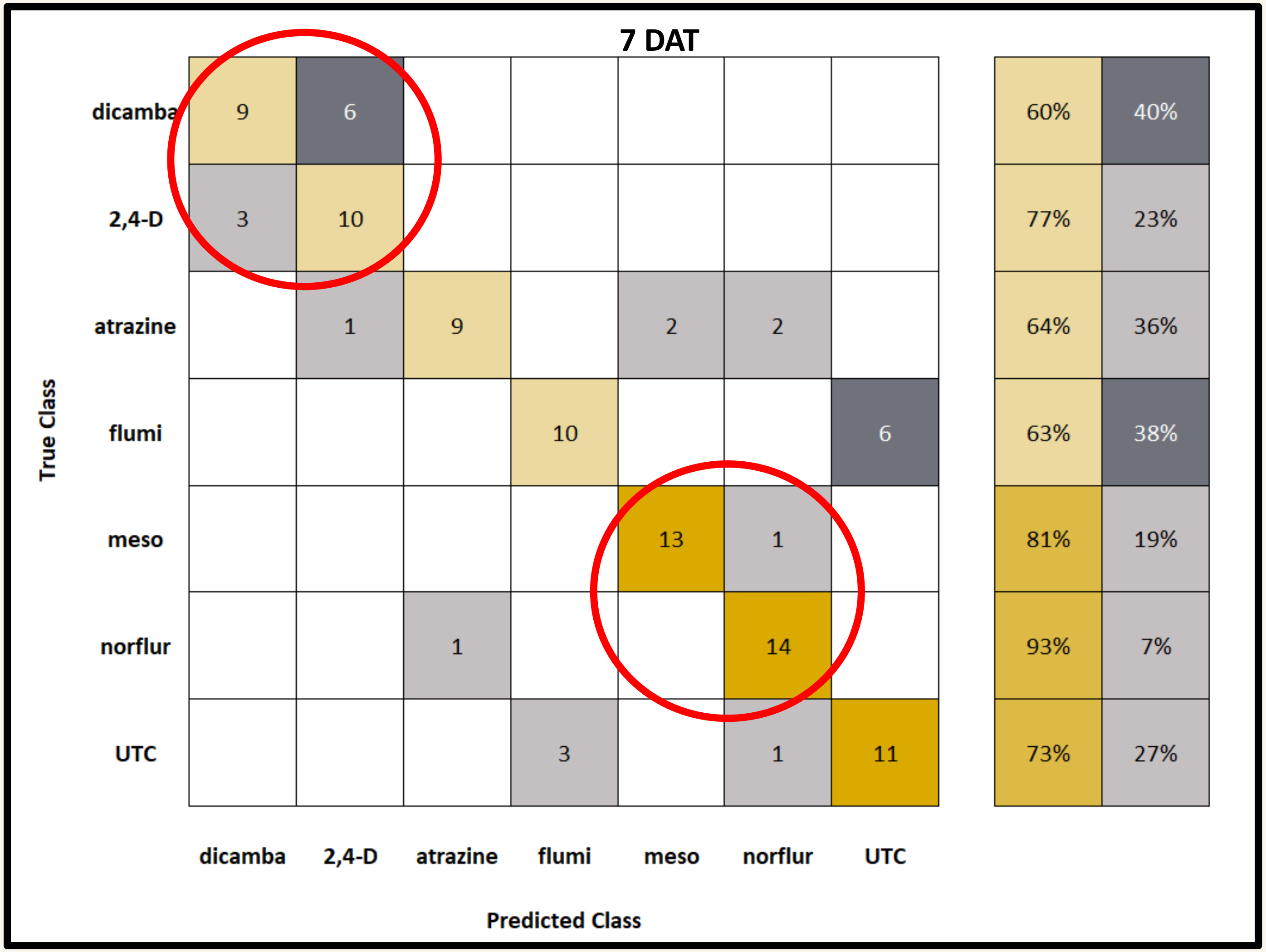
7 DAT

True Class

	dicamba	2,4-D	atrazine	flumi	meso	norflur	UTC		
dicamba	9	6						60%	40%
2,4-D	3	10						77%	23%
atrazine		1	9		2	2		64%	36%
flumi				10			6	63%	38%
meso					13	1		81%	19%
norflur			1			14		93%	7%
UTC				3		1	11	73%	27%

Predicted Class

Waterhemp Confusion Matrix



7 DAT

	dicamba	2,4-D	atrazine	flumi	meso	norflur	UTC		
dicamba	9	6						60%	40%
2,4-D	3	10						77%	23%
atrazine		1	9		2	2		64%	36%
flumi				10			6	63%	38%
meso					13	1		81%	19%
norflur			1			14		93%	7%
UTC				3		1	11	73%	27%

True Class

Predicted Class

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Waterhemp Classification Accuracy

- ❖The highest accuracies at 1 DAT were 2,4-D and mesotrione with 75% accuracy.
- ❖Atrazine classification accuracy increased over time with 50% accuracy 1 DAT compared to 88% accuracy 4 DAT.
- ❖Greater than 70% accuracy was achieved in classifying herbicides with similar symptoms.

Conclusions

❖Hyperspectral imaging with LeafSpec can predict the herbicide applied before injury symptoms are visible with a high level of accuracy.

❖LeafSpec imaging allowed for earlier detection of injury symptoms from dicamba compared to visual evaluation.

❖Distinguishing herbicides with different sites of action is possible with LeafSpec imaging though accuracies varied by herbicide and time after application.

Implications / Future Research

- LeafSpec technology would have utility in early herbicide discovery to discern if a molecule is similar to a known SOA group.

- Capabilities and limitations found in these results will be used to build improved imaging models combining spectral, morphological, and textural features.

- Spectral index heatmaps will be used to increase model classification accuracies of auxin herbicide applications.

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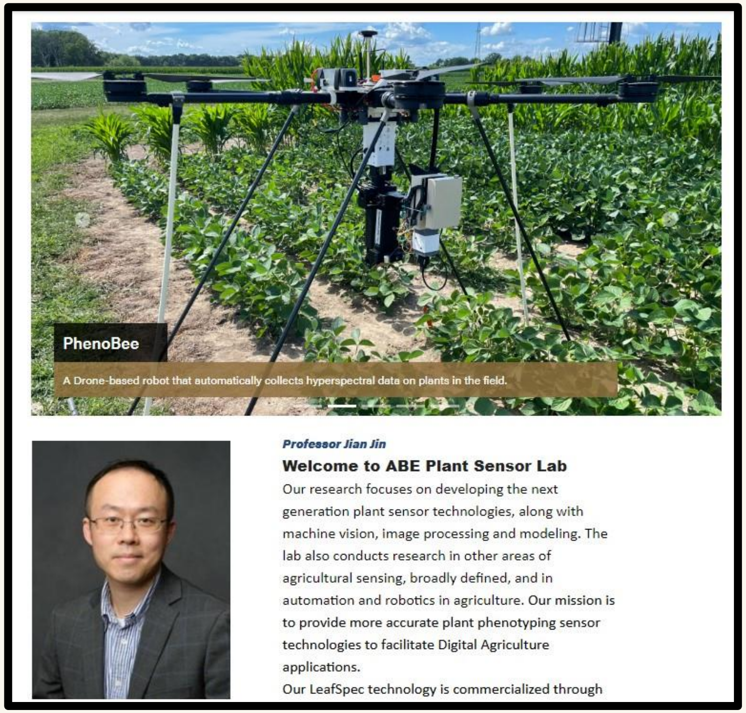
- ❖Purdue Weed Science Team
- ❖Purdue Jian Jin Lab



PhenoBee

A Drone-based robot that automatically collects hyperspectral data on plants in the field.

Professor Jian Jin

Welcome to ABE Plant Sensor Lab

Our research focuses on developing the next generation plant sensor technologies, along with machine vision, image processing and modeling. The lab also conducts research in other areas of agricultural sensing, broadly defined, and in automation and robotics in agriculture. Our mission is to provide more accurate plant phenotyping sensor technologies to facilitate Digital Agriculture applications.

Our LeafSpec technology is commercialized through

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Questions?

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