### **Problem definition**

### What is Phenotyping? (Netherlands Plant Eco-phenotyping Centre)

- "Root phenotyping is the study of observable root traits, such as structure, growth, and environmental interaction."
- "In NPEC's case, it involves tracking traits like root length, branching using image analysis
- "At NPEC, seeds of Arabidopsis thaliana are sown in Petri dishes, imaged daily as roots grow, and exposed to microbes or treatments."
- "These images are used to quantify key traits and analyze plant-microbe interactions."

# **Insights Phenotyping Provides:**

- "Phenotyping reveals how roots grow and adapt under different conditions, such as limited water or varying microbial environments."
- "It supports breeding for resilient crops, enabling sustainable and improved agricultural practices."

### **Challenges in Phenotyping:**

#### 1. Complex Root Systems:

 "Overlapping and crisscrossing roots make manual segmentation time-consuming and error-prone."

#### 2. Precision in Inoculation:

 "Microbial treatments require pinpoint accuracy to ensure consistent experimental outcomes."

### 3. Scalability:

 "Processing 10,000+ seedlings manually is impractical and creates a bottleneck in data analysis."

### 4. Time and Resources:

 "Current manual methods are slow and require significant human labour, limiting research efficiency."

### In conclusion, automation is needed...

# Overview of the proposed solution:

### **Solution Components:**

- Computer vision: root analysis → segmentation, length, tips.
- Robotics: inoculation → precision, consistency.

#### **Benefits to NPEC:**

- Streamlines root phenotyping → less manual effort.
- Scalable → processes thousands of seedlings daily.
- Reliable data → root traits, growth, plant-microbe studies.
- Accelerates research → genotypes, stress resistance, nutrient uptake.

### **Business Impact:**

- Faster experiments, larger-scale studies.
- Supports sustainable agriculture → resilient crops.

### **Results & Evaluation:**

### **Results (Computer Vision):**

- U-Net → root detection accuracy with 82.18 F1 score on a validation set.
- 12.47 (% error score) sMAPE → high accuracy in root length detection.
- $\circ$  Tip detection  $\rightarrow$  essential for inoculation pipeline input.
- $\circ$  Root segmentation  $\rightarrow$  identifies roots (1–5) in complex Petri dish images.

#### Reinforcement Learning:

- o working on RL PPO model for robotic inoculation.
- o RL pipeline will integrate with CV for complete automation.

#### Business Value:

- Accurate root analysis → scalable phenotyping for 10,000+ seedlings.
- Consistent and reliable data → accelerates genotype identification.
- o Directly supports plant-microbe studies and sustainable agriculture goals.

#### Transition to Next Slide:

 "Next, I'll walk you through the development and iterations that led to these results."

# **Error Analysis and Iterations of the CV pipeline:**

### **Iteration 1:**

### • Initial Steps (Constants):

- o Image preprocessing: crop, pad, patchify, and unpatchify.
- U-Net model trained on 2023 dataset (11k images split into train/val).

### Challenges/Error Analysis:

- Poor segmentation results → F1 score of 76.88 on validation set.
- Missing large portions of roots → insufficient for downstream tasks (e.g., root segmentation and analysis).
- Could not attempt individual root segmentation.

### Impact on the Project:

- o Identified key issues with training data or model setup.
- Provided a baseline for improvement → next iteration needed better performance.

#### • Transition to Next Slide:

 "This iteration helped identify the limitations of the initial dataset and model. In the next iteration, I implemented changes to improve segmentation accuracy."

### **Iteration 2:**

### Improvements from Iteration 1:

- Combined datasets → larger, more diverse training data.
- F1 score improved from 76.88 to 82.18, enabling clear masks.
- Introduced morphological operations to reduce noise and close gaps.

### **Root Segmentation Workflow:**

- Divided masks into **5 vertical sections** → bounding boxes identify roots.
- Skeletonised roots and calculated lengths using Dijkstra's algorithm.
- Assigned IDs to roots → accounted for missing or non-contiguous plants.

#### **Challenges and Learnings:**

- Vertical sectioning caused some roots to be missegmented (cut off).
- Small plants not identified → revealed the need for more adaptable segmentation methods.

#### **Business Value:**

- Enabled detailed root phenotyping by accurately identifying and measuring roots.
- Provided data critical for analysing root traits and advancing genotype research.

### **Transition to Next Slide:**

 "This iteration brought significant improvements, but there were still challenges to address. In the next iteration, I focused on refining segmentation further to capture smaller and more complex root systems."

### **Iteration 3:**

### **Improvements from Iteration 2:**

- Retained inference model → consistent, high-quality masks.
- Dynamic sectioning → adaptive segmentation captured edge cases more effectively.
- Enhanced ability to retain roots in challenging scenarios (e.g., plants at the edges of the Petri dish).

### **Workflow Consistency:**

- $\bullet \quad \text{Skeletonized roots} \rightarrow \text{calculated lengths using Dijkstra's algorithm}.$
- Assigned root IDs based on dynamic sections.

### **Challenges and Learnings:**

- Very small plants still missed → significant relative error in sMAPE due to their small size.
- Improvement in sMAPE (12.47) showed progress but highlighted the need for better detection of tiny plants.

# **Assumptions:**

**5 plants max** per dish.

Plants seeded in 5 sections.

Seeds placed in **top quarter** of the dish.

Assumptions guided segmentation process.

# Limitations:

Model not perfect but accurate.

**Overlapping roots** → misinterpretations.

**Tiny roots** mistaken for scratches/noise.

# **Further Steps:**

Most cases identified correctly.

#### Inference Model:

- Improve segmentation accuracy → tiny roots and overlapping areas.
- Address the current model's edge-case failures.

### Preprocessing:

- Better noise filtering to detect all roots, even very small ones.
- Adapt to challenging plant positions for better segmentation.

### **Reinforcement Learning Integration:**

- PPO hyperparameter tuning → optimizing n\_steps.
- Use CV root tip detection as the pipette goal.
- Automate inoculation fully through RL-CV integration.

### **Pipeline Refinement:**

- Focus on accurate root development (length) analysis.
- Ensure robust and scalable solutions for phenotyping.

### **Future Applications:**

- Apply pipeline to other phenotyping tasks.
- Enable large-scale experiments to advance genotype research.

### The End...