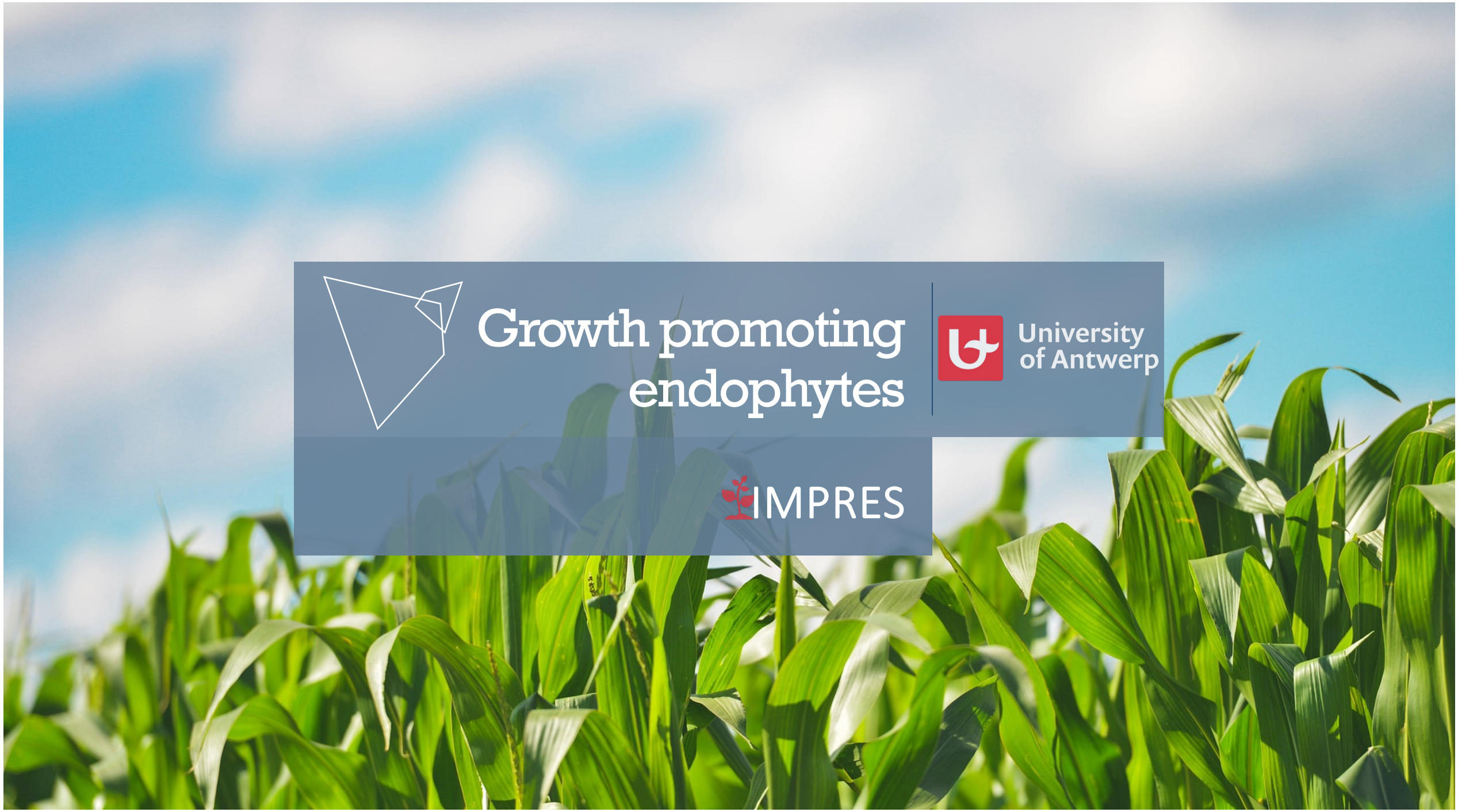
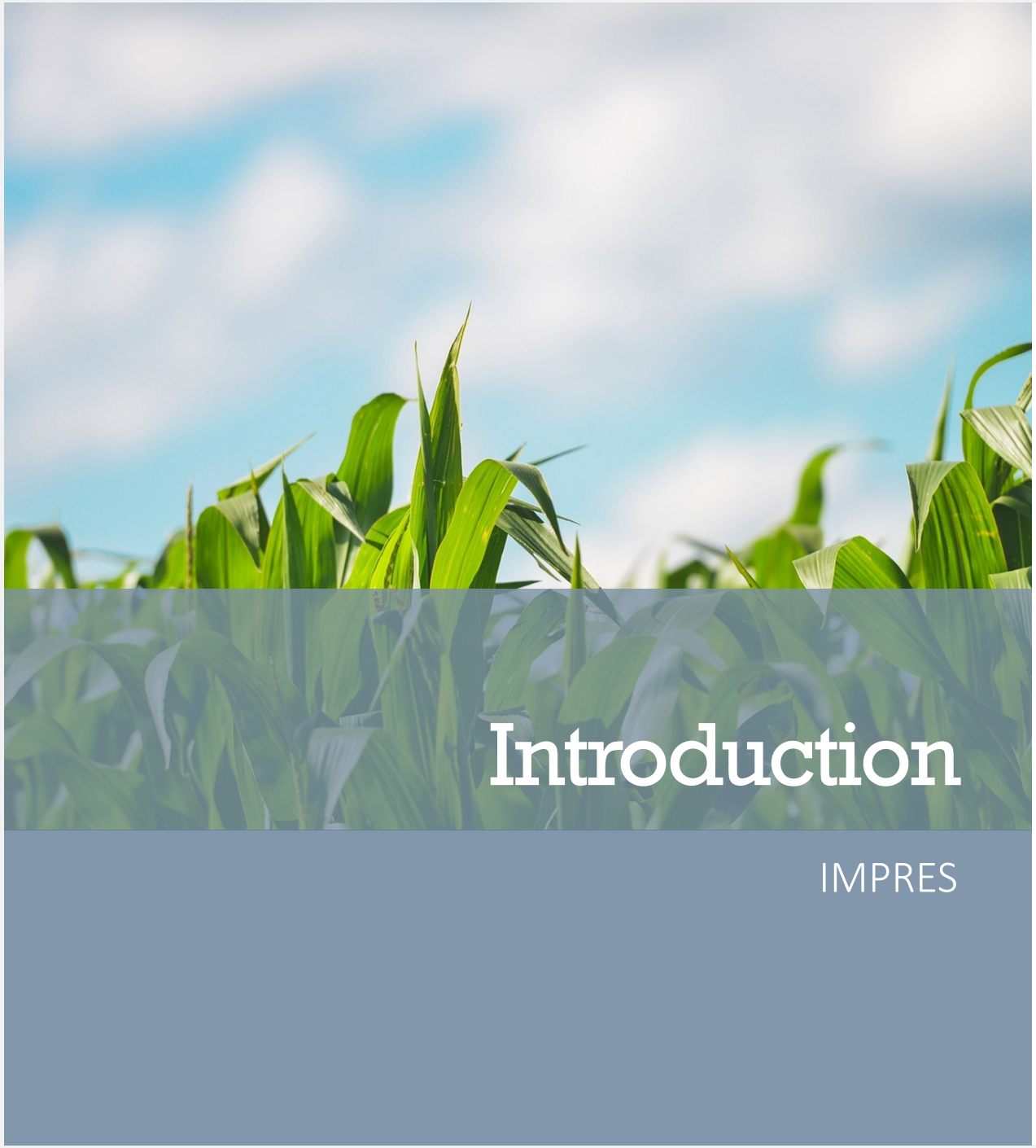

Growth promoting endophytes
University of Antwerp
IMPRES



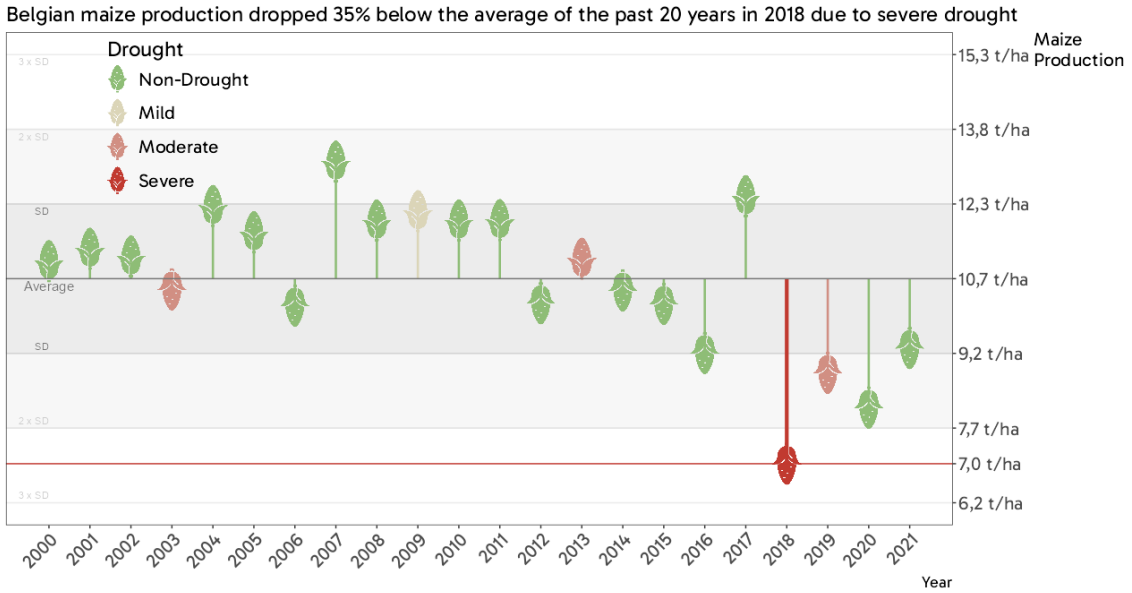
Introduction

IMPRES

- Studying endophytes under drought in maize
- How do we elucidate their function?
  - At the microbe-level
    - Fluorescence microscopy
    - qPCR
    - Genomic assembly and analysis
  - At the plant-level
    - Kinematic analysis
    - Leaf length tracker
    - Transcriptomic analysis
    - Follow-up analyses (metabolites, enzymes,…)

Studying endophytes under drought in maize

- An **endophyte** = an endosymbiont, often a bacterium or fungus, that lives within a plant without causing apparent disease
- They have often proven to be a valuable solution to promote plant growth under different conditions
- Due to climate change, drought frequency and intensity are set to increase
- Severe drought can greatly impact maize yield
- **The goal of this research project is thus to elucidate the known positive effect some endophytes have on maize growth under drought stress**
- Research interests
  1. Do these endophytes act in a specific plant tissue?
  2. How are different processes affected at the plant-level?
  3. Which metabolites and/or proteins are produced that mediate crosstalk between the endophytes and the host-plant?

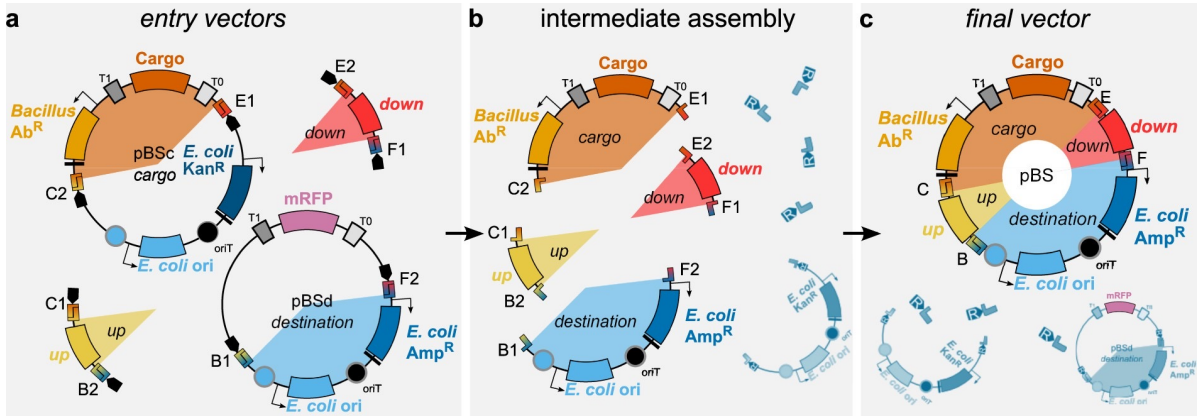


Data: FAOSTAT & Global Drought Monitor

Bacterial strains – Specific analyses

- Two bacterial strains labelled E7 and E14
  - *Bacillus paralicheniformis* and *Bacillus velezensis*
  - Obtained from a collection of microbes extracted from grasses grown in arid environments through collaborations with universities in Egypt and Saudi-Arabia
- **Fluorescent labelling** of the two *Bacillus* strains to obtain information on (sub-cellular) localisation and abundance *in planta*
  - Colonization experiments
  - Fluorescence microscopy
  - Quantification using qPCR

Vector cloning is currently in progress in order to create fluorescently labelled strains



Fluorescence microscopy

Quantification using qPCR

Fungal strain – Specific analyses

- One fungal strain labelled *Trichoderma harzianum* ESALQ1306
  - Commercially available and used to promote maize growth in Brazil
- **Genome sequencing, assembly, and annotation** of a *Trichoderma* strain
  - Further analysis of this genome can help explain the findings *in planta*

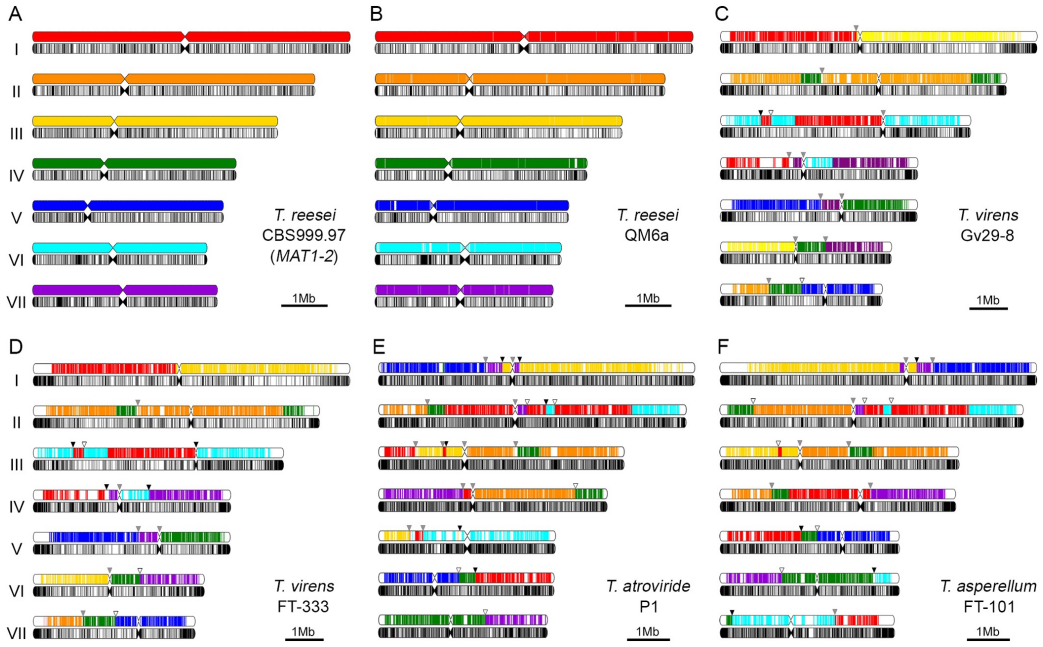
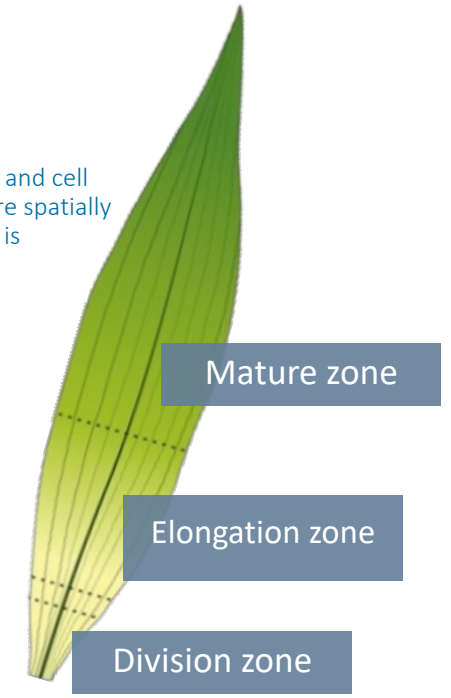


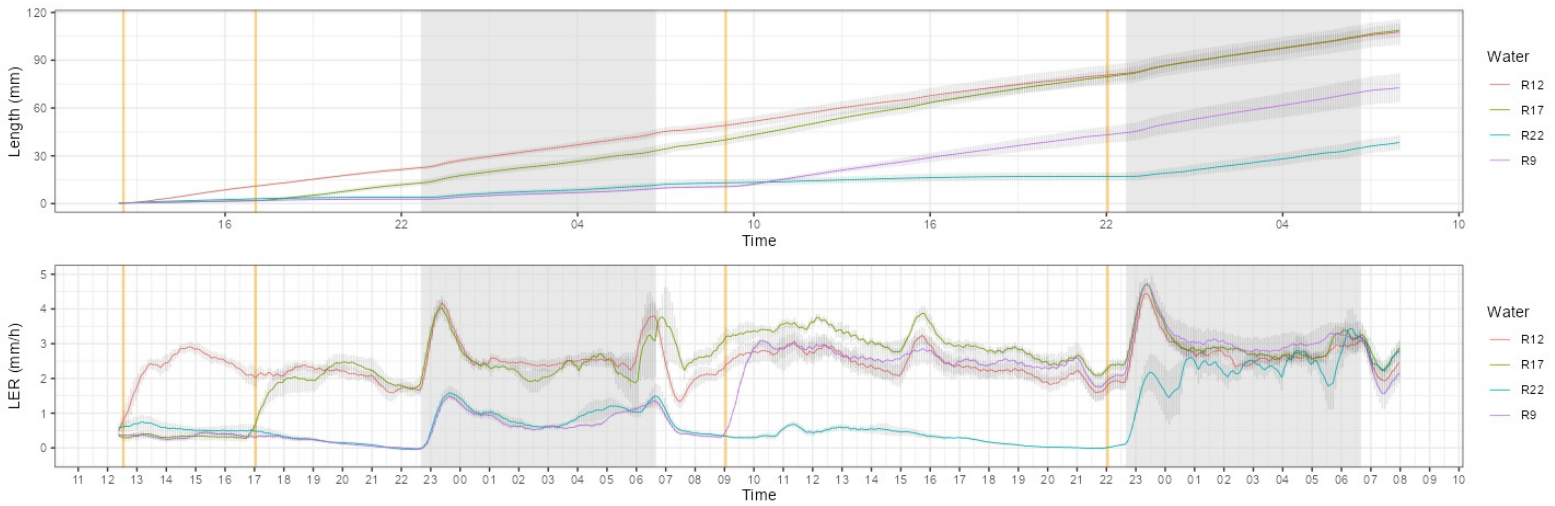
Image from Li et al. (2021) illustrating the chromosomal organisation of different *Trichoderma* species

General *in planta* experiments and analyses

- **Kinematic analysis** to determine the contribution of cell division and cell expansion to maize leaf growth
  - Already performed for both bacterial strains and the fungal strain, but can be redone in context of mutant lines
- How is leaf-growth affected over time?
  - Study the drought-response over the diurnal cycle using our newly obtained **leaf-length tracker** (LLT)!

Growth is a result of cell division and cell expansion, two processes that are spatially separated in the maize leaf. This is something we can exploit!





Study of dynamic changes using the LLT, e.g., yellow-marked timepoints indicate re-watering of drought-stressed plants; increase in leaf elongation rate (LER) occurs almost instantaneously

General *in planta* experiments and analyses

- Strategic sampling of the important tissues to study the interaction in more detail (determination of important tissues is based on kinematic analysis results)
  - Transcriptomic analysis
    - Already performed for the fungal strain, but can be redone in context of mutant lines; will be performed for E7 and E14
  - Metabolite profiling
  - Validation experiments (quantification of enzyme activity, ROS, and various compounds; use of mutant maize lines,…)

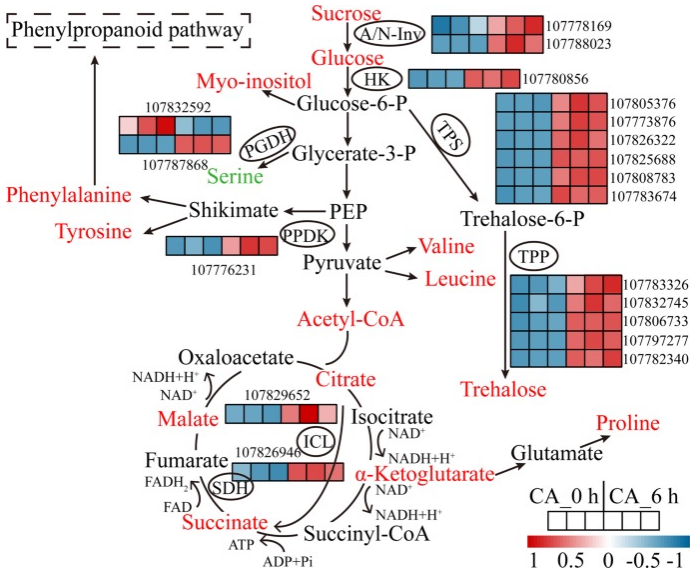
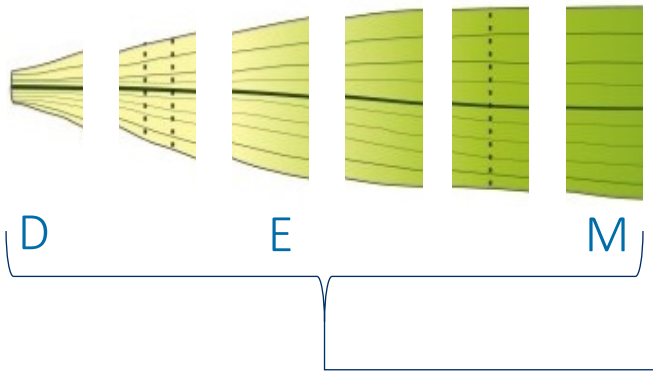
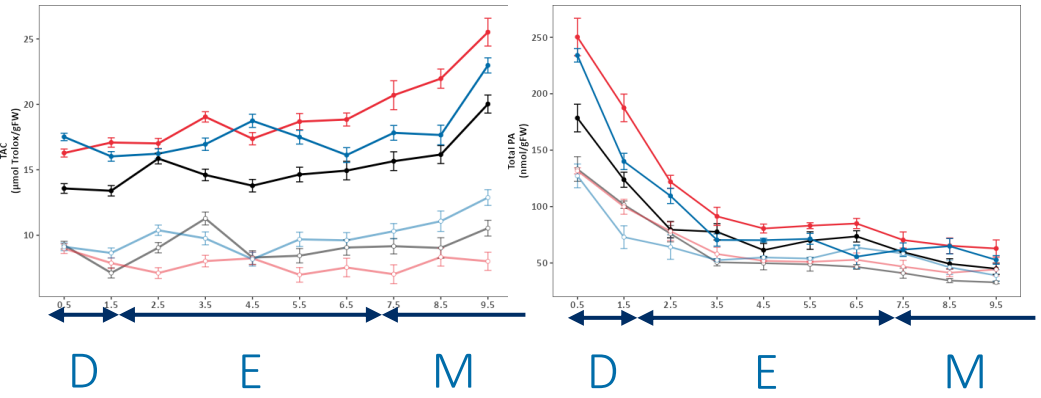
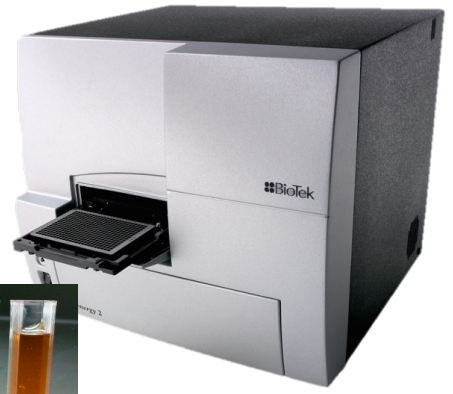


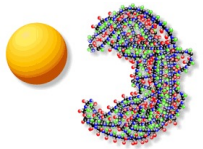
Image from Xu et al. (2020) illustrating the use of transcriptomics to study the effect of cold in tobacco



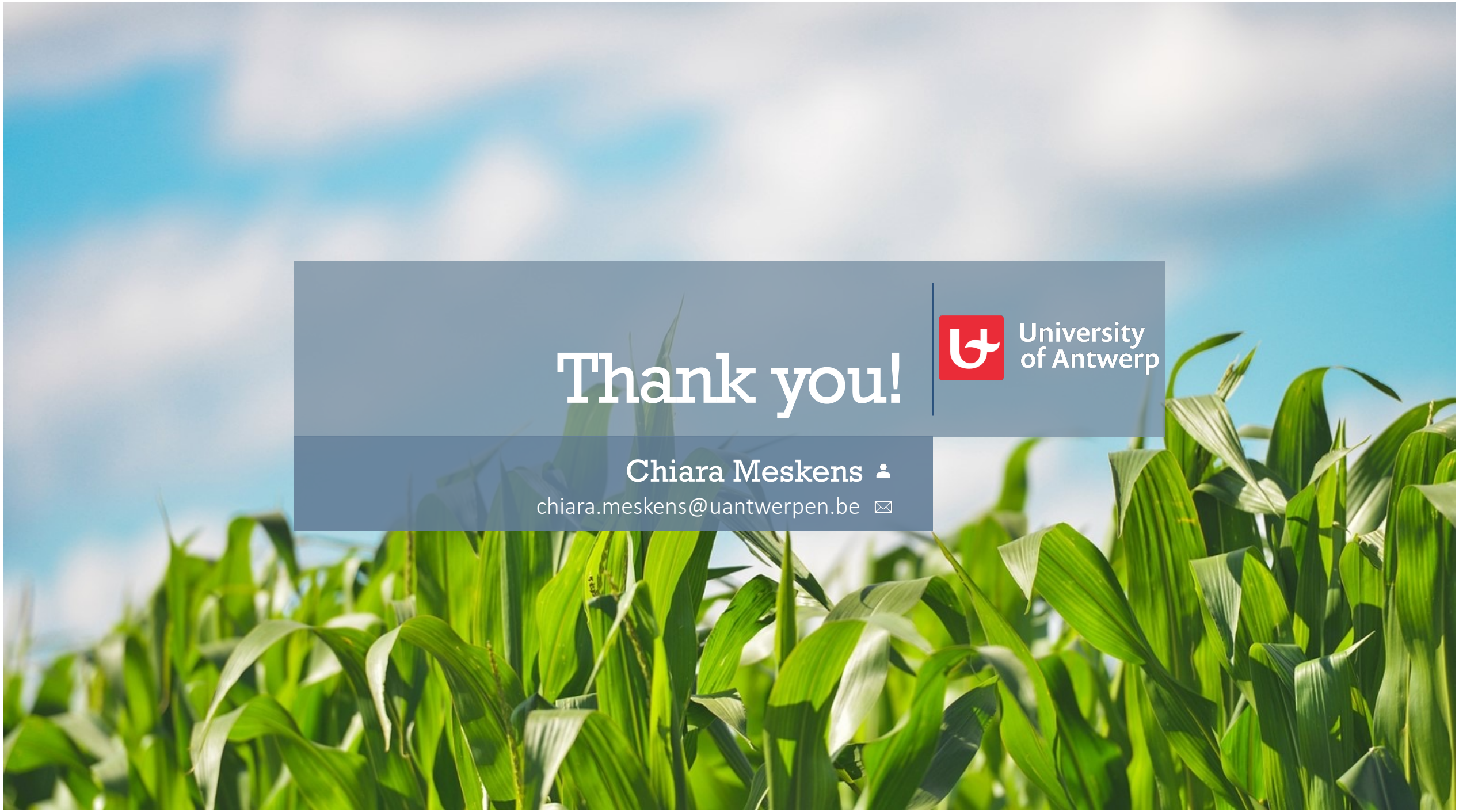
Study zone-specific effects in the maize leaf







Colour-based assays to perform various quantifications using a platereader


Thank you!
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