Symposium:

Future proof seeds: innovative crop protection solutions

Organized by:

Program

13:30-13:35	Opening	Gea Bouwman, Plantum
13:35-14:00	SUCSEED - Deciphering and boosting seed defense mechanisms to avoid seed pathogen transmission and damping-off	Jerome Verdier, INRAE IRHS
14:00-14:20	Green on Seed – the quest for new ‘green’ crop protection solutions	Frans van Tetteroo, Vertify
14:20-14:45	Plasma Activated Water (PAW) – Cleaning seeds without chemicals	Mark van Boxtel en Ineke Wijkamp, Vital Fluid
14:45-15:00	Discussion and closing	Gea Bouwman, Plantum

seedmeets technology

SUCSEED project

Stopping the Use of –Cides in SEEDs

@SUCSEED_Project

IRHS Seed Lab

jerome.verdier@inrae.fr

SUCSEED project ID

> Coordination INRAE

> Starting date: 1st January 2021 (6 years)

> Total cost: 11.3 M€ (2.9 M€ subvention) – French consortium (16 partners)

> Fundamental research (TRL 1-2) with applied outcomes (TRL 3-5)

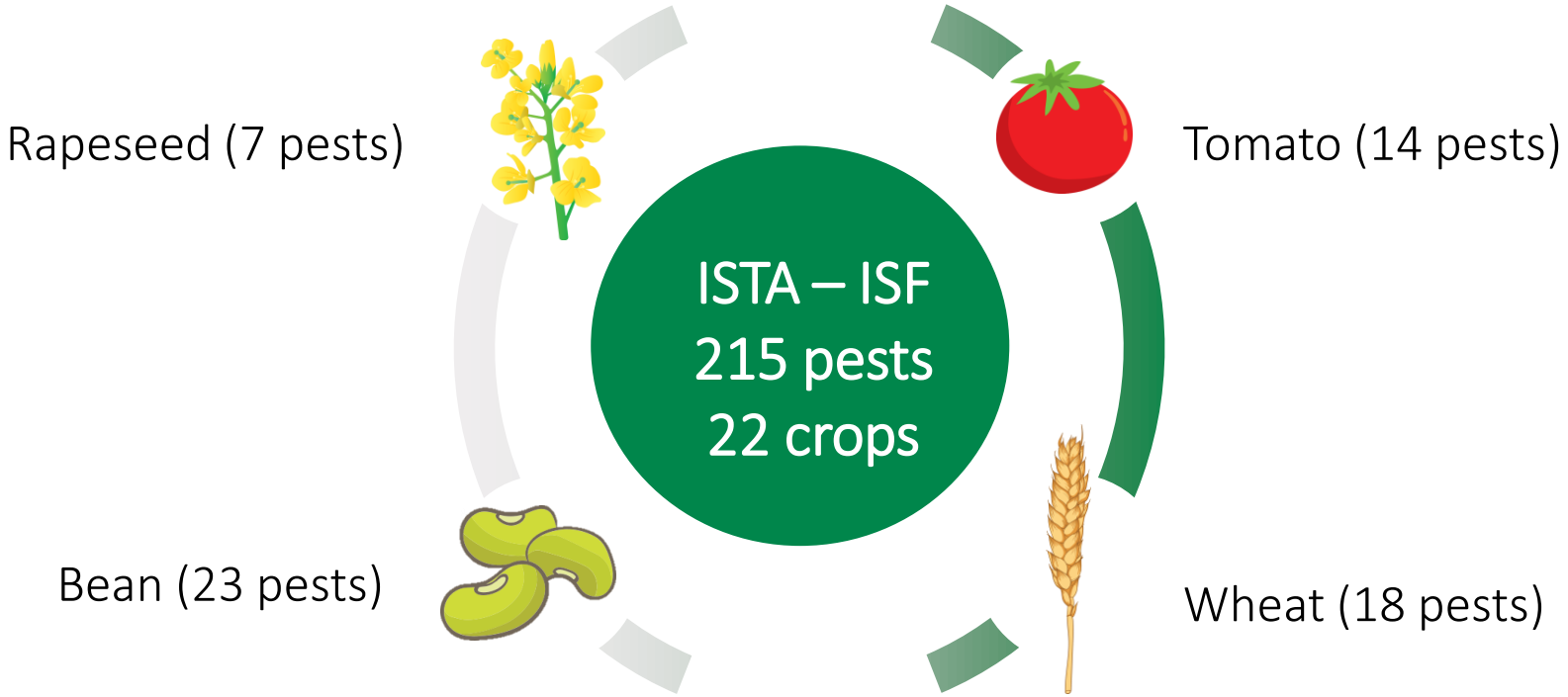
IRHS Institut de Recherche en Horticulture et Semences — INRAE

Matthieu BARRET (INRAE Angers)
Scientific coordinator
Matthieu.barret@inrae.fr

@SUCSEED_Project

Problem 1:

Seed as trading material : **Carrier of plant pathogens**



Securing seed health to avoid plant disease emergence

Problem 2:

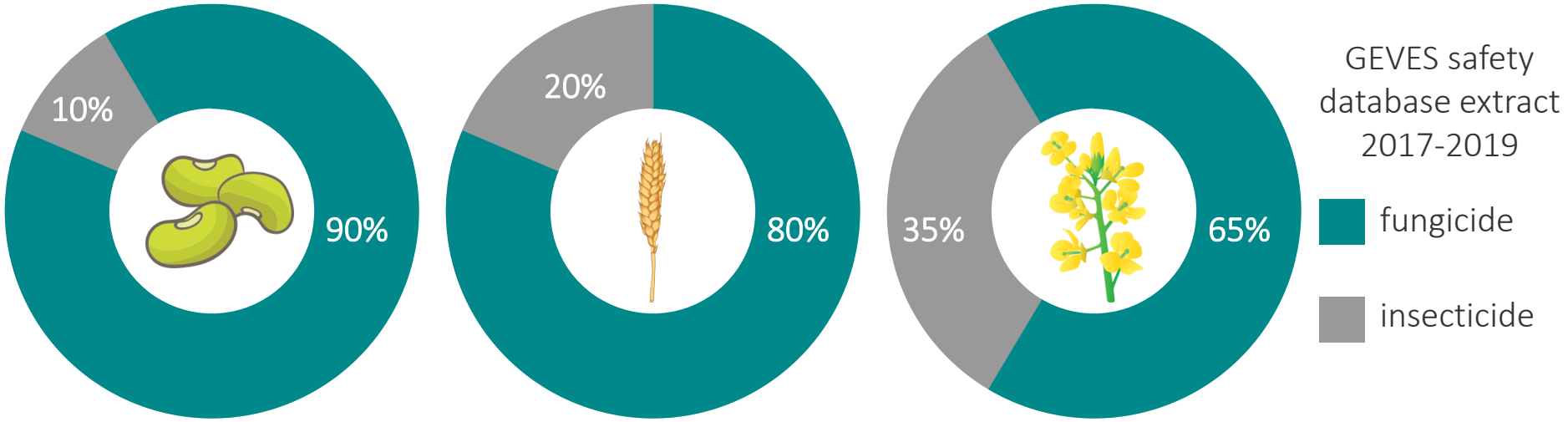
Damping-off is due to seed- and soil-borne pathogens

@ A. Dorrance (APSnet)

Incidence on crop establishment / yield: 5 - 80% (Lamichhane, 2017)

Current solutions:

Secure seed quality through **seed treatment** ($6.7 Billion, 2020)[1]



But reduction of available chemical plant protection products

[1] IMARC, Seed Treatment Market Size, Share, Trends and Forecast 2021-2026 (imarcgroup.com) – 13/01/2022

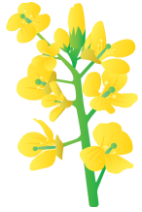
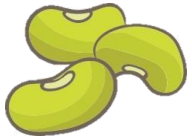
SUCSEED Objectives

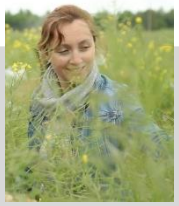
Innovative and bio-inspired alternatives to pesticides adapted to seed to

- Prevent seed-transmitted pathogens (bacteria + fungi)
- Limit damping-off (fungi + oomycetes)
- Promote growth/vigor under stress conditions

Towards the development of **biocontrol and biostimulant solutions**

SUCSEED Workflow

WP1 – PATHOSYSTEM DEVELOPMENT
(N. Nesi, IGEPP)

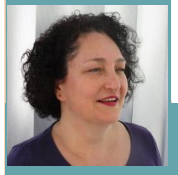
WP2 - ENHANCEMENT OF SEED DEFENSES
(J. Verdier, IRHS)

WP3 - SEED MICROBIOME ENGINEERING
(M. Simonin, IRHS)

WP4 - SEED LOCAL ENVIRONMENT ANALYSIS
(L. Rajjou, IJPB)

WP5 - SEED FORMULATION
(H. North, IJPB - P. Saulnier, MiNT)


WP7



REGULATORY ECONOMIC & SOCIAL CONTEXTS ANALYSES
(RM Borges, IRPI)

WP6 - DEPLOYMENT OF SOLUTIONS ON A RANGE OF GENETIC RESOURCES & ENVIRONMENTS
(T. Langin, GDEC)

WP8

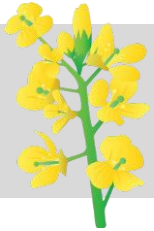
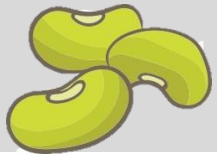
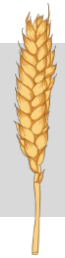
PROJECT MANAGEMENT DISSEMINATION & TRAINING
(M. Barret, IRHS)

WP1 Development of studied pathosystems

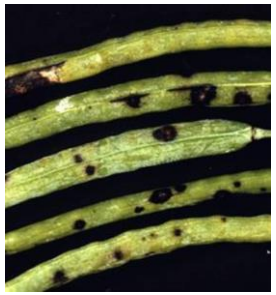
Nathalie NESI
Nathalie.nesi@inrae.fr

Selection of 8 cultivars per species

Plant





Susceptibility tests of genotypes against corresponding pathogens

Pathogens

Clavibacter *Pythium*	*Alternaria* *Rhizoctonia*	*Xanthomonas*	*Fusarium*

=> Use of contrasted susceptibility of genotypes versus pathogens

WP2 Alternative solutions:

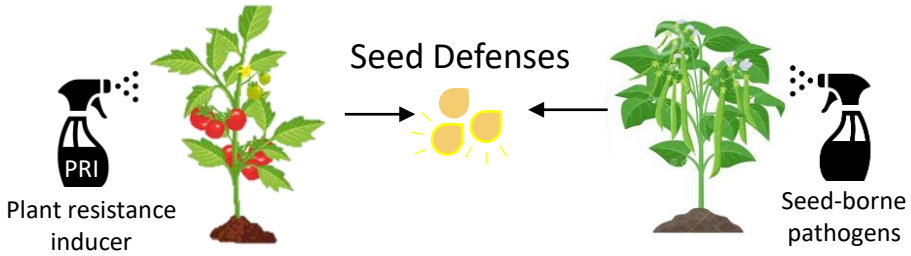
Jerome VERDIER
Jerome.verdier@inrae.fr

Exploring and understanding the complexity of SEED immunity / defense

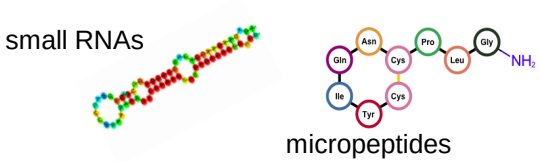
Development of pathosystems

ENHANCEMENT OF SEED DEFENSES

METHOD OLOGY



Expected SOLUTIONS



WP3 Alternative solutions:

Marie SIMONIN
marie.simonin@inrae.fr

Understanding and controlling the biotic environment of plants

Development of pathosystems

ENHANCEMENT OF SEED DEFENSES

SEED MICROBIOME ENGINEERING

METHOD OLOGY

Boosting Seed Defenses

Protection
Vigor

Core microbiota

Beneficial SynCom

Expected SOLUTIONS

small RNAs

micropeptides

SynCom

WP4 Alternative solutions:

Unravelling composition and functionality of seed exudates released during germination

Loic RAJJOU
loic.rajjou@agroparistech.fr

Development of pathosystems

ENHANCEMENT OF SEED DEFENSES

SEED MICROBIOME ENGINEERING

SEED LOCAL ENVIRONMENT ANALYSIS

METHOD OLOGY

Boosting Seed Defenses

Protection
Vigor

Core microbiota

Beneficial SynCom

Protection
Vigor

Expected SOLUTIONS

small RNAs

micropeptides

SynCom

proteins

metabolites

WP4 Seed formulation

Helen NORTH
helen.north@inrae.fr

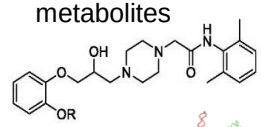
Patrick SAULNIER
patrick.saulnier@univ-angers.fr

Development of pathosystems

small RNAs

micropeptides

metabolites



SynCom

proteins

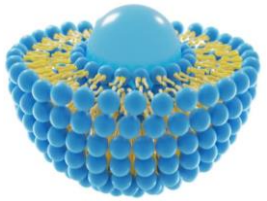
Alternative solutions

Seed formulation: promote molecular stability and bioavailability

"Classical" seed formulation

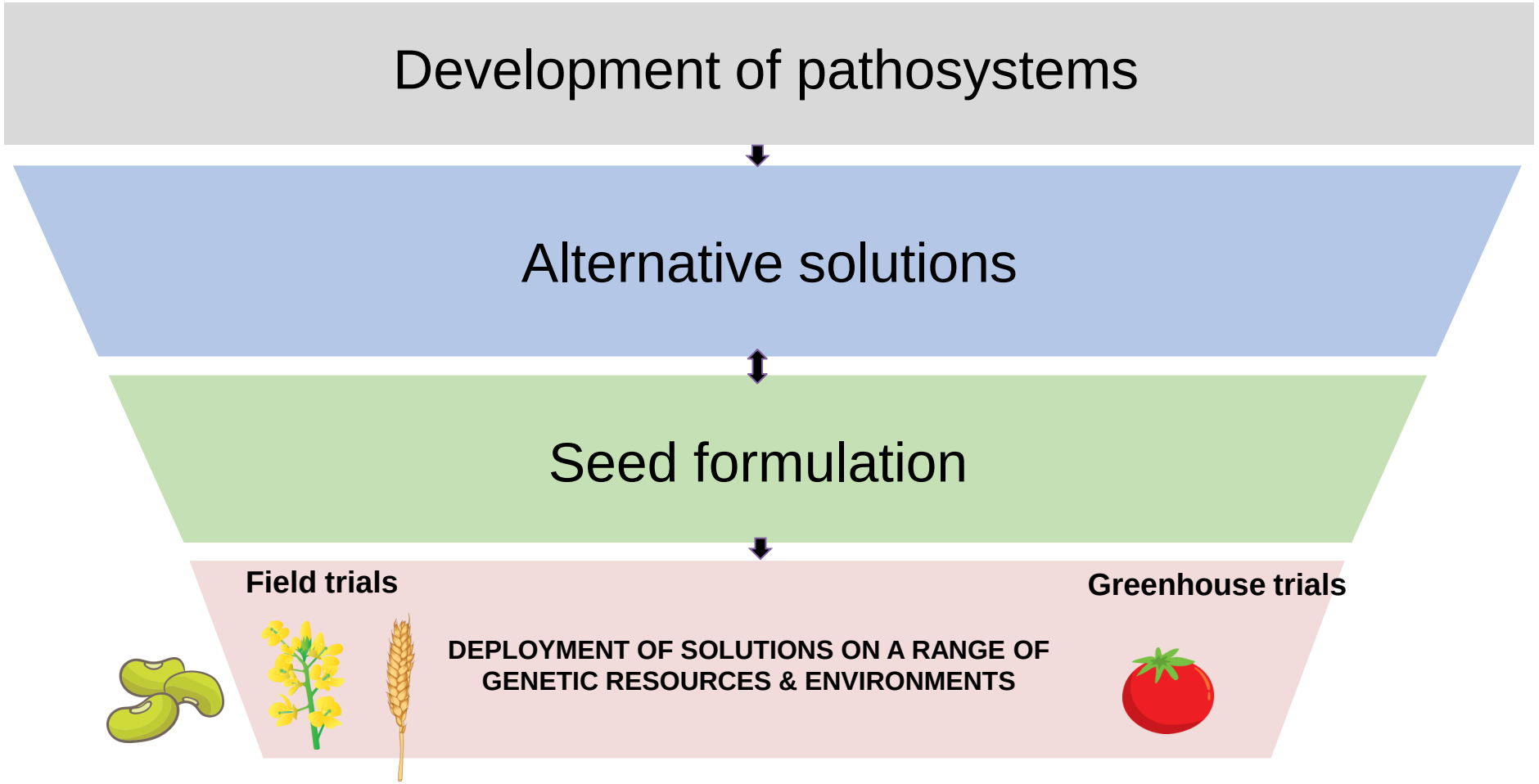
Novel pharmaceutical formulation
(emulsion and nanoencapsulation)

(Derived from human health)



WP5 Field validation

Thierry LANGIN
Thierry.langin@inrae.fr



Human and social sciences (HSS)

Rose Marie BORGES
r-marie.borges@uca.fr

Identify the obstacles to the development

↓

Inform public and political decisions

Regulations

Market players

Survey

Development of pathosystems

Alternative solutions

Seed formulation

Validation

REGULATORY ECONOMIC & SOCIAL CONTEXTS ANALYSES

Education / Management

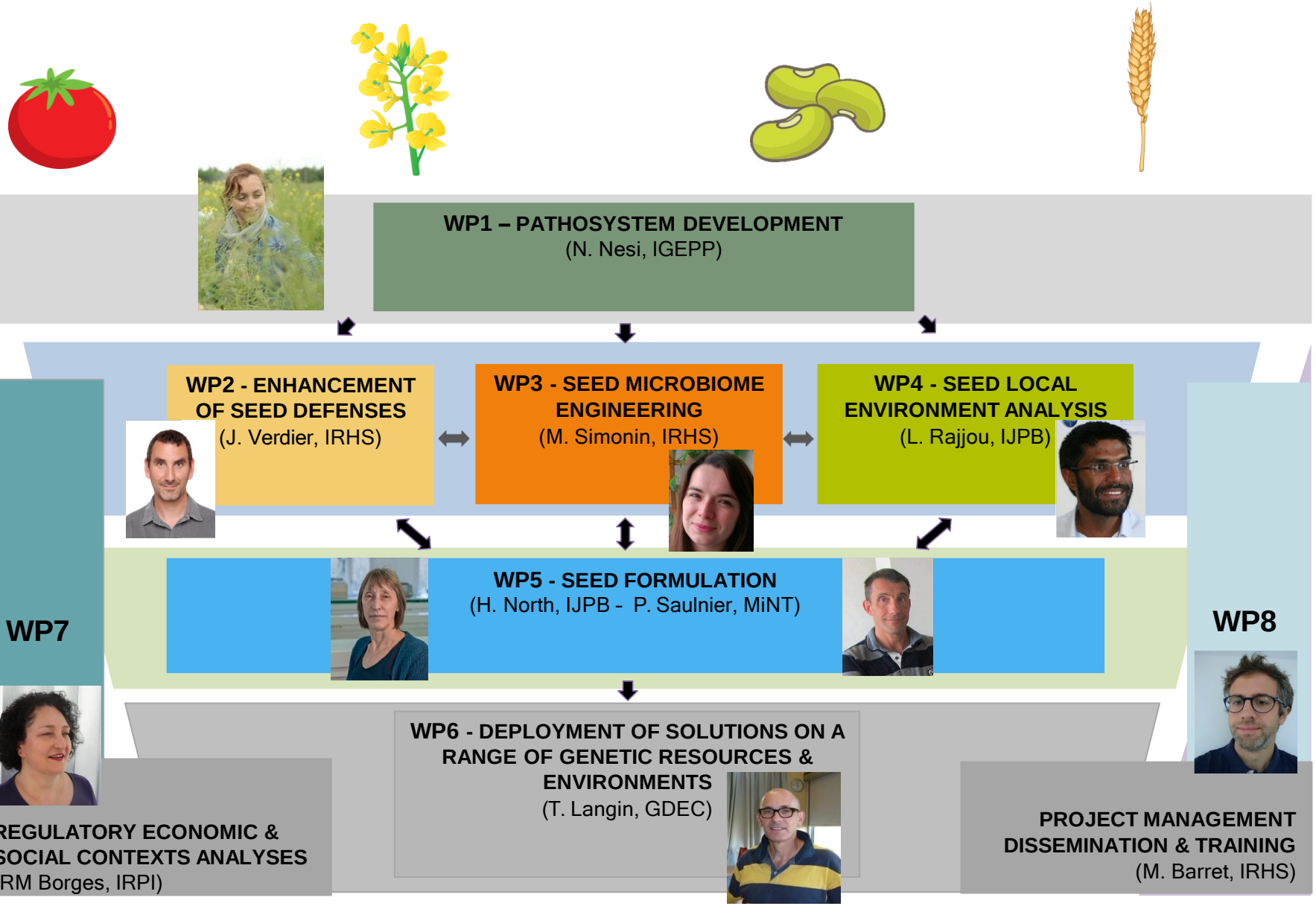
université angers

AgroParisTech

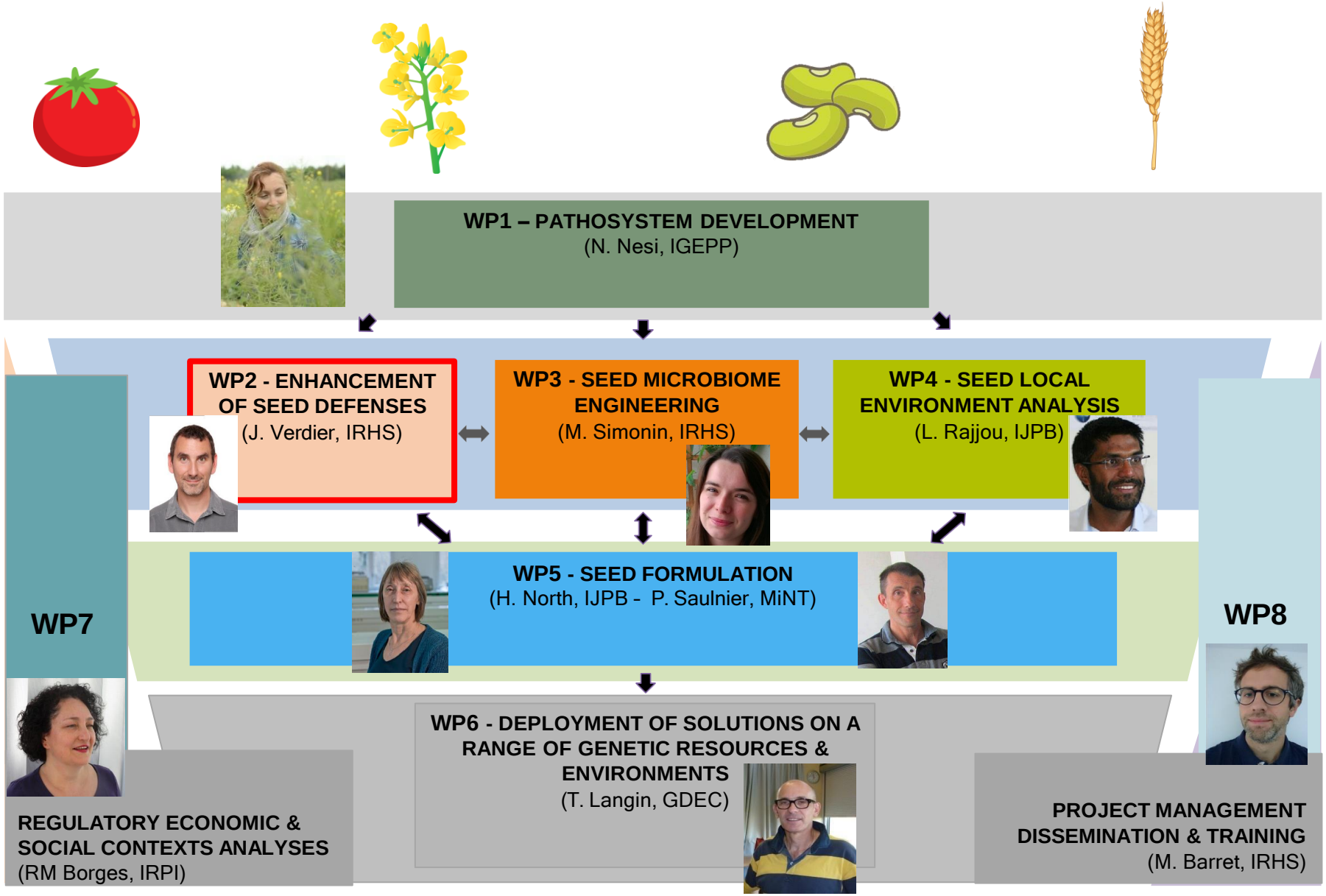
University Angers & AgroParis tech

=> development of modules dedicated to seed defenses

SUCSEED Current Status (in the 2nd year out of 6 years)



SUCSEED Current Status (in the 2nd year out of 6 years)



WP2: Boosting Seed defense
Example of Bio(seed) priming

Seed priming => increase seed germination vigor and homogeneity

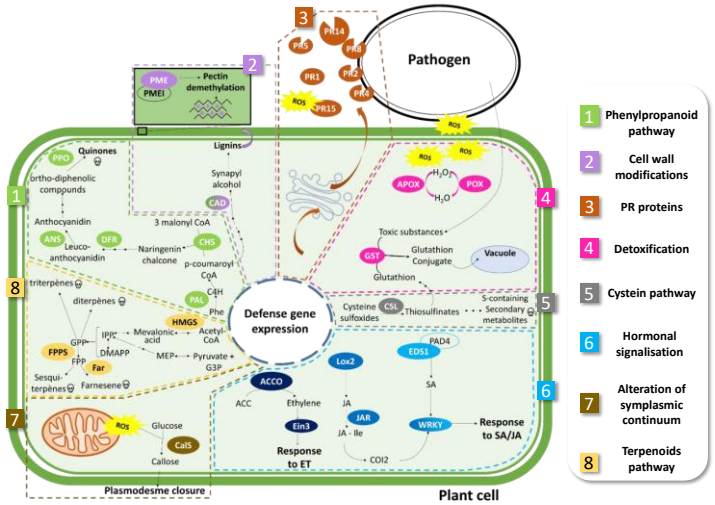
HYDRO- or
OSMO-priming

+

Defense priming => prime defense

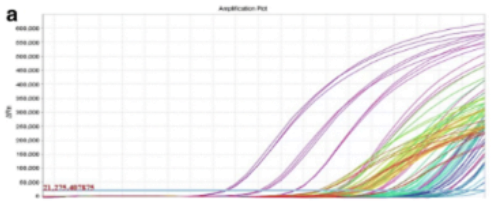
Tested PRIs (Plant Resistance Inducers)
⇒ MeJA, INA, BABA, Bion®,….

Can PRI induce Defense priming in SEED?



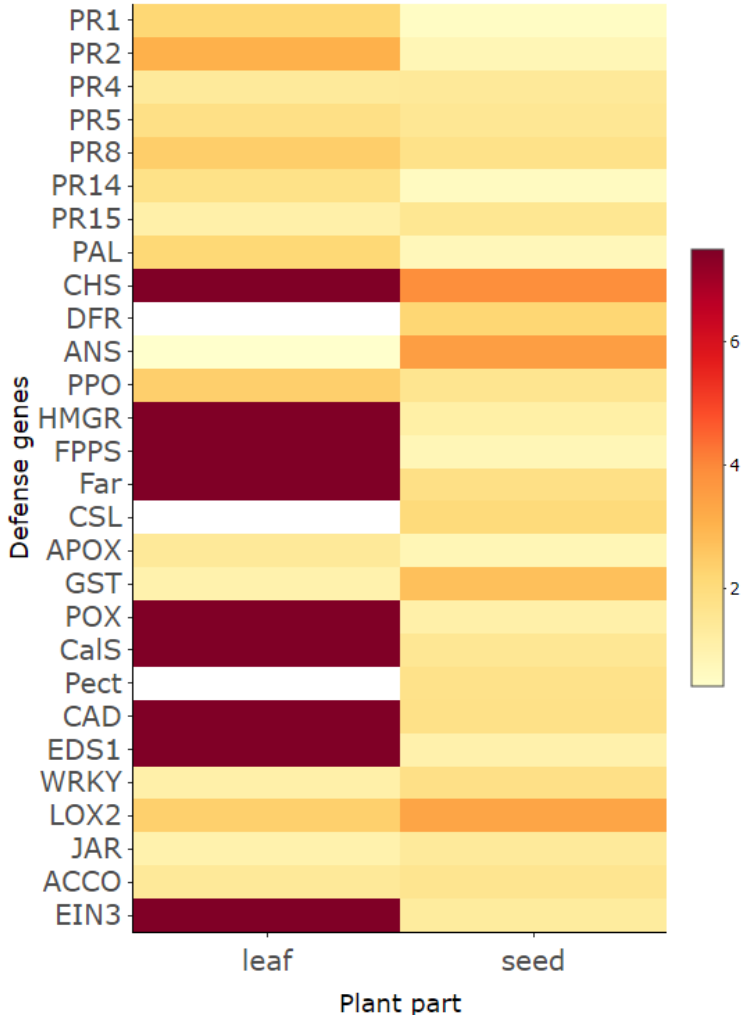
28 genes representing all defense pathways

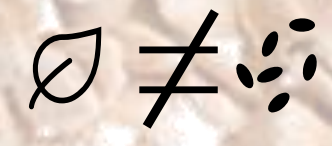
qPCR (qPFD®)



MN Brisset (IRHS)

PRI1 treatement on tomato tissues:



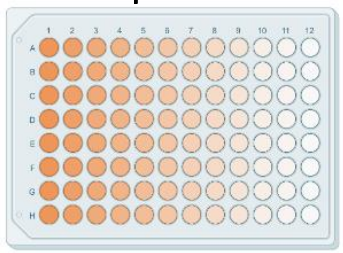


=> Yes, Activation of different defense pathways in seeds

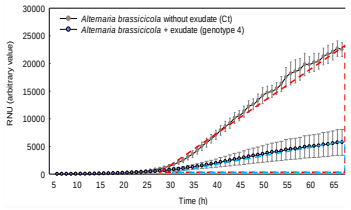
Quantitative impact of PRIs on seed defense

Fungi (*Alternaria Brassisicolae*)

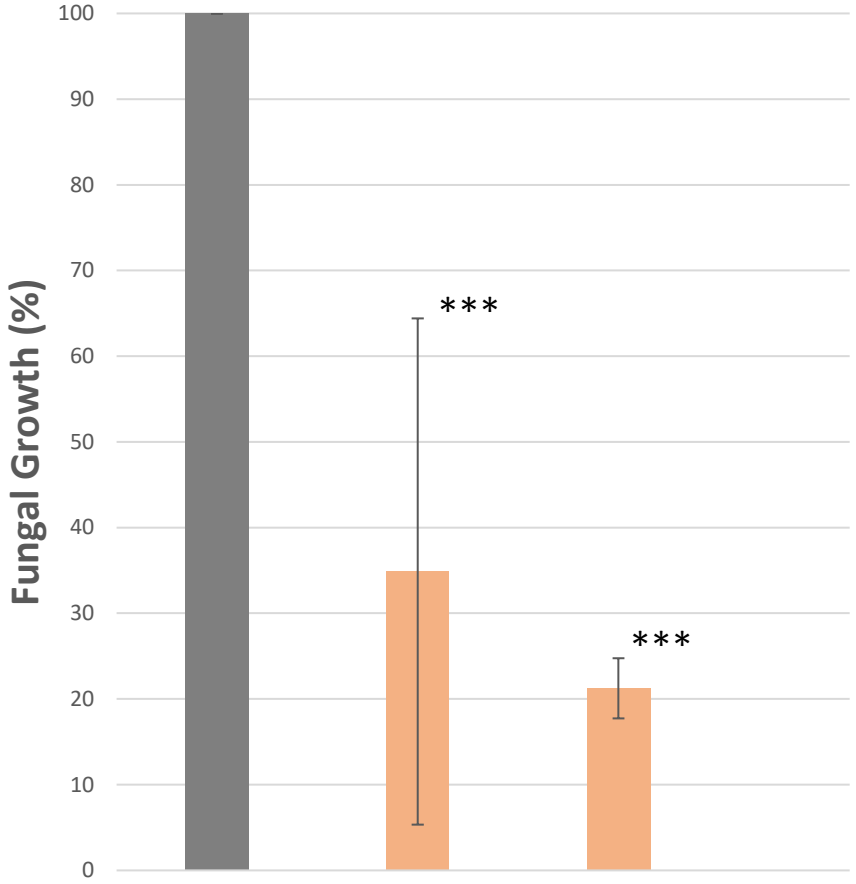
Bio-primed seed exudates



Nephelometry

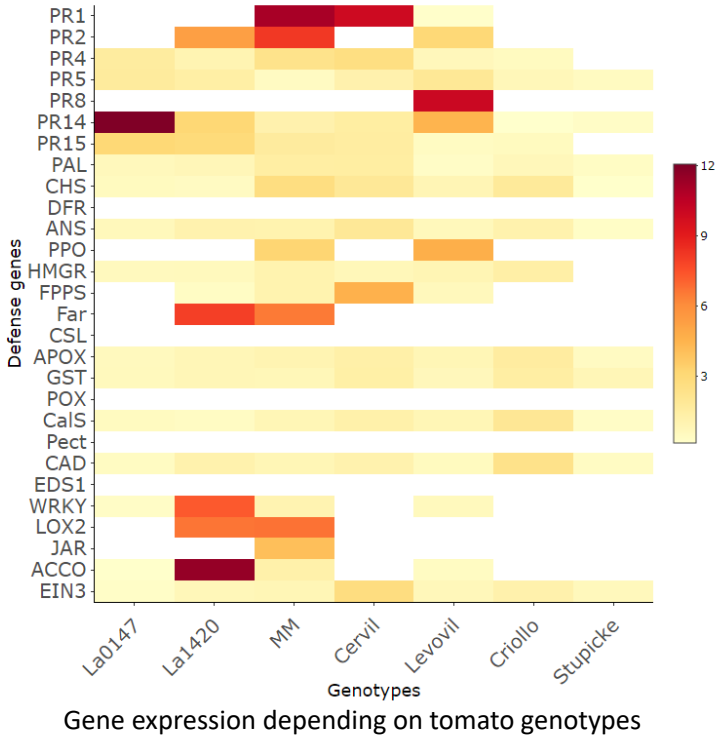


= fungal growth



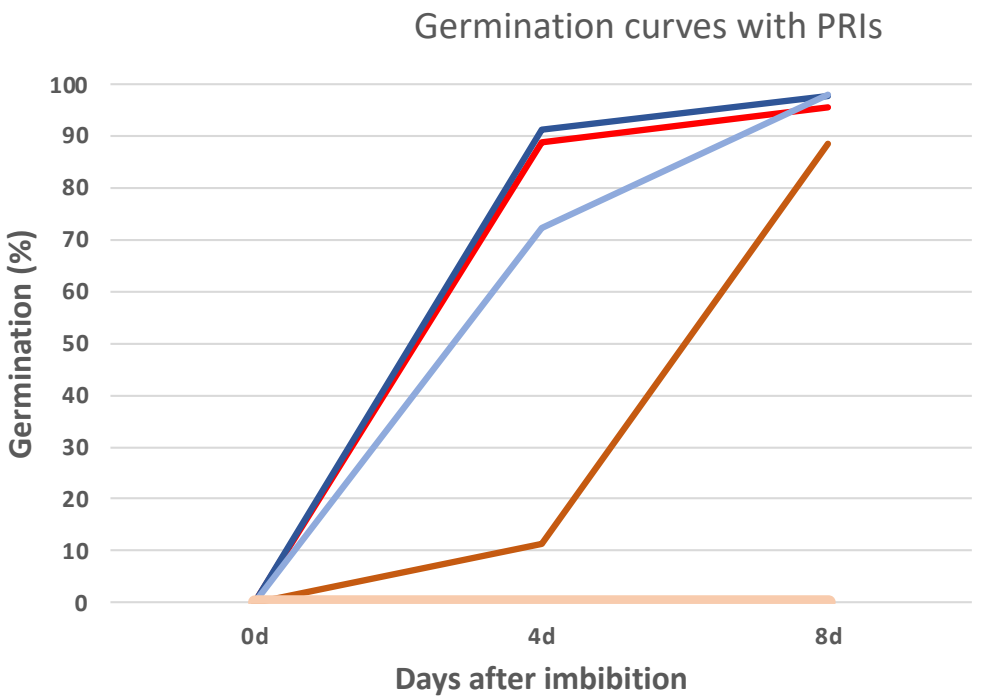
Genotype	Tomato Genotype 1	
Treatment	No PRI	Seed-treated by PRI2
Fungal concentration	+++	+

Can we directly use of PRI to boost seed defenses?

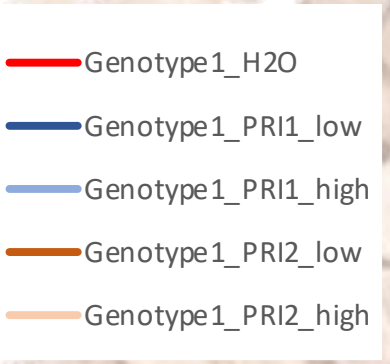


Gene expression depending on tomato genotypes

1- genotype-dependent



2- delayed germination

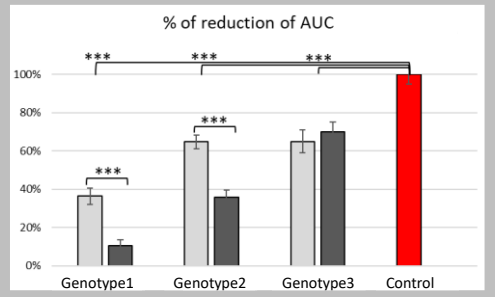


3- Dosage dependent
4- environment dependent
.....

Not really.... need to identify downsteam molecules that are induced by PRIs and involed in seed defense

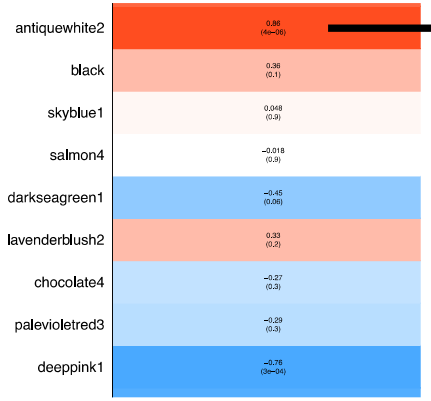
Identification of defense molecules induced by PRI in seeds

Contrasted efficiency of PRI in different genotypes



RNAseq analysis

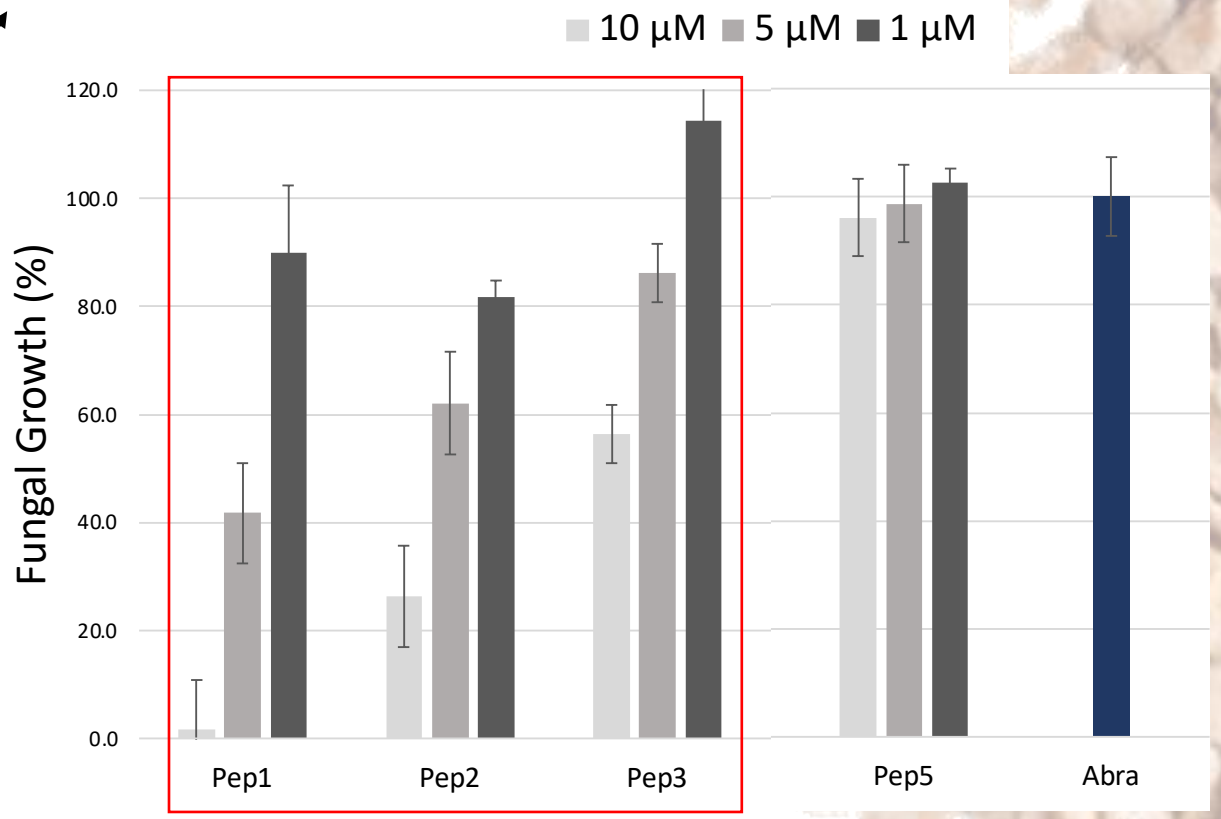
Gene Correlation Network analysis



Genes/molecules highly correlated with activation of defense

(Micro)peptides

Synthetic peptides biomimicking plant peptides identified in seed defense



....seeds produced peptides following PRI treatment

=> peptides use as biocontrol solution coated to seeds

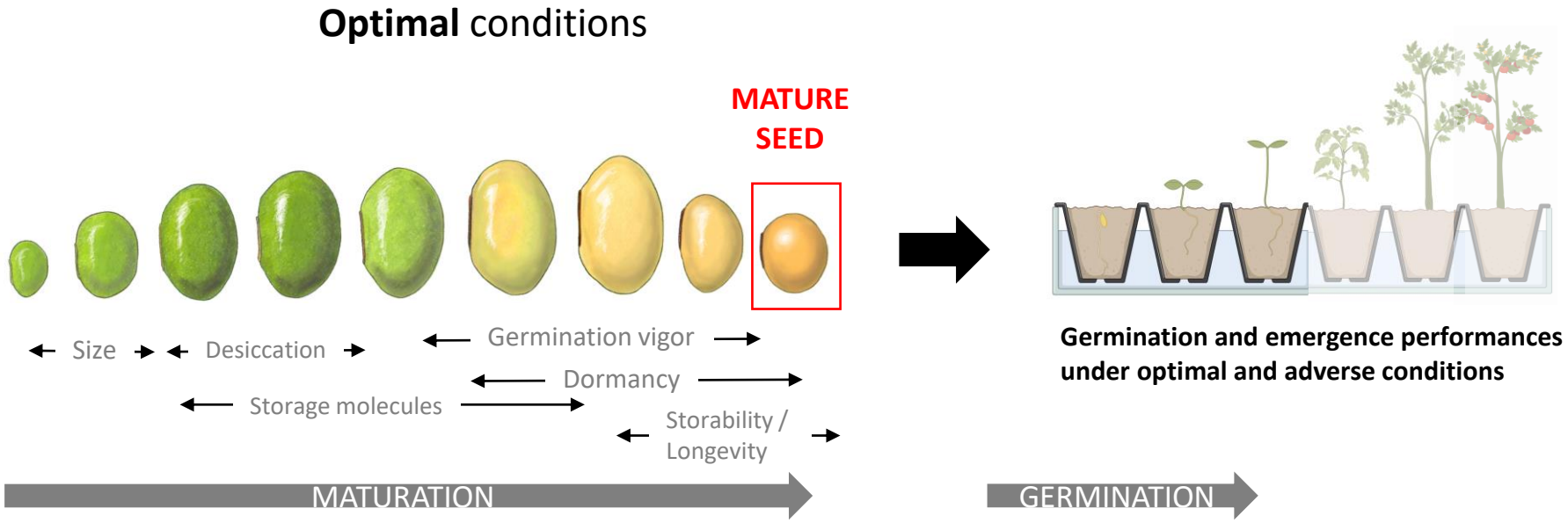
Currently tested on a wide range of host and non-host pathogens

Research team located in Angers (France)

IRHS Seed Lab

SEED lab (Seed, Epigenetic, Environment, Development)

1. Understanding SEED MATURATION PROCESSES involved in **seed qualities**

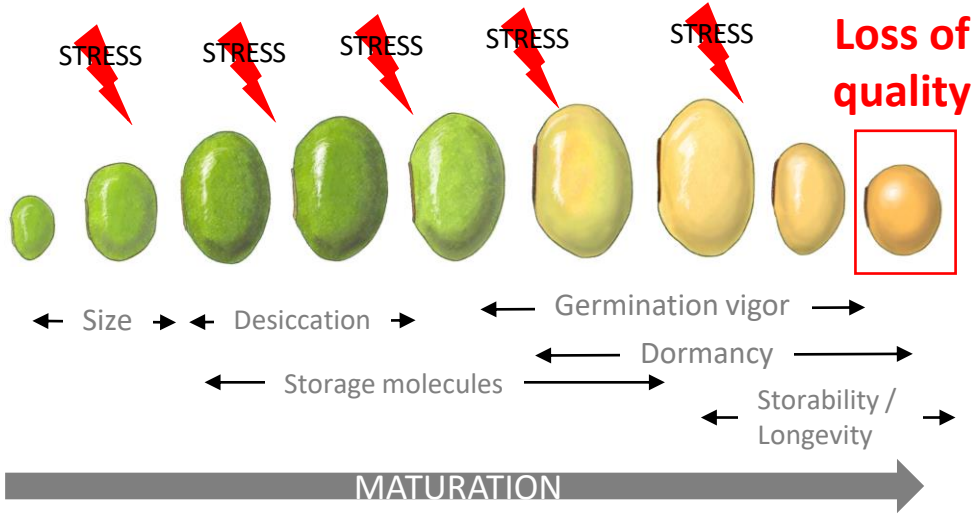


Research team located in Angers (France)

IRHS Seed Lab

SEED lab (Seed, Epigenetic, Environment, Development)

1. Understanding SEED MATURATION PROCESSES involved in **seed qualities**
2. Understanding SEED RESPONSES to BIOTIC and ABIOTIC stresses (=> bio-sourced biocontrol/biostimulation)



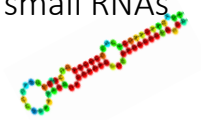
IMPACT on Germination and emergence performances

GERMINATION

Solutions:

Genetic Engineering

small RNAs



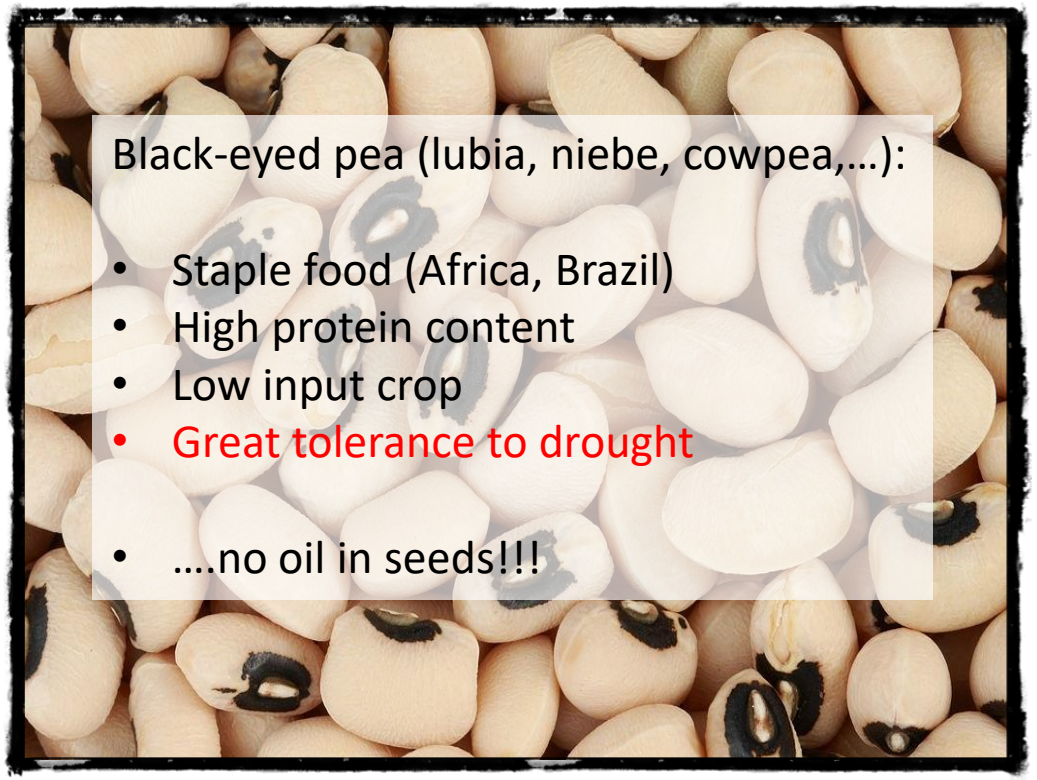
micropeptides

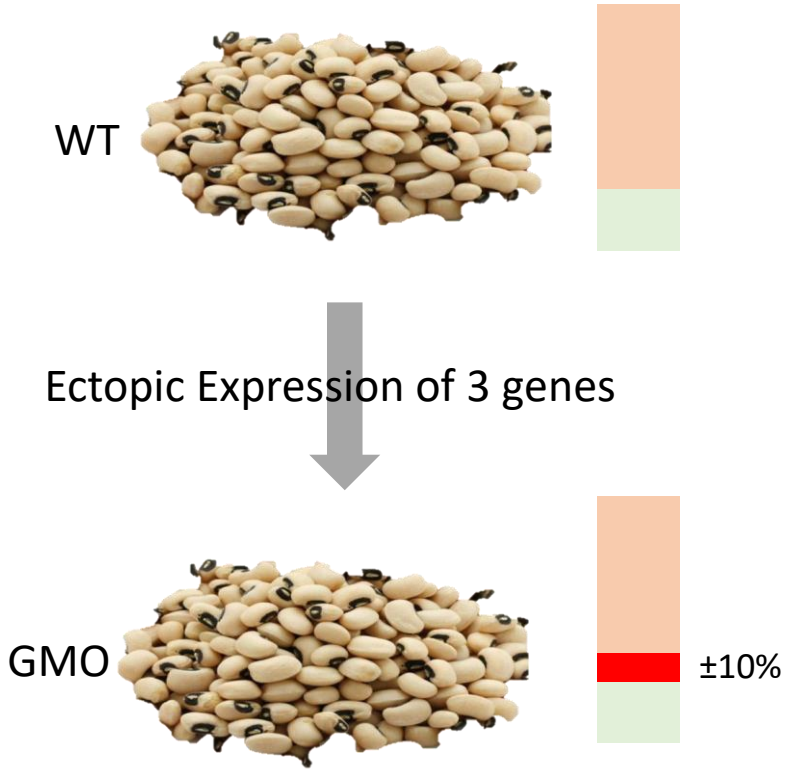
Easy/cheap synthesis

Current studied plants : many Legumes (beans, soybeans,…), tomato, Arabidopsis,…

Using genetic engineering (1)

how to improve seed nutritional quality

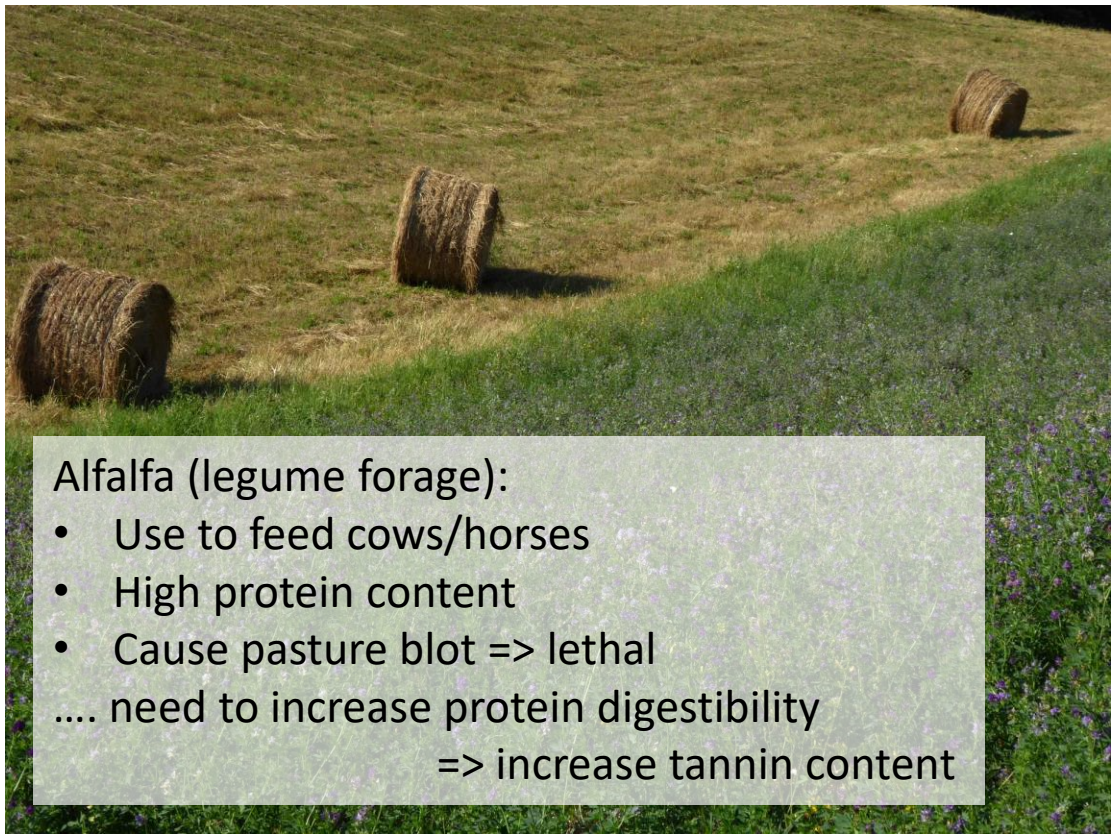




Collaboration with CSIRO (Australia), philanthropic project

Using genetic engineering (2)

how to use seed characteristics to improve plants



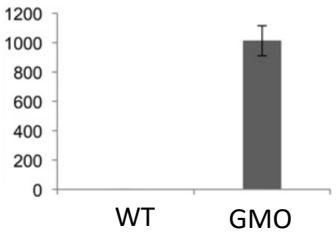
Alfalfa (legume forage):

- Use to feed cows/horses
- High protein content
- Cause pasture blot => lethal

…. need to increase protein digestibility
=> increase tannin content

Leaves = no tannin

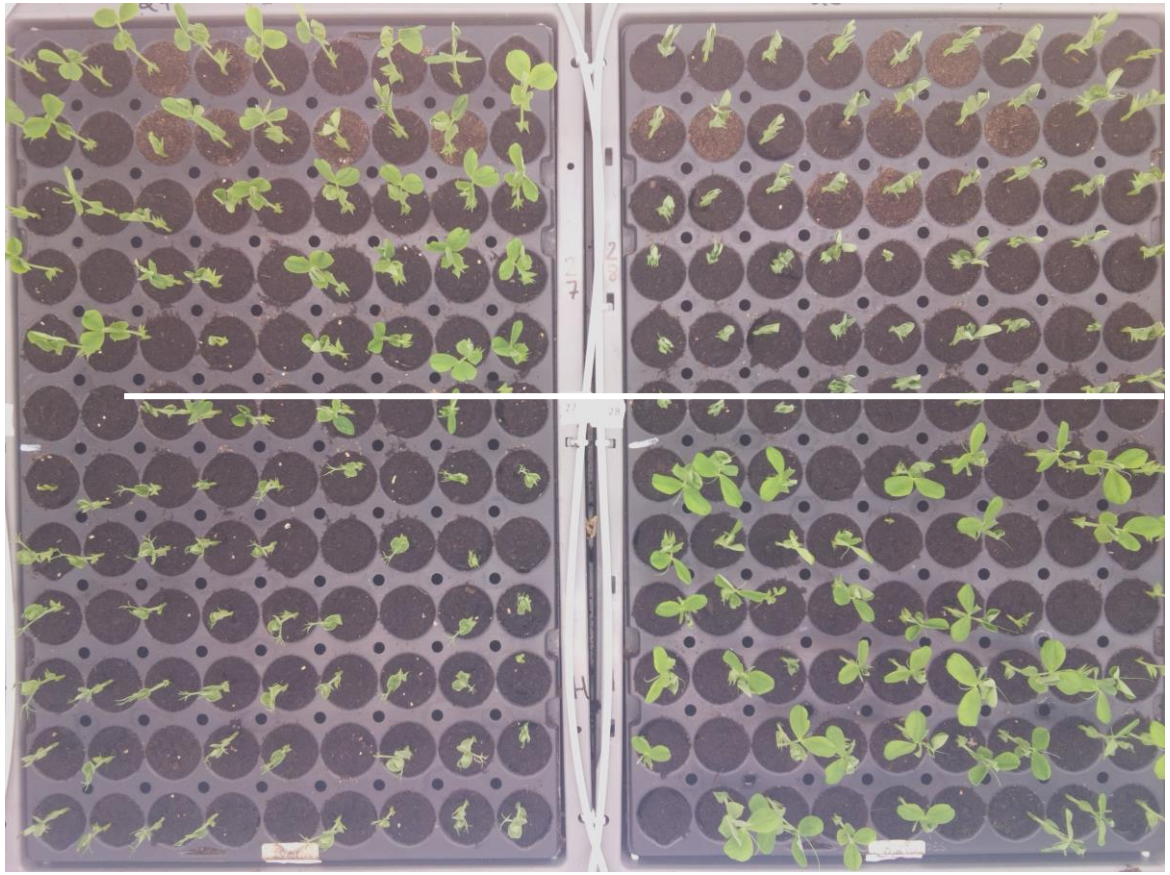
Seeds = high tannin

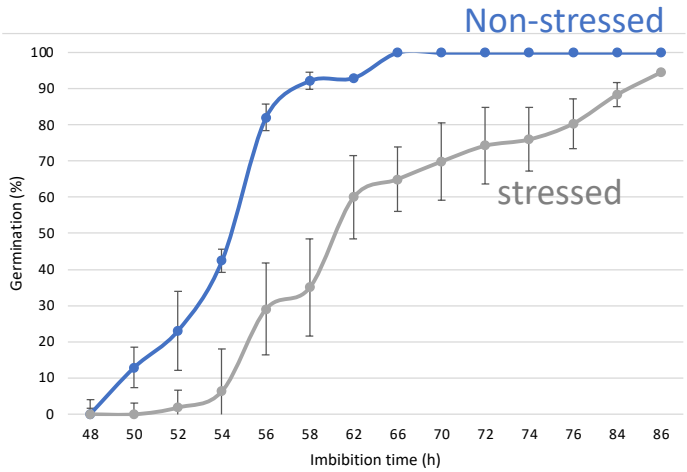


Collaboration with Forage Genetics (USA)
Verdier et al. 2012 (PNAS), Patents (2012, 2014)

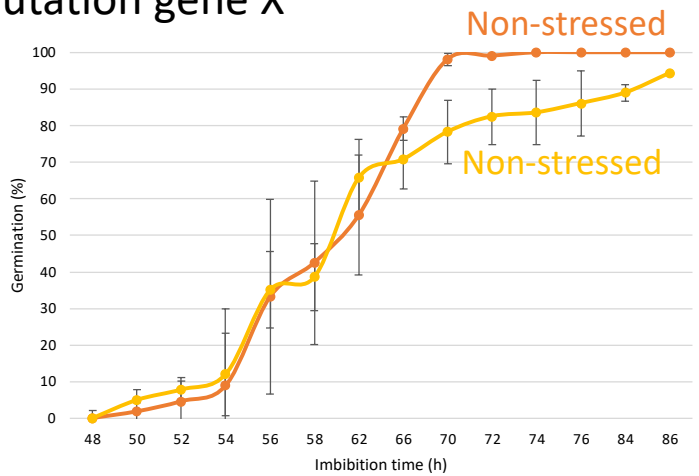
How to attenuate loss of germination vigor due to stress?

Low-quality seed lot produced under stress conditions





Mutation gene X



Heterogeneity of germination

=> asynchronous plant development

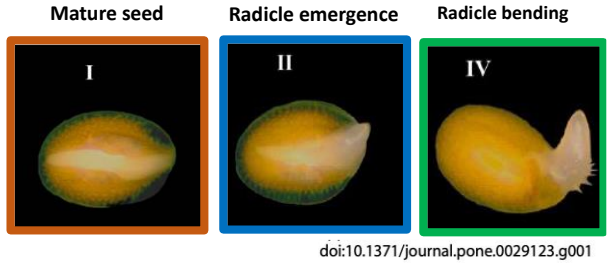
Identification of a **molecule to reset the impact of heat stress during seed production**… testing in progress...

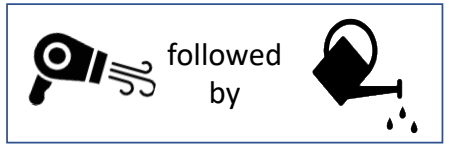
Declaration of invention (DI-RV-20-0020) INRAE 2023

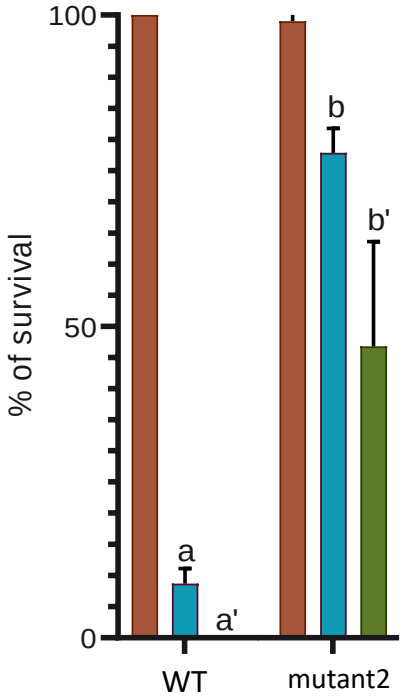
How to make seedling more tolerant to heat/drought stress?

Germination is very sensitive to drought…

….. leading to seedling death => loss of yield!!







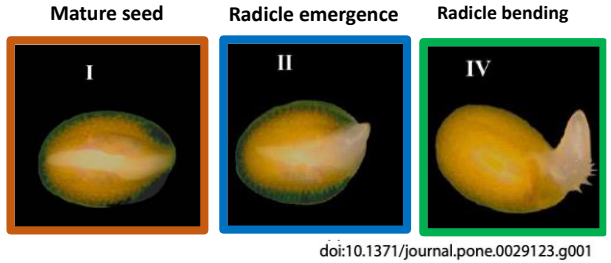
=> regulated via endogenous peptide (17aa)

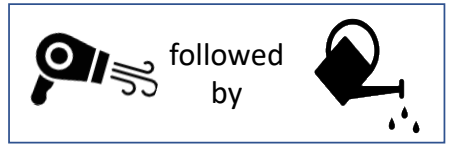
Patented by INRAE (Sept 2023)

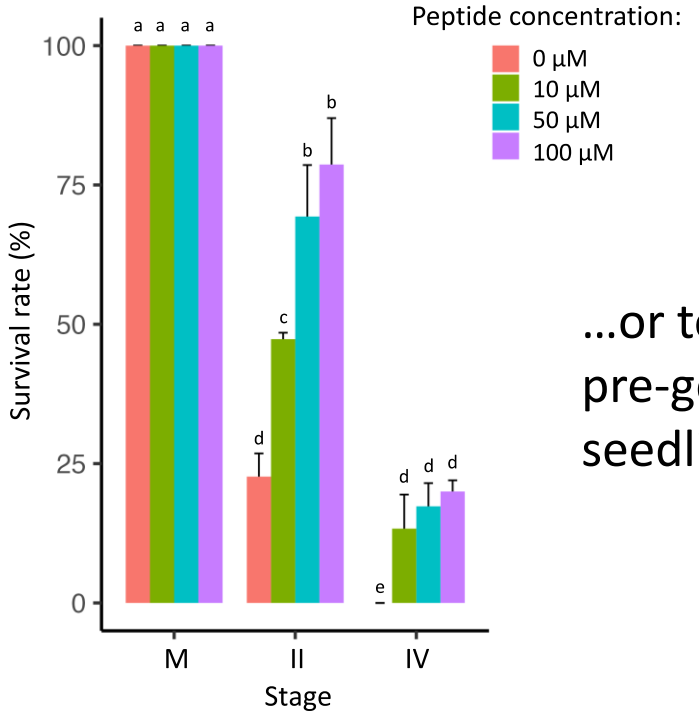
How to make seedling more tolerant to heat/drought stress?

Germination is very sensitive to drought…

….. leading to seedling death => loss of yield!!



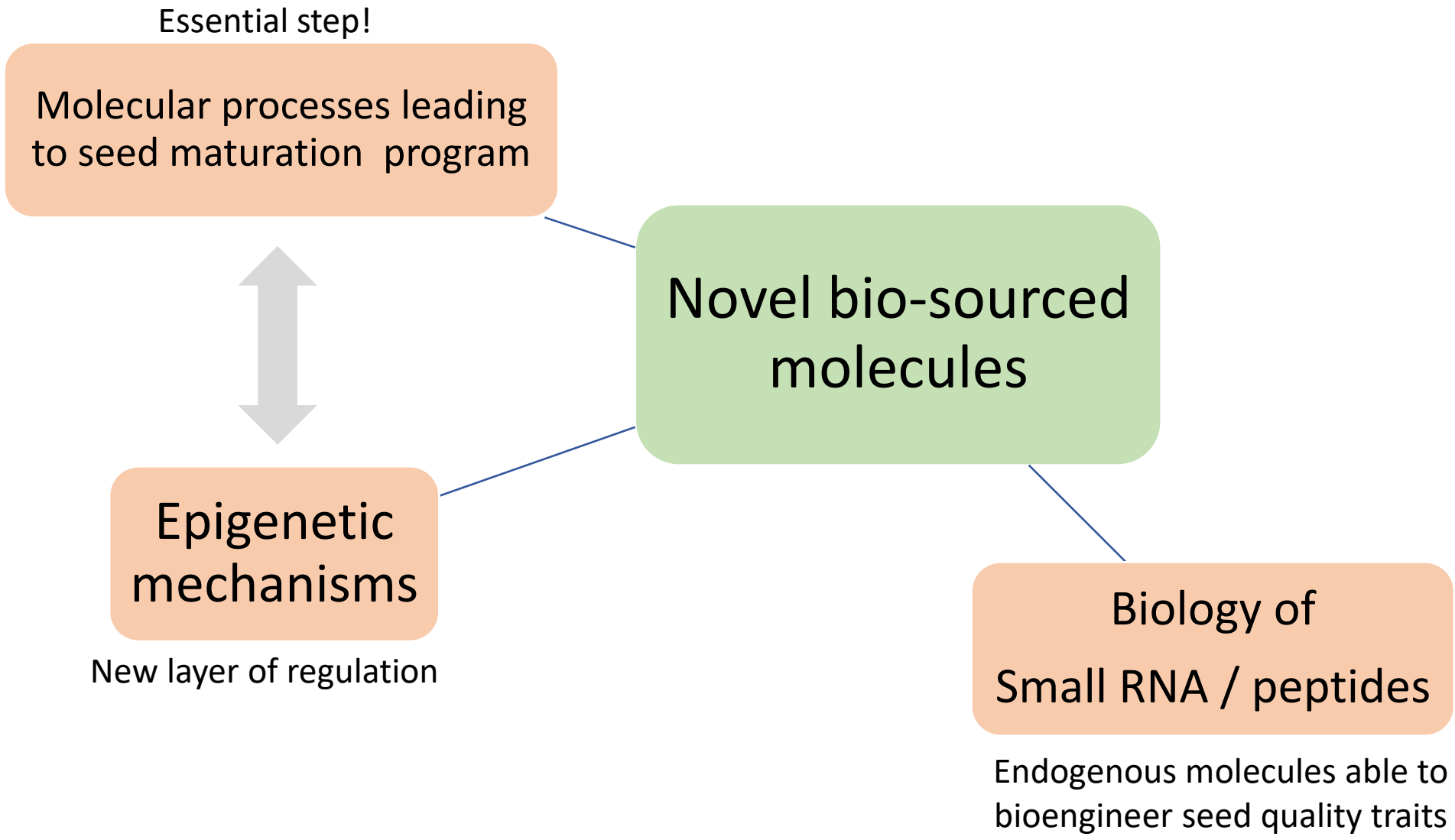




…or to pellet dried pre-germinated seedling ?

Patented by INRAE (Sept 2023)

Take Home Message



Thank you for your attention!

All the members of the SEED team (IRHS Angers)

Thanks to ….

All the collaborators of the **SUCSEED** project from IRHS, IJPB, GDEC, GEVES, MiNT,….

We Will SUCSEED

Contact: jerome.verdier@inrae.fr

16 Partners - *Service providers

ACADEMIC PARTNERS

- Institut de Recherche en Horticulture et Semences (Angers)
- Institut Jean-Pierre Bourgin (Versailles)
- Génétique Diversité Ecophysiologie des Céréales (Clermont Fd)
- Institut de Génétique, Environnement et Protection des Plantes (Rennes)
- Institut des Sciences des Plantes - Paris-Saclay (Paris)
- Institut de Biologie de l'École Normale Supérieure (Paris)
- Micro et Nanomedicine translationnelle (Angers)
- Génétique et Amélioration des Fruits et Légumes (Avignon)
- Science Action Développement - Activités Produits Territoires (Paris)
- Institut de Recherche en Propriete Intellectuelle (Paris)
- Clermont Recherche Management (Clermont Fd))

ASSOCIATIONS

Union Française des semenciers

Syndicat professionnel des supports de culture, fertilisants organiques et biostimulants

International Biocontrol manufacturer association

* Fédération Nationale des Agriculteurs Multiplicateurs de Semences

Groupe d'Etude et de contrôle des Variétés Et des Semences

PRIVATE SECTOR

- FRAYSSINET
- * iMEAN
- * KAPSERA
- * MilliDrop

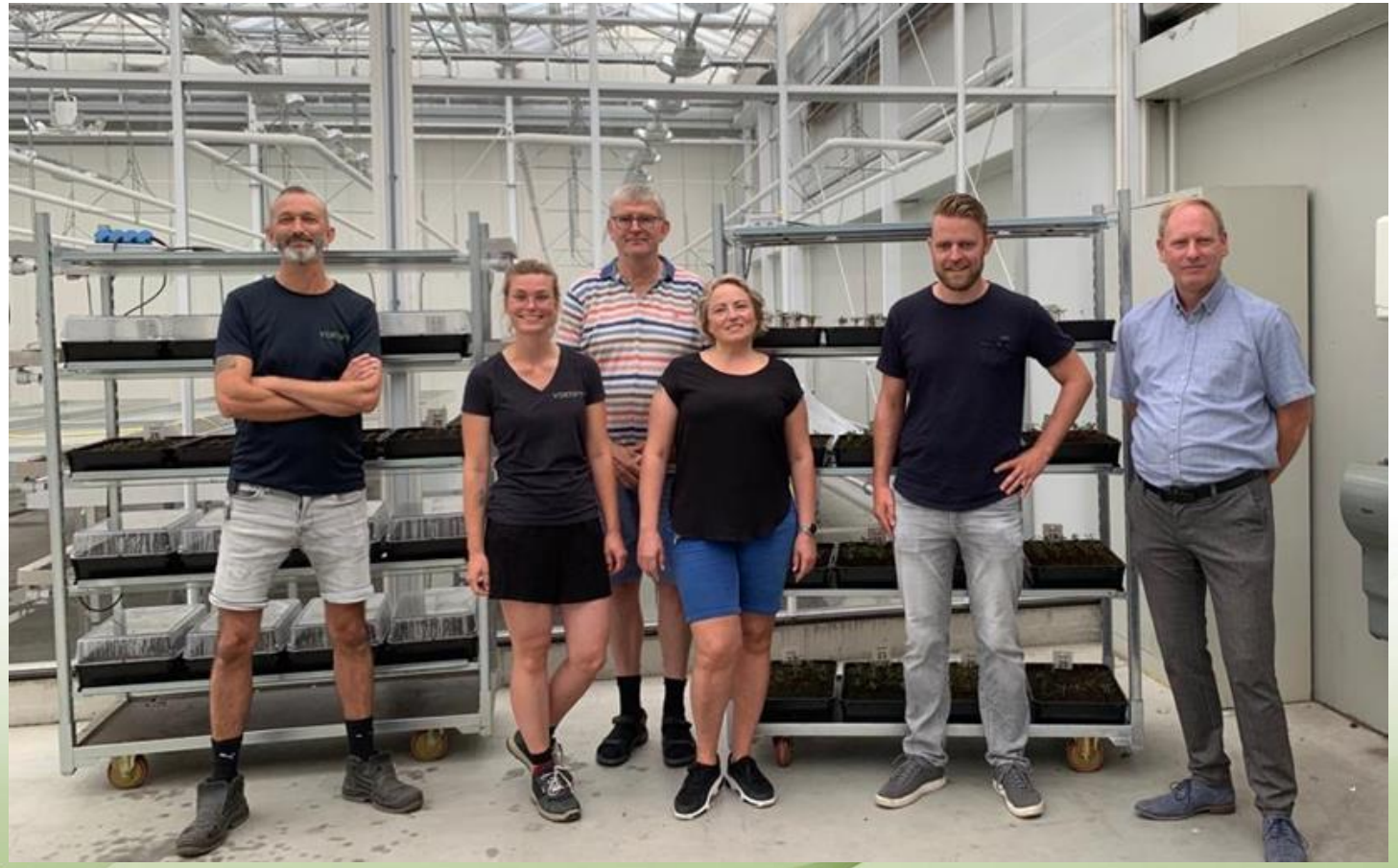
Groen op Zaad

Symposium ‘Future proof seeds: innovative crop protection solutions‘

Seed meets Technology

28-09-2023

Introduction: the team



Remco Heijne,
Renske Reus,
Frans Tetteroo,
Olga Bot-Matveyeva,
Jasper Schermer,
Cor Oostingh.

Consortium of partners to come closer to sustainable and resilient seedlings

- 4-year project (2021-2024)
- Private Public Partnership
- 50% funded by consortium partners
- 50% subsidy of the Dutch Government (TKI)
- In kind contributions of partners

Why?

- Gain knowledge on sustainable protection of seedlings
- Determine alternatives for conventional seed coating treatments
- Development of consistent tests for further testing on wide genetic varieties
- Combining knowledge and experience based on model crop/pathogen combinations

How?

Work Packages are defined:

1. Evaluation of worldwide possible seed treatment opportunities
2. Defining test protocols and evaluation of alternatives on protection of seedlings
3. Evaluation of efficacy on
   - Genetic variation
   - Commercial applicable coatings
   - Direct and indirect seeding crops

In kind contributions:

- Seed companies: seeds, quality testing and CFU counts
- Incotec and Centor: Seed treatments
- Vertify and IRS: Testing facilities

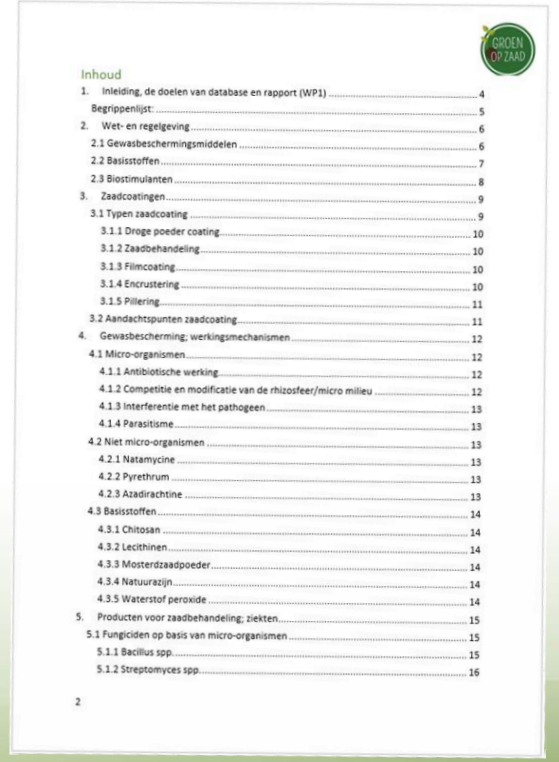
Possible Low Risk opportunities

- Wide range of bacteria from natural sources with fungal and insecticide efficacy (single strain)
  - Bacillus spp (f+i)
  - Pseudomonas spp (f)
  - Streptomyces spp (f+i)
- Wide range with fungi from natural sources with fungal and insecticide efficacy (single strain)
  - Trichoderma spp. (f)
  - Saccharomyces spp. (f)
  - Pythium spp. (f)
  - Clonostachys spp. (f)
  - Beauveria spp. (i)
- Basic substances
- Extracts from plants
- Organic compounds

Work package 1: Literature study

- Registration process
  - EU regulation 1107/2009
  - EFSA
  - CtgB (NL)

- Seed application techniques (esp. for use with micro-organisms)
- Mode of action of actives
  - Antibiotic
  - Competition in rhizosphere
  - Interference
  - Parasitism
- Wide range single strain micro-organisms



Inhoud

Work package 2:

- Focus on soil born diseases

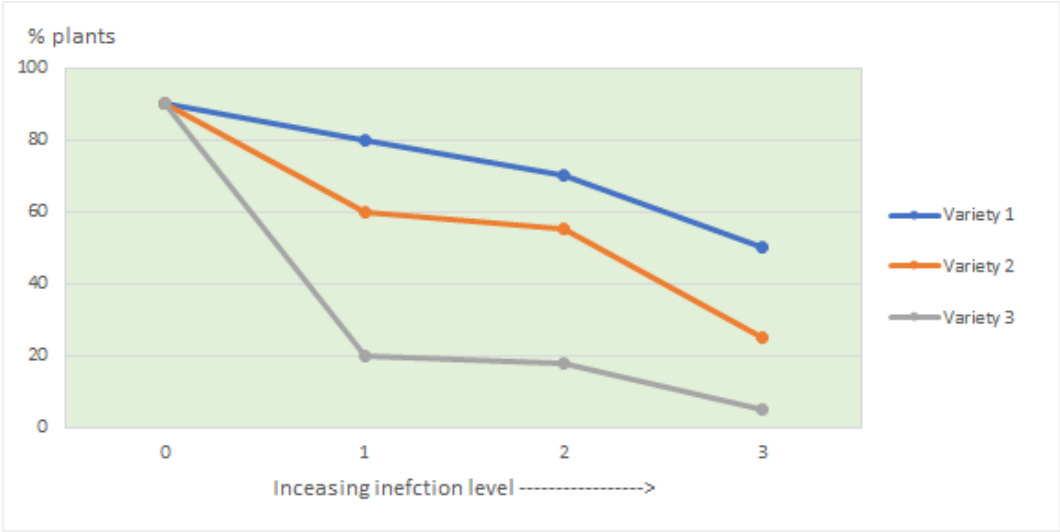
Pathogen	1st test layer (climate room)	2nd test layer (climate room/greenhouse)
Rhizoctonia	Lettuce	Cauliflower
Pythium	Red beet	Sugar beet
Pythium	Spinach	Gherkin
Fusarium	Spinach	Lettuce

Work package 2

Protocol development

- Strains to use
- Acceptable disease pressures
- Genetic variation

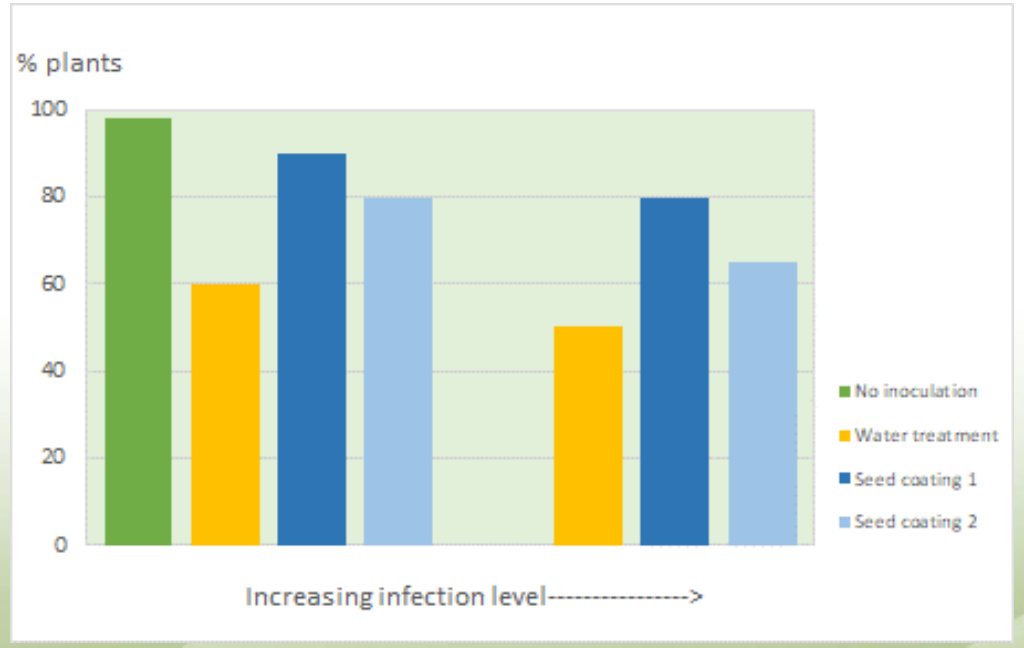
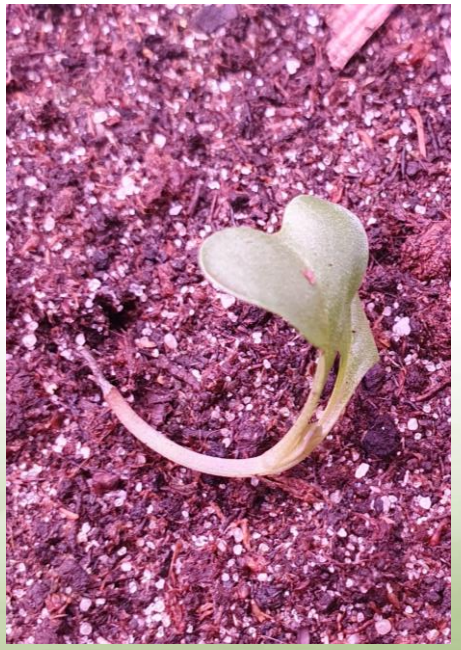
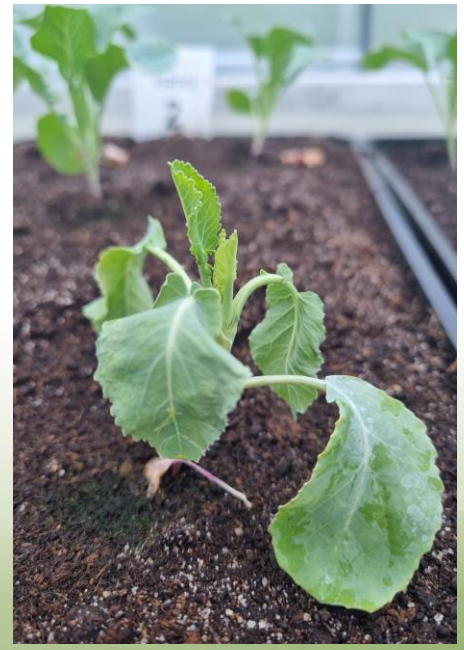
Evaluation of active ingredients



AG 4 AG 1-1B AG 2-2T AG 2-1 AG 3

Work package 3

- Commercial applicable coatings
- Direct / indirect seeding
- Use three best active ingredients


% plants
100
80
60
40
20
0
No inoculation
Water treatment
Seed coating 1
Seed coating 2
Increasing infection level---------->



What we have learned so far:

Many factors are critical for success:

- Genetic variation → susceptibility of varieties for individual situations
- Coating techniques → adapt to product type
- Products → control, prevent or suppress, quality
- Consistency of trial results → caused by …

Future proof seeds:

Genetic variation

Crop protection

Biostimulants

Storability

Product Quality

Coating techniques

Growing substrate

Nursery

Legislation

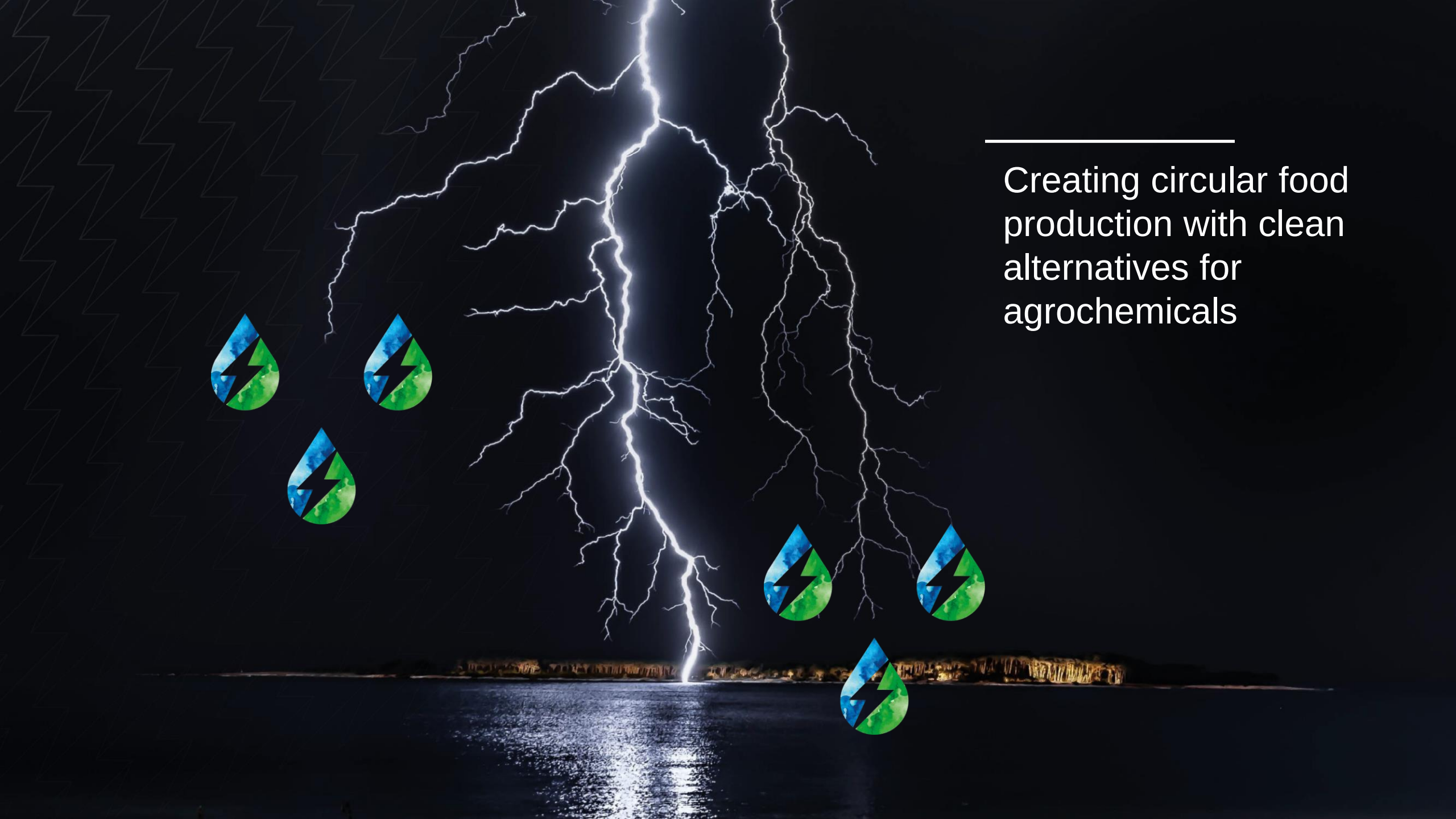
Crop rotation

Seed space

VitalFluid

Global Leader in
Plasma Activated Water
Applications

- EU active substance - 322mkg/yr
- Resistence, resurgence
- Huge biodiversity loss
- Residues in water and on food



Creating circular food production with clean alternatives for agrochemicals

History

The foundation of VitalFluid

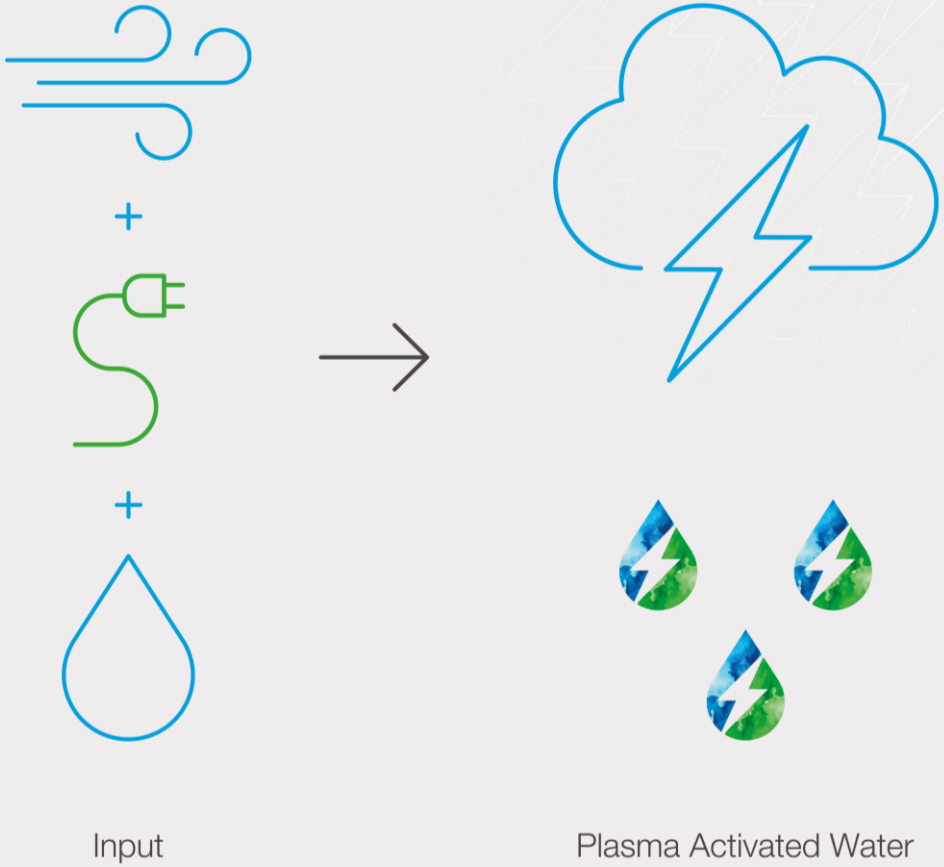
- Founded in 2014 by Paul Leenders and Polo van Ooij, Robertjan Zonneveld joined as investor in 2019
- From to TU/e -> Garage box -> HTC
- From 1 FTE (2018) -> 10 FTE (2019) -> 30 FTE (2022)
- First commercial deal 2021, now 10 machines sold
- Initial funding with grants EU Horizon2020 and REACT EU, backed by investment (BOM and VDL)

VitalFluid enables sustainable agriculture

Plasma Activated Water – Lightning in a box

Using the electric force of nature

- Inputs air, water and electricity
- Temporary disinfecting properties



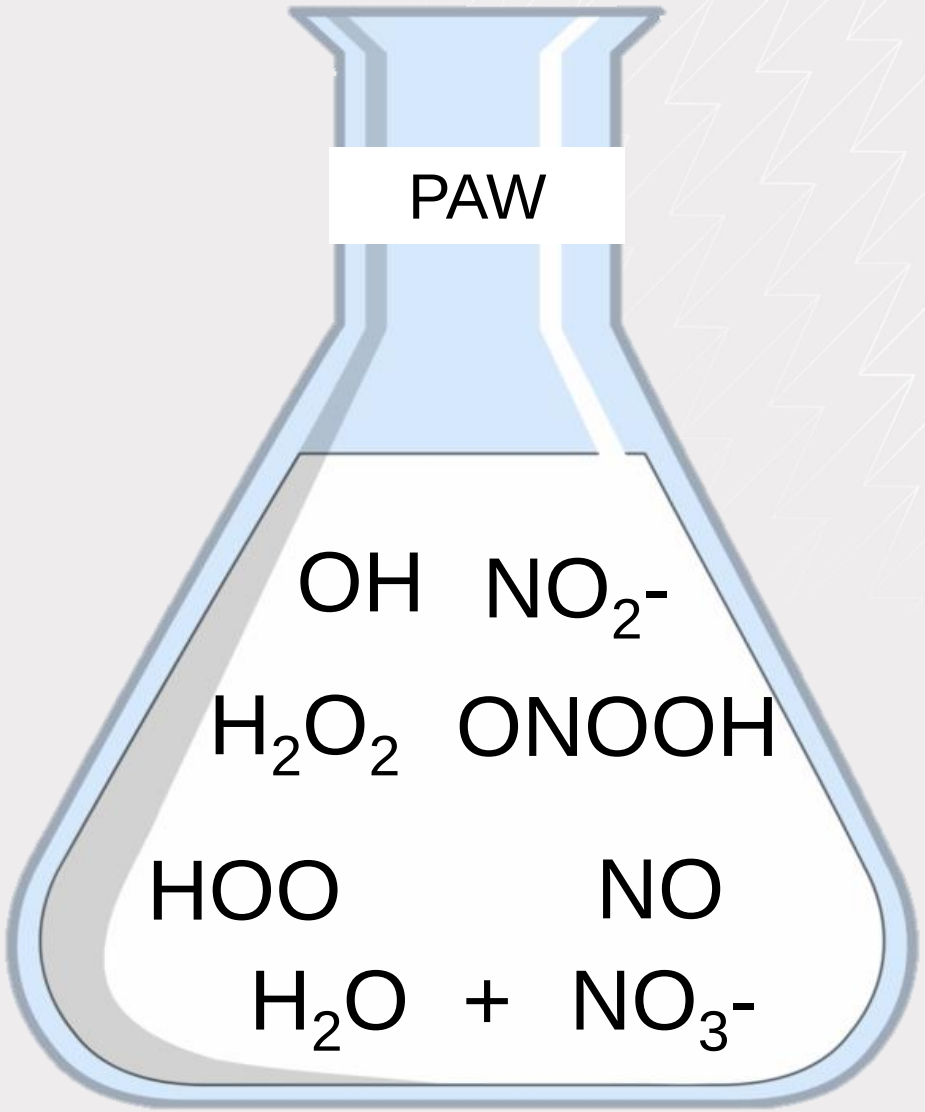
Sustainable

Not harmful to the climate

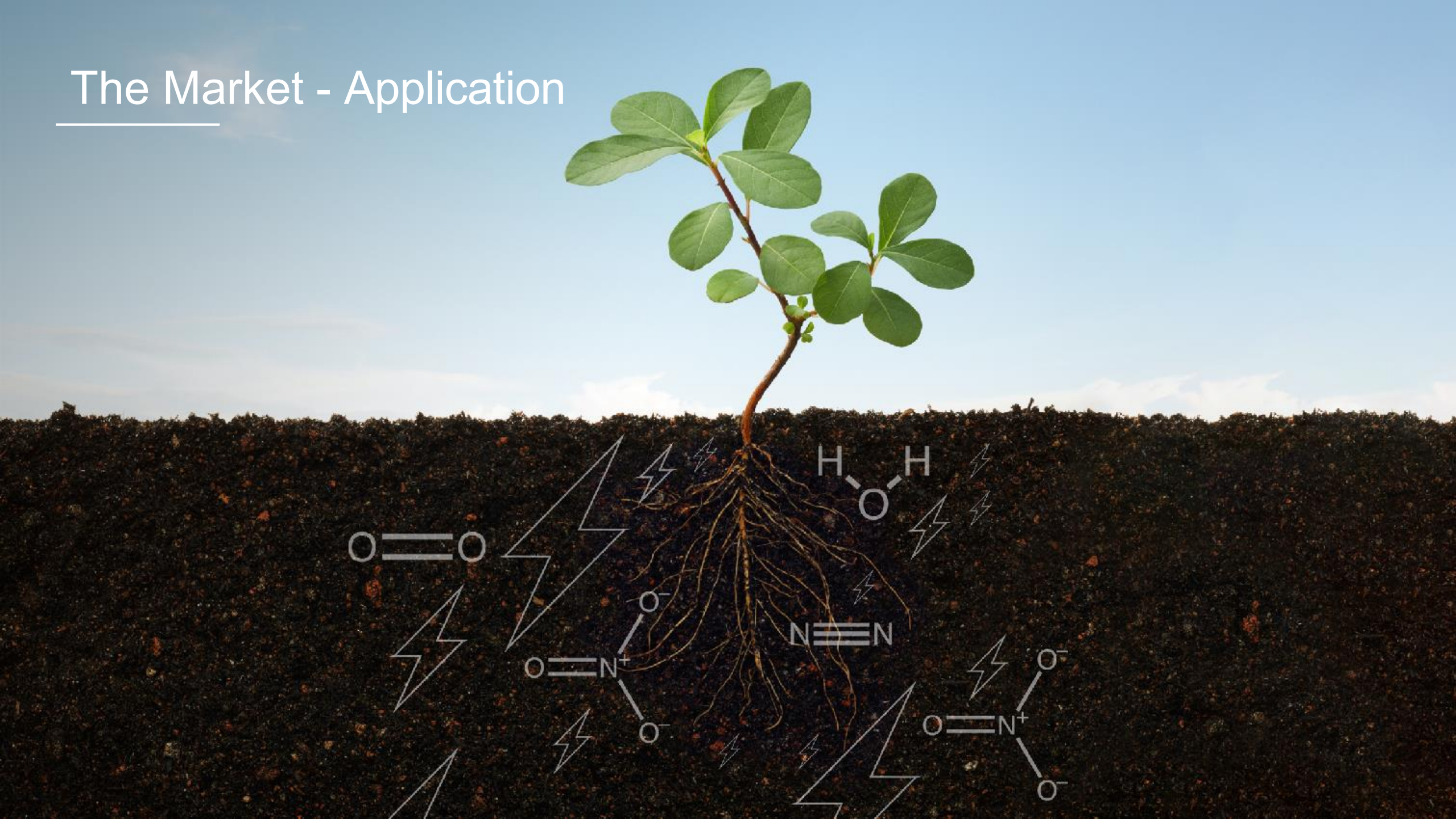
- Nature based solution
- Water, air and electricity
- On site inputs only
- No contaminants, residues

Sustainable

- On site production, no supply of raw materials
- Carbon Neutral when powered by green energy
- No chemicals

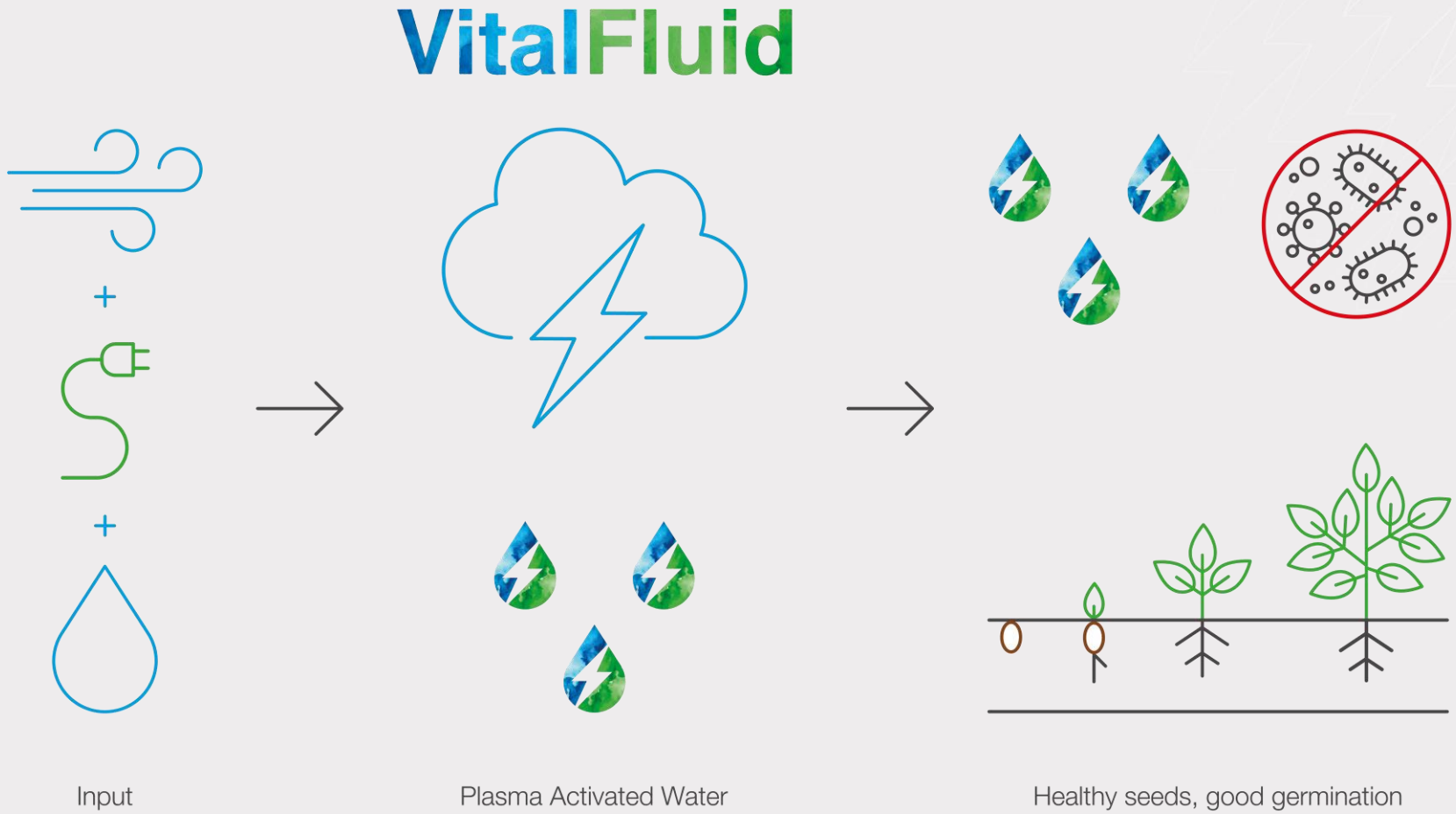


The Market - Application



VitalFluid Seed Treatment

Infographic



VitalFluid Seed Treatment

Clean and healthy seeds

- No more chemicals
- No more pathogens
- Increased germination

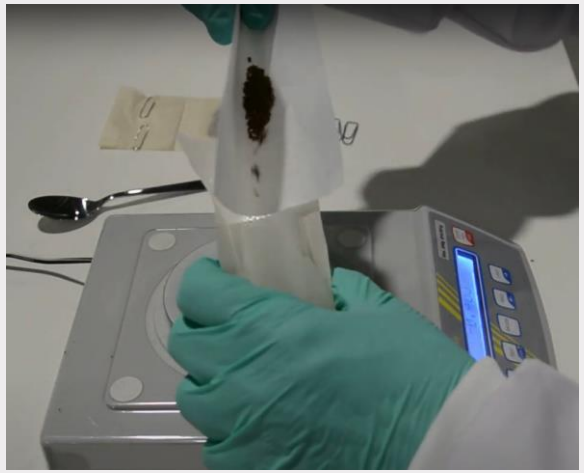
Trials disinfecting - priming

- 14 different seed Breeders
- > 40 seed - pathogen varieties

Trials

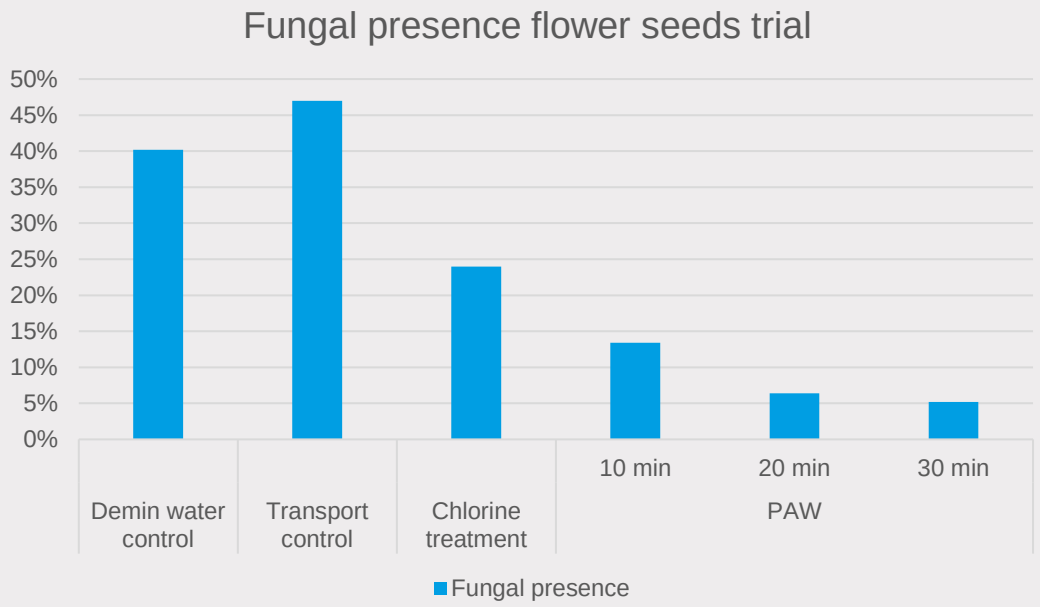
Different Treatments

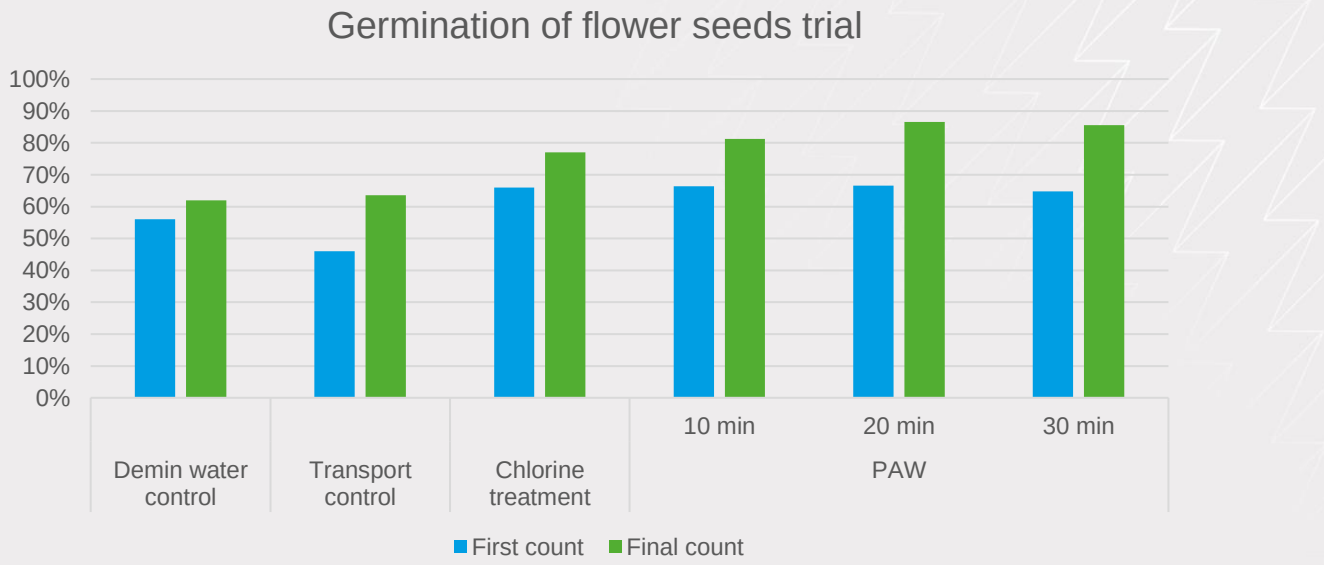
- Variety of concentrations
- Different incubation times



VitalFluid Seed Treatment

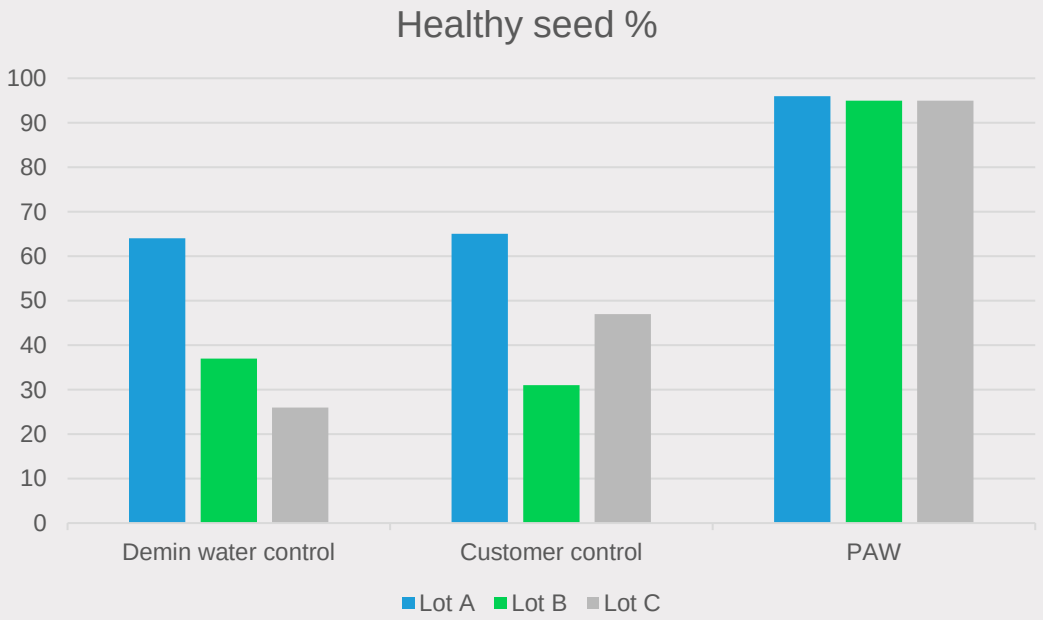
Trial Flower seeds

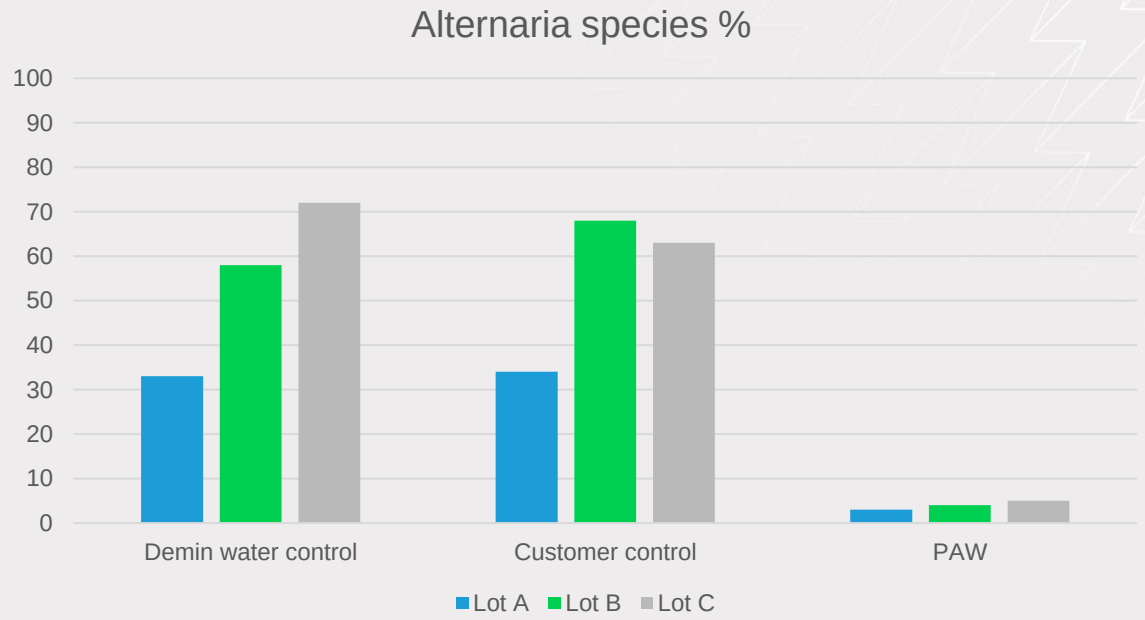




VitalFluid Seed Treatment

Trial Chicory seeds





VitalFluid Seed Treatment

Upscaling

- Spraying
- Rotary coater

Regulations

Regulations

Regisration Worldwide

- USA vs EU

- EU - long process
- EU - big investment

Plasma Equipment Development

VitalFluid Units

Aegir PAW disinfecting unit

- 1 kW system
- 2 L per batches
- Different concentrations

Freya unit

- 15 kW system
- 21,5 mol N / hr
- 300 mmol N / Ltr
- 1,6 x 1,2 x 2,2 mtr (WxDxH):

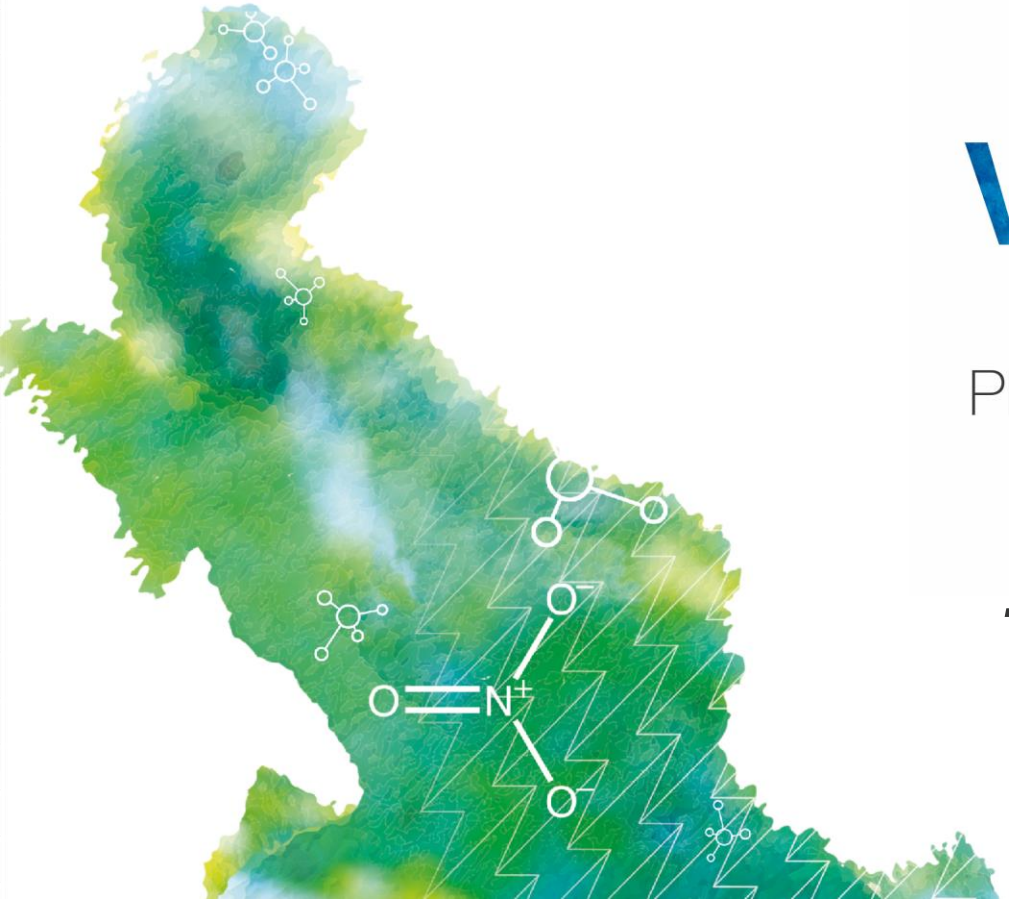
We create Lightning

Plasma in action

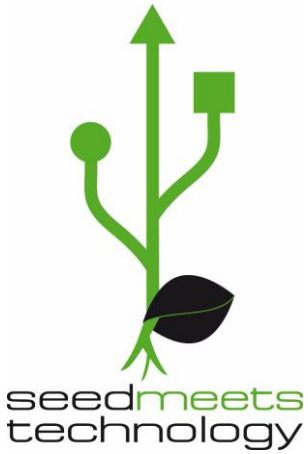
VitalFluid

Global Leader in
Plasma Activated Water
Applications

Thank you!



Discussion and closing



Thank you for your attention!

VERTIFY
EXPLOR&XPLAIN

Plantum