

# Biostimulants Unveiled: A Diversity of Types



# Biostimulants: What Are They?

## NO

No fertilizer

No pesticide

No direct effect against a disease or pest

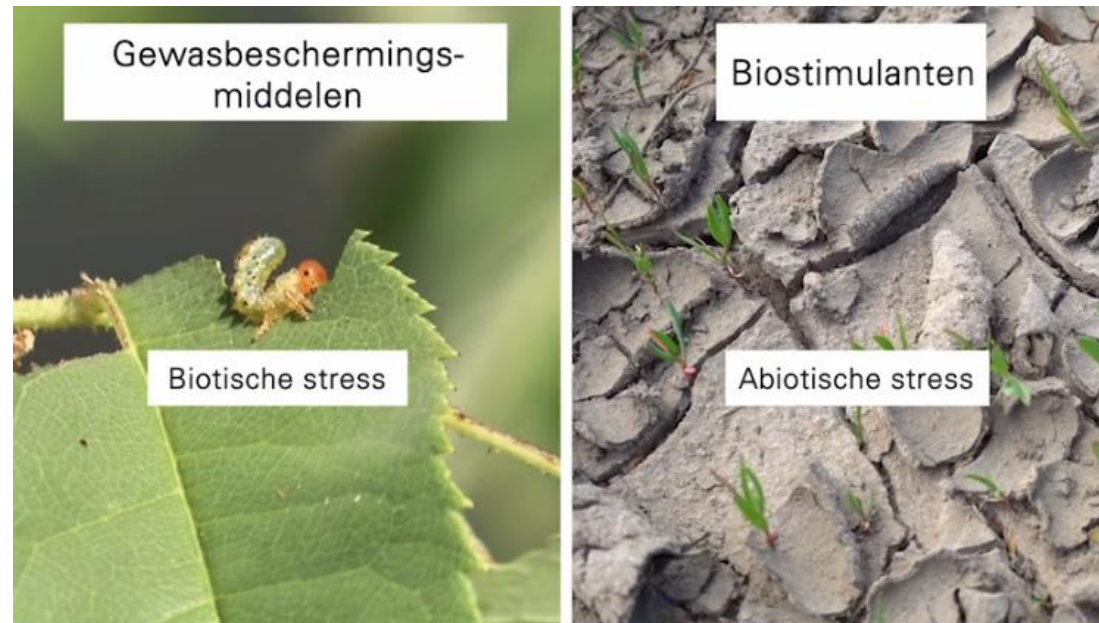
No soil improver

## YES

Improves nutrient uptake

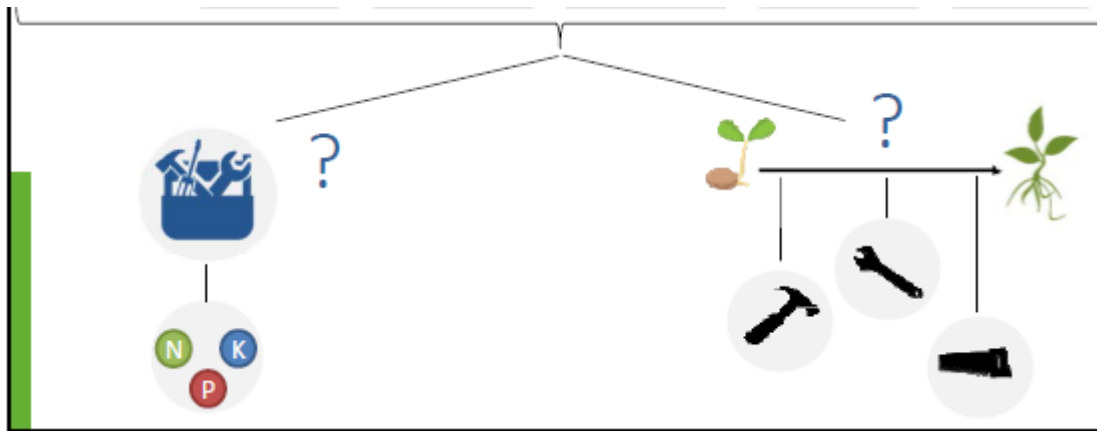
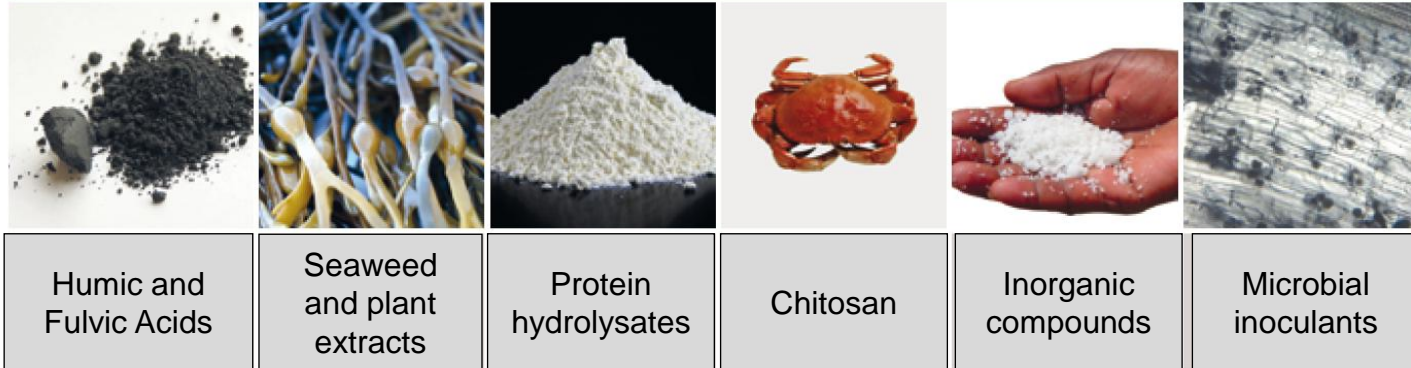
Enhances tolerance to abiotic stress

Improves crop quality



# Biostimulants: What Are They?

## MAJOR CATEGORIES OF BIOSTIMULANTS



### Composition Optima root:

- ✓ seaweed extract
- ✓ Humic acid
- ✓ Amino acids



### Composition Nutricin:

- ✓ willow bark extract
- ✓ seaweed extract
- ✓ 20 plant-based amino acids
- ✓ orthosilicic acid



# Humic and fulvic acids

## MAJOR CATEGORIES OF BIOSTIMULANS



Humic and Fulvic Acids



Seaweed and plant extracts



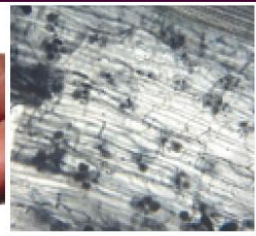
Protein hydrolysates



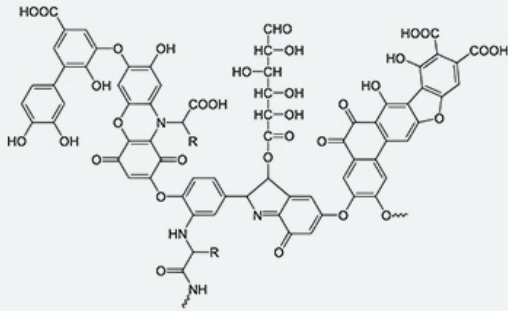
Chitosan



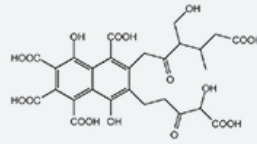
Inorganic compounds



Microbial inoculants



Humic acid molecule



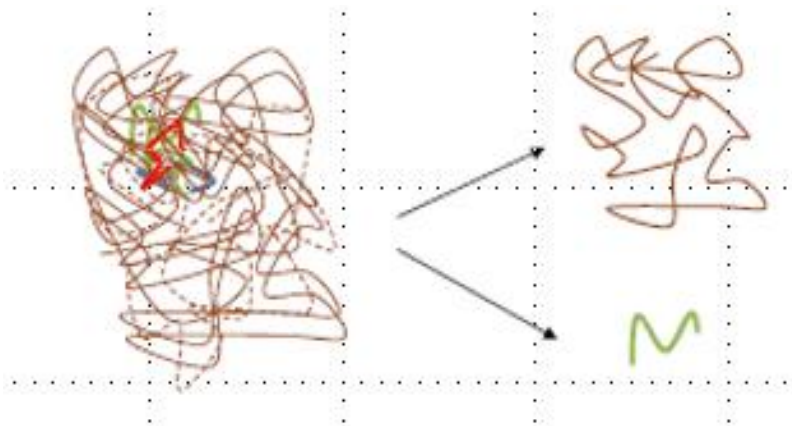
Fulvic acid molecule

### Humic Acids

- Insoluble in acidic environment
- Soluble in alkaline environment
- "High" MW (Molecular Weight)

### Fulvic Acids

- Soluble in acidic & alkaline environment
- "Low" MW (Molecular Weight)



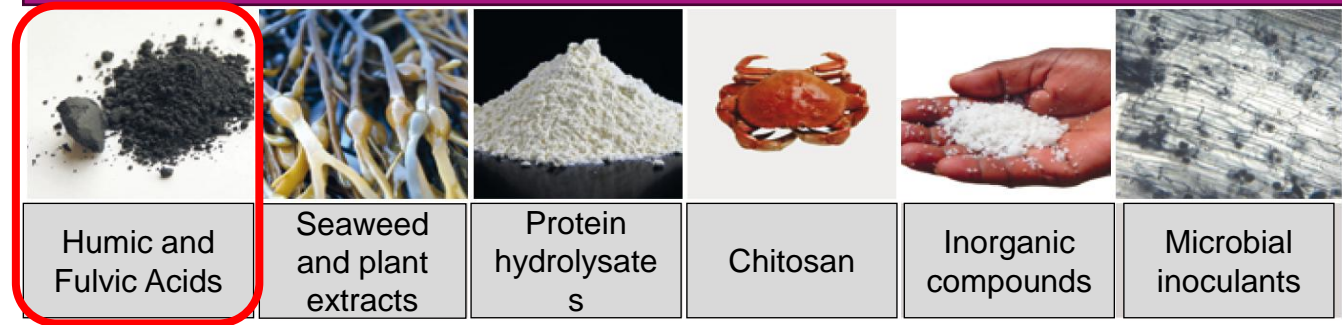
### Humins

- Insoluble



# Humic and fulvic acids

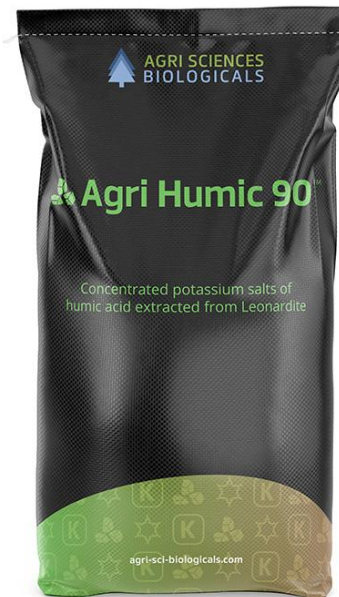
## MAJOR CATEGORIES OF BIOSTIMULANS



Composition: Humic acids: 3% w/w, Fulvic acids: 17%



Composition: Total humic extract content: 15% w/w (165 g/L), Humic acids: 12% w/w (132 g/L), Fulvic acids: 3% w/w (33 g/L)



Composition: Humic acids: 51% Fulvic acids: 40% (pH 10-11)

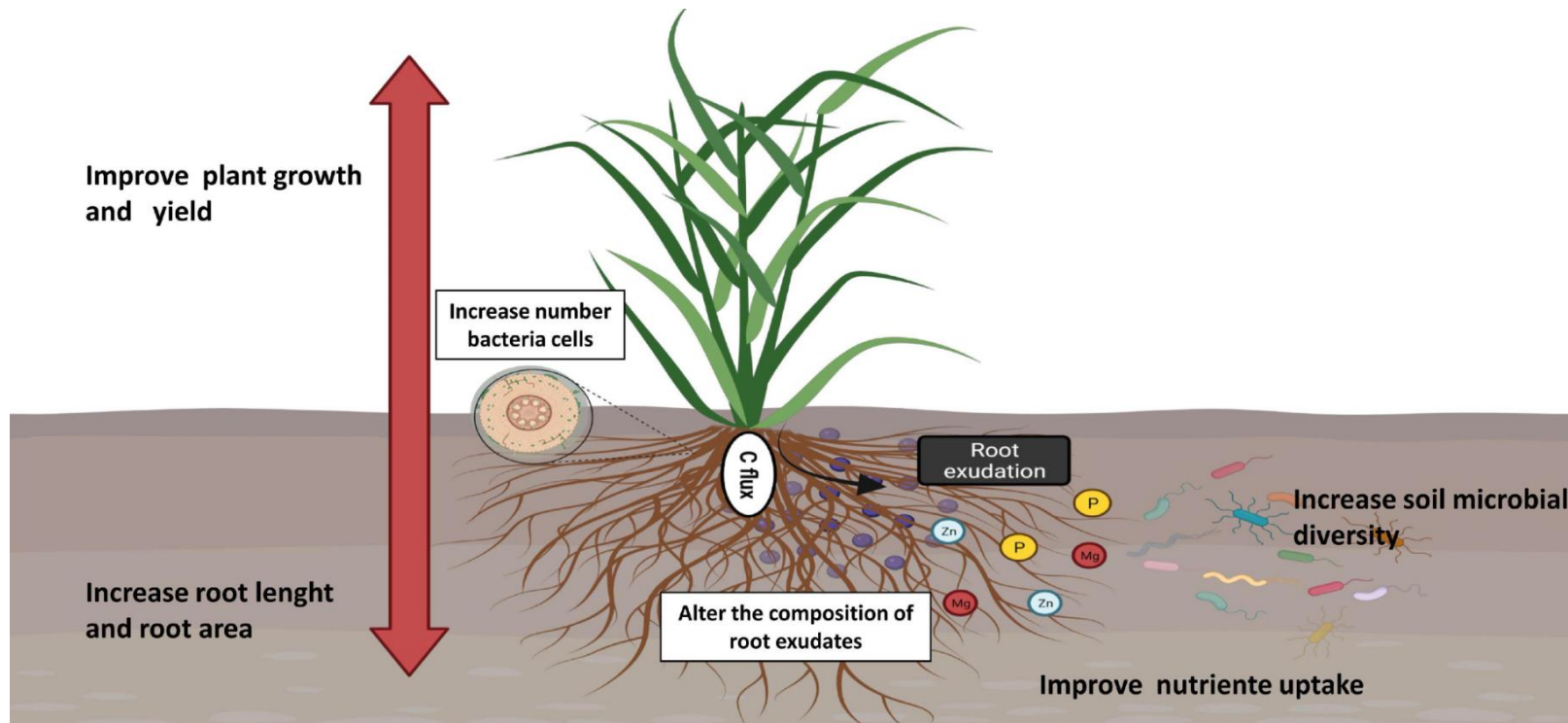
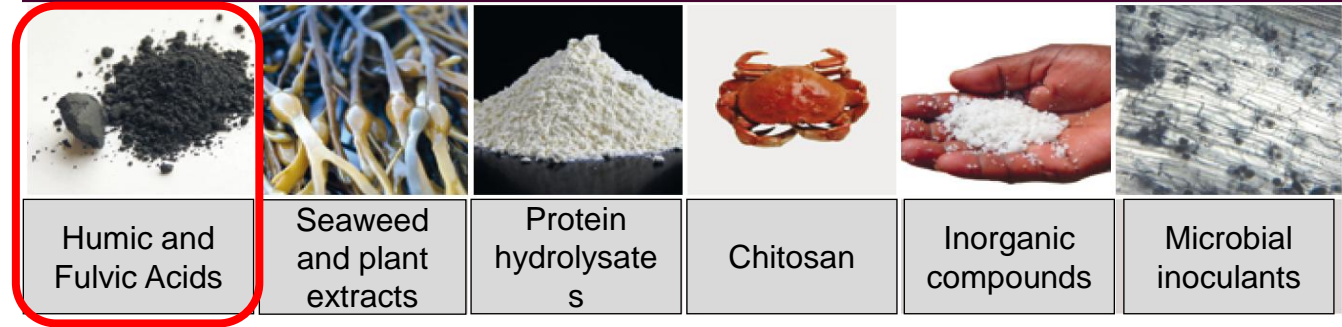
Humic acids: large molecules function best in soil, providing an optimal growing environment

Fulvic acids: similar effects on crops, but the mineralization fulvic acids faster than humic acids  
→ better availability for plants

Fulvic acids smaller than humic acids, due to smaller size:  
→ remain longer in the soil under various pH and salinity levels  
→ a prolonged effect

# Humic and fulvic acids

## MAJOR CATEGORIES OF BIOSTIMULANS



Can bind with heavy metals

Improve tolerance to abiotic stress through the production of polyphenols

- Salt stress
- Drought tolerance

<https://www.frontiersin.org/journals/microbiology/articles/10.3389/fmicb.2021.719653/full>

# Seaweed and plant extracts



## ***Ascophyllum nodosum* (Knotted Wrack):**

Most popular seaweed used in biostimulants products  
contains a wide range of natural plant growth regulators, including cytokinins, auxins, and gibberellins  
Promote root development, nutrient uptake, and overall plant growth  
Increased tolerance to environmental stresses  
Provides essential micronutrients like iodine and zinc

• ***Ecklonia maxima* (Brown Seaweed):** *Ecklonia maxima* is known for its high content of natural antioxidants, which help plants cope with abiotic stressors like drought and heat






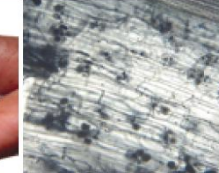
• ***Laminaria spp.* (Kelp):** Kelp extracts, rich in natural compounds and essential nutrients, stimulating root development, nutrient uptake efficiency, and flowering.

• ***Sargassum spp.*:** boasts high levels of amino acids

• ***Ulva spp.* (Sea Lettuce):** *Ulva*-based biostimulants offer a balanced mix of nitrogen, phosphorus, and potassium

• ***Fucus spp.* (Bladderwrack):** boosts cell division, root elongation, and lateral bud development, leading to optimal plant architecture

## MAJOR CATEGORIES OF BIOSTIMULANS

					
Humic and Fulvic Acids	Seaweed and plant extracts	Protein hydrolysates	Chitosan	Anorganic compounds	Microbial inoculants





# Seaweed and plant extracts

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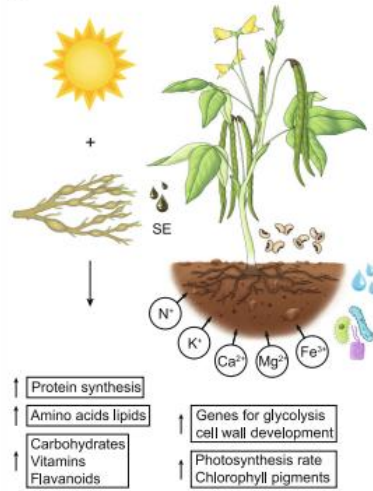
WILEY-**Plant, Cell & Environment**

DEOLU-AJAYI ET AL.

(a) Normal conditions



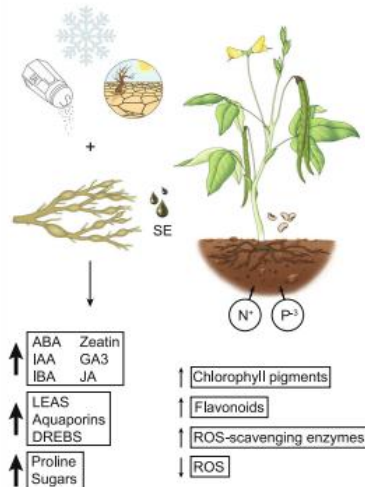
(b) Normal conditions + SE



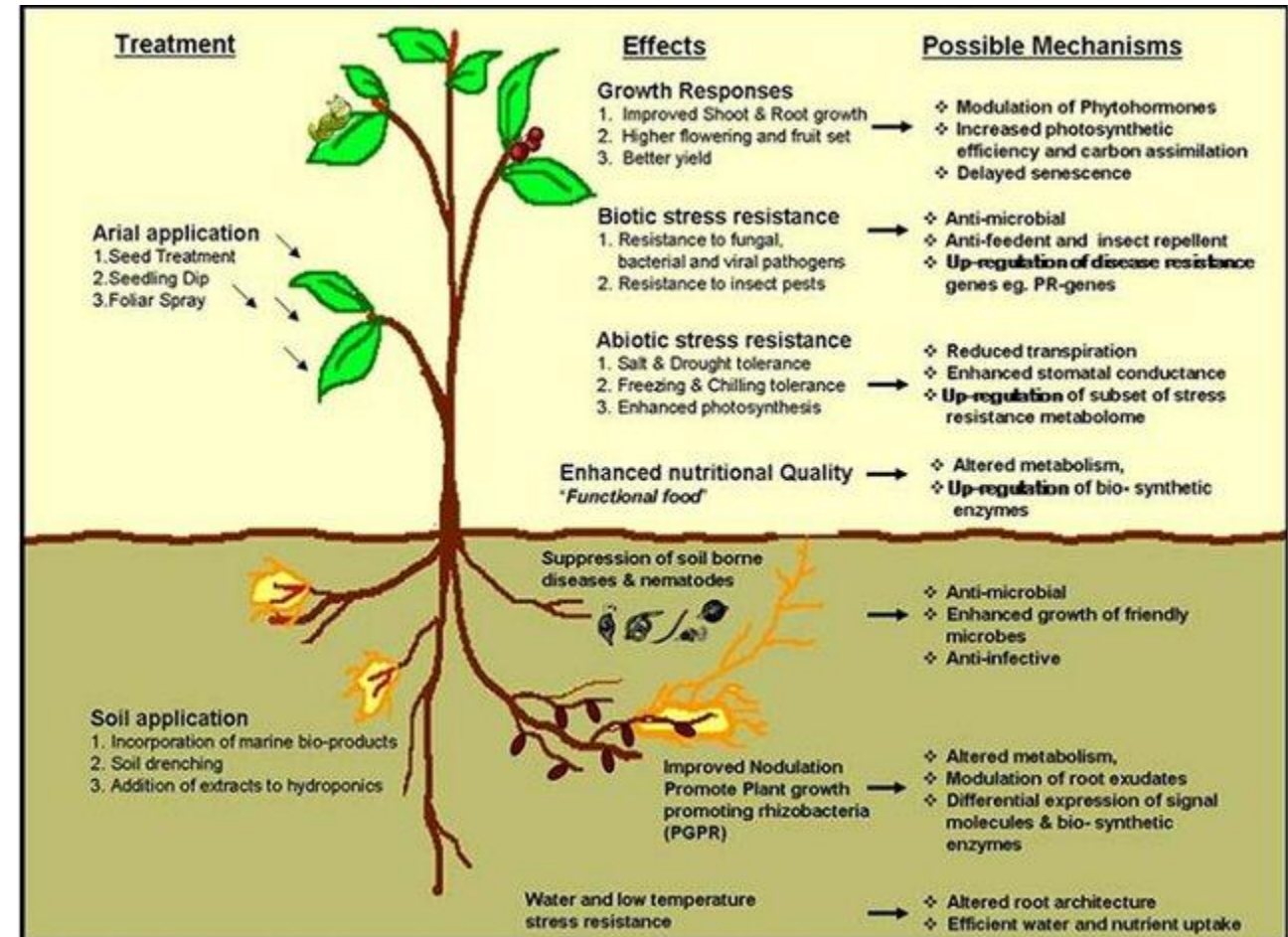
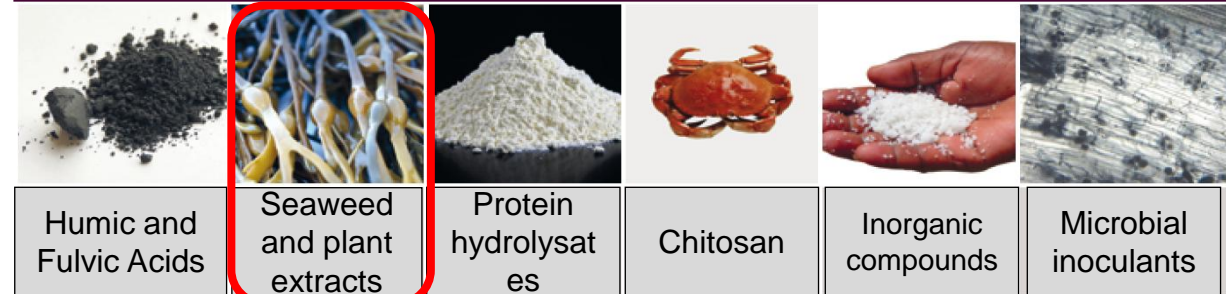
(c) Stress conditions



(d) Stress conditions + SE

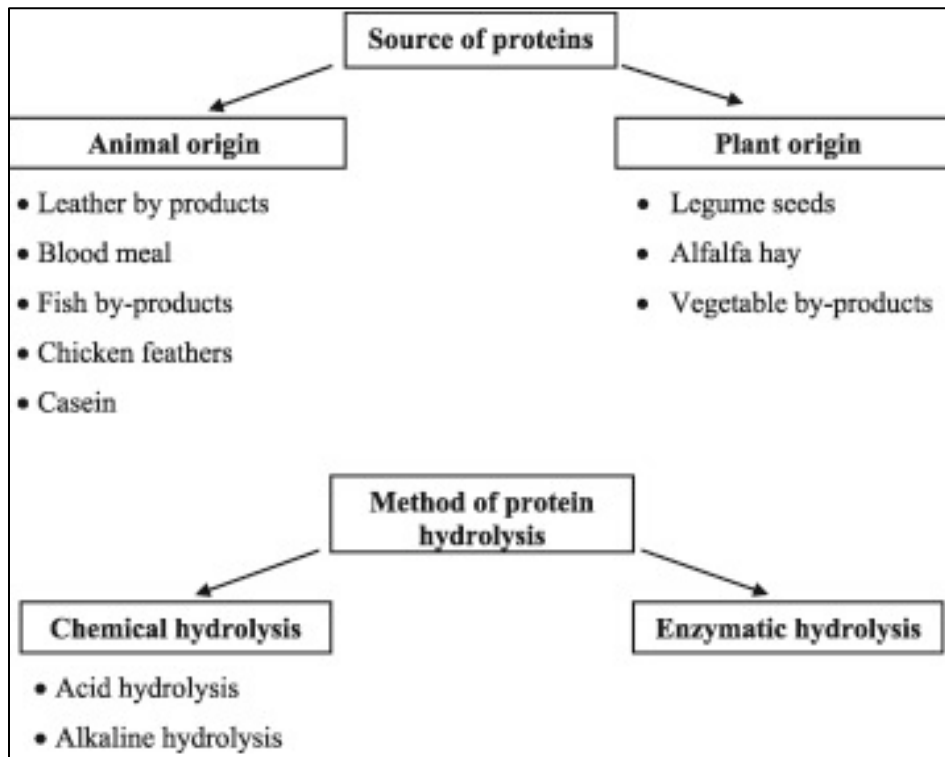
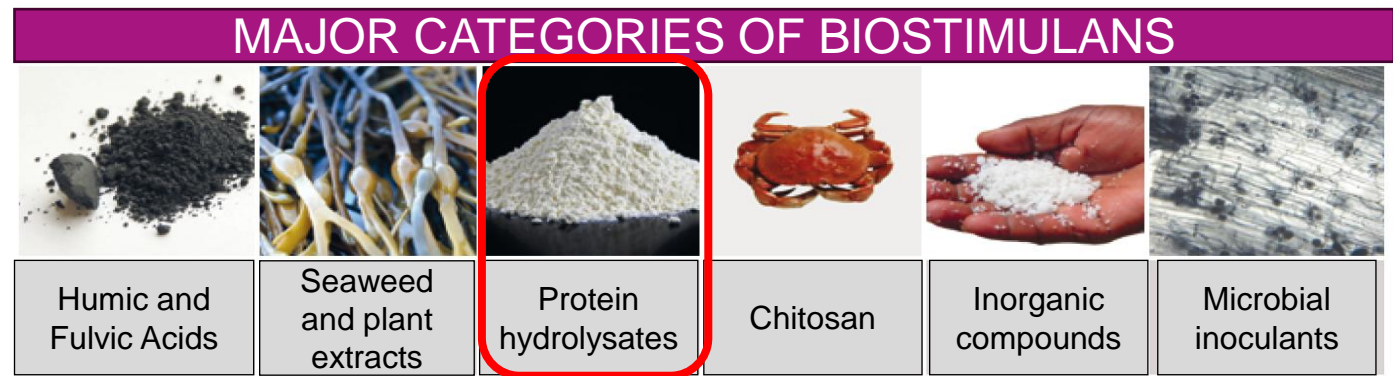


## MAJOR CATEGORIES OF BIOSTIMULANS





# Protein hydrolysates



## Types:

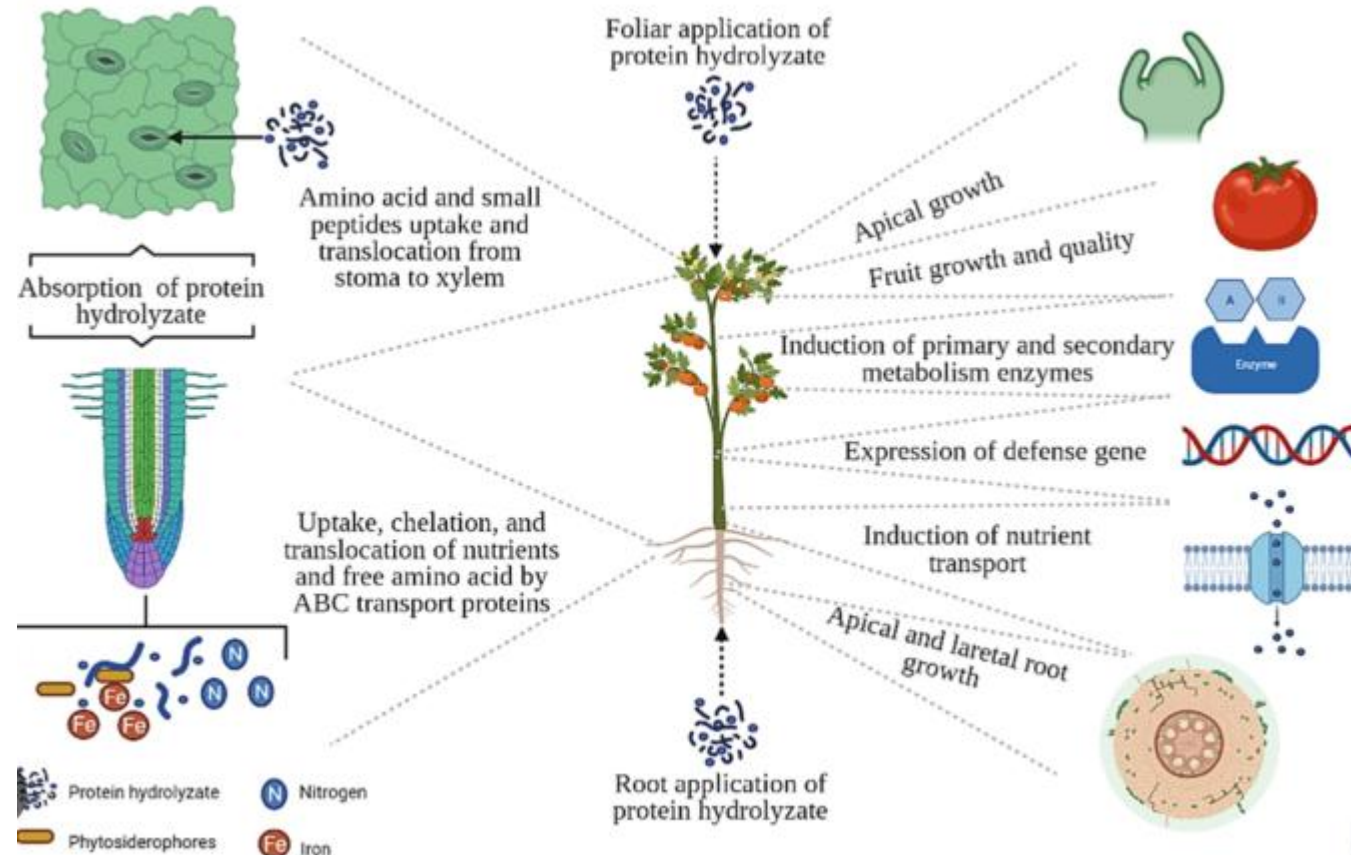
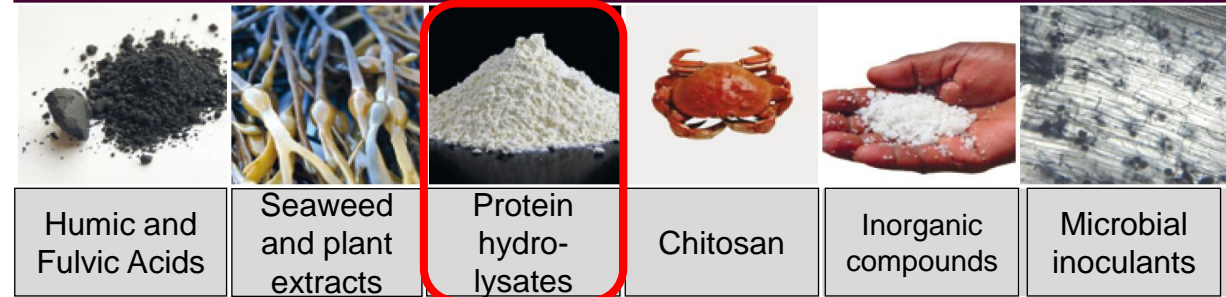
- Protein hydrolysates: mix of amino acids and peptides
- Individual amino acids: e.g., glutamate & betaine, proline, histidine

## Origin:

- Plant material or animal by-products
- Chemical hydrolysis or enzymatic hydrolysis
- Purified components or synthesized single compounds

# Protein hydrolysates

## MAJOR CATEGORIES OF BIOSTIMULANTS



# Chitin and Chitosan

Includes polymers of biological origin such as polysaccharides, cellulose, peptides, and starch

## MAJOR CATEGORIES OF BIOSTIMULANS



Humic and Fulvic Acids



Seaweed and plant extracts



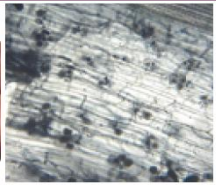
Protein hydrolysates



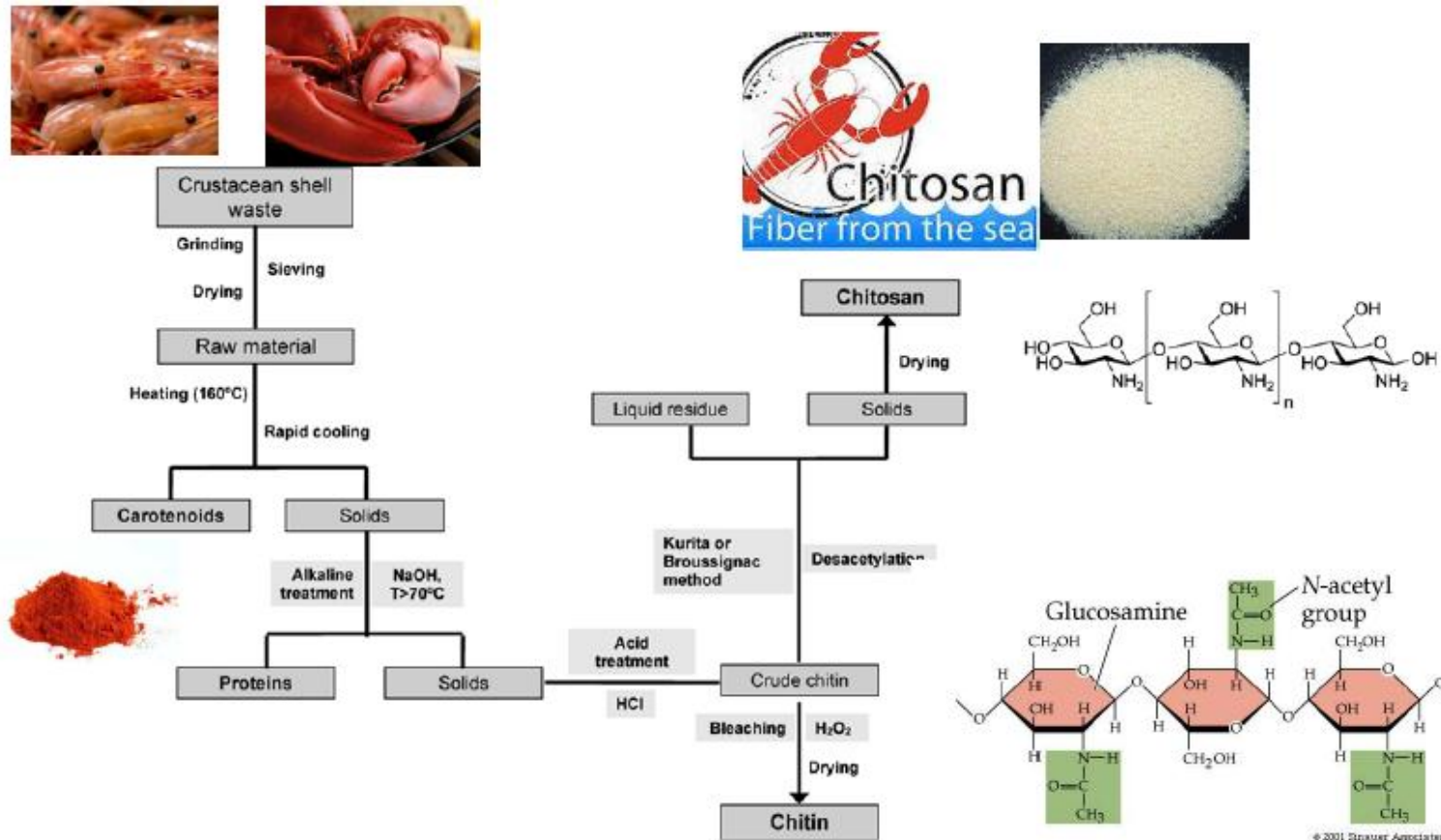
Chitosan



Inorganic compounds



Microbial inoculants

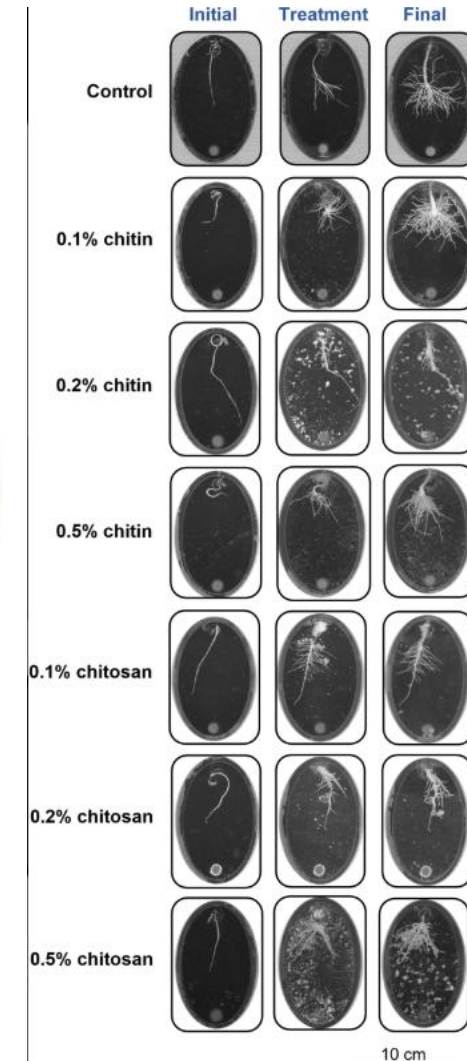
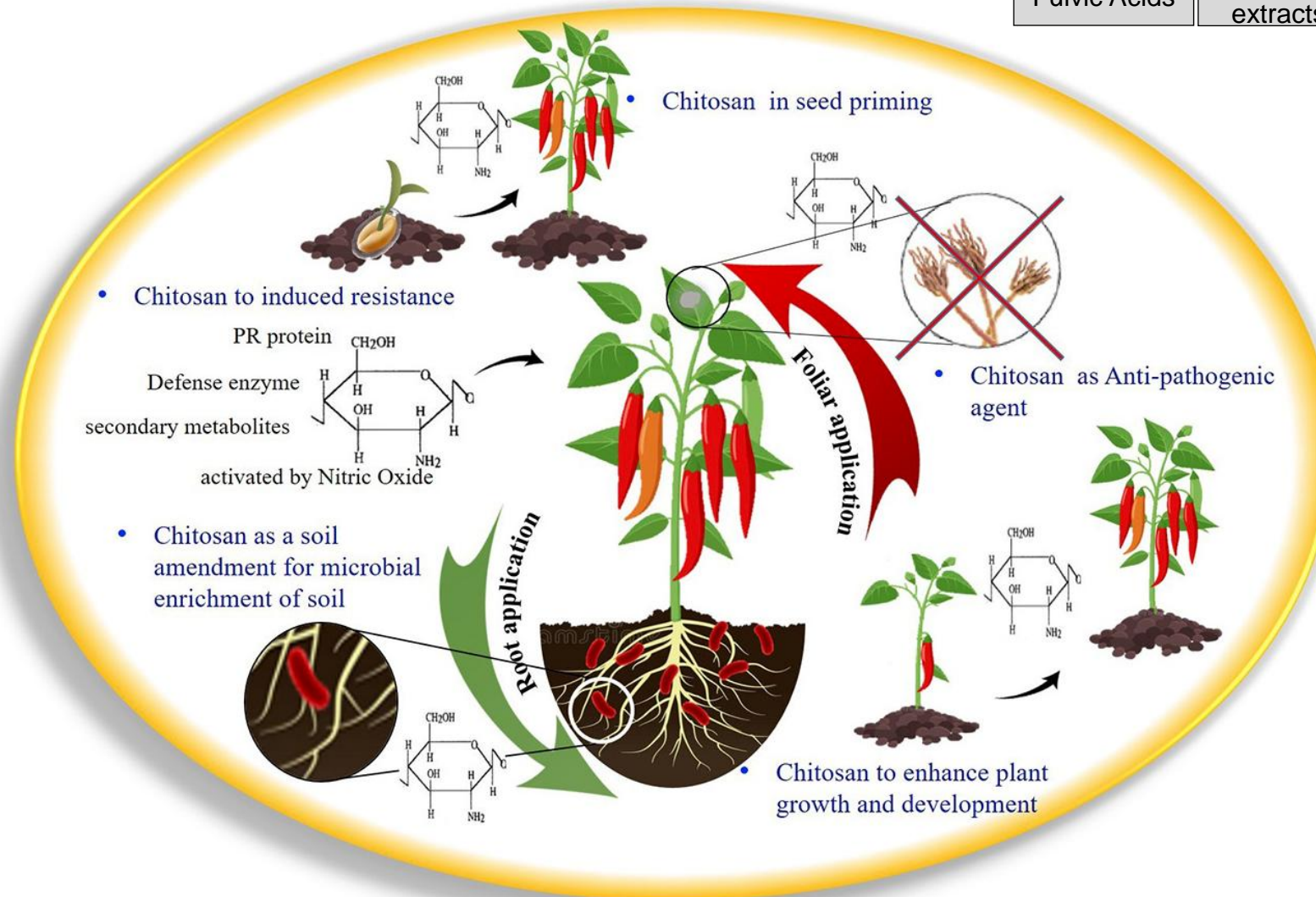
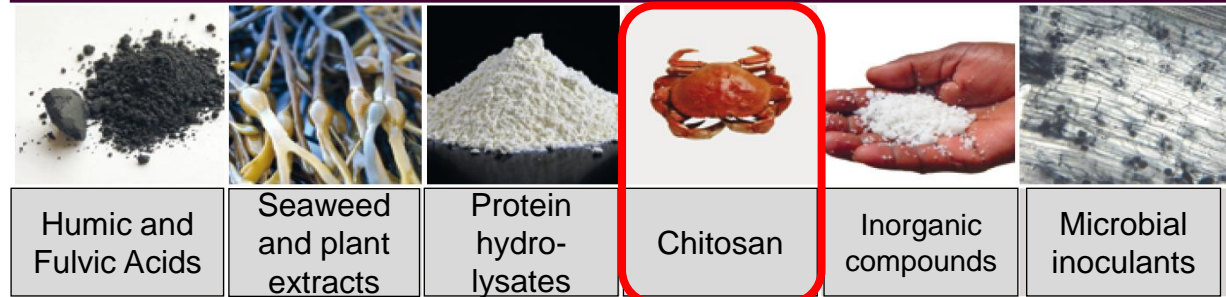


Chitosan preparations can differ in molecular mass, and degree of deacetylation – these differences can greatly biological function



# Chitosan

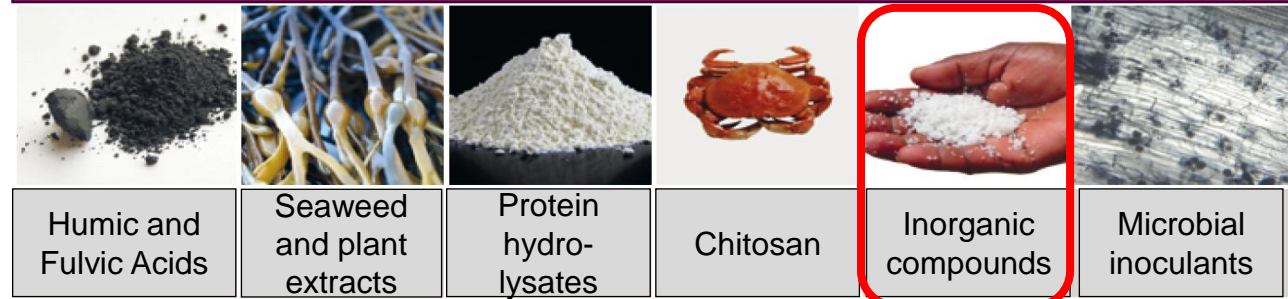
## MAJOR CATEGORIES OF BIOSTIMULANS



<https://onlinelibrary.wiley.com/doi/full/10.1002/pei3.10106>

# Inorganic compounds

## MAJOR CATEGORIES OF BIOSTIMULANS



Inorganic compounds with main beneficial elements such as Al, Co, Se and which promote plant growth, the quality of plant products, and abiotic stress tolerance.

### SILICON

- Works in various ways against stress and fungi
- Strengthens the cell wall
- Improves the uptake of calcium and potassium
- **Water solubility** is important (orthosilic acid :  $\text{H}_4\text{SiO}_4$ )!
- Various types available on the market:
  - Based on sodium/potassium silicates: (pH 12-14): not miscible with pesticides
  - Stabilized silicates
  - Nano silicates

#### Silacon

NPK: 2-3-11 +14.7%  $\text{SiO}_2$  +0.75% hydrolyzed seaweed

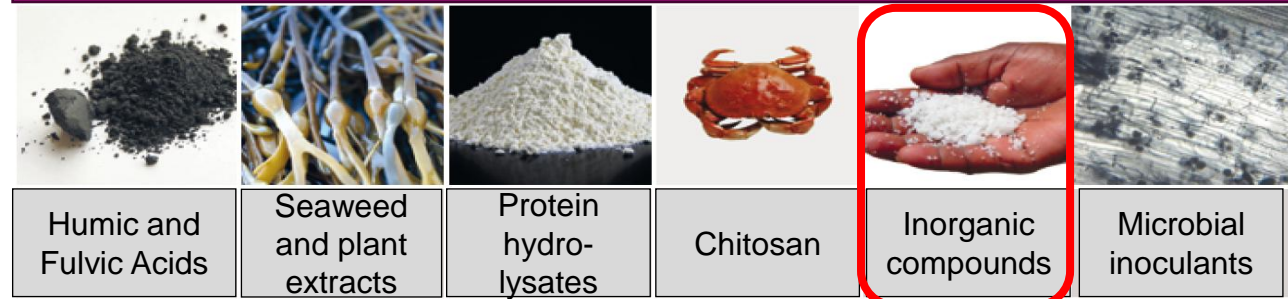
- ✓ High concentration of directly available silicium
- ✓ Silicium helps to form a physical barrier against biotic and abiotic stress
- ✓ Rich in potassium to make crops more resistant to drought
- ✓ Increases sugar content in fruits
- ✓ Promotes the uptake of calcium and micronutrients
- ✓ Can be applied in agriculture, in fertigation systems, or used as a foliar spray



3% orthosilic acid

# Inorganic compounds

## MAJOR CATEGORIES OF BIOSTIMULANS



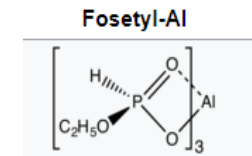
Inorganic salts of beneficial and essential elements: chlorides, phosphates, phosphites, silicates and carbonates have been used as fungicides.

Influence osmotic regulation, pH, hormone signalling and enzymes involved in stress response (e.g. peroxidases).

Their function as biostimulant:  
Influence plant growth  
acting on nutrition efficiency  
abiotic stress tolerance

### Phosphate versus Phosphonate

- Phosphate ( $\text{HPO}_4^{2-}$ ) = fertilizer
- Phosphonate ( $\text{HPO}_3^{2-}$ ) = no fertilizer, is a pesticide
  - Aliette






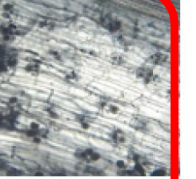


- Potassium phosphite



# Bacterial and fungal inoculants

## MAJOR CATEGORIES OF BIOSTIMULANTS

					
Humic and Fulvic Acids	Seaweed and plant extracts	Protein hydrolysates	Chitosan	Inorganic compounds	Microbial inoculants

**Microbial Biostimulants stimulate the growth of beneficial microorganisms improving and restoring soil biodiversity and Soil health:**



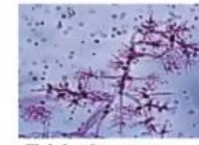
Bacteria

PGPR that colonize plant rhizosphere solubilizing nutrients and participating in Nitrogen and other elemental cycles



AM fungi

It is the name of the plant-fungi symbiosis. They increase the root absorption and provide plant nutrients and water



Trichoderma spp.

Saprophyte fungi that easily colonize the rhizosphere, competing with other organisms and improving plant growth

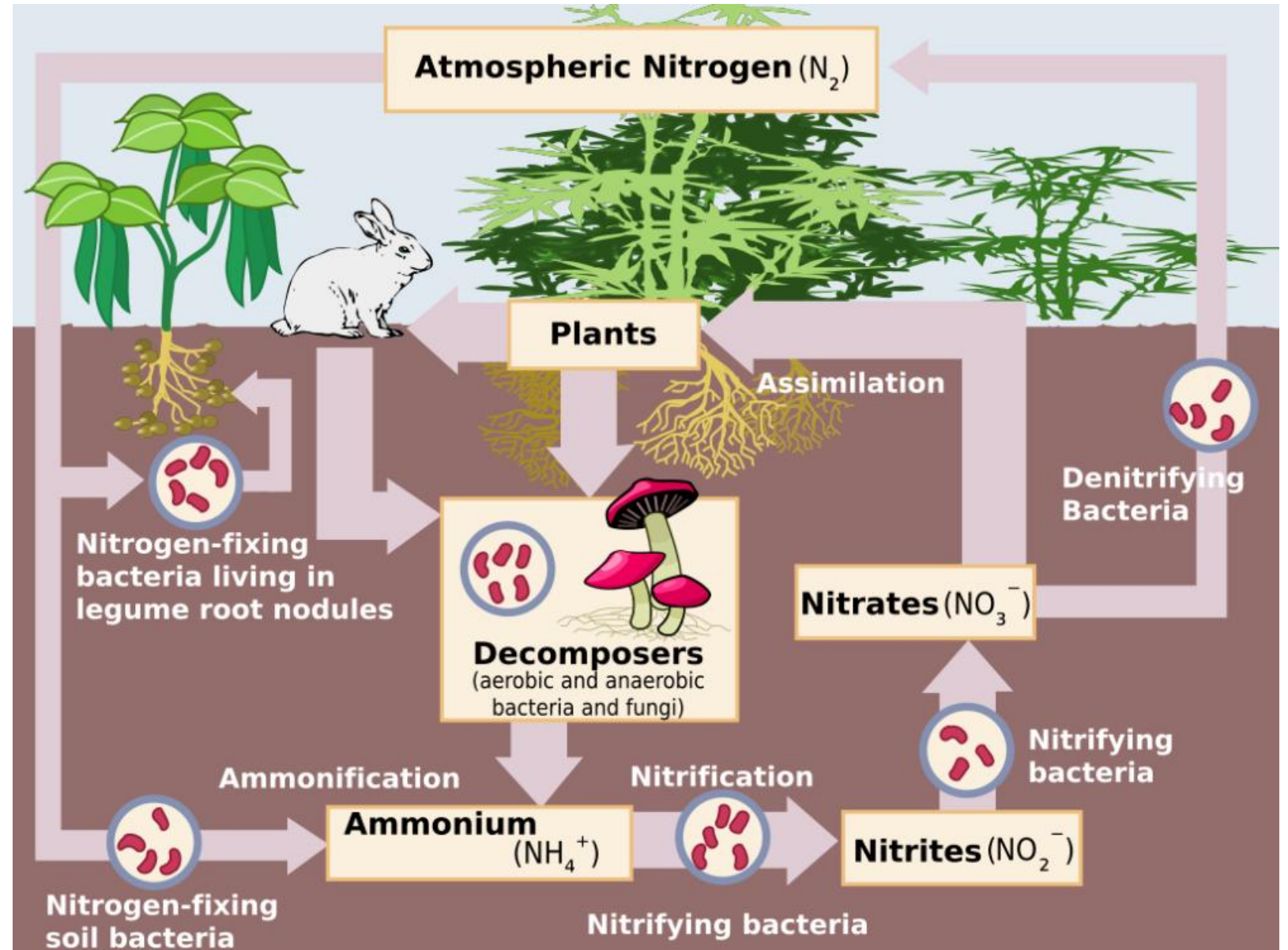
Better Soil Structure and Root Biostimulation

### THE WINNING APPROACH

An approach combining micro-organisms, organic fertilizers and other types of biostimulants optimally contributes to preserve and/ or restore soil health.

# Bacteria

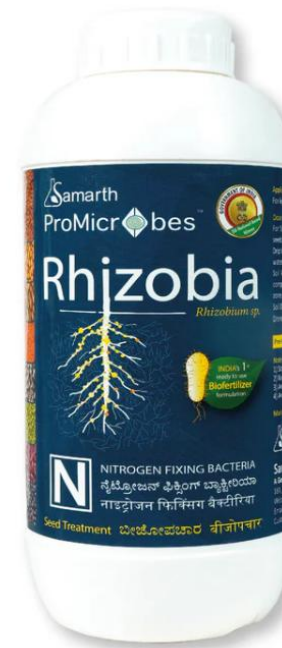
Nitrifying and denitrifying bacteria



# Bacteria

## Nitrogen-Fixing Bacteria

- There are many bacteria that enable nitrogen fixation from the air, categorized into different classes.
- All of them use the nitrogenase complex to convert atmospheric  $N_2$  into a form that plants can absorb:
  - **Symbiotic bacteria** (*Rhizobium* or *Bradyrhizobium*)
    - These bacteria form an essential structure (root nodules + bacteria that can fix  $N_2$ ).
  - **Diazotrophic bacteria** (*Azotobacter*, *Pseudomonas*, or *Azomonas*)
    - These bacteria are located in the rhizosphere; they generally fix  $N_2$  when another nitrogen source is absent and when oxygen is scarce.
  - **Endophytic bacteria** (*Methylobacterium*, *Azospirillum*)
    - Bacteria that are accepted by the plant's immune system and spread through the plant tissue (leaves and/or roots).



*Rhizobium*



*Methylobacterium*



*Azotobacter salinestris* Stamm CECT 9690

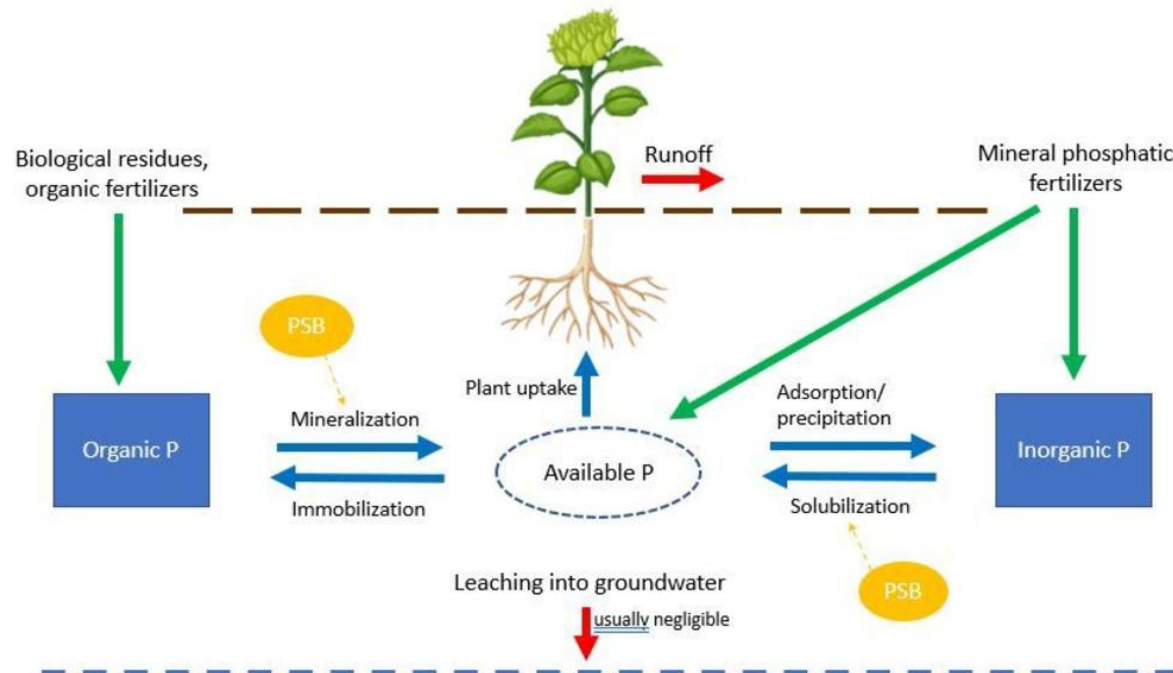


# Bacteria

## Phosphorus-solubilizing bacteria

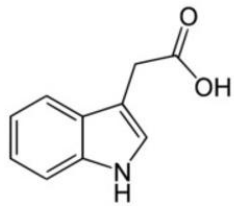
Bacterial strains (*Pseudomonas*, *Bacillus*, *Rhizobium*, *Burkholderia*, *Achromobacter*, *Agrobacterium*, *Micrococcus*, *Aerobacter*, *Flavobacterium* and *Erwinia*)

Ability to solubilize insoluble inorganic phosphate (mineral phosphate) (such as tricalcium phosphate, dicalcium phosphate, hydroxyl apatite and rock phosphate)

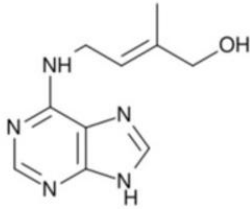


# Bacteria

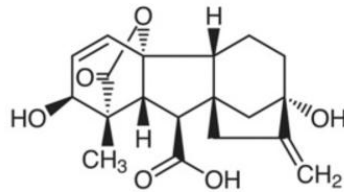
Phytohormone Production, also produced by plant growth-promoting rhizobacteria (PGPRs)

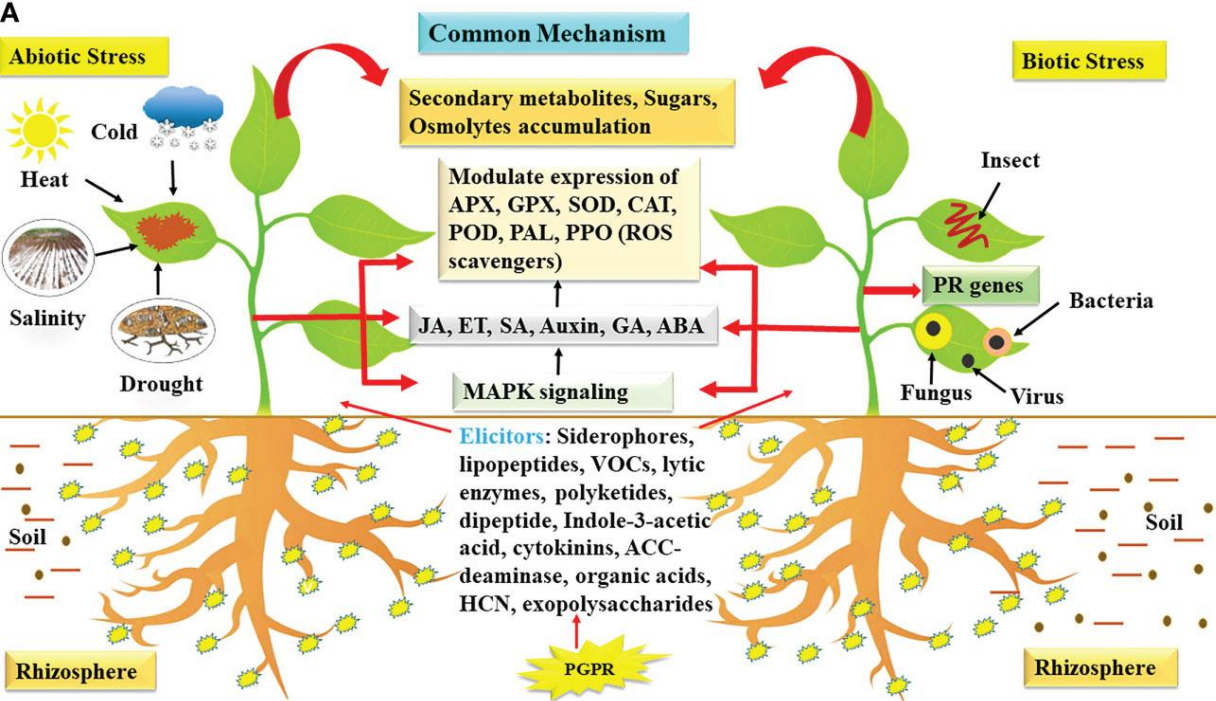


Auxin



Cytokinin

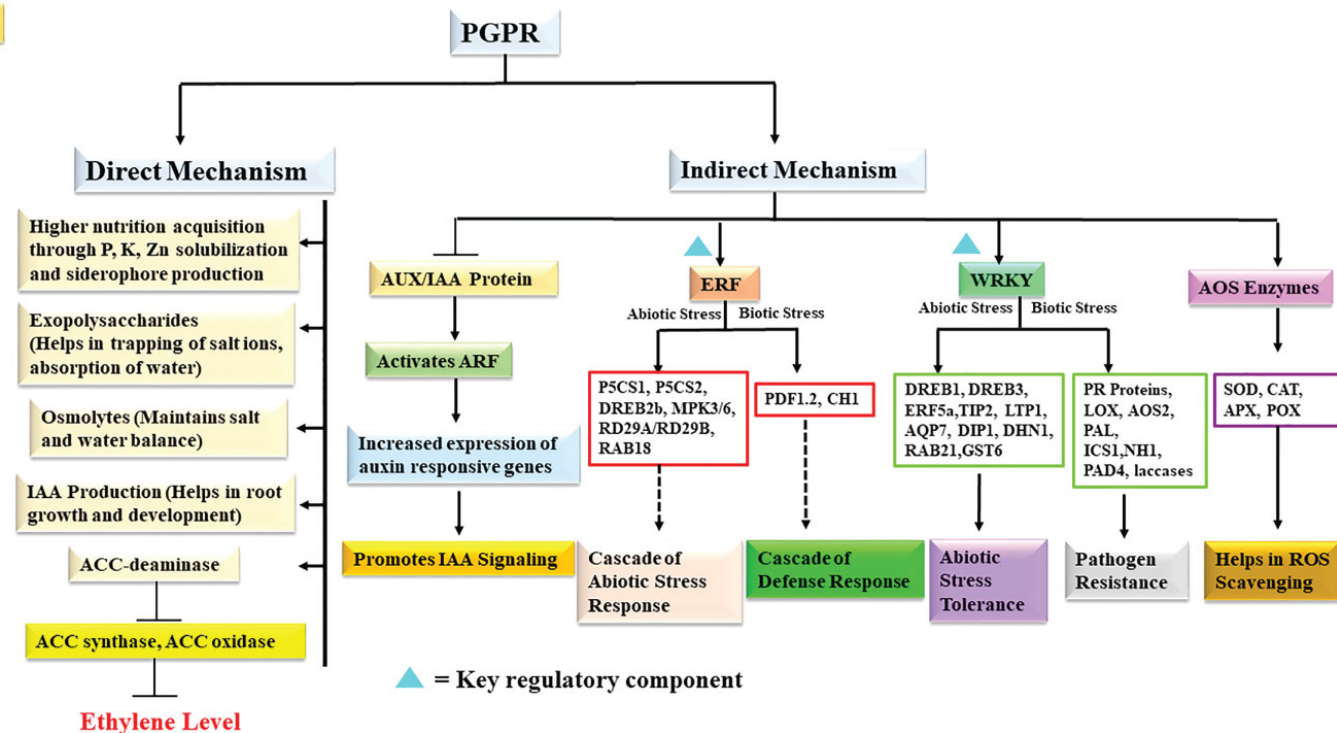




*Bacillus sp.*








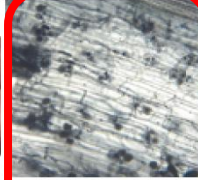
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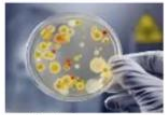


# Bacterial and fungal inoculants

## MAJOR CATEGORIES OF BIOSIMULANS

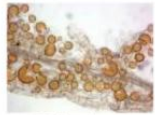
					
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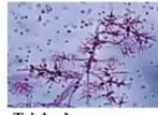
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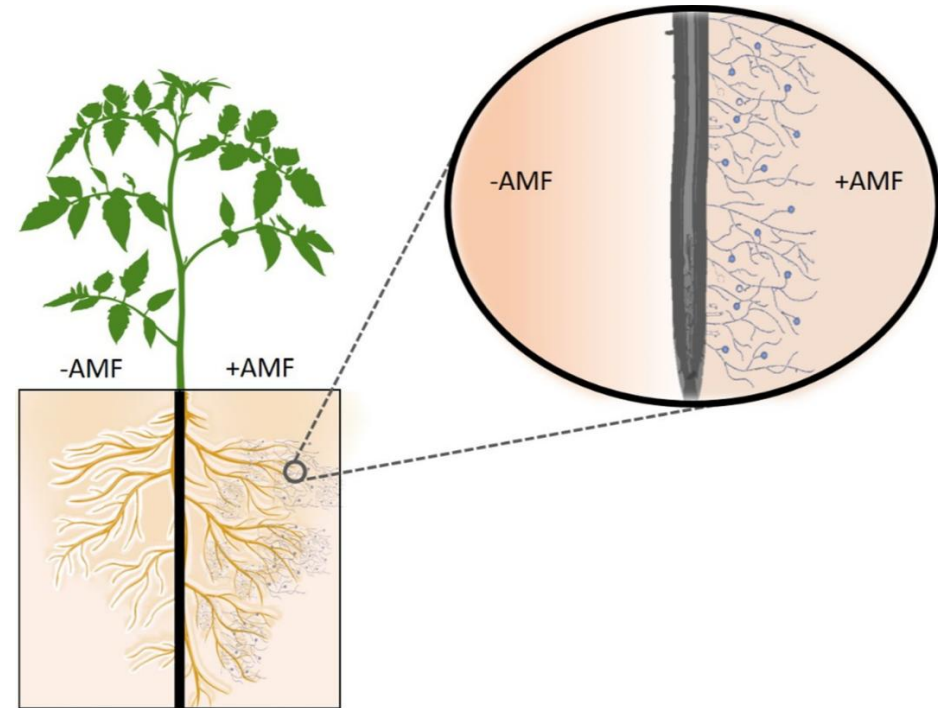
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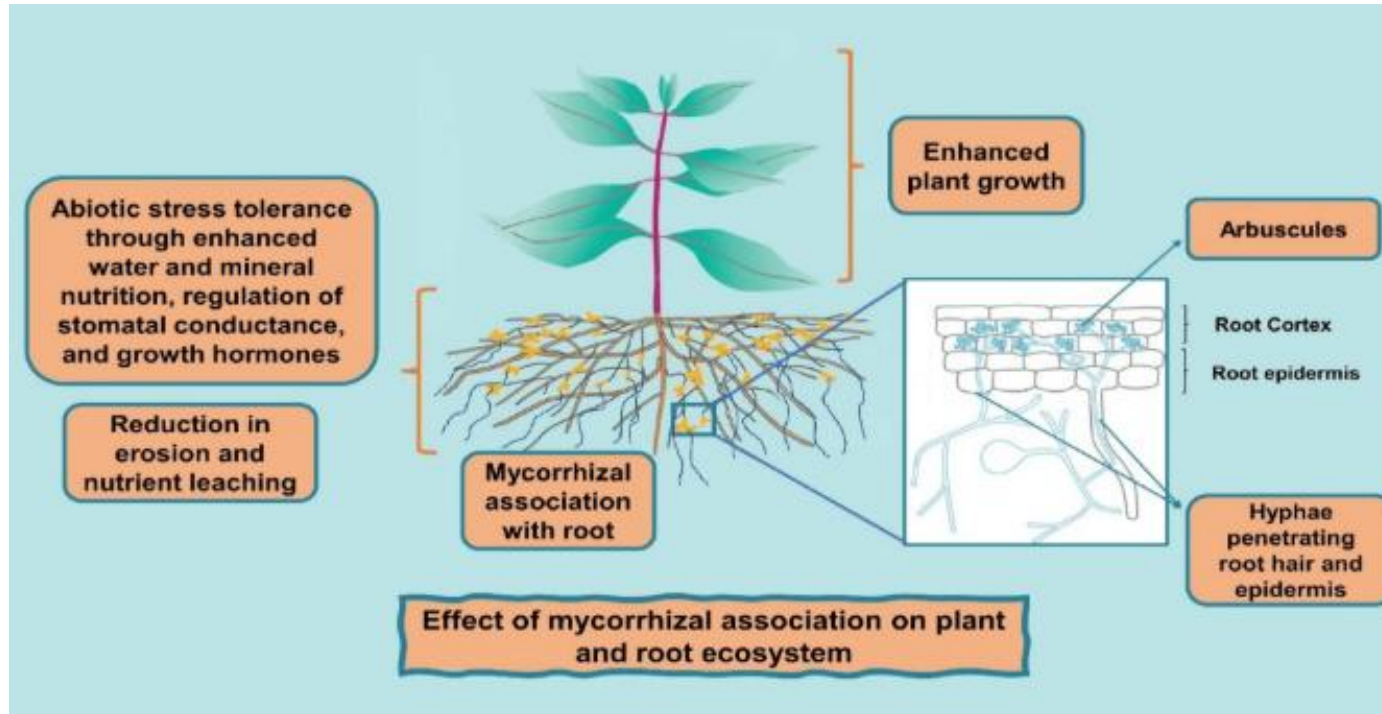
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Castigilone *et al.*, 2021



# Arbuscular mycorrhizal fungi (AMF)



## MycoApply® EndoMaxx®

Biostimulants and Related Products – MycoApply






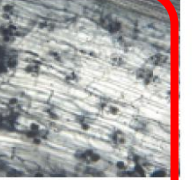
– Arbuscular Mycorrhizal Fungi

MycoApply EndoMaxx is a dry formulation containing arbuscular...

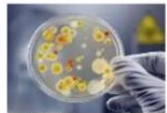


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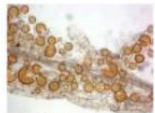
					
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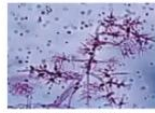
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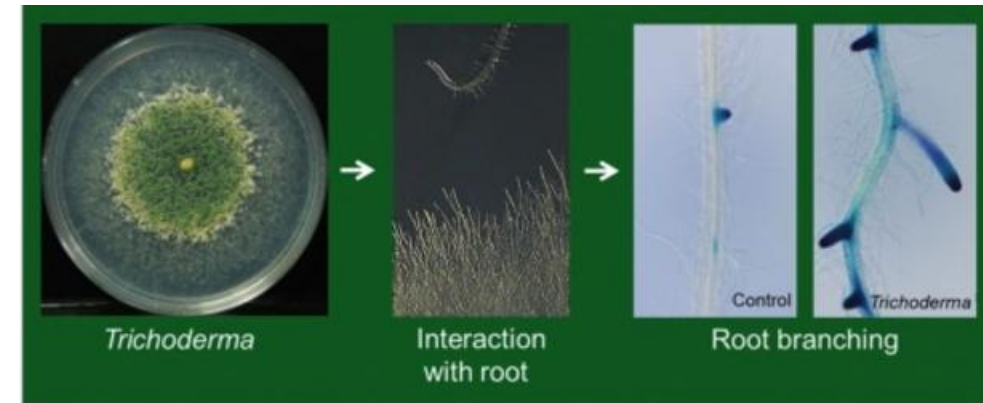
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Castigilone *et al.*, 2021





# *Trichoderma* spp.

## — IT IS A NUTRIENT SOLUBILIZER

In fact, it helps a lot to solubilize phosphorus, which is a fundamental nutrient for plants. And acidifying the soil encourages better absorption of this macronutrient, which is very important in the growth of strong plants.

## — PROMOTES ROOT DEVELOPMENT

Contributes to the formation of stronger roots. In addition, deeper and, therefore, solid rooting is achieved. It is a guarantee for a healthier plant.

## — PROMOTES PLANT GROWTH

It promotes the production of hormones such as auxins, which activate the vigorous growth and size of the plant.

## — IT HAS AN IMPORTANT BIOCONTROL PROPERTY

The symbiotic relationship that the trichoderma fungus establishes with the plant through the root acts as a preventive antibiotic. In this way, it is favored to have more resistance to viruses, bacteria... and prevents certain pathogens from attacking the root.

## — PROTECTS THE PLANT FROM ABIOTIC STRESS

Trichoderma fungus protects the plant from abiotic stress, by being an extra support for it, which thus adapts better to situations such as changes in temperature or lack of water, factors to which, as we know, every crop is especially sensitive.



# Experimental designs trials 2023-2024 indoor ornamentals: effect on rooting

- Plant material: Hedera, Spathiphyllum, Viola and Primula
- Randomized block design, 4 repetitions
- Products:
- 2-weekly measurements:
  - Plant height and diameter
  - FW and DW
  - Plant quality
  - Root score
  - WET-sensor



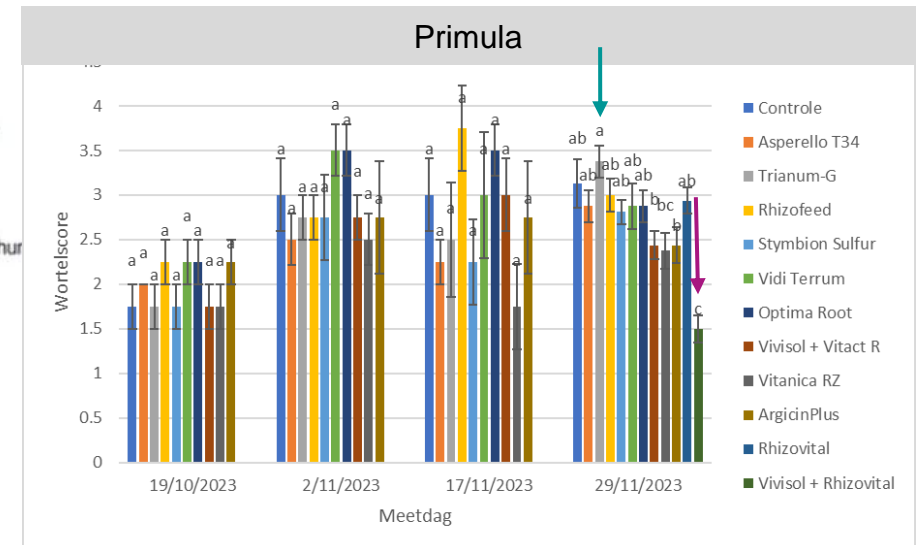
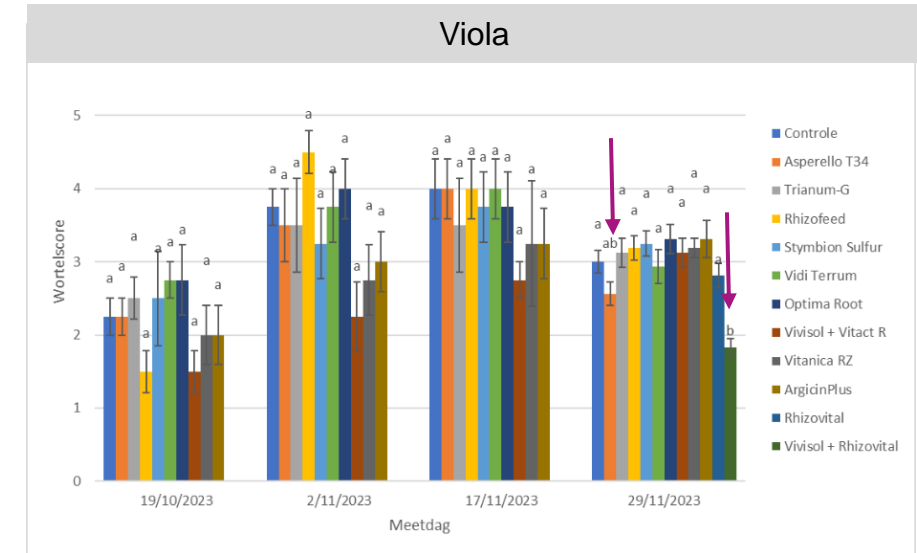
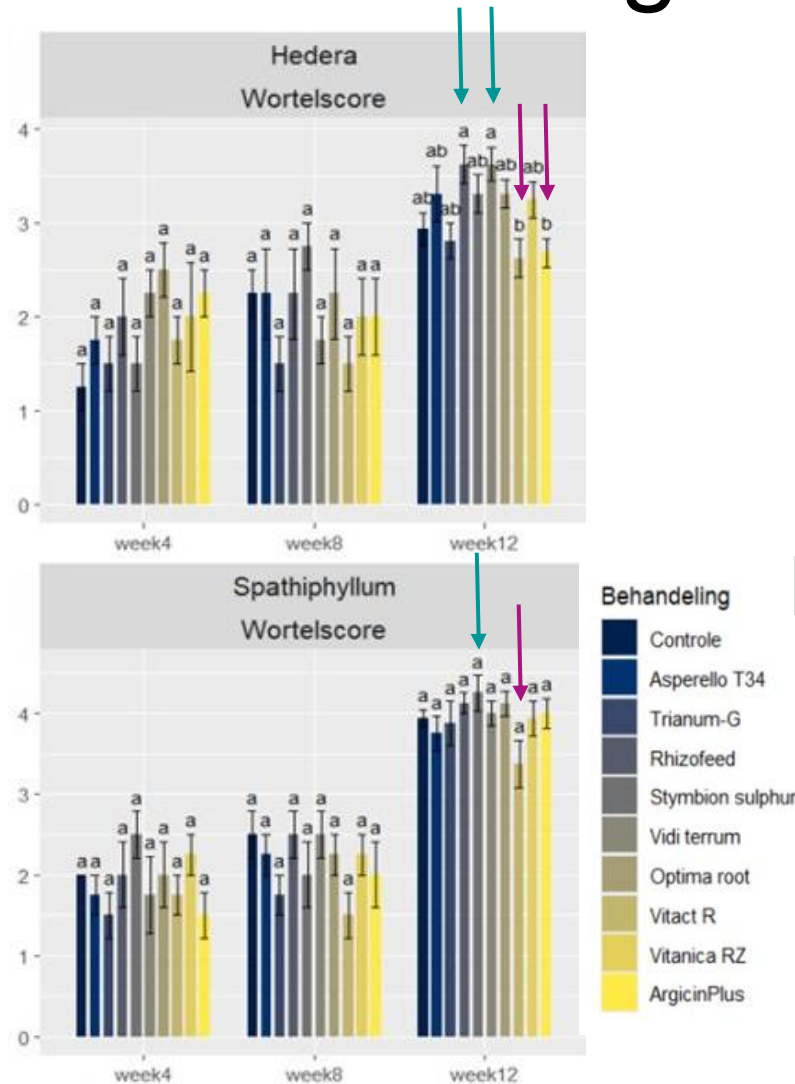
Product	Company	Type	Application mode	Frequency	Dose
Untreated control	/	/	/	/	/
Asperello T34	Biobest	MO	Pouring	2 days after potting	0.1 g/L
Triatum-G	Koppert	MO	Mixed in substrate	During potting	75 g/100 L substrate
RhiZoFeed	Intergrow	MO	Foliar spray	Just after potting + 4 weeks later	1 g/L
Stymbion Sulphur	Bion	AA	Foliar spray	Just after potting + every 2 weeks	3 mL/L
Vidi terrum	Koppert	AA	Foliar spray	Just after potting + every 2 weeks	5 mL/L
Optima root	Soiltech	Seaweed extract + humic acid	Foliar spray	Just after potting + 2 weeks later	10 mL/L
Vivisol + Vitact R	DCM	MO	Mixed in substrate Pouring	During potting 2 weeks after potting + 4 days later	1 g/L 1 mL/L
Vitanica RZ	Compo Expert	Seaweed extract + MO	Pouring	2 days after potting + every 2 weeks	0.8 mL/L
ArgicinPlus	PlantoSys	Anorganic elements + plant extract	Pouring + foliar spray	Pouring 2 days after potting + spraying every week	First 2 mL/L; 1 mL/L

# Experimental designs trials 2023-2024 indoor ornamentals: effect on rooting

**Optima Root & Rhizofeed:**  
Positive effect on growth and root development

**Vivisol + Vitact R:** more compact growth and less root development

But, difference in response depending on plant species for the other products





# Experimental design trials 2023-2024 indoor ornamentals: effect on drought stress

- Plant material: Salvia, Pelargonium, Rose and Cordyline
- Randomized block design, 4 repetitions

## - Products:

## - Measurements before, during and 1 week after stress:

- Plant height and diameter
- FW and DW
- Plant quality
- Root score
- WET-sensor
- Porometer

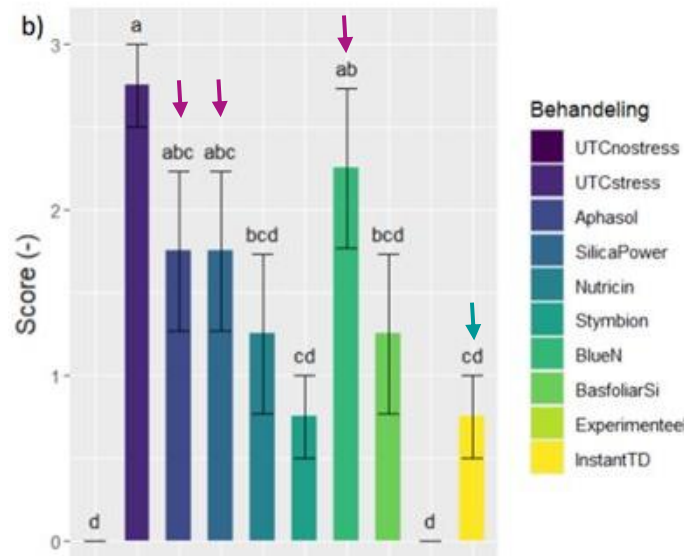


Product	Company	Type	Application mode	Frequency	Dose
Irrigated untreated control	/	/	/	/	/
Stressed untreated control	/	/	/	/	/
Aphasol	Aphasol	AA	Foliar spray	2 weeks after potting, 1-2 days before stress, 2-3 days after stress period	3 mL/L
SilicaPower	PlantoSys	Si	Foliar spray	Every 7 days	1 mL/L
Nutricin	PlantoSys	Anorganic elements + plant and seaweed extract	Foliar spray	Every 7 days	4 mL/L
Stymbion	Bion	AA	Foliar spray	Every 14 days	3 mL/L
Blue-N	Corteva	MO	Foliar spray	2 weeks after potting	0.33 g/L
Basfoliar Si	Compo Expert	Anorganic elements	Foliar spray	2 weeks and 2-3 days before stress	5 mL/L
Experimental product	Syngenta	Seaweed extract	Pouring	Every 7 days, starting 2 weeks after potting	5 mL/L
Instant TD + Impuls TD	DCM	MO	Mixed with substrate + pouring	During potting + pouring 2 weeks before stress	1 g/L + 0.25 g/L

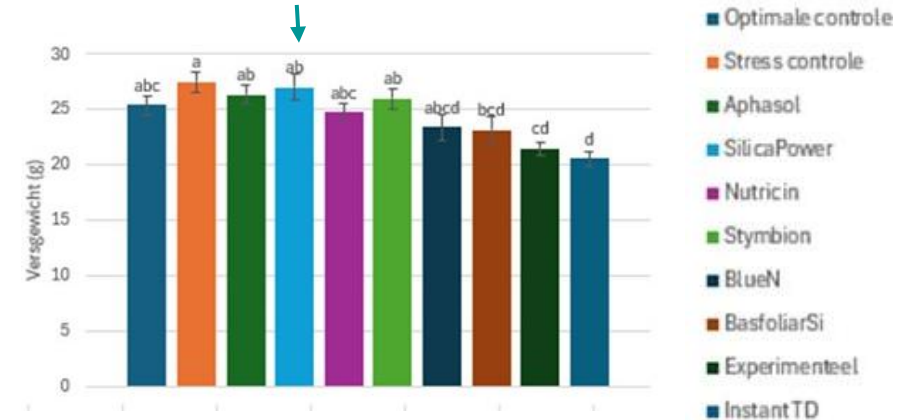
# Experimental design trials 2023-2024 indoor ornamentals: effect on drought stress

- ➔ Positive of Instant TD on drought stress tolerance of Rose and Cordyline, but more compact growth afterwards
- ➔ SilicaPower & Nutricin: more wilting but afterwards good growth and quality
- ➔ But, difference in response depending on plant species for the other products

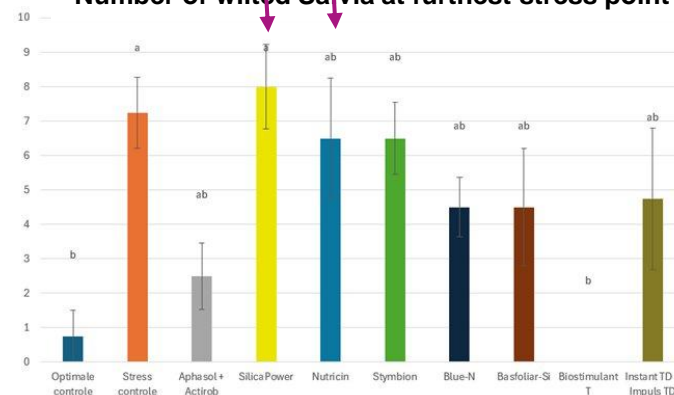
Wilt score at furthest stress point in potted rose



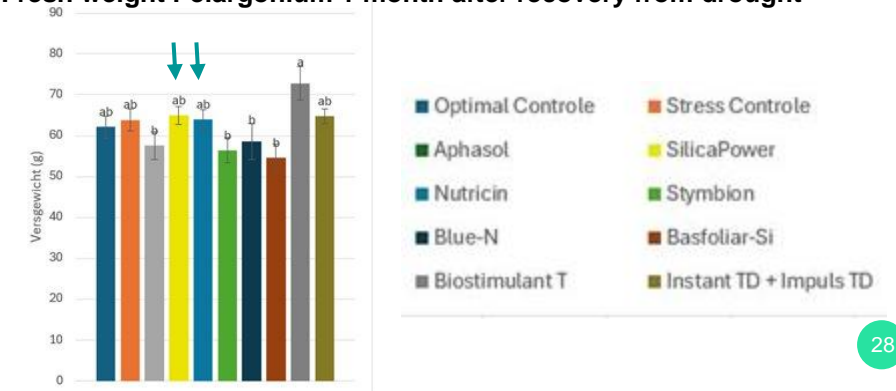
Fresh weight Cordyline 1 month after recovery from drought



Number of wilted Salvia at furthest stress point



Fresh weight Pelargonium 1 month after recovery from drought



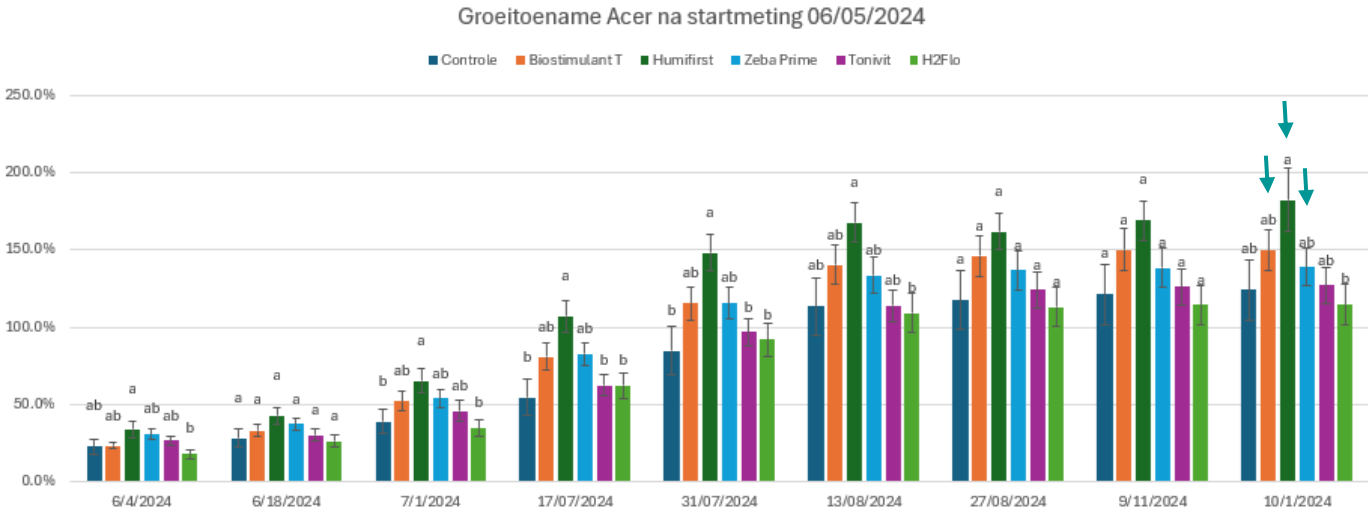
# Experimental design trials 2023-2024 outdoor ornamentals: trial on forest trees

- Plant material: *Acer pseudoplatanus*
- Randomized block design, 3 repetitions
- Products:
- Measurements every 2 weeks:
  - Plant height
  - Pressure chamber
  - Wet-sensor
  - Spectroradiometer to measure leaf reflectance

- Results:

+ Positive effect of Humifirst on growth of Acer, followed by Biostimulant T and Zeba Prime

Product	Company	Type	Application mode	Frequency	Dose
Untreated control	/	/	/	/	/
Biostimulant T	Syngenta	Seaweed extract	Pouring	Every 2 weeks	10 L/ha
Humifirst	Tradecorp	Humic and fulvic acids	In planting hole in soil	During planting	50 g/100 L soil
Zeba Prime	UPL	Soil improver - Starch	In planting hole in soil	During planting	2 g/tree
Tonivit	UPL	AA	Pouring	Once after planting	10 L/ha
H2Flo	ICL	Wetting agent	Pouring	Montly	2.4 L/ha





# Experimental design trials 2024 outdoor vegetables: trial on onion resilience (REFLECHI)

Trt	Product	Dose/ha		Application code
1	Untreated			
2	Quantis	2.000	I	CDE
3	Hamerol	3.000	I	BEH
4	BioForge	1.000	I	ACEG
5	Aphasol	2.000	I	BCDEF
6	SilicaPower	0.500	I	ABCDEFGH
7	Actisil	0.500	I	CDEF
8	Avitar	2.000	I	BCDEF

Viaverda 2024



# Experimental design trials 2024 outdoor vegetables: trial on onion resilience

Table: notation 10/06/2024

	Uniformité	Volume	Couleur	Santé
1	7	7,25	7	7
2	7	6,75	7	7
3	7	7,25	7	7
4	6,75	6,75	7	7
5	6,75	6,75	7	7
6	6,75	6,75	7	7
7	7	7,25	7	7
8	7	7,5	7	7
1=	hétérogène	petit	pâle	malsain
9=	uniforme	grand	foncée	sain

Table: notation 02/07/2024

	Uniformité	Volume	Couleur	Santé	Croissance
1	6,8	7,0	6,8	7,3	7,0
2	6,3	6,8	6,8	7,3	7,3
3	6,8	7,3	7,3	7,5	7,3
4	6,8	6,8	7,5	7,8	7,8
5	6,5	6,8	7,8	7,5	7,5
6	6,3	7,0	7,0	7,3	7,0
7	7,8	7,3	7,3	8,0	7,8
8	7,0	7,5	7,3	7,8	7,5
1=	hétérogène	petit	pâle	malsain	peu
9=	uniforme	grand	foncée	sain	beaucoup

Table: notation 12/08/2024

	Uniformité	Volume	Couleur	Santé	Croissance
1	7,5	6,8	7,0	6,0	6,8
2	7,5	7,3	7,0	6,0	7,0
3	7,8	7,5	7,0	6,3	7,3
4	7,5	7,3	7,0	6,0	7,0
5	7,5	7,3	7,0	6,3	7,3
6	7,3	7,3	7,0	6,0	7,0
7	7,5	7,5	7,0	6,5	7,5
8	7,5	7,8	7,0	6,0	7,0
1=	hétérogène	petit	pâle	malsain	peu
9=	uniforme	grand	foncée	sain	beaucoup

# Experimental design trials 2024 outdoor vegetables: trial on onion resilience

Trt		Gross yield (tonnes/ha)
1	Untreated	55,8
2	Quantis	58,1
3	Hamerol	56,1
4	BioForge	56,0
5	Aphasol	55,7
6	SilicaPower	54,1
7	Actisil	61,5
8	Avitar	55,1
tonnes/ha		56,5





# Conclusions

Group	Possible mechanisms	Subcategory causing this effect
Microbial	Activation of the plant defense (e.g. polyphenols)	M, B
	Increased or limited production of plant hormones (e.g., jasmonic and salicylic acid, ethylene, auxins, cytokinins, gibberellins)	M, B
	Competition for food and space with pathogens	B
	Production of antagonistic substances (direct or indirect elimination of diseases and pests)	B
	Activation of specific plant genes	M, B
	Increased physical defense	B
	Changes in soil structure	B
	Accelerated breakdown of organic matter	M, B

Translation from: <https://research.wur.nl/en/publications/biostimulanten-soorten-en-werkingsmechanismen>

M = Mycorrhiza, B = Bacteria

# Conclusions

Group	Possible mechanisms	Subcategory causing this effect
Non-microbial	Activation of the plant defense (e.g. polyphenols, tanins)	P, C, D
	Increased or limited production of plant hormones (e.g., jasmonic and salicylic acid, ethylene, auxins, cytokinins, gibberellins)	P, C, D, A, N
	Food source for beneficial microorganisms	P, C, D
	Production of antagonistic substances (direct or indirect elimination of diseases and pests)	P, C, A, N
	Activation of specific plant genes	P, C, N
	Increased physical defense	P, A
	Distribution of nutrients (within the plant)	C, D
	Changes in soil structure and soil microbiome	P, C, D, N
	Accelerated breakdown of organic matter	P, C, N
	Change in plant physiology	C, N

Translation from: <https://research.wur.nl/en/publications/biostimulanten-soorten-en-werkingsmechanismen>

P = Plant and seaweed extracts, C = Compost (humic and fulvic acids), D = Digestates from fresh crops and food industry by-products, N = Nutrient polymers, A = Inorganic compounds

# Thanks for your attention

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Questions?

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