






**AGROHUB**  
III Andalucía IBVF - Sevilla 2026

III Andalucía Agro-Hub PhD students meeting  
Instituto de Bioquímica Vegetal y Fotosíntesis (IBVF) (CSIC-US)  
Sevilla, 12-13<sup>th</sup> March 2026



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2 SYT1 Ca<sup>2+</sup>-related interactors

*eca1/4* mutant generation

*eca1/4* x ColP<sup>10</sup> (SYT1 & SYT2) + *eca1/4* (SYT1 & SYT2)

crossing

*eca1/4* (SYT1 & SYT2) + ColP<sup>10</sup> (SYT1 & SYT2)

MAGENTA Ca<sup>2+</sup> cytosol

YELLOW GFP:ER

wt

*syf1*





**Nutritional nitrate in**

**Introduction**

**Results**

**Conclusions**



## INTRODUCTION

The asparagus is a vegetable highly valued for its organoleptic and nutritional quality, moreover, it contains a large amount of bioactive compounds



- SAPONINS
- FRUCTANS
- FLAVONOIDS
- INSOLUBLE FIBER

- ↓ Cholesterol
- Anticarcinogenic activity
- Prebiotics
- Improve digestive health
- Antioxidants
- Antioxidant
- Prevents colon cancer



Asparagus cultivation by-products have a higher content of bioactive compounds than the spear, and they are useful in the food sector as functional ingredients, in agriculture as biostimulants and biofungicides, and in the nutraceutical sector as supplements

### ⚠️ Problems of Conventional Cultivation

- Costly harvesting
- Washing and drying processes
- Seasonal cultivation

Decline and replanting due to *Fusarium* infections and the accumulation of allelopathic substances → reduce productivity

### 💡 Alternative: Hydroponic Cultivation

- The plants grow in a controlled environment
- Continuous production throughout the year
- Higher productivity and sustainability
- Facilitates access to the roots
- Enables the study of allelopathy



## OBJECTIVES AND WORK PLAN

**1**  
Design and development of hydroponic cultivation



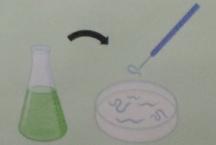
The production of roots, fronds and shoots and the extraction of bioactive compounds

**2**  
Incorporation of bioactive compounds into foods



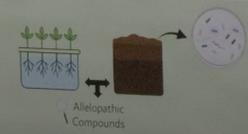
Development of healthier foods with improved shelf life

**3**  
Evaluation of the biological activity of extracts of bioactive compounds



Use of *C. elegans* models of human diseases. Alzheimer's, Parkinson's, and obesity

**4**  
Study of allelopathic compounds and soil microbiota



Allelopathic Compounds

**5**  
Study of the effect of biochar addition on soil microbiota and the reduction of allelopathic substances

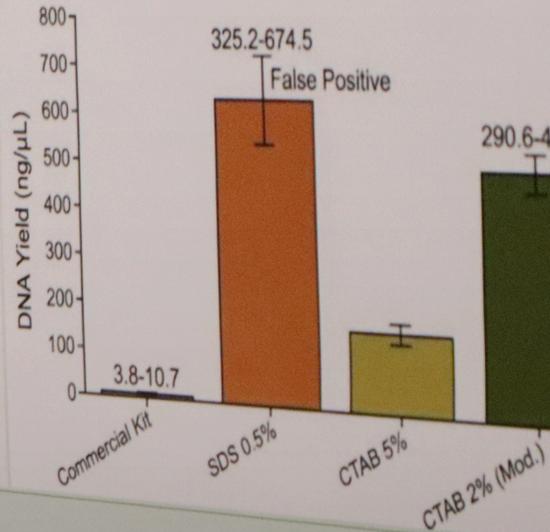
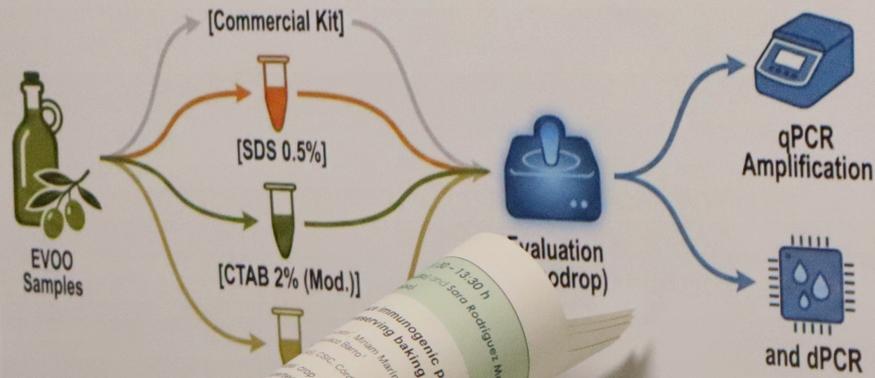


Biochar produced from lignocellulosic residues of asparagus roots and fronds through controlled pyrolysis





# METHODOLOGY



## CONCLUSIONS AND FUTURE PERSPECTIVES

CTAB protocol (especially at 2%) is established as the most effective and reproducible extraction method for lipid matrix. In contrast to the false positives generated by the anionic detergent SDS (on the basis of PCR reactions carried out afterwards), the implementation of additional steps, including magnetic purification is essential to further assessment of DNA quality by using specific methods (i.e. qubit) is essential.

- **Next Steps (Thesis Framework):** The successful isolation of high-quality DNA lays the foundation for the upcoming validation using advanced amplification methods.
- **Industry Impact:** The consolidation of this molecular tool could potentially provide the sector with a robust and verifiable method for the authentication and traceability, with the aim of placing safe food on the market and protecting public health.

**ACKNOWLEDGEMENTS**  
This doctoral thesis work is funded by the Project: "Artificial Intelligence for Food Safety and Quality Control".





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**ACRO HUB**

**OBJECTIVES**

- To elucidate the genetic basis of the interaction between *l. pneumophila* and *Cratichneumon* identifying orthologous genes related to the *nodD2* regulatory gene of *Rhizobium leguminosarum*.
- To evaluate the potential role of those genes in regulating the expression of gene involved in *Nod*-like factor biosynthesis and in establishing symbiosis with rice roots.

**ANALYSIS**

| Identity (%) | Score (bits) | E-value |
|--------------|--------------|---------|
| 52.1         | 79           | 0.02    |
| 27.3         | 69           | 0.14    |
| 37.3         | 65           | 0.77    |

**MUTANTS IN SYMBIOSIS**

Co-inoculation

**CSIC**

**Teez**

**MaX**

**Extracytoplasmic function sigma factors ( $\sigma^{ECF}$ ) in the regulation of the type III secretion system (T3SS) in *Pseudomonas***

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 Dept. of Biotechnology and Environmental Protection, Estación Experimental del Zaidin-CSIC, Granada

Pathogenic bacteria of the *Pseudomonas* genus like the phytopathogen *Pseudomonas syringae* and the human pathogen *Pseudomonas aeruginosa* use the type III secretion system (T3SS) to inject toxic effectors directly into host cells, promoting virulence and host colonization (Fig. 1). In *P. syringae* was longer known that production of T3SS requires the function of an extracytoplasmic function  $\sigma$  factor ( $\sigma^{ECF}$ ), which are regulatory proteins that binds to the RNA polymerase promoting transcription of target genes in response to specific signals. By transcriptomic analyses, we recently identified a novel  $\sigma^{ECF}$  factor of *P. aeruginosa*, named  $\sigma^{Rcd}$ , that controls T3SS production in this pathogen (Table 1). Using green fluorescence protein (*gfp*) transcriptional fusions to T3SS promoter regions (Fig. 2A) and by Western-blot using an antibody directed to the T3SS protein PcrV (Fig. 2B), we confirmed T3SS induction in response to  $\sigma^{Rcd}$ . The  $\sigma^{Rcd}$  factor belong to the ECF293 group the activity of which is activated in response to oxidative stress by a signaling cascade that also involves a zinc-containing anti- $\sigma$  factor encoded by a gene located downstream the *rctS* gene (named RcsR) (Fig. 3). By AlphaFold, we generated an interaction model of the  $\sigma^{Rcd}$ /RcsR complex (Fig. 4A), which was confirmed by BACT two hybrid analyses (Fig. 4B). Besides T3SS genes, our transcriptomic analyses showed that the  $\sigma^{Rcd}$  factor promotes production of the methionine sulfoxide reductase MsrP and MsrQ, which have been recently shown to be expressed in response to hypochlorous acid (HOCl), a compound produced by neutrophils in response to pathogens. Using *gfp* transcriptional fusions to T3SS promoters we demonstrated that the *P. aeruginosa* T3SS system is induced by HOCl in a  $\sigma^{Rcd}$ -dependent manner (Fig. 5).

**I. Transcriptomic analyses in *P. aeruginosa***

56 protein coding genes > 5-fold up-regulated in response to  $\sigma^{Rcd}$

| Gene ID | Gene Name  | Accession | Enrichment | Enrichment | Enrichment | Enrichment | Enrichment | Enrichment |
|---------|------------|-----------|------------|------------|------------|------------|------------|------------|
| PA0201  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0202  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0203  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0204  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0205  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0206  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0207  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0208  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0209  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0210  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0211  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0212  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0213  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0214  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0215  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0216  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0217  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0218  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0219  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0220  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0221  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0222  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0223  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0224  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0225  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0226  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0227  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0228  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0229  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0230  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0231  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0232  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |

**II. *gfp* transcriptional fusions to T3SS genes and Western blot**

**A**

**B**

**III. Genetic context of the  $\sigma^{Rcd}$  and RcsR  $\sigma^{ECF}$**

**IV. Interaction between the  $\sigma^{Rcd}$  and RcsR proteins**

**A**

**B**

**Table 1. Result of transcriptomic analysis showing the genes that are induced in response to the  $\sigma^{Rcd}$  factor**

| Gene ID | Gene Name  | Accession | Enrichment | Enrichment | Enrichment | Enrichment | Enrichment | Enrichment |
|---------|------------|-----------|------------|------------|------------|------------|------------|------------|
| PA0201  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0202  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0203  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0204  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0205  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0206  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0207  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0208  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0209  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0210  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0211  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0212  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0213  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0214  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0215  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0216  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0217  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0218  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0219  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0220  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0221  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0222  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0223  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0224  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0225  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0226  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0227  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0228  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0229  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0230  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0231  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |
| PA0232  | <i>gfp</i> | U00001    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    | 1488.53    |

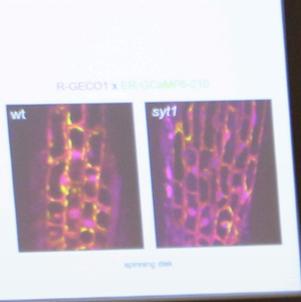
**V. The**

**Figure 5. Transcriptional fusion of T3SS promoter region of the *gfp* in *P. aeruginosa* wt and  $\Delta$ rcsR, exposed to 0  $\mu$ M and 500  $\mu$ M concentrations.**

**CSIC**

**Teez**

**MaX**



# Industrial Thermal Processing Modulates Protein Damage and Gastric Digestion in Liquid and Powdered Infant Formula

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 Dept. of Nutrition and Sustainable Animal Production, Estación Experimental del Zaidin (EEZ-CSIC), Granada.  
 leticia.benavides@eez.csic.es

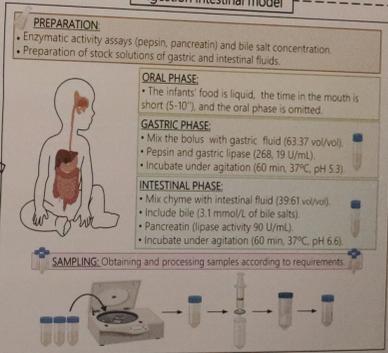
## INTRODUCTION

Infant formula (IF) serves as the primary nutritional substitute and an essential alternative to breast milk when natural breastfeeding is not feasible. However, to ensure the **microbiological safety** required by international standards, these IFs undergo rigorous **industrial thermal treatments**. Despite being necessary, such processing has been shown to induce various **protein modifications** which have the potential to compromise **nutritional quality** and the bioavailability of essential amino acids<sup>1</sup>. In the current market, IFs are primarily available in **liquid and powdered** formats, each involving significantly different manufacturing and drying technologies<sup>2</sup>. Due to these technological disparities, it is fundamental to conduct a comparative analysis of their **nutritional profiles** and, particularly, to evaluate their **gastrointestinal digestion** behavior under simulated physiological conditions that specifically mimic the **infant digestive tract**. **The aim** of this study was to evaluate and compare the impact of the physical format (liquid vs. powder) of the same infant formula on its nutritional profile and gastrointestinal digestion behavior.

## METHODOLOGY

The **BIOMILK** project aims to implement a standardized gastrointestinal digestion methodology for infants to evaluate protein digestibility and the bioaccessibility of nutrients and bioactive compounds in IF. With guidance from the INFOGEST network, a digestion procedure optimized for newborns (2 to 5 months) has been developed. To validate this methodology, the effects of industrial processing were studied using four formulas: two powdered (IF01, IF03) and their respective liquid counterparts of identical composition (IF02, IF04).

### Digestion intestinal model



- Initial characterization of infant formulas**
- CIELab Color ( $L^*$ ,  $a^*$  and  $b^*$ )
  - Thermal damage markers
  - Nutrient composition
  - Protein profile (SEC)

- Digestion and obtaining bioaccessible fractions**
- Protein digestibility (OPA method)
  - Total bioaccessible peptides
  - Protein profile (SEC)

## RESULTS

Figure 1. 3D CIELAB Colour of IFs

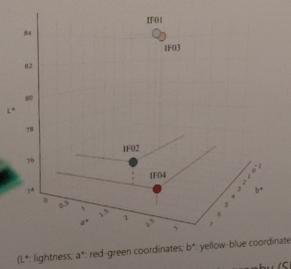


Figure 2. Thermal damage markers

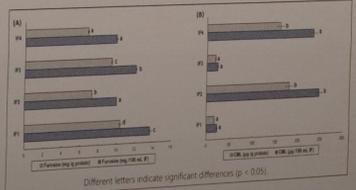


Figure 3. Protein digestibility of IFs

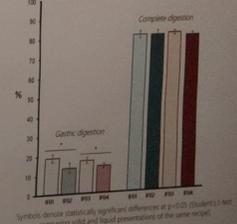
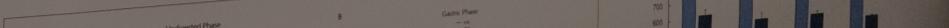


Figure 4. Size-exclusion chromatography (SEC) profiles of liquid and powdered infant formulas after *in vitro* complete intestinal digestion of IFs







CENTRO DE INVESTIGACIONES CIENTÍFICAS ISLA DE LA CARTUJA