MASTER THESIS OFFER OF CEPLAS
(FOR COMPUTATIONAL MASTER STUDENTS FROM TECHNICAL UNIVERSITY OF MADRID, UPM)

Title of Master Thesis

Identification of a citronellal biosynthetic pathway from Lemon eucalyptus comparative metabolomic and transcriptomic analyses

Description of student´s tasks

The student will develop a bioinformatic project at the National Biotechnology Centre (CSIC). The student will analyze high-throughput short- and long-read sequencing data to generate *de novo* transcriptomic data. Differential gene expression, phylogenetic and variant-calling analyses will be performed on these data.

Prerequisites: *(languages, informatics skills, bioinformatic skills, other knowledge, etc)*

Student in Biosciences Grade (Biotechnology, Biology, etc.). Previous bioinformatic skills and knowledge of languages would be an asset.

Training Project

EXTERNAL PRACTICES/MASTER THESIS. The fundamental goal of the external practices is to guide the student in applying his previously acquired knowledge to real tasks in a group work environment the realistically represents the work conditions the students will encounter in their future roles. In this way, the student will be able to get familiar with a working environment (work schedule, responsibility, attitude, organization, etc.), and with the adequate working methodology in professional reality, contrasting and applying the acquired academic knowledge.

Activities that will be performed in the academic internship/ Master Thesis:

Plants have played a vital role in human history, providing us with food and medicines. Even today, some of the most effective treatments for certain diseases are derived from plant extracts. However, the low natural yield of these compounds poses a challenge to their widespread use. Thanks to recent advances in synthetic plant biology, we can now turn plants into biofactories, boosting the accumulation of natural compounds or even producing molecules new to nature, such as human vaccines.

Plant biofactories have several advantages over classical bacterial or yeast bioreactors. Perhaps the most important is their potential for scalability, as they are autotrophic and do not require sterile conditions for growth in the open field. Plants also possess intracellular and intercellular compartmentalization, especially important when the desired product or its intermediates are toxic and need to be stored or processed in specialized cellular compartments or organs such as flowers, roots or trichomes. In addition, some of these organs naturally secrete plant metabolites and can be modified to release the desired product, which greatly facilitates the purification of the

compound produced. In addition, products from plant biofactories are generally safer and require less processing for use in humans, as they do not harbor human or mammalian pathogens.
However, to date, no plant biofactory chassis has been established in which different metabolic pathways (similar to *E. coli* or *Saccharomyces cerevisiae*) can be inserted. The aim of this project is to develop a plant biofactory platform for the sustainable production of terpenes, the largest family of plant secondary metabolites, with numerous uses in medicine and in the fragrance and flavor industries. We will use these plants to produce an insect repellent (PMD) that is currently chemically synthesized from citronellal, a plant monoterpene. This process generates hazardous waste and is not environmentally friendly. We will produce an alternative, sustainable source of this repellent.
To do this, we will first create tobacco plants (*Nicotiana tabacum*) with increased levels of metabolic precursors of terpenes and trichome density. In parallel, **we will identify the citronellal synthesis pathway by comparative metabolomics and transcriptomics of two varieties of lemon eucalyptus (*Corymbia citriodora*) that differ only in their citronellal production capacity.** We will also identify an enzyme that transforms citronellal into PMD from a fungus (*Penicillium paxilli*) that is able to produce this biotransformation very efficiently.

The student joining the project will participate in the comparative multi-omic analysis of *Corymbia citriodora*.

Objectives:
1- Metabolomic profiling of essential oil from different *Corymbia citriodora* varieties.
Essential oil extracted from different *Corymbia citriodora* varieties will be obtained and analyzed by Gas Chromatography-Mass Spectrometry. The student will perform a comparative analysis of the terpenoid profile.
2- *De novo* transcriptomic analyses of citronellal-producing *Corymbia citriodora* varieties.
Currently, only the genome of a non-citronellal producing variety has been sequenced. The student will generate a *de novo* transcriptomic assembly of the varieties that produce the highest amount of citronellal.
3- Identification of *Corymbia citriodora* genes potentially involved in citronellal synthesis.
Candidate genes involved in citronellal synthesis will be identified by comparative transcriptomic analysis of citronellal versus non-citronellal *Corymbia citriodora* varieties. Genes with potential enzymatic roles involved in the pathway (alcohol dehydrogenase, reductases, etc.) will be identified and differential gene expression, phylogenetic and variant-calling analyses will be performed on these datasets.

Nº of positions offered:	**1**
Has the student dealings with underage persons?	**No**
Starting date:	**1/10/2023**
Fecha de fin: **(End date)**	**1/07/2024**
Horas semanales: **(Weekly hours)**	**Between 20 and 35 weekly hours.**
Horario jornada laboral: **(Working hours)**	**Adaptable between 9:00 and 19:00.**
Importe Ayuda/Bolsa de estudio: **(Amount of fellowship / remuneration)**	**Application to external funding will be supported (e.g., JAE-intro).**
Tutor académico: (Academic tutor (UPM)) Email:	
Departamento tutor académico: **(Dept. of academic tutor)**	

Tutor empresa: **(External tutor)**	**Eduardo González Grandío**
Email tutor empresa: **(Email external tutor)**	**Eduardo.gonzalez@cnb.csic.es**
Departamento tutor empresa: **(Dept. of external tutor)**	**Departamento de genética molecular de plantas.**
Ubicación de la estancia de las practicas **(Location of the internship)**	**Calle Darwin 3, 28049, Madrid**
ENTIDAD COLABORADORA: **(Collaborating Entity)**	**Centro Nacional de Biotecnología.**
A cumplimentar por Oficina Prácticas ETSIAAB: **Créditos a reconocer (Nº ECTS)**:	