DOCTORAT ECOLOGIE
BRETAGNE GEOSCIENCES
LOIRE AGRONOMIE ALIMENTATION

PhD PROPOSAL FOR THE DOCTORAL SCHOOL « Ecologie, Géosciences, Agronomie, ALimentation »

GENERAL INFORMATION

Thesis title: regulation of leaf PRImary MEtabolism and sink/source relationships during acclimation to drought stress in Brassicaceae
Acronym: PRIMABRA
Disciplinary field 1: Agronomy **Disciplinary field 2:** Food sciences
Three keywords: physiology, metabolism, drought
Research unit : IGEPP (UMR 1349, INRAE-Institut Agro-Université de Rennes 1), Institute for genetics, environment and plant protection (https://www6.rennes.inrae.fr/igepp/)
Name of the thesis director: Alain Bouchereau **Email address of the thesis director :** alain.bouchereau@univ-rennes1.fr **Name of the thesis co-supervisor 1 (if** applicable): Younès DELLERO **Email address of the thesis co-supervisor 1 (if applicable):** younes.dellero@inrae.fr
Thesis grant (funding origin and amount): **50% CDE UR1** and 50% ARED
Contact(s) (mailing address and E-mail): Alain Bouchereau, UMR IGEPP, Domaine de la Motte, BP 35327,35653 Le Rheu cedex, France, alain.bouchereau@univ-rennes1.fr
Recruitment process: Recruitment process depends on thesis funding. To select the corresponding recruitment process, please visit the EGAAL website here. This information is needed for proposal publication. ☒ **Doctoral school contest** ☐ **Interview** ☐ **Other (indicate) :**

All sections must be filled. Once filled, please save the proposal form in pdf format using the following naming: Supervisor Name_Unit_Subject Acronym_EN.pdf

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SCIENTIFIC DESCRIPTION OF THE PhD PROJECT

Socio-economic and scientific context : (10 lines)

OilSeed Rape (OSR) is a major oleaginous crop cultivated in the world, with important applications for human and animal nutrition and biofuel production. However, the yield potential of winter OSR (largely grown in Europe) is threatened today by increasing climate change and in particular by drought stress in the first stages of the growth cycle (automnal vigor). Indeed, long-term drought stress strongly affects biomass accumulation and resource allocation between organs in OSR at the vegetative stage with further strong consequences on seed yield establishment . In this context, contrasted regulations for metabolism and its dynamic must proceed according to the developmental stage of leaf tissues, and contribute to optimize carbon and nitrogen resource allocation and distribution from source tissues (old) to sink tissues (developping). However, these ultimate metabolic adjustments require a specific regulation of metabolic fluxes within plant primary metabolism, which remained to be dissected. Leaf primary metabolism comprises different metabolic pathway strongly entangled together: photosynthesis, photorespiration, glycolysis, tricarboxylic acid cycle, nitrogen assimilation. The PRIMABRA PhD project aims to unraveling the modifications of metabolic fluxes within leaf primary metabolism supporting osmotic, energetical and nutritional adjustments of OSR during acclimation to drought stress. We are also interested in studying the inter-specie variability associated to these metabolic regulations in brassicaceae in order to promote a potential future utilization of this diversity for crop improvement.

Assumptions and questions (8 lines)

The progressive apparition of water shortage rapidly induces stomatal closure that negatively affects photosynthesis. Consequently, carbon and nitrogen lost by the photorespiration process increase, similarly to the properties to retain carbon and nitrogen required for osmotic potential adjustments, which subsequently affects carbon and nitrogen transport by xylem and phloem tissues (additional osmotic stress). Therefore, oilseed rape acclimation to drought stress relies on its capacity to reorchestrate the functionning of different pathway of leaf primary metabolism and their interconnections through the regulation of metabolic fluxes, in order to maintain energy production (tricarboxylic acid cycle) for cellular growth process in young leaves and nutrient remobilisation from old leaves. The PRIMABRA PhD project aims to answer the following questions:

- What is the impact of drought stress on leaf primary metabolism efficiency and source-sink relationships in oilseed rape ?
- Which metabolic steps from the tricarboxylic acid cycle and its connection areregulated during acclimation to drought stress with respect to nutrient remobilisation ?
- Is there any interspecie variability within brassicaceae associated to these metabolic regulations during drought stress ?

The main steps of the thesis and scientific procedure (10-12 lines)

The PhD will be separated into three tasks. For each task, plants for oilseed rape (*Brassica napus*), chinese mustard (*Brassica juncea*) and ethiopian mustard (*Brassica carinata*) will be grown for few weeks (developmental seed vigor evaluation) in a growth chamber and a moderate drought stress will be applied during two weeks (40% of field capacity). Sample harvesting will be scheduled to consider spatial and temporal evolution of plant response notably through the separation of young and old leaves.

Task 1: Impact of drought stress on the physiological status of sink and source leaves of brassicaceae.

- ⇨ Measurements of leaf water content, leaf osmotic potential, leaf gas exchanges (photosynthesis/photorepsiration/mitochondrial respiration) and photosystem II fluorescence.

Task 2: Impact of drought stress on the functionning of the tricarboxylic acid cycle in sink and source leaves of brassicaceae.

- ⇨ Measurements of enzymatic activities and metabolite contents in relation with the tricarboxylic acid cycle
- ⇨ Measurements of levels and ratios of co-factors linked to energetical and redox cell status (ATP/ADP, NADH/NAD, NADPH/NADP, GSH/GSSG, Ascorbate/dehydroascorbate)

Task 3: Modeling metabolic flux within the tricarboxylic acid cycle in sink and source leaves of brassicaceae

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⇨ Labeling expriements with ^{13}C-metabolic probes, measurements of ^{13}C-enrichment within metabolites linked to tricarboxylic acid cycle activity, Software-assisted modelling of metabolic flux (ScalaFlux and Influx).
Methodological and technical approaches considered (4-6 lines) The PhD candidate will learn how to apply, manage and evaluate the impact of a controlled drought stress on a crop species. The PhD candidate will also use several targeted and complementary approaches within the field of plant eco-physiology, enzymology, metabolomic and ^{13}C-based fluxomic. To perform these approaches, the PhD candidate will benefit from our network of scientific collaborations with the metabolomic/fluxomic platform P2M2 from our IGEPP laboratory (Rennes), the IPS2 laboratory (Paris-Saclay), the Bordeaux metabolome platform and the Toulouse metabolomic and fluxomic platform "MetaToul".
Scientific and technical skills required by the candidate We ar looking for a PhD candidate with a strong knowledge in plant physiology and metabolism. Skills in scientific communication, writing and english speaking are also essential. We strongly encourage applicants owning a first experience in the management of any abiotic stress to plants, the quantification of leaf primary metabolites (sugars/organic acids/amino acids) or the measurement of leaf gas exchange parameters. Some knowledge in statistical treatment tools will be a real advantage for the PhD candidate. Capacities to work in a team will be valuable.

THESIS SUPERVISION[1]

Unit name: IGEPP	**Team name:** RCA (Yield under abiotic challenges)
Unit director name: Maria MANZANARES-DAULEUX	**Team director name:** Nathalie NESI
Mailing address of the unit director: maria.manzanares-dauleux@inrae.fr	**Mailing address of the team director:** nathalie.nesi@inrae.fr
Thesis director Surname, first name: Alain Bouchereau Position: Professor Obtained date of the HDR (Habilitation thesis to supervise research): July 2001 Employer: Université de Rennes 1 Doctoral school affiliation: EGAAL Rate of thesis supervision in the present project (%): 50 % Total rate of thesis supervision in ongoing theses (supervisions and co-supervisions) (%): 0 % Number of current thesis supervisions/co-supervisions: 0	
Thesis co-supervisor 1 (if applicable)	

[1] **In EGAAL Doctoral School, if only one scientist in thesis supervision = 100% of supervision rate; if 2 people involved in thesis supervision = from 50% to 70% of supervision rate for the director; if 3 people involved in thesis supervision = 40% / 30% / 30% of supervision rate distribution among supervisors.**

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Surname, first name: Younès DELLERO

Position: Researcher

Habilitation thesis to supervise research ☐ yes ☒ no If yes, date diploma received:

Employer: INRAE

Doctoral school affiliation: EGAAL

Rate of thesis supervision in the present project (%): 50 %

Total rate of thesis supervision in ongoing theses (supervisions and co-supervisions) (%): 0 %

Number of current thesis supervisions/co-supervisions: 0

Professional status of previous PhD students supervised by both director and co-supervisors (from 5 years)

Please provide the following information for each PhD students supervised

Surname, first name: Sylvain DECHAUMET

Date of PhD beginning and PhD defence: 02/11/2014-18/05/2018

Thesis supervision: Alain BOUCHEREAU

Professional status and location: Project manager, Medday Pharmaceuticals, Paris

Contract profile (post-doc, fixed-term, permanent):

List of publications from the thesis work:

Dellero Y, Clouet V, Marnet N, Pellizzaro A, **Dechaumet S**, Niogret MF, **Bouchereau A**. 2020. Leaf status and environmental signals jointly regulate proline metabolism in winter oilseed rape. J Exp Bot 71, 2098-2111. DOI : 10.1093/jxb/erz538

Poret M, Chandrasekar B, van der Hoorn RAL, **Dechaumet S**, **Bouchereau A**, Kim TH, Lee BR, Macquart F, Hara-Nishimura I, Avice JC. 2019. A Genotypic Comparison Reveals That the Improvement in Nitrogen Remobilization Efficiency in Oilseed Rape Leaves Is Related to Specific Patterns of Senescence-Associated Protease Activities and Phytohormones. Front Plant Sci 10, 46. DOI: 10.3389/fpls.2019.00046

Five main recent publications of the supervisors on thesis subject:

Dellero Y, Heuillet M, Marnet N, Bellvert F, Millard P, **Bouchereau A**. 2020. Sink/Source Balance of Leaves Influences Amino Acid Pools and Their Associated Metabolic Fluxes in Winter Oilseed Rape (Brassica napus L.). Metabolites 10, 16. . DOI : 10.3390/metabo10040150

Dellero Y, Clouet V, Marnet N, Pellizzaro A, Dechaumet S, Niogret MF, **Bouchereau A**. 2020. Leaf status and environmental signals jointly regulate proline metabolism in winter oilseed rape. J Exp Bot 71, 2098-2111. DOI : 10.1093/jxb/erz538

Poret M, Chandrasekar B, van der Hoorn RAL, Dechaumet S, **Bouchereau A**, Kim TH, Lee BR, Macquart F, Hara-Nishimura I, Avice JC. 2019. A Genotypic Comparison Reveals That the Improvement in Nitrogen Remobilization Efficiency in Oilseed Rape Leaves Is Related to Specific Patterns of Senescence-Associated Protease Activities and Phytohormones. Front Plant Sci 10, 46. DOI: 10.3389/fpls.2019.00046

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Dellero Y, Lamothe-Sibold M, Jossier M, Hodges M. 2015. Arabidopsis thaliana ggt1 photorespiratory mutants maintain leaf carbon/nitrogen balance by reducing RuBisCO content and plant growth. Plant J 83, 1005-1018. DOI: 10.1111/tpj.12945

Hatzig S, Zaharia LI, Abrams S, Hohmann M, Legoahec L, **Bouchereau A**, Nesi N, Snowdon RJ. 2014. Early osmotic adjustment responses in drought-resistant and drought-sensitive oilseed rape. J Integr Plant Biol 56, 797-809. DOI: 10.1111/jipb.12199

THESIS FUNDING

Origin(s) of the thesis funding: 50% ARED and 50% CDE UR1
Gross monthly salary: 1757 €
Thesis funding state : Non acquired
Funding beginning date/Funding ending date: September 2021 – 3 years

Date: 24 March 2021

Name, signature of unit director: IGEPP

Maria MANZANARES-DAULEUX P/O Christophe MOUGEL

Christophe MOUGEL
Directeur Adjoint UMR IGEPP

Name, signature of team director: RCA

Nathalie NESI

Name, signature of thesis project director:

Alain BOUCHEREAU