AMPHIPHILES BIO-SOURCÉS : SYNTHÈSE ET CARACTÉRISATION COLLOÏDALE

FULLY BIO-SOURCED AMPHIPHILES: SYNTHESIS AND COLLOIDAL CHARACTERIZATION

Etablissement **Avignon Université**

École doctorale **Agrosciences et Sciences**

Spécialité **CHIMIE**

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Encadrement de la thèse **GREGORY DURAND**

Financement du 01-10-2024 au 30-09-2027 *origine* **Ecole doctorale** *Employeur* **Avignon Université**

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Amphiphiles, natural products, detergents, terpenes, biosources, green chemistry

Description de la problématique de recherche - Project description

The aim of the thesis project is to design and develop new amphiphiles derived from biomass by eco-friendly methods. In order to expand alternatives to classical detergents widely used in daily products and industrial applications, we propose to promote natural building blocks and renewable resources. Our strategy for the design of bio-sourced amphiphilic structures will be based on:
(a) Identifying relevant pairing of hydrophilic and hydrophobic moieties based on bioavailable materials,
(b) Investigation of innovative mild synthetic approaches for modification and coupling conditions (i.e. microwaves assisted synthesis, green solvents, low carbon impact purifications methods, ...)
(c) Carry out a complete characterization of their colloidal properties
(d) Complete characterization of their colloidal properties.

Among the bio-sources, two will be considered : (1) the apolar terpens obtained from fruit and vegetable sources, and (2) the polar bio-polymer, inulin, a fructan-type oligosaccharide extracted from chicory.

Thématique / Domaine / Contexte

This project falls within the themes of AXIS II of the EUR Implanteus: Preservation, extraction, transformation and formulation of vegetable matter. with the objective of designing and synthesizing added values products from bioavailable resources mainly from vegetable origin. This project represents a great opportunity for students to develop interdisciplinary scientific profile.

According to Marketsandmarkets, surfactants market reached $42.1 billion in 2020 and it is estimated to continue growing up to $52.4 billion in 2025. However, most of these synthetic surfactants are obtained from petrochemicals, their production as well as their elimination undergo chemical processes that release toxic wastes. With increasing awareness of the potential adverse impact of conventional surfactants on the environment and human health, it seems essential that bio-based surfactants emerge to offer safer, more affordable, available in abundance, and biodegradable alternatives (1). Therefore, the conception of surfactants based on renewable bio-resources with “greener” methods becomes a highly important issue (2) Indeed, the global market of such surfactants is expected to grow from $6.9 to nearly $14 billion by the end of 2030 as reported by the latest analysis by Virtue Market Research. Among the various applications of surfactants, one can cite food industry, (3) cleaning products (household or laundry), cosmetics (4) or pharmaceuticals. All these industries are therefore actively seeking to biocompatible and greener alternative surfactants.

Objectifs

Our lab has a strong expertise in the development of synthetic surfactants and amphiphilic polymers and their full colloidal characterization with biomedical applications. With this project, we want to combine agro-food derived materials with low environmental impact chemistry to create added-value bio-sourced detergents.

We will consider two families of natural compounds widely available to conceive valuable library: (a) terpenes as hydrophobic source and (b) carbohydrates as a supply of simple to complex hydrophilic moieties.

The main challenge will be to work on the proper association between them to finely tune the hydrophobic/hydrophilic balance. Mild and eco-friendly synthetic approaches will be considered.

Méthode

The research program will be as follows (A Gantt diagram is also attached):

1/ State of art summary

To initiate properly the project prior to start experimental activities, constitution of an exhaustive bibliography will be necessary. This part will benefit from preliminary bibliographic study conducted by a Master 1 student from IMAS program (EUR Implnateus) . During the state of art evaluation, some clarifications will emerge regarding several factors:
(a) structural parameters of existing bio-sourced detergents and use in industry,
(b) among known modification's methodologies, selection of « green » compatible strategies in order to minimize the use of classical methods (often time and power consuming, not eco-friendly, requiring harsh conditions and/or polluting solvents, reagents or starting materials),
(c) use of computational methods to simulate several parameters related to hydrophobicity such as HLB or log P to predict surfactants behavior (solubility, shape of auto-assembly...) and applications (emulsifier o/w or w/o, detergent, solubilizing agent).

2/ Development of bio-sourced amphiphiles: "head-to-tail" terpene-based surfactants

Hydrophobic terpenes will be used as an apolar tail simple model to allow introduction of non-ionic and ionic polar heads. We plan to use citronellol, generally extracted from stems and leaves of any specific species of Cymbopogon, known as Java Citronella. Citronellol is commonly applied in the perfumery and cosmetic industries as a source of flavor and its bioactive properties are also exploited as insect repellent, antibacterial or antifungial. Due to its presence of the primary hydroxyl group in its structure, it can be used as a chemical reagent and provide a useful platform for preparation of esters. Those are already applied in field of materials as monomer for bio-sourced polymers (5) or in perfume industry as pro-fragrances components (6) This work will stand as comparison to develop more efficient and green methods of synthesis. Synthesis optimizations will be transposed to adequate more complex and more innovative architectures.

To increase the hydrophobic contribution, sesquiterpenes or diterpenes could be considered. Among them, phytol seems to be a good candidate. It is considered as the most abundant acyclic isoprenoid present in the biosphere of our planet. This diterpene member of the long-chain unsaturated acyclic alcohols is produced by almost all photosynthetic organisms, including algae, plants and bacteria as a part of the chlorophyll molecule. While primarily used as a fragrance constituent, its significant biological properties have recently drawn the attention for possible application in pharmaceutical fields (7). Phytol has also been used to build sacrificial surfactant used as herders on oil spill mitigation (8)

3/ Development of bio-sourced amphiphiles: Inulin-Based oligomeric surfactants

Fructo-oligosaccharides (FOS) or fructans are natural biopolymers found in chicory root, garlic, onion and other fruits. Among them, inulin is linear polymer composed of fructose units connected with each other by β(2□1) glycosidic linkages with a single terminal α(1□2) bonded D-glucose. It is mainly extracted from chicory, Jerusalem artichoke and dahlia and its degree of polymerization (DP) ranges from 2 to about 60, depending on its source. In this project, we want to use inulin obtained from chicory as starting material for hydrophobic functionalization on the native or oxidized polymeric structure. Primary and secondary alcohols (in purple) will be used for terpene grafting on native backbone to build amphiphilic nature. Inulin can also be selectively oxidized (in red) in order to generate carboxylic acid suitable for hydrophobic derivatization.

Inulin has received "generally recognized as safe" status by the Food and Drug Administration (FDA) due to its several outstanding properties including biodegradability, renewability, nontoxicity and therapeutics properties (9). In addition, the biopolymer witnesses many advantages:
(a) – ability to handle variety of chemical modifications (10),
(b) – tunable scaffold to control degree of substitution and so amphiphilic balance,
(c) – versatility in potential uses such as emulsifiers, functional food (11) or surfactant tools to study membrane proteins (12), that could be beneficial to price final applications.

Moreover, chicory and inulin draw increasing interest from vast societal program with deep impacts. The H2020 European €7.3 million CHIC (for "Chicory Innovation Consortium") project promoted chicory culture to produce dietary inulin and bioactive terpenes for a bio-based economy (13).

4/ Purification and Characterization of biosurfactants

A large part of this work will consist in the purification and characterization of the synthesized molecules. We will try to avoid as much as

possible classical purification methods and use more eco-friendly and scalable processes such as size exclusion chromatography. Dialysis will also be considered for purification as the lab has large scale dialysis equipment. For the inulin, we will focus our attention to the small FOS to allow purification by HPLC using HILIC column.
Once purified, the new structures will be fully characterized using spectroscopic techniques such as 1H, 13C and 2D NMR, and mass spectroscopy. In addition, gas chromatography could be a powerful tool to analyze terpene derivatives as well as FTIR for final characterization of modified inulin.

5/ Colloidal Characterization of biosurfactants
Colloidal properties will be studied, to understand how those molecules self-assemble in solution in terms of critical micellar concentration (CMC), water solubility, size and shape of the micelles. The CMC/CAC is the most important parameter characterizing a surfactant. We will use a range of complementary techniques to determine CMC values, including surface tension (ST), spectrofluorimetry (Fluo), and 1H NMR. Additionally, on promising molecules, analytical ultracentrifugation (AUC) and small-angle X ray or neutron scattering (SAXS and SANS, respectively) will be performed using the FRISBI platform in Grenoble.

Dynamic light scattering (DLS) provides a quick and inexpensive means of characterizing the average hydrodynamic size, dispersity and zeta-potential. The homogeneity and aggregation number will be probed by SEC-MALLS through the CPER platform, and more detailed information could be gleaned from AUC, SAXS or SANS.

Références bibliographiques

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Précisions sur l'encadrement - Details on the thesis supervision

We will have formal weekly meetings with the PhD student and daily oral and email interactions. The PhD will present the result of the week with a short and concise powerpoint presentation.
The PhD student will present his/her work to the lab on a monthly basis.
Every trimester we will discuss the progress made by the PhD student and whether it fits with the calendar. Ajustement in the prioritary lines of research will be done when necessary.

The PhD student will be trained in the lab by Marine Soulié (Research Engineer) and Dr Pierre Guillet.
Dr Gregory Durand and Marine Soulié will provide their expertise in organic synthesis and colloidal characterization
Dr Pierre Guillet will provide his expertise in colloidal characterization and polymer science
Dr Maher Abla will provide his expertise in polymer post-functionnalization and HILIC characterization

Conditions scientifiques matérielles et financières du projet de recherche

The host laboratory has a well-equipped organic chemistry lab, in which the proposed research can be conducted without difficulty. It has state-of-the-art analytical facilities for the analytical and physical chemical characterization of the synthesized compounds. Available instruments include 400 MHz Bruker NMR spectrometer, Krüss K100 tensiometer, UV and IR spectrometers. UPLC-MS and Mass spectrometry is possible through the common facilities of the Institute the lab belongs to.

More info on the Lab can be find at: http://s2cb.univ-avignon.fr/

Ouverture Internationale

As part of a Student Exchange Program, the homogeneity and aggregation number of the self-assemblies could be probed by SEC-MALLS in Graz University in the lab of Pr Sandro Keller.

As part of a Student Training Program, selected promising molecules could further studied by , analytical ultracentrifugation (AUC) and small-angle X ray (SAXS) at the FRISBI platform in Grenoble

Objectifs de valorisation des travaux de recherche du doctorant : diffusion, publication et confidentialité, droit à la propriété intellectuelle,...

The multidisciplinary nature of the project will provide to the doctoral student a full and complete experimental training as he/she will work on: (1) organic synthesis of amphiphiles; (2) full chemical characterization and thorough purification of the molecules produced and; (3) thorough colloidal characterization of the self-assemblies.
The project involves therefore several majors such as organic synthesis, physical-chemistry and analytical chemistry. The PhD program will involve other scientific tasks such as scientific reporting and oral dissemination as well as scientific writing (publication and possibly patent). The PhD doctorate will interact with collaborators on a regular basis which will allow him/her to develop team work abilities.

Collaborations envisagées

We have initiated a collaboration with Dr. Sumit Bhawal and Dr. Sandeep R. Patil, from Navrachana University, India. This preliminary work will be helpful to adjust relevant targets in term of colloidal properties and could be strengthened during the doctoral project.

Dr. Maher Abla has joined the team S2CB as an associated member and he will provide his expertise on biopolymers functionalization and characterization.

We will collaborate with the group of Pr. Sandro Keller (University of Graz) on the characterization of the colloidal properties of the self-assemblies formed by our molecules and on the determination of their detergency i.e. the ability to solubilize membrane and lipids.

We will apply to the training courses organized by FRISBI platform so that the candidate will be able to conduct experiment at the synchrotron in Grenoble.

Profil et compétences recherchées - Profile and skills required

• Excellent Master’s degree in chemistry, specialization in organic chemistry is preferable
• Strong interest in research and significant first experience in experimental activities in the lab
• Background or interest in bio-ressources modifications, green chemistry or physico-chemistry could be a plus
• Ability to work effectively in a team and good communication skills (spoken and written)
• Intrinsic curiosity, rigor et adaptability are sought qualities

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