



EXTRACTION OF BIOACTIVES USING DEEP EUTCTIC SOLVENTS FROM MARINE BY-PRODUCTS FOR THEIR POTENTIAL APPLICATION IN THE DRY EYE DISEASE TREATMENT

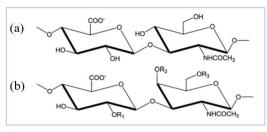
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A novel green technique is developed to extract bioactive compounds from the marine biomass for their potential application in the treatment of the dry eye disease (DED). Natural deep eutectic solvents (DES) are used for the extraction of bioactives using a green lowcost process. The obtained extracts are characterized to assess their composition in hyaluronic acid (HA) and chondroitin sulfate (CS), proteins, lipids and ash. DES and the isolated bioactives are physically and chemically characterized. In addition, bioassays are also performed to assess the biocompatibility, anti-inflammatory, antioxidant and antimicrobial activity to ensure their potential application on DED treatment.



HA and CS have shown to be effective compounds in the treatment of DED. Their conventional extraction techniques are highly complex and time-consuming, as they include the use of organic solvents, enzymes and detergents to break the cellular structures of the tissues. Therefore, the application of a green methodology that is based on the use of alternative solvents is strongly recommended. In this study, DES are applied as low cost, novel and green solvents to extract HA and CS. Different combinations of DES at specific ratios are evaluated to obtain the most suitable solvents to dissolve HA and CS from marine sources, at optimal extraction conditions. Hence, marine biomass is used as the source for the biopolymers, as it is a inexpensive and highly abundant renewable raw material. The isolated extracts obtained using the designed DES are characterized to study their composition.



Chemical Structure of a) Chondroitin Sulfate (R=H or COCH $_3$), b) Hyaluronic acid



The extraction porcedure is based on the use of different combinations of DES to isolate the biopolymers. The DES are analyzed theoretically and experimentally to evaluate their thermodynamic and physicochemical properties. The isolated biopolymers are characterized to assess their composition in HA, CS, lipids, proteins and ash. *In vitro* studies are done on human corneal epithelial cell line and on DED-associated microorganisms.



- Simulation of the thermodynamic behavior of designed DES; synthesis of the novel solvent systems; theoretical and experimental physicochemical characterization.
- 2. Characterization of extracts obtained from marine by-products using DES.
- 3. *In vitro* studies of the cellular viability, antioxidant, anti-inflammatory and antimicrobial activities of selected compounds to assess their potential application for the DED treatment.









