

Snail Extracted Mucus and Cosmeceutical Applications: Strategies To Increase Its Beneficial Effects



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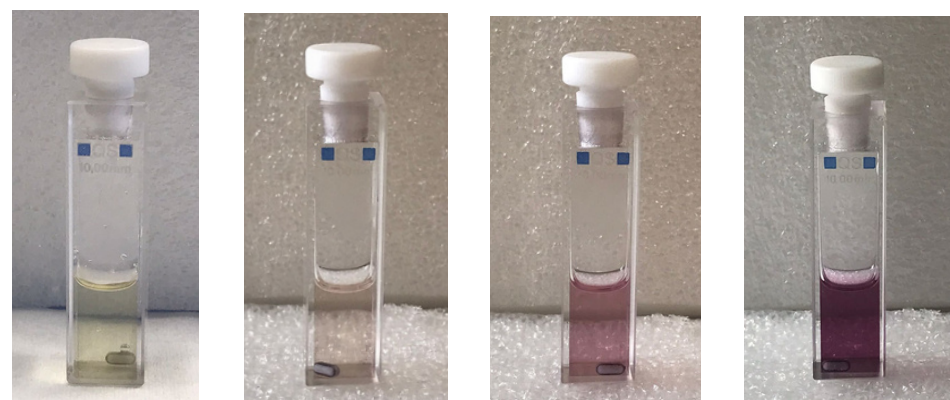
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Introduction

Nowadays, several studies have highlighted the ability of snail mucus in maintaining healthy skin conditions due to its emollient, regenerative and protective properties. In particular, mucus derived from *H. aspersa muller* has already been reported to have beneficial properties such as antimicrobial activity and wound repair capacity. In order to enhance beneficial effects of snail mucus two different approaches were exploited: enrichment with antioxidant compounds derived from edible flowers wastes (*Acmella oleacera*, *Centaurea Cyanus*, *Tagetes erecta*, *Calenda officinalis*, *Moringa Oleifera*) and development innovative nanoparticles with added therapeutic value.



SM-NP formulation development

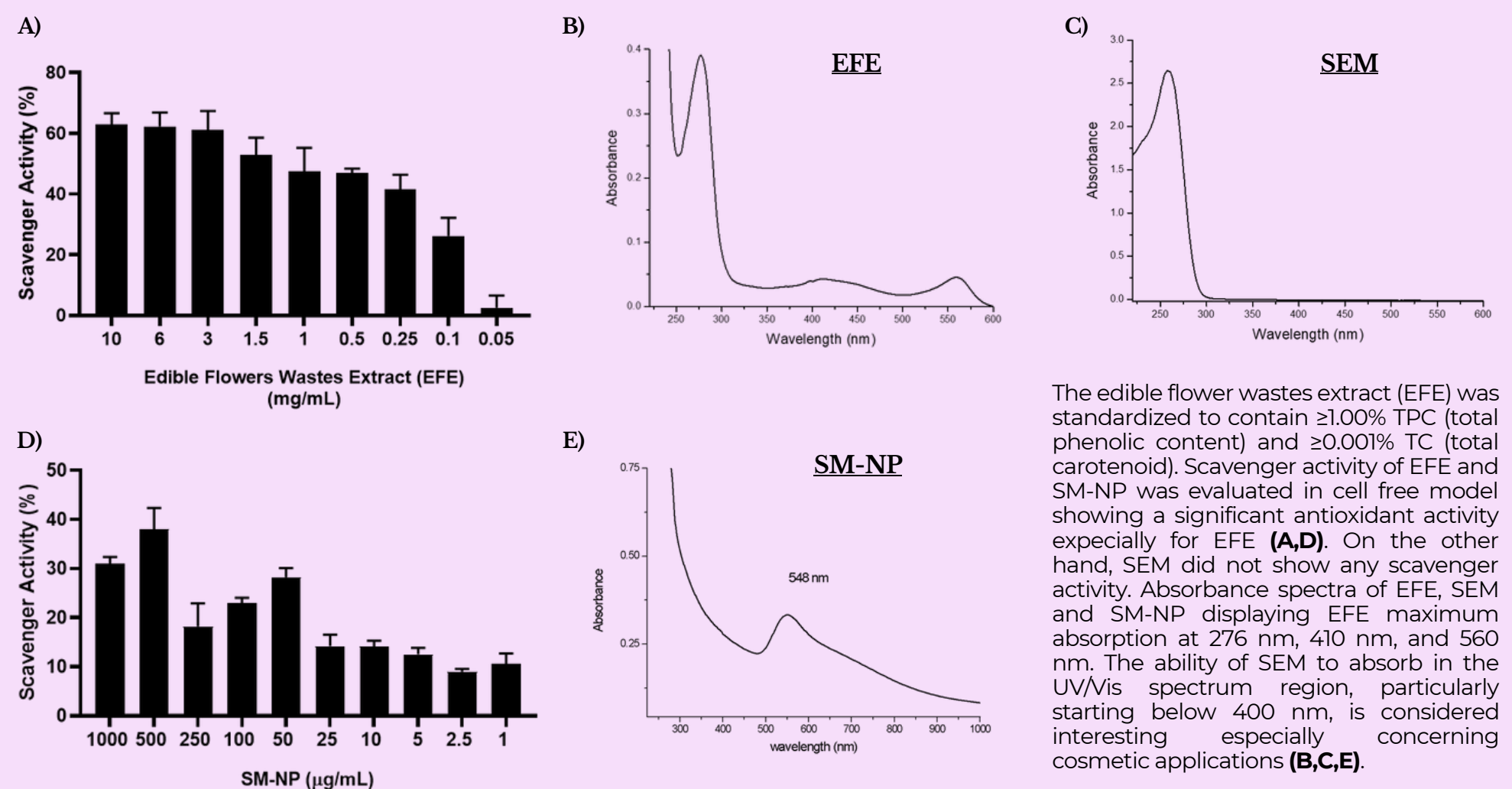
Methodology

UVB-damage ($\lambda = 280\text{--}300\text{ nm}$) and LPS-induced inflammation were used as models to investigate *in vitro* the cytoprotective effects of edible flowers wastes extract (EFE), snail extracted mucus (SEM) and nanoparticles in keratinocytes (NCTC 2544 cells).

Edible flowers wastes extract's polyphenols were used to boost antioxidant activity of snail mucus providing cytoprotective effects in previously described models. Cellular antioxidant response was evaluated by measuring GSH content, ROS and LOOH levels. Snail mucus and edible flowers wastes extract were tested alone and in combination. Additionally, in order to enhance antioxidant activity of snail mucus, an extract was obtained by dehydration and used to develop snail mucus-coated gold nanoparticles (SM-NP) which were tested for their potential anti-inflammatory and antioxidant properties, through qRT-PCR analysis.

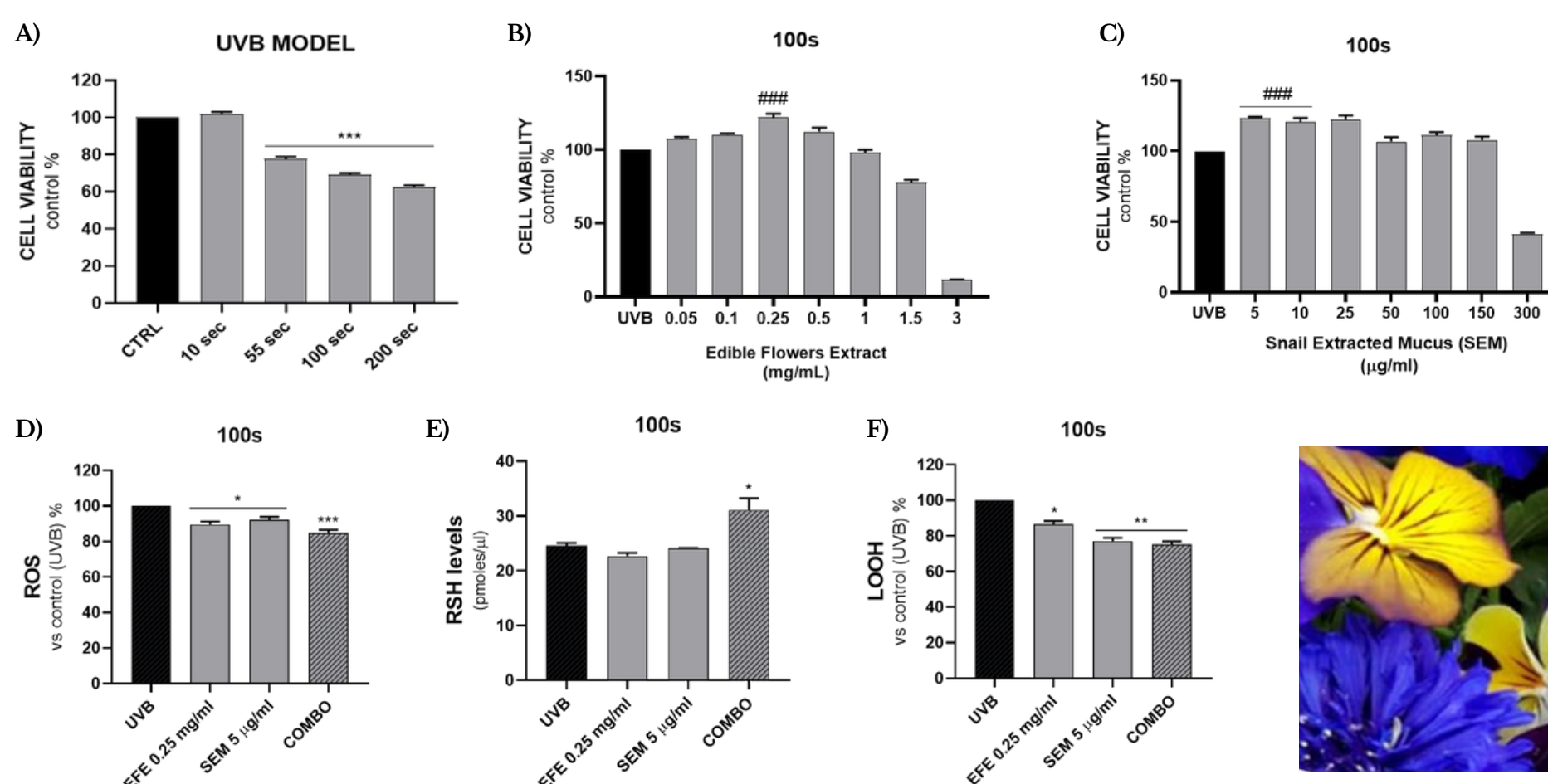
Results

Edible Flowers Wastes Extract, Snail Mucus and SM-NP characterization



The edible flower wastes extract (EFE) was standardized to contain $\geq 1.00\%$ TPC (total phenolic content) and $\geq 0.001\%$ TC (total carotenoid). Scavenger activity of EFE and SM-NP was evaluated in cell free model showing a significant antioxidant activity especially for EFE (A,D). On the other hand, SEM did not show any scavenger activity. Absorbance spectra of EFE, SEM and SM-NP displaying EFE maximum absorption at 276 nm, 410 nm, and 560 nm. The ability of SEM to absorb in the UV/Vis spectrum region, particularly starting below 400 nm, is considered interesting especially concerning cosmetic applications (B,C,E).

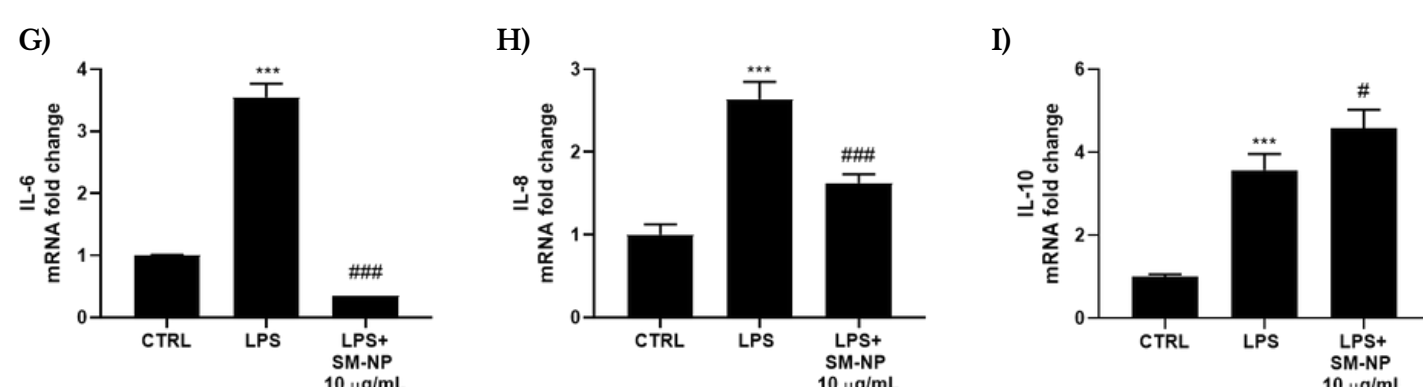
In vitro cytoprotective effect of EFE and SEM against UVB-damage



Preliminary cytotoxicity assays showed no significant effect of EFE and SEM at most concentrations tested, except for the highest (3 mg/ml and 300 µg/ml respectively). In order to induce UVB-damage, keratinocytes were exposed to UVB light at different time points and subsequently 100 sec was the time point chosen for further experiments (A). Cytoprotective effect was observed at 0.25 mg/ml for EFE and at 5 µg/ml for SEM (B,C). In order to assess antioxidant capacity of EFE and SEM alone and in combination ROS levels, GSH cellular content and lipid peroxidation (LOOH levels) were evaluated (D-F), highlighting a synergistic effect especially for ROS decrease and concomitantly GSH recovery. Cytotoxicity tests were also performed using different concentrations of SM-NP (1-250 µg/ml) showing no effect on cell viability. Ultimately, novel synthesized SM-NP were tested in NCTC 2544 challenged with LPS to evaluate their potential anti-inflammatory activity. Results showed SM-NP capacity to reduce IL-6 and IL-8 pro-inflammatory cytokines transcription (G, H) and increase anti-inflammatory IL-10 (I) even at low concentrations (10 µg/ml).



SM-NP anti-inflammatory activity on keratinocytes



Conclusions

In this work we demonstrated that flowers wastes can be considered valid candidates for cosmeceutical applications to enrich the snail mucus based anti-age products as they showed valid antioxidant properties. Moreover, we proved that snail mucus is suitable for creating innovative formulations as SM-NP which exhibited effective anti-inflammatory activity.