SITELF · SOCIETÀ CHIMICA ITALIANA

2° SITCOSM

CONFERENCE

Dall'Accademia all'Industria
per lo Sviluppo del Settore Cosmetico

BOOK OF ABSTRACTS



AURUM · Pescara, Italia

27 – 28 ottobre 2026

SITELF Società Italiana di Tecnologia e Legislazione Farmaceutiche

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Book of Abstracts

2° SITCOSM Conference

Dall'Accademia all'Industria per lo Sviluppo del Settore Cosmetico

Aurum, Pescara (Italia), 27–28 ottobre 2026

A cura del Comitato Organizzatore della 2° SITCOSM Conference
Promosso da SITELF – Società Italiana di Tecnologia e Legislazione Farmaceutiche e Società Chimica Italiana
Con l'Università "G. d'Annunzio" di Chieti-Pescara

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CHI SIAMO

Promotori di questa iniziativa sono ricercatori, ricercatrici e docenti universitari specializzati in cosmetologia, associati alla SITELF (Società Italiana di Tecnologia e Legislazione Farmaceutiche) e alla Società Chimica Italiana. A partire dallo scorso anno, ci siamo impegnati in modo sistematico in attività volte ad accrescere la visibilità scientifica del settore cosmetico nel panorama accademico e industriale italiano, perseguendo come obiettivo strategico il consolidamento di un dialogo strutturato tra i due mondi.

LA NOSTRA MISSIONE

Il settore cosmetico rappresenta un comparto industriale di rilievo, caratterizzato da una forte componente scientifica e tecnologica che merita un riconoscimento accademico adeguato. La nostra missione è quella di promuovere e consolidare un cross-talking autentico tra accademia e industria, favorendo lo scambio di conoscenze, metodologie e approcci innovativi. Crediamo che solo attraverso una sinergia concreta tra ricercatori e professionisti dell'industria sia possibile elevare la qualità della ricerca cosmetologica nazionale e rispondere in modo efficace alle sfide regolatorie, scientifiche e di mercato che il settore affronta.

L'EVENTO

La 2° SITCOSM Conference nasce da questa visione e rappresenta la conferma e il consolidamento di un appuntamento che intendiamo rendere annuale. Il convegno costituisce uno spazio di confronto scientifico aperto, in cui ricercatori accademici, professionisti dell'industria cosmetica e figure regolatorie possono dialogare, presentare i propri lavori di ricerca e gettare le basi per nuove collaborazioni. La scelta di Pescara come sede della 2ª edizione riflette l'impegno dell'Università "G. d'Annunzio" di Chieti-Pescara nell'essere parte attiva di questa rete di connessioni.

Accademia e industria cosmetica: costruire insieme il futuro scientifico del settore.

La 2° SITCOSM Conference è il luogo in cui questa visione prende forma: un momento di confronto scientifico libero, aperto e costruttivo, ripetuto ogni anno per costruire una comunità scientifica cosmetologica sempre più coesa e riconosciuta.

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SEDE

Aurum, Pescara

Martedì 27 ottobre 2026

08:30	Registrazione dei partecipanti
09:00–09:10	Saluti istituzionali **Prof.ssa Paola Minghetti** · *Presidente SITELF*
09:10–09:15	Introduzione al convegno **Prof.ssa Piera Di Martino** · *Presidente del gruppo SITCOSM*
09:15–09:45	**Plenary Lecture** — From Research to Business: the role of Technology Transfer. The Materias experience **Aniello Cammarano** · *Materias*
09:45–11:05	**Sessione 1 — Biologia cutanea ed efficacia** *Chair: A. Casiraghi (Unimi), C. Villa (Unige)*
09:45	O1 Future-Proofing the Skin: Transcriptomic Insights & Advanced Testing Models for Preventive Skincare **Vincenzo Nobile** · *Complife Italia*
10:05	O2 *Hyssopus officinalis* as a Modulator of Epidermal Lipid Metabolism for Skin Barrier Homeostasis **Lucia Ricci** · *Università di Napoli Federico II*
10:25	O3 Attività riparatrice e antinfiammatoria sinergica dell'associazione astaxantina–fenilcapsaicina su modello di epidermide umana ricostituita **Antonio Mazzucco** · *Università di Siena*
10:45	O4 Natural antioxidant creams against blue light induced skin damage: a new strategy for daily photoprotection **Margherita Accardo** · *Università della Basilicata*
11:05–11:25	**Coffee break**
11:25–12:25	**Sessione 2 — Eco-design, regolatorio e qualità formulativa** *Chair: M. Centini (Unisi), S. Tampucci (Unipi)*
11:25	O5 An Instrumental Approach to Eco-Design Reformulation of Cosmetic Products: Replacing Microplastics and High-Impact Ingredients **Giovanni Tafuro** · *Unired / Unifarco / Università di Padova*
11:45	O6 Formulation of Body Cleansers with Alternative Opacifying Agents to Replace Synthetic Polymer Microparticles **Lorella Giovannelli** · *Università del Piemonte Orientale / MIRATO*
12:05	O7 Stability analysis of cosmetic formulations using Turbiscan Lab Expert **Valentino Laquintana** · *Università di Bari "Aldo Moro"*
12:25–14:30	**Light lunch + Sessione Poster (P1–P13)**
14:30–15:50	**Sessione 3 — Sostenibilità e valorizzazione di ingredienti naturali** *Chair: R. Cortesi (Unife), L. Giovannelli (UPO)*
14:30	O8 From Saffron Flower Beauty to Skin Care: Upcycling Saffron Flower By-Products into Multifunctional Cosmetic Emulsions **Tiziana Esposito** · *Università di Salerno*
14:50	O9 Development of a Pickering O/W Emulsion Containing an EVO-Based Carotenoid Extract for Cosmetic Applications **Anna Imbriano** · *Università di Perugia*
15:10	O10 Development and Evaluation of NADES-Based Extracts Hydrogels **Carla Villa** · *Università di Genova*
15:30	O11 Identification of bioactive compounds in botanical ingredients of cosmetic interest: spilanthol in *Acmella* species and bisabolol in Chamomile varieties **Federica Colombo** · *Skylab Energia / UPO*
15:50–16:10	**Pausa**
16:10–17:10	**Tavola rotonda I — Efficacia, sostenibilità e regole: cosa chiede l'industria alla ricerca** *Moderatrice: P. Di Martino (UdA) · Panelisti: S. Cudrig (I&S Srl), E. Procino (Cosmetica Italia), V. Nobile (Complife), G. Tafuro (Unired)*
17:10	Chiusura della prima giornata
≈ 20:00	**Cena Sociale — Ristorante Da Bacone, Lungomare G. Matteotti 54, Pescara**

Mercoledì 28 ottobre 2026

08:30	Registrazione dei partecipanti
09:00–11:00	**Sessione 4 — Sistemi innovativi di veicolazione** *Chair: B. Ruozi (Unimore), M.L. Manca (Unica)*
09:00	O12 Waste to Beauty: integrated nanoformulation strategies for the sustainable valorization of agri-food side streams **Ines Castangia** · *Università di Cagliari*
09:20	O13 Aspasomal Systems for the Cutaneous Delivery of Depigmenting Oligopeptides **Sofía Gisella Brignone** · *Università di Pisa*
09:40	O14 Cutaneous delivery of rutin with supramolecular nanosystems **Rita Cortesi** · *Università di Ferrara*
10:00	O15 Development and characterization of clay-stabilized Pickering emulsions for dermo-cosmetic application obtained from natural and sustainable ingredients **Samuele Bonafè** · *Università di Perugia*
10:20	O16 Spray to film gellan gum-chitosan-sericin hydrogels for the topical delivery of plant-derived bioactives in cosmetic formulations **Maria Rosa Gigliobianco** · *Università "G. d'Annunzio" Chieti-Pescara*
10:40	O17 Fermented *Opuntia ficus-indica* cladode extract as a new functional ingredient in a topical cosmetic formulation **Camilla Elena Di Bella** · *Università "G. d'Annunzio" Chieti-Pescara*
11:00–11:20	**Coffee break**
11:20–12:25	**Tavola rotonda II — Dalla materia prima al prodotto finito: scalabilità e trasferimento industriale** *Moderatrice: L. Perioli (Unipg) · Panelisti: Pharmacosm Polli, ASSICC, T. Esposito (Unisa), F. Garavaglia (Skylab Energia), L. Giovannelli (UPO), V. Laquintana (Uniba)*
12:25–12:40	**Premiazione poster e conclusioni**
12:40	**Fine dei lavori**

Sessione 1 — Biologia cutanea ed efficacia

27 ottobre 2026, 09:45–11:05

Chair: A. Casiraghi (Unimi), C. Villa (Unige)

O1	Future-Proofing the Skin: Transcriptomic Insights & Advanced Testing Models for Preventive Skincare *Vincenzo Nobile · Complife Italia*	10
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O4	Natural antioxidant creams against blue light induced skin damage: a new strategy for daily photoprotection *Margherita Accardo · Università della Basilicata*	14

Future-Proofing the Skin: Transcriptomic Insights & Advanced Testing Models for Preventive Skincare

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Introduction

The cosmetic industry is shifting from correcting visible damage towards identifying and modulating the early molecular events that determine long-term skin resilience. Transcriptomics can reveal these clinically silent responses, but its application in volunteer studies is limited by the invasiveness and poor scalability of conventional skin biopsies. A repeatable, non-invasive sampling method capable of recovering both host- and microorganism-derived nucleic acids could connect mechanistic research with real-world product evaluation and enable integrated multi-omics studies [1].

Aim of the work

The aim of this work was to establish a translational platform combining *in vitro* stress transcriptomics with non-invasive in vivo molecular sampling. The primary objective was to identify a volunteer-friendly sampling approach suitable for longitudinal host transcriptomics and simultaneous microbiome and metatranscriptomic investigation, thereby supporting biomarker-based preventive, resilience and skin-longevity claims.

Materials and Methods

In vitro mechanism-discovery model: A skin-relevant model was exposed to ultraviolet (UV) stress and evaluated with or without cosmetic treatment. Global gene-expression and pathway analyses were used to map responses related to cellular stress, antioxidant defence, extracellular-matrix maintenance and inflammatory regulation.

Non-invasive sampling study: Swab, tape stripping, microneedle collection, oil-blotting film and microbiopsy were compared in a within-subject pilot study (n=3) using matched facial sites and randomized device order. RNA yield and integrity were evaluated by NanoDrop, Qubit and TapeStation, including RNA integrity number equivalent (RINe), DV200 and ribosomal RNA peak assessment.

Multi-omics analytical workflow: Long-read cDNA sequencing was used to preserve transcript-level information and to characterize human and microbial RNA recovered from the same sample [2]. Differential-expression and pathway analyses generated mechanism hypotheses and candidate biomarker panels, which were designed for orthogonal confirmation by droplet digital PCR (ddPCR) [3].

Results and discussion

UV exposure altered approximately 3,000 genes, whereas treatment-versus-UV comparison identified 71 treatment-responsive genes. Of these, 65% changed in the opposite direction to the UV-induced response, indicating partial molecular rescue and supporting pathways associated with stress normalization, antioxidant defence and extracellular-matrix preservation. Among the sampling approaches, tape stripping and oil blotting provided the strongest host-RNA quality, with RINe/DV200 values of 8.2/85.7% and 6.8/75.3%, respectively. Tape stripping generated a predominantly human profile (89.5% human and 10.5% microbial RNA), making it particularly suitable for epidermal host-transcriptomic endpoints. In contrast, oil blotting recovered a balanced molecular signal (47.1% human and 52.9% microbial RNA). This dual representation is strategically relevant because one non-invasive sample may support host transcriptomics, microbial community profiling and assessment of microbial transcriptional activity. Exploratory gene-panel analysis further indicated that oil blotting captured signals associated with the epidermis, sebaceous glands, hair follicles and sweat-gland-related biology, suggesting a broader biological window than sebum measurement alone. From an industrial perspective, the method is rapid, repeatable and compatible with longitudinal volunteer trials; from a scientific perspective, it enables host-microbiome interactions to be examined within a unified analytical framework.

Conclusion

The integration of controlled *in vitro* stress models with non-invasive, multi-omics-compatible skin sampling provides a practical bridge between mechanism discovery and human cosmetic evaluation. Oil blotting emerged as a promising enabling technology because it combines acceptable host-RNA integrity with balanced microbial recovery from a single skin-surface sample. Coupled with long-read transcriptomics, pathway analysis and ddPCR confirmation, this approach can translate otherwise silent preventive effects into measurable biomarkers and scientifically grounded claims related to protection, resilience and skin longevity. The pilot findings require confirmation in larger, product-specific studies, but they establish a scalable academy-industry framework for next-generation cosmetic substantiation.

References

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Hyssopus officinalis as a Modulator of Epidermal Lipid Metabolism for Skin Barrier Homeostasis

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Keywords: skin barrier homeostasis; plant cell culture; *Hyssopus officinalis*

Introduction

The skin barrier is a highly organized structure that preserves tissue hydration and homeostasis, primarily through the stratum corneum, where corneocytes are embedded in a lipid matrix composed mainly of ceramides, cholesterol, and fatty acids [1]. Impairment of this organization leads to increased transepidermal water loss (TEWL) and compromised skin integrity. Ceramides are key determinants of epidermal barrier function, and their biosynthesis is regulated by enzymes including serine palmitoyltransferase (SPT) and β-glucocerebrosidase (GBA) [2]. Accordingly, approaches aimed at supporting endogenous lipid metabolism are gaining interest in cosmetic research. In parallel, green biotechnology approaches such as plant cell culture offer sustainable and reproducible sources of bioactive specialized metabolites [3,4].

Aim of the work

This study aimed to investigate *Hyssopus officinalis* (*Lamiaceae*) cell cultures as a sustainable source of bioactive compounds supporting skin barrier homeostasis.

Materials and Methods

A hydroalcoholic extract obtained from *Hyssopus officinalis* cell cultures was chemically characterized by LC-HESI-HRMS to define its metabolite profile. Biological activity was investigated through an integrated workflow combining *in vitro* keratinocyte models, *ex vivo* human skin explants, and *in vivo* clinical study. Skin barrier function was assessed by evaluating endogenous ceramide-related pathways, the expression of key structural proteins, and transepidermal water loss (TEWL) in mechanically stressed skin.

Results and discussion

H. officinalis cell culture extract significantly reduced intracellular ROS levels (−20%, $p < 0.01$). HoEx increased β-glucocerebrosidase (GBA) activity *in vitro* and *ex vivo* (Fig. 1), upregulated GBA, SPT, INV, and FLG gene expression (Fig. 2), and increased epidermal ceramide immunostaining (+36%, $p < 0.01$; Fig. 3). In volunteers with mechanically stressed skin, GBA activity increased by 30% versus baseline and was significantly higher than with placebo ($p < 0.05$; Fig. 4a). Moreover, TEWL decreased by 15% after one week ($p < 0.05$ versus baseline; $p < 0.001$ versus placebo; Fig. 4b).

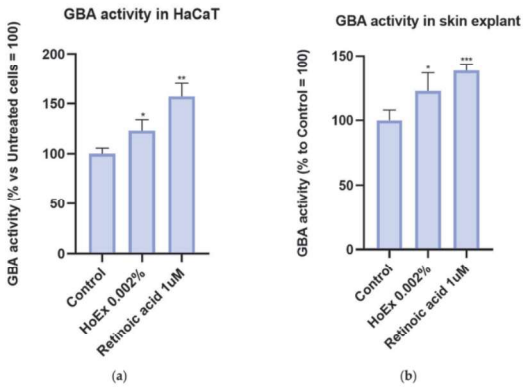


*Fig. 1. GBA activity in HaCaT cells (a) and skin explants (b). Mean ± SD (n = 3 independent experiments). * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$ versus control.*

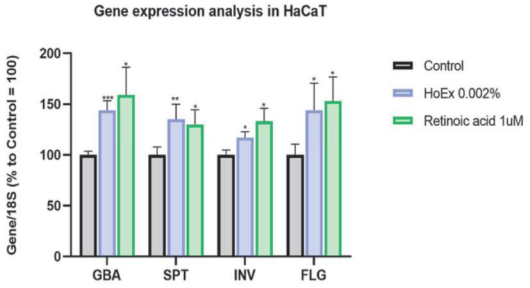


*Fig. 2. GBA, SPT, INV and FLG gene expression in HaCaT cells. Mean ± SD (n = 3 independent experiments). * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$ versus control.*

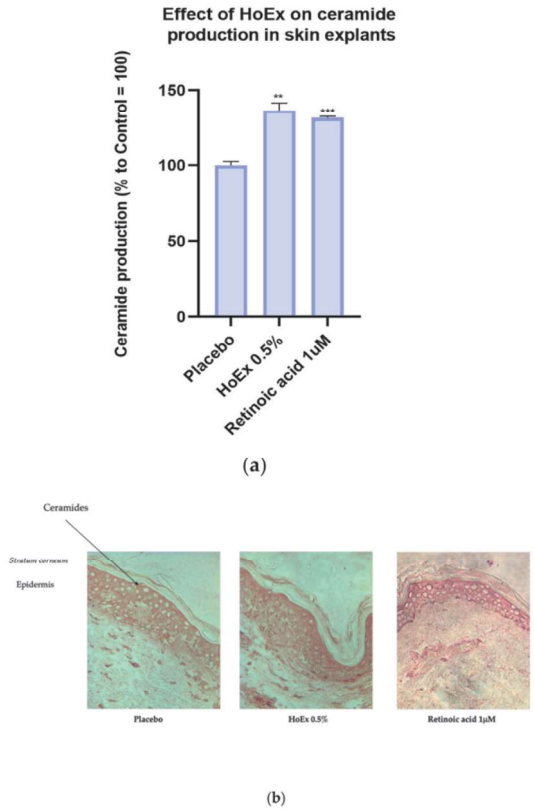


*Fig. 3. Epidermal ceramide immunostaining after 72 h: quantification (a) and representative images of skin explants (b). Placebo = 100%. Mean ± SD (n = 3 independent experiments). ** p < 0.01; *** p < 0.001 versus placebo.*

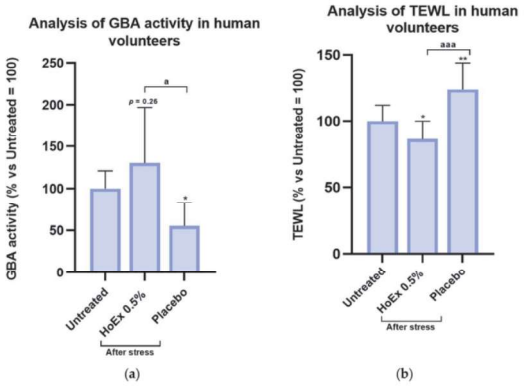


*Fig. 4. Clinical GBA activity (a) and TEWL (b) after one week (n = 10). Pre-stress baseline ("Untreated") = 100%. Mean ± SD. * p < 0.05; ** p < 0.01 versus baseline; a p < 0.05 and aaa p < 0.001 versus placebo.*

Conclusion

Overall, these findings indicate that *H. officinalis* cell culture extract supports skin barrier homeostasis by modulating key pathways involved in epidermal lipid metabolism and differentiation. The modulation of GBA and SPT-related pathways, together with increased epidermal ceramide levels and reduced TEWL following mechanical stress, supports a multifaceted activity contributing to the maintenance of epidermal barrier integrity.

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Attività riparatrice e antinfiammatoria sinergica dell'associazione astaxantina–fenilcapsaicina su modello di epidermide umana ricostituita

Antonio Mazzucco
UNISI – Dipartimento di Farmacia

Introduzione. Gli stimoli irritanti danneggiano i cheratinociti: ne riducono la vitalità e inducono il rilascio di mediatori pro-infiammatori come l'IL-1α, uno dei principali motori dell'inflammaging cutaneo. Astaxantina e capsaicinoidi hanno note proprietà antiossidanti e antinfiammatorie, ma il loro effetto combinato per via topica non è stato ancora caratterizzato.

Scopo. Valutare l'attività riparatrice e antinfiammatoria di astaxantina (AX), capsaicina (CAP) e del suo analogo sintetico fenilcapsaicina (PhCAP), da sole e in associazione binaria, su epidermide umana ricostituita (RhE) danneggiata.

Metodi. I tessuti EpiDerm™ (EPI-200, MatTek) sono stati danneggiati con Triton X-100 all'1% per 1 h e poi trattati per 18 h (37 °C, 5% CO_2) con gli ingredienti commerciali disciolti in olio d'oliva: AX (Astaplancton® HA), CAP (estratto di *Capsicum*, ≥2% capsaicina) e PhCAP (aXivite®). Ogni ingrediente è stato testato da solo all'1% e al 2% p/p, e in associazione allo 0,5+0,5% e all'1+1% p/p. La vitalità cellulare è stata misurata con saggio MTT (OD_{570}), il rilascio di IL-1α nel terreno con ELISA. I risultati sono stati normalizzati sul veicolo (olio d'oliva). La sinergia è stata definita come scarto percentuale tra effetto osservato ed effetto atteso, quest'ultimo calcolato come media dei singoli ingredienti alla stessa dose totale (2%). Lo studio è stato condotto in un centro di saggio certificato BPL.

Risultati. Il Triton X-100 ha ridotto la vitalità al 45,3% rispetto ai tessuti non trattati. Tutti i trattamenti hanno aumentato la vitalità rispetto al veicolo. AX 1% + PhCAP 1% ha dato il recupero più alto: +38,1% contro un atteso di +22,3% (sinergia +70,7%); AX 1% + CAP 1% si è fermata a +30,7% (sinergia +13,2%). La stessa associazione AX + PhCAP ha dato la riduzione più forte di IL-1α: −30,7% rispetto al veicolo (87,0 vs 125,6 pg/mL), mentre i singoli ingredienti al 2% hanno ridotto l'IL-1α solo del 2,6–9,3%. Allo 0,5+0,5% le associazioni hanno dato circa +20% di vitalità con sinergia marginale, a indicare che l'effetto dipende dalla concentrazione.

Conclusioni. All'1% ciascuna, astaxantina e fenilcapsaicina in associazione mostrano un'attività riparatrice e antinfiammatoria sinergica *in vitro*, superiore all'associazione astaxantina–capsaicina. Questo supporta il loro impiego in prodotti topici contro il danno da irritanti e l'inflammaging. Servono conferme con più repliche, altri stimoli (es. UV) e studi clinici.

Parole chiave: astaxantina; fenilcapsaicina; capsaicina; epidermide umana ricostituita; IL-1α; sinergia; inflammaging

Natural antioxidant creams against blue light induced skin damage: a new strategy for daily photoprotection

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Introduction

Growing evidence indicates that blue light (380-500 nm), emitted from both sunlight and screens plays a significant role in skin photoaging. Blue light exposure triggers oxidative stress and ramps up reactive oxygen species (ROS) production in skin cells, which accelerates visible aging [1].

As a result, researchers and cosmetic companies have increasingly turned their attention to natural bioactive compounds derived from plants and botanical sources as promising functional ingredients for skincare formulations.

Aim of the work

In this context, hydroxycitrate (HCA), a bioactive compound derived from *Garcinia cambogia*, and red wine powder (RWP) obtained from Aglianico del Vulture (*Vitis vinifera* L.), both known for their interesting antioxidant and anti-inflammatory properties, represent promising natural ingredients for the development of topical formulations aimed at protecting the skin from blue light-induced oxidative damage [2].

Accordingly, the aim of this study was to formulate and characterize two oil-in-water cosmetic creams, enriched with HCA or RWP, and to evaluate their physicochemical stability, cytocompatibility, and protective efficacy against blue light-induced oxidative stress in human keratinocytes.

Materials and Methods

Materials: HCA and RWP were incorporated into oil-in-water emulsion-based topical formulations, which included skin-compatible emollients, humectants and functional cosmetic excipients [2].

Preparation and in vitro evaluation: The creams were formulated as oil-in-water emulsions and characterized through physicochemical and microbiological stability tests, including pH, viscosity, centrifugation resistance, thermal stress, and microbial quality under accelerated storage conditions [3]. Their biological performance was then evaluated *in vitro* using HaCaT human keratinocytes to assess cytocompatibility and antioxidant activity by measuring cell viability and intracellular ROS production following blue light exposure in the presence or absence of the HCA- and RWP-enriched formulations [4].

Results and discussion

Both formulations showed had good physicochemical and microbiological stability throughout accelerated storage conditions while maintaining appropriate rheological properties and skin-compatible pH (Fig. 1).



Fig. 1. Evaluation of the cream's texture and its sensory properties on the skin.

Neither formulation affected the basal viability of HaCaT keratinocytes, demonstrating good cytocompatibility. Following blue light exposure, intracellular ROS levels significantly increased, whereas pretreatment with both formulations effectively attenuated oxidative stress. Among the tested formulations, showed the greatest antioxidant efficacy, reducing ROS levels almost to those observed in untreated cells. Interestingly, the formulation with Aglianico del Vulture red wine extract showed a greater antioxidant potential than that based on hydroxycitrate. It was shown that hydroxycitrate had protection at its highest concentration. These findings suggest that both plant-derived bioactives are suitable candidates for incorporation into multifunctional cosmetic formulations designed to counteract blue light-induced oxidative damage.

Conclusion

Creams containing natural antioxidants from hydroxycitrate and Aglianico del Vulture red wine extract showed good stability, cytocompatibility, and ability to protect from oxidative stress induced by blue light exposure. These findings support the potential application of HCA and RWP enriched formulations as multifunctional cosmetic products for daily protection against blue light-induced skin oxidative stress and premature photoaging.

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Sessione 2 — Eco-design, regolatorio e qualità formulativa

27 ottobre 2026, 11:25–12:25

Chair: M. Centini (Unisi), S. Tampucci (Unipi)

O5	An Instrumental Approach to Eco-Design Reformulation of Cosmetic Products: Replacing Microplastics and High-Impact Ingredients *Giovanni Tafuro · Unired / Unifarco / Università di Padova*	17
O6	Formulation of Body Cleansers with Alternative Opacifying Agents to Replace Synthetic Polymer Microparticles *Lorella Giovannelli · Università del Piemonte Orientale / MIRATO*	19
O7	Stability analysis of cosmetic formulations using Turbiscan Lab Expert *Valentino Laquintana · Università di Bari "Aldo Moro"*	21

An Instrumental Approach to Eco-Design Reformulation of Cosmetic Products: Replacing Microplastics and High-Impact Ingredients

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Introduction

The transition towards sustainability and eco-design in cosmetic formulation is increasingly driven by both environmental concerns and regulatory requirements. In particular, the adoption of Commission Regulation (EU) 2023/2055, restricting intentionally added synthetic polymer microparticles, i.e. microplastics, is accelerating the replacement of conventional and effective ingredients such as nylon-12 and polymethyl methacrylate (PMMA).[1] For leave-on cosmetic products, the placing on the market of products containing these ingredients will be progressively restricted according to the transitional periods established by the Regulation. At the same time, formulators are encouraged to reduce the environmental footprint of cosmetic products by replacing other high-impact ingredients, including acrylic rheology modifiers and silicone-based emollients, with more sustainable alternatives.[2]

Aim of the work

This study presents an instrumental approach to the eco-design reformulation of cosmetic products by identifying natural and biodegradable alternatives to synthetic texturizers classified as microplastics, followed by the replacement of acrylic rheological modifiers and silicone emollients while preserving the stability, texture and sensory properties of the original formulations.

Materials and Methods

Materials: Three oil-in-water skin care emulsions on the market have been reformulated with an eco-design perspective: F1 is an acne-prone skin serum, F2 is an eye-contour gel-cream and F3 is a body butter. Synthetic texturizers X (Nylon-12), Y (PMMA) have been replaced with natural alternatives: Z (Zea mays starch), B (Distarch phosphate), M (Maltodextrin), D (Hordeum Vulgare Seed Flour), E (Cellulose), F (Calcinated kaolin) S (Silica) H (Mica) and I (Talc).
The acrylic rheological modifiers have been replaced with associations of polysaccharides: t (sclerotium gum), v (xanthan gum), k (diutan gum), r (succinoglycan), and g (carrageenan). The silicon oils and polymers have been replaced with plant-based emollients CU (Undecane, Tridecane) and DSG (Ethyl olivate, triheptanoin) and a biodegradable elastomer P (Dilinoleic Acid/Butanediol Copolymer, Castor Oil/IPDI Copolymer).
Rheological tests: performed both in continuous and oscillatory flow condition using a rotational rheometer, to characterize the samples' flow behavior and viscoelastic properties. i.e elastic G' and viscous g2 moduli.[3]
Texture analysis: the texture parameters, such as firmness, consistency, adhesiveness, and stringiness, were measured through an immersion/de-immersion test conducted with a Texture Analyzer.[4]
Sensory test: a double-blind sensory test was performed using a paired samples t-test to evaluate the products' applicative and sensorial properties.

Results and discussion

Instrumental characterization enabled the identification of natural texturizers exhibiting physico-mechanical properties comparable to nylon-12 and PMMA. Starch (Z) and maltodextrin-based materials (M) proved particularly suitable for fluid emulsions, whereas silica (S) showed superior performance in richer formulations. The selected alternatives were successfully incorporated into finished products, maintaining stability and sensory acceptance. Polysaccharide-based systems with sclerotium gum (t), xanthan gum (v) and diutan gum (k), conferred high spreadability and viscoelastic properties to products F1 and F2, although slight differences in structural strength (G*, tanδ) and adhesiveness were observed (Figure 1). For the F3 the association of carrageenan (g) and sclerotium gum (t) allowed preserving the rich and elastic texture, however resulting in an increase in stringiness and tanδ compared to the reference product. The replacement of silicone ingredients with natural emollients further contributed to improving the environmental profile of the formulations while maintaining desirable application properties.

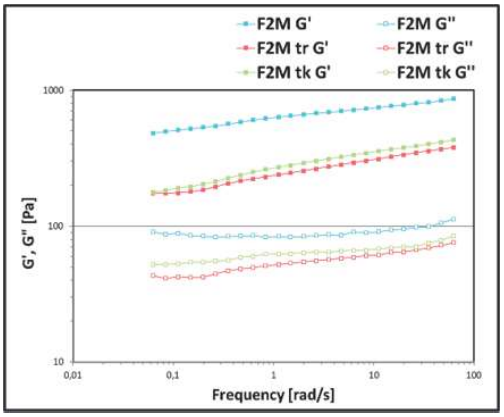


Fig. 1. Trend of elastic G' and viscous G'' moduli in function of the oscillation frequency for product F2 in which the synthetic texturizers and the acrylic polymer were replaced with combinations of polysaccharides.

Conclusion

This work demonstrates that a systematic instrumental approach combining rheology, texture analysis provides an effective strategy for eco-design reformulation. The methodology enables the replacement of microplastic texturizers, acrylic polymers and silicone ingredients with natural and biodegradable alternatives, supporting compliance with current European legislation and sustainability goals while preserving the performance and consumer acceptability of cosmetic products.

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Formulation of Body Cleansers with Alternative Opacifying Agents to Replace Synthetic Polymer Microparticles

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Introduction

Regulation (EU) 2023/2055 amended Annex XVII of the REACH Regulation by introducing restrictions on the placing on the market of synthetic polymer microparticles, on their own or intentionally added to mixtures, including those added to cosmetic products after the applicable transitional periods [1,2]. Following the applicable transitional period, the restriction will apply to rinse-off cosmetic products from 17 October 2027. Cosmetic manufacturers must therefore reformulate products containing synthetic polymer-based ingredients that meet the regulatory definition of synthetic polymer microparticles.

In body cleansers, synthetic polymer-based opacifiers are primarily used to produce a uniform opaque appearance, often perceived by consumers as richer and creamier. Their replacement is technologically challenging because alternative ingredients must provide the desired optical properties while maintaining the physicochemical stability, viscosity, skin-compatible pH, performance, and economic sustainability of the formulation. The identification and evaluation of suitable alternatives are therefore particularly relevant for high-volume rinse-off cosmetic products, such as shampoos and bath or shower cleansers [3].

Aim of the work

This study aimed to formulate and evaluate body cleansers containing alternative opacifying or pearling agents as substitutes for a conventional synthetic opacifier (SO), a styrene/acrylates copolymer-based opacifier.

Materials and Methods

The experimental research was conducted at the laboratories of MilMil 76 SpA, which is part of the MIRATO SpA Group.

Materials: Surfactants, fragrances, preservatives, humectants, functional substances, pH modifiers, rheology modifiers, opacifying and pearling agents.

Preparation of body cleansers:
Different kinds of surfactants were used to formulate body cleansers. As the primary surfactant, sodium laureth sulfate was selected, as it is economical, foaming, and has high cleansing capacity. Its irritant potential was reduced through a secondary amphoteric surfactant, such as cocamidopropyl betaine, and mild, plant-based, non-ionic surfactants, including lauryl-glucoside and coco-glucoside glyceryl oleate. These surfactants were combined with emollients like glyceryl oleate to improve product dermo-compatibility.
Initially, body cleansers were formulated with SO or alternative opacifying or pearling agents. Three brands from the company's portfolio were evaluated, each with increasing concentrations of active surfactant matter (ASM), defined as the total content of ionic, non-ionic, and amphoteric surfactants (Fig. 1), and varying percentages of SO:
• ENTRY (6.5% ASM): 0.3%, 0.6%, and 0.9% SO
• MEDIUM (9% ASM): 0.4%, 0.8%, and 1.2% SO
• PREMIUM (12% ASM): 1.3%, 2.0%, and 2.5% SO.
Forty formulations were developed using different pearling agents (PB, PP, PK, each with cocamidopropyl betaine) and opacifier blends (O2, O3, O4, each with glycol distearate) as alternatives to SO. Their performance was compared with products without an opacifier or pearling agent, as well as with those containing SO.

Characterization of body cleansers: the organoleptic properties (appearance, color) and physicochemical parameters (pH and viscosity) of the cleansers were evaluated immediately after their preparation, during stability studies at room temperature under both light and dark conditions, and during accelerated stability tests in a climatic chamber at 40 °C for 6 months as well as after a thermal cycling test at 40 °C and 4 °C.
UV spectrophotometry was used to assess the opacity trends conferred by opacifying/pearling agents in the most stable surfactant formulations. Calibration curves for each opacifying/pearling agent were obtained, but the assessment was only semi-quantitative because the composition of the alternative opacifiers was complex and not fully known.

Results and Discussion

Cleansers without opacifiers were clear and transparent (Fig. 1a), whereas those with opacifiers or pearling agents ranged from translucent to opaque.
All formulations had pH values compatible with skin and viscosities typical of rinse-off products, consistent with company specifications for brands (Fig. 1b). Formulations with SO, PB, PP, and PK remained stable for at least six months and during accelerated stability testing, unlike those with O2, O3, and O4, which showed physical instability. The opacifying effect increased proportionally with the amount of pearling agent used, and UV spectrophotometry confirmed this

trend (Fig. 2). The correlation between absorbance and the concentration of SO and pearling agents was strong for SO and PP, good for PK, and moderate for PB.

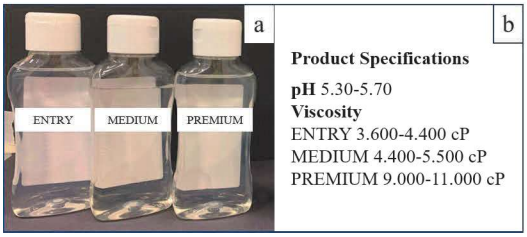


Figure 1. Cleansers without opacifier (a) and product specifications of the three company brands (b).

The opacifying effect increased proportionally with the amount of pearling agent used, and UV spectrophotometry confirmed this trend (Fig. 2). The correlation between absorbance and the concentration of SO and pearling agents was strong for SO and PP, good for PK, and moderate for PB.

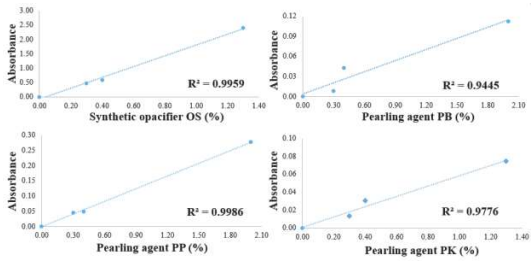


Figure 2. Opacity trends of cleansers containing SO, PB, PP, and PK measured at 260 nm.

Alternative pearling agents were less effective than SO. Remarkable opacifying effects occurred only at higher concentrations of these agents (Fig. 3).

Conclusion

The results demonstrate the technical feasibility of replacing the conventional styrene/acrylates copolymer-based opacifier with alternative pearling agents in body cleanser formulations.

Among the tested alternatives, PB, PP, and PK showed the most promising performance, maintaining formulation stability under the investigated storage conditions. However, higher concentrations were required to achieve an opacifying effect comparable to that of the conventional ingredient, resulting in increased formulation costs. Overall, these findings support the potential applicability of PB, PP, and PK in the reformulation of high-volume rinse-off cosmetic products. Nevertheless, further optimization is necessary to improve their opacifying efficiency and economic viability.

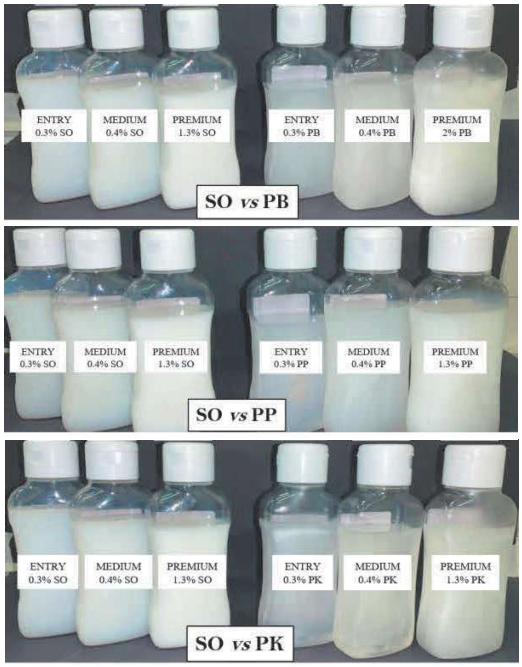


Figure 3. Body cleansers formulated with synthetic opacifier (SO, left) and with pearling agents (PB, PP, or PK, right).

References

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Stability analysis of cosmetic formulations using Turbiscan Lab Expert.

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Introduction

For cosmetic and pharmaceutical companies, assessing the stability of formulations is one of the most important issues, ensuring the production of safe products that meet stringent quality requirements. Cosmetic formulations are often characterised by complex systems based on dispersed materials, such as creams, emulsions and colloidal dispersions. To meet the growing demand for the rapid development of these types of formulations, it is desirable to be able to analyse dispersion over time with minimal modifications to the test setup. The Turbiscan optical analyser (Formulaction, L'Union, France) is used for this purpose. This instrument uses multiple light scattering technology with a near-infrared light source to examine a sample vertically (from bottom to surface) via repeated scans over time. The presence of two fixed detectors enables transmission and backscatter signals to be acquired as a function of position (height) and time. These signals depend on particle (or globule) size and concentration, in accordance with Mie's theory. The result is a series of intensity profiles that monitor the evolution of the sample's physical characteristics over time. This provides an early indication of instability phenomena, such as globule and/or particle migration (sedimentation and creaming), and variations in particle size (coalescence and flocculation). This is especially useful in the initial phase, when these phenomena may not be detectable using other techniques or by the naked eye [1,2].

Aim of the work

This study aims to examine the long-term stability of various cosmetic formulations by presenting case studies that evaluate the optical transmission and photon backscatter profiles of different samples. The products studied included creams, cleansers, chemical sunscreen-based sunscreens, mouthwashes and dry shampoos.

Methods

The different cosmetic formulations were placed in cylindrical glass tubes and subjected to a stability analysis using a TurbiscanLab® Expert. This involved using a pulsed near-infrared LED with a wavelength of 880 nm to acquire spectra as changes in transmission (ΔT) and backscatter profiles (ΔBS). Furthermore, the cosmetic formulations underwent accelerated physical stability studies in a climate chamber for one month at a temperature of 40°C and a relative humidity of 60%, as well as via centrifugation at 3800 rpm for 30-minute cycles at 25°C.

Results and discussion

The stability of the different cosmetic formulations was assessed using Turbiscan technology, which measures transmission and backscattering profiles over a period of up to 24 hours. The Turbiscan Stability Index (TSI) was also calculated. The TSI can range from 0 upwards; however, stable systems exhibit an overall TSI between 0 and 2, representing the change in TSI over time across different scans.

The formulations were selected and monitored using traditional techniques, such as accelerated ageing tests in a climate chamber or centrifuge, complemented by Turbiscan analysis. The products prepared included creams, gentle facial cleansers, mouthwashes, rinse-off products such as dry shampoos based on plant extracts or essential oils, and medium- to high-SPF sun protection creams containing chemical filters. For instance, a standardised protocol was adopted for the preparation of the mouthwashes, which involved dispersing powders into an aqueous or hydroalcoholic medium via magnetic stirring for 30 minutes at room temperature (20–25 °C). The citrate buffer (0.1 M) was formulated to achieve a pH close to 7 (pH 6.6 ± 0.5), in accordance with the procedure outlined in FU XII.

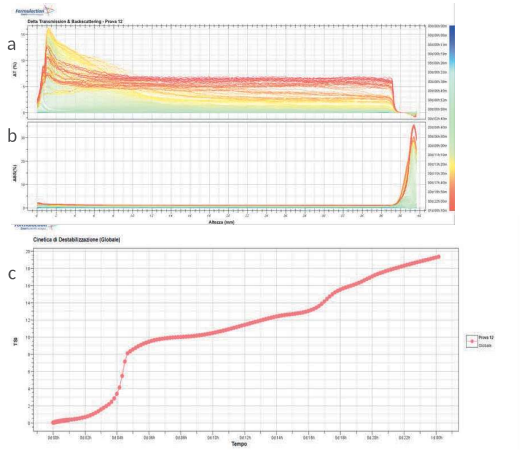


Fig. 1. Examples of Turbiscan profiles showing changes in the (a)transmission, (b) backscattering and (c) TSI over 24 hours.

Figure 1 shows a mouthwash formulation undergoing flocculation and the formation of a surface layer

(creaming). The TSI value remained below 4.0 for the first 4 hours, but then rose rapidly to exceed 20.0 within 24 hours. The ΔT and ΔBS values indicate that flocculation occurs concurrently with creaming. Overall, the formulation is considered unstable over the 24-hour period.

In contrast, Figure 2 displays the global TSI diagram (with a value below 2) and the transmittance and backscattering profiles of a stable cream. Analysis shows that the TSI remains below 1.0 for the first 2 hours, rising to a value of 1.6 over the subsequent 16 hours. The curve's profile indicates very slight sedimentation and flocculation phenomena occurring over time, reflecting a state of mild instability. This is confirmed by the transmittance (ΔT) and backscattering (ΔBS) variation curves. Overall, the formulation can be considered stable over the 18-hour period.

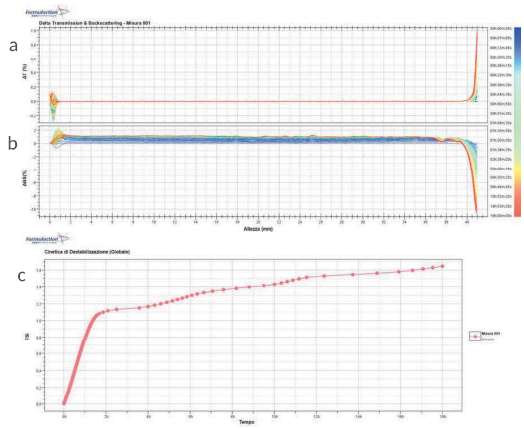


Fig. 2. Examples of Turbiscan profiles showing changes in the (a)transmission, (b) backscattering and (c) TSI over 18 hours.

Conclusion

When time is critical, a rapid screening approach, such as that offered by the Turbiscan analyser, is essential for selecting cosmetic formulation candidates. With the aim of rapidly identifying stable formulations, we evaluated the applicability of this analysis on cosmetic formulations comprising systems that differ significantly in terms of components, preparation methods and physicochemical characteristics. These included preparations based on plant-derived products. Once the samples have been selected, they can be monitored for as long as necessary.

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Sessione 3 — Sostenibilità e valorizzazione di ingredienti naturali

27 ottobre 2026, 14:30–15:50

Chair: R. Cortesi (Unife), L. Giovannelli (UPO)

O8	From Saffron Flower Beauty to Skin Care: Upcycling Saffron Flower By-Products into Multifunctional Cosmetic Emulsions *Tiziana Esposito · Università di Salerno*	21
O9	Development of a Pickering O/W Emulsion Containing an EVO-Based Carotenoid Extract for Cosmetic Applications *Anna Imbriano · Università di Perugia*	29
O10	Development and Evaluation of NADES-Based Extracts Hydrogels *Carla Villa · Università di Genova*	29
O11	Identification of bioactive compounds in botanical ingredients of cosmetic interest: spilanthol in *Acmella* species and bisabolol in Chamomile varieties *Federica Colombo · Skylab Energia / UPO*	30

From Saffron Flower Beauty to Skin Care: Upcycling Saffron Flower By-Products into Multifunctional Cosmetic Emulsions

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Introduction

The growing demand for effective, innovative, and sustainable cosmetics is encouraging cosmetic companies to identify new competitive ingredients that combine biological activity, technological performance, and a strong sustainability profile. In this context, valorising agro-food by-products is an attractive strategy for obtaining high-value cosmetic ingredients while reducing the environmental impact associated with biomass disposal. Saffron production generates large amounts of floral residues after the manual separation of the stigmas. Although saffron flowers are visually appealing and rich in valuable phytochemicals, they are generally discarded as waste at the end of the harvesting process. These by-products contain phenolic and flavonoid compounds with antioxidant and potential photoprotective properties, making them promising candidates for developing multifunctional cosmetic products. However, incorporating them into cosmetic formulations requires delivery systems that preserve both physicochemical stability and biological activity [1,2]. Oil-in-water (O/W) emulsions represent attractive topical carriers because of their skin compatibility, favorable sensory properties and ability to incorporate compounds with different polarities.

Aim of the work

This study aimed to develop and characterize O/W emulsions containing a hydroalcoholic extract from saffron flower by-products (*Crocus sativus* L.; ZAF) and to evaluate its potential as a multifunctional cosmetic ingredient. In particular, the study examined how ZAF incorporation affected the emulsion's technological and rheological properties and assessed the formulation's ability to preserve the extract's chemical stability and bioactivity under accelerated storage conditions (Figure 1).

Materials and Methods

Materials: ZAF was obtained from dried saffron flowers (*Crocus sativus* L.) collected in the Navelli Plateau, Abruzzo, Italy, an area traditionally associated with the production of Zafferano dell'Aquila PDO. The flowers were subjected to ultrasound-assisted extraction using 70% aqueous ethanol. RP-HPLC-DAD was used to investigate the extract's phytochemical profile, while antioxidant activity was evaluated using DPPH and ABTS assays and total phenolic content determination.
An O/W cosmetic emulsion containing 1% w/w ZAF (Z) was prepared and compared with the corresponding blank formulation (B). The systems were evaluated for appearance, pH, centrifugation stability, rheological and viscoelastic behavior, droplet-size distribution and zeta potential at the initial time point, after storage at 25 °C and following thermal-shock cycles. Chemical and functional stability were further investigated under accelerated conditions at 40 °C/75% RH by monitoring Kaempferol-3-O-sophoroside (K3OS) content via HPLC-DAD and assessing DPPH radical-scavenging activity.

Results and discussion

The extraction yield was 52.5% w/w. Kaempferol-3-O-sophoroside (K3OS) was the main quantified metabolite (9.50 ± 0.10% w/w), while ZAF showed a high total phenolic content (230.28 ± 7.81 mg K3OS equivalents/g) and marked antioxidant activity (DPPH EC_{50}: 101.44 ± 1.54 µg/mL). The incorporation of 1% w/w ZAF into the O/W emulsion (Z) produced a homogeneous and acceptable formulation compared with the blank emulsion (B), without adversely affecting pH, centrifugation stability or droplet-size distribution. Both B and Z showed shear-thinning and thixotropic behavior. Oscillatory tests revealed a predominantly elastic response, with G′ higher than G″ within the linear viscoelastic region, while thermal rheological loops showed near-complete recovery of the initial structure after heating and cooling cycles. These findings confirmed that ZAF did not compromise the emulsion's mechanical and physical stability. The accelerated stability studies showed that the emulsion matrix effectively protected the extract, with K3OS retention above 95% after three months at 40 °C/75% RH, demonstrating the extract's stability and reliability for cosmetic use.

Conclusion

This study demonstrates that saffron flower by-products can be effectively upcycled into a functional cosmetic ingredient with antioxidant properties. Their incorporation into O/W emulsions resulted in stable formulations with suitable technological and rheological properties, while also improving the preservation of the extract's main phytochemical

marker and antioxidant activity under accelerated storage conditions. These findings underline the potential of saffron floral residues as high-value ingredients for sustainable cosmetic innovation and support their further use in developing multifunctional skin-care formulations.



Fig. 1. Figure 1: From dried *Crocus sativus* L. petals to a hydroalcoholic extract (ZAF) incorporated into O/W emulsions with antioxidant and UV-filter-boosting potential (ZF).

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Development of a Pickering O/W Emulsion Containing an EVO-Based Carotenoid Extract for Cosmetic Applications

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Introduction

The growing demand for sustainable and naturally derived cosmetic products has stimulated the development of innovative formulations able to improve the stability and efficacy of bioactive compounds. Carotenoids exhibit antioxidant, anti-aging and photoprotective properties, but their application in topical formulations is limited due to their susceptibility to oxidation and photodegradation [1]. Pickering emulsions, stabilized by solid particles instead of conventional surfactants, represent a promising strategy for improving the stability and safety profile of cosmetic formulations containing carotenoids [2].

Aim of the work

The aim of this study was to develop and optimize an oil-in-water (O/W) Pickering emulsion containing a carotenoid-enriched extra virgin olive oil extract (EVOO_CE) obtained from pumpkin (*Cucurbita maxima L.*) peel and to evaluate its suitability for cosmetic applications.

Materials and Methods

Materials. 2,2′-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid) diammonium salt (ABTS), (±) -6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (Trolox), Nanoclay hydrophilic bentonite were obtained from Sigma-Aldrich (Milano, Italy). Isopropanol (IPA), water (H_2O), methanol (MeOH), and methyl tert-butyl ether (MTBE) of HPLC grade were acquired from Carlo Erba Reagents (Milano, Italy). Extra Virgin Olive Oil (Bertolli, Florence, Italy), named EVOO, was purchased from a local retail supermarket (Perugia, Italy); hyaluronic acid sodium salt (1500-1800 kDA) was purchased from (A.C.E.F, s.p.a, Piacenza, Italy). β-carotene (>97.0%) standard was purchased from TCI (Tokyo Chemical Industry) chemicals (Toshima, Tokyo, Japan).
The other solvents (hexane, ethanol, acetone and petroleum ether) were purchased from VWR (Milano, Italy). Ultrapure water was generated by Synergy® UV Water Purification System (Millipore Sigma, St. Louis, MI, USA).
Extraction and Characterization. Pumpkin peel from *Cucurbita maxima L.* (Hokkaido variety) was dried, milled, and used for carotenoid extraction. A carotenoid-enriched extra virgin olive oil (EVOO_CE) was prepared through green extraction approaches based on dynamic maceration (40°C, 1 h) [3]. Extracts were characterized by spectrophotometric determination of total carotenoid content (TCC) and antioxidant activity (ABTS assay), while carotenoid profiling was performed by HPLC-DAD, also supported by HPLC-MS data.
Preparation of Pickering Emulsion. An oil-in-water (O/W, 10:90 v/v) Pickering emulsion containing EVOO_CE as the dispersed phase was prepared using hydrophilic bentonite as a particle stabilizer and ultrasonication as the emulsification technique (Figure 1). The emulsion was subsequently converted into an emulgel by incorporating sodium hyaluronate (0.5%, w/w).

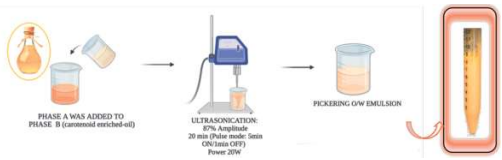


Fig. 1. HPU Emulsification Process

Pickering emulsion characterization. The formulation was characterized by dynamic light scattering (DLS) for droplet size distribution and ζ-potential. Carotenoid loading was determined spectrophotometrically. The emulgel was further investigated for spreadability, storage stability and photostability under ICH-compliant conditions. Biocompatibility and antioxidant activity were evaluated on HaCaT keratinocytes using MTT viability and intracellular ROS assays.

Results and discussion

The green extraction approach based on dynamic maceration in EVOO yielded a carotenoid-enriched oil characterized by a TCC of 570.08 ± 17.26 µg β-carotene equivalents/g dried pumpkin peel and an antioxidant activity of 180.55 ± 1.96 µg Trolox equivalents/g dried pumpkin peel. HPLC-DAD analysis identified fifteen carotenoids, including free and esterified xanthophylls and β-carotene, confirming the complexity and functional value of the extract. The developed Pickering emulsion exhibited suitable physicochemical characteristics and effectively protected carotenoids against environmental stress conditions. Furthermore, the formulation showed good

biocompatibility on HaCaT keratinocytes and demonstrated antioxidant potential by reducing intracellular ROS levels, supporting its suitability as a cosmetic platform for natural photoprotective compounds.

Conclusion

The combination of a green extraction strategy and Pickering technology enabled the development of a stable carotenoid-loaded emulgel with antioxidant and photoprotective potential. Therefore, the proposed formulation represents a promising sustainable cosmetic platform for skin application of natural bioactive compounds.

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DEVELOPMENT AND EVALUATION OF NADES-BASED EXTRACTS HYDROGELS

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Introduction

Natural Deep Eutectic Solvents (NADES) are emerging and promising green solvents for the extraction of bioactive compounds from botanical matrices, with considerable advantages over classical solvents, such as biodegradability, absence of toxicity, and general sustainability. Moreover, they show a wide and tuneable polarity range and high solubilization strength for different compounds. Their safety and intrinsic active properties allow to obtain functional and ready-to-use ingredients (consisting of the NADES-based extractive itself) of potential interest in food, cosmetic and pharmaceutical fields [1].

Aim of the work

In the present study, with the aim of preparing natural simple, safe, and active formulations, different NADES-based hydrogels were obtained from botanical matrices (sage, citrus, and bergamot). NADES solvents were prepared by microwave activation (MW) and then evaluated as extractants and carriers of different bioactive compounds, assessing their radical scavenging properties (RSA%) and antimicrobial activity. Their rheological behavior, in combination with different natural polymeric systems (Sodium hyaluronate HA; Xanthan gum XG; carboxymethylcellulose CMC; hydroxyethylcellulose HEC) was then investigated to obtain stable hydrogels for different applications.

Materials and Methods

MW NADES preparation

A hydrogen bond acceptor (Betaine) with hydrogen bond donors (glycerol, lactic acid) were mixed together in opportune ratio and stirred at 70°C for 2 min [2].
N1 - Betaine /Glycerol (1:2 molar ratio)
N2 – Betaine/Lactic Acid/Glycerol (2:1:1 molar ratio)

MW NADES extraction

The natural matrix (sage leaves S, citrus peel C, and bergamot peel B) and the previously prepared NADES solvents (N1, N2), were mixed (1:10 w/w) and processed under at 70°C for 5 min. The mixtures were then purified by centrifugation and the NADES-based extracts of interest (S-N1, S-N2, C-N1, C-N2, B-N1, B-N2) were collected and analysed without further purifications.

Radical Scavenging Activity (RSA%)

Radical Scavenging Activity of NADES-based extracts was determined by DPPH assay (Table 1), using Trolox as the standard, according to the literature [3].

Antimicrobial Activity

Minimum Inhibitory Concentrations (MICs) of all the NADES-based extracts were assessed on different clinical bacterial strains (*S. aureus, S. epidermidis, E. faecium, E. faecalis*) through the microdilution procedure (Table 2) described by the European Committee for Antimicrobial Susceptibility Testing (EUCAST) [4].

NADES-based extracts hydrogels preparation

Hydrogels were simply prepared at room temperature by rapid dispersion of 1% - 2% (w/w) gelling polymer in the NADES-based extracts, then adding water to obtain gelled network. (Figure 1)
HA - Sodium hyaluronate 2%
CMC - Carboxymethylcellulose 2%
HEC - Hydroxyethylcellulose 2%
XG- Xanthan gum 1.5%

NADES- hydrogels rheological behavior

NADES-hydrogels were evaluated in regard to their rheological behavior, applying a continuous shear method by means of a Brookfield viscosimeter (Figures 1-2).

Results and discussion

In this work, food grade natural compounds (betaine, glycerol, lactic acid) with intrinsic cosmetic functionalities were combined to obtain efficient natural deep eutectic solvents, allowing the recovery of bioactive compounds with different properties, from all the botanical matrices considered.
NADES-based sage extracts S-N1 and S-N2 seem to be very promising ingredients, showing good antioxidant and antimicrobial activities. Bergamot and citrus extracts (B-N and C-N) did not present any antimicrobial activity, anyway their radical scavenging activity is noteworthy, along with a pleasant scent (Table 1).
All the formulated hydrogels showed excellent rheological behaviour, with pseudoplastic flow and progressive viscosity reduction as shear rate increased. This behaviour is particularly advantageous for topical formulations, improving spreadability while maintaining formulation stability.

Table 1 - Radical Scavenging and Antimicrobial activities of NADES-based extracts

Sample	RSA% (± SD)	ANTIMICROBIAL ACTIVITY			
		E. faecalis	*E. faecium*	*S. aureus*	*S. epidermidis*
S-N1	60±0.6	3%	3%	3%	3%
S-N2	54±0.8	0.75%	0.75%	0.75%	0.75%
C-N1	43±0.9	-	-	-	-
C-N2	36±2.1	-	-	-	-
B-N1	30±0.9	-	-	-	-
B-N2	14±0.1	-	-	-	-

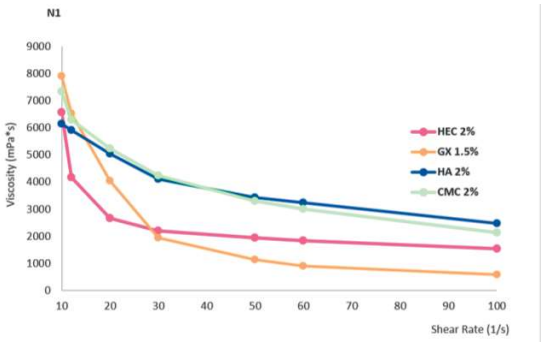


Fig. 1. Rheological behavior of NADES-hydrogels with NADES solvent N1

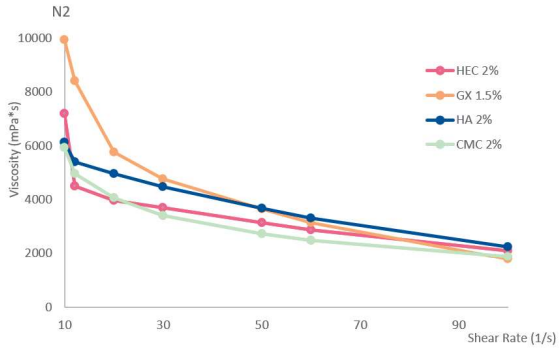


Fig. 2. Rheological behavior of NADES-hydrogels with NADES solvent N2

Conclusion

On the basis of the data obtained, NADES-based extracts hydrogels could be considered promising natural carriers of bioactive compounds, helpful in developing new natural, safe, and bioactive hydrogel cosmetic and pharmaceutical topical formulations with antimicrobial and antioxidant properties.

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Identification of bioactive compounds in botanical ingredients of cosmetic interest: spilanthol in *Acmella* species and bisabolol in Chamomile varieties

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Introduction

The growing demand for cosmetic products formulated with naturally derived ingredients has increased interest in botanical raw materials as sources of bioactive compounds with potential cosmetic applications. The chemical characterization of plant-derived ingredients is therefore a critical aspect of quality assessment, as the identification and quantification of specific bioactive constituents yield valuable information on the composition and consistency of these raw materials.

The genus *Acmella* comprises several plant species that are increasingly studied for cosmetic applications, primarily due to their alkamide content, with spilanthol recognized as the principal characteristic constituent. Since phytochemical composition can differ among species, quantitative determination of spilanthol represents an important tool for comparative evaluation and quality assessment of botanical ingredients. Spilanthol has attracted particular attention for its reported anti-aging, skin-smoothing, and tensor properties, as well as its potential as a naturally derived cosmetic ingredient. Studies on topical formulations containing *Acmella oleracea* extracts have demonstrated improvements in skin smoothness and reduction in roughness and wrinkle appearance, thereby supporting the potential of spilanthol-containing botanical ingredients in cosmetic applications [1,2].

Matricaria chamomilla is widely used in cosmetic formulations and represents a natural source of bisabolol, a sesquiterpene alcohol recognized for its soothing, anti-inflammatory, and antimicrobial properties. Since bisabolol concentration can vary depending on cultivar and plant material, quantitative determination of this compound yields valuable data for comparative chemical characterization and quality assessment of chamomile-derived ingredients [3,4].

Beyond its biological properties, bisabolol is considered a marker compound for the chemical profiling of chamomile-derived ingredients. Fluctuation in bisabolol content may reflect differences in genotype, cultivation conditions, geographical origin, and processing techniques. Therefore, quantitative determination of bisabolol represents a useful analytical approach for comparative evaluation, quality assessment, and standardization of botanical raw materials used in cosmetic applications.

Aim of the work

The aim of this study was to quantitatively characterize and compare spilanthol content in three *Acmella* species, namely *Acmella species 1*, *Acmella species 2* and *Acmella species 3*, and bisabolol content in four Chamomile varieties, namely *Chamomile variety 1*, *Chamomile variety 2*, *Chamomile variety* 3 and *Chamomile variety 4*, providing an analytical assessment of bioactive compounds relevant to botanical ingredients in cosmetic applications.

Materials and Methods

Materials: Methanol, n-hexane, sodium sulfate cartridge, PTFE membrane filters. Purified analytical standards of spilanthol and bisabolol.

Extraction of spilanthol
Acmella species (50 mg) were extracted twice with methanol by sonication for 10 min. After centrifugation and PTFE filtration, the resulting extracts were analyzed by liquid chromatography with diode-array detection (LC-DAD) using a gradient of acetonitrile, methanol, and 0.1% formic acid in water at flow rate of 0.2 mL/min. Spilanthol was identified by its retention time (13.2 min) and quantified using a purified analytical standard.

Extraction of bisabolol
Matricaria chamomilla samples (0.5 g) were extracted with 20 mL of n-hexane by sonication for 30 min. The resulting extracts were purified using a sodium sulfate cartridge to remove residual water and subsequently analyzed by gas chromatography-mass spectrometry (GC-MS) in splitless mode, at a flow rate of 1.31 mL/min. Data were acquired in scan mode using a quadrupole mass analyzer. Bisabolol was identified by its mass spectrum and quantified using a purified analytical standard.

Results and discussion

Quantitative analysis revealed differences in the content of the target bioactive compounds among the botanical materials investigated.

Spilanthol concentrations in *Acmella* species ranged from 0.7% to 1.5%, with *Acmella species 1 (Fig. 1)* showing the highest content, followed by *Acmella species 2* and *Acmella species 3*. The concentration observed in *Acmella species 1* was consistent with the 0.92–1.45% range previously reported in the literature

for *Acmella oleracea* capitula [5], although differences in plant material and analytical procedures should be considered when comparing these data.

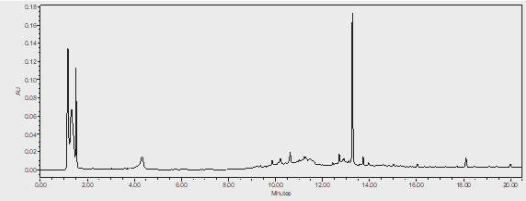


Figure 1: LC-DAD spectrum of Acmella species 1.

Bisabolol concentrations among the four Chamomile varieties ranged from 15 to 90 mg/kg. *Chamomile variety 2* showed the highest bisabolol content, followed by *Chamomile variety* 3, *Chamomile variety 4*, and *Chamomile variety 1*. The GC-MS spectrum of *Chamomile variety 2*, showing the characteristic mass spectral profile of bisabolol, is reported in *Fig. 2*. While direct comparison with literature values is limited by differences in analytical approach and expression of results, previous studies have demonstrated substantial variability in the sesquiterpenic profile of chamomile according to cultivar, geographical origin, and chemotype [6].

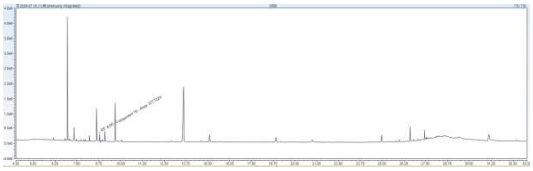


Figure 2: GC-MS spectrum of Chamomile variety 2.

Overall, the observed variability in the concentrations of target bioactive compounds highlight the necessity of quantitative analytical characterization for the selection, quality assessment, and standardization of botanical ingredients used in cosmetic applications.
The results demonstrated a clear variability in the content of the bioactive compounds investigated among the analyzed botanical materials. In *Acmella*, differences in spilanthol concentrations were observed among the species studied. *Chamomile varieties* showed distinct levels of bisabolol.
These results highlight that both botanical origin and varietal characteristics influence the concentration of bioactive constituents and, consequently, the chemical profile of plant-derived raw materials.
The application of chromatographic techniques, namely LC-DAD for spilanthol and GC-MS for bisabolol, enabled the targeted identification and quantitative determination of these compounds. This approach provides objective and reproducible data to support cosmetic companies in the characterization, comparison and selection of botanical raw materials according to their specific bioactive profiles.

Quantitative chemical characterization supports the standardization and quality control of botanical ingredients in cosmetic technology, enabling more informed raw material selection and improved consistency in cosmetic formulations. In an industrial context, identifying and selecting botanical sources with defined target compounds content can also enhance the efficient use of plant resources, thereby reducing variability and minimizing unnecessary material consumption throughout the production chain.

Conclusion

Overall, these findings highlight how analytical characterization can be a useful tool not only for quality assessment but also for supporting the rational selection and standardization of botanical ingredients. This approach promotes the valorization of plant resources and supports the development of more economically and environmentally sustainable methods in cosmetic production.

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Sessione 4 — Sistemi innovativi di veicolazione

28 ottobre 2026, 09:00–11:00

Chair: B. Ruozi (Unimore), M.L. Manca (Unica)

O12	Waste to Beauty: integrated nanoformulation strategies for the sustainable valorization of agri-food side streams *Ines Castangia · Università di Cagliari*	33
O13	Aspasomal Systems for the Cutaneous Delivery of Depigmenting Oligopeptides *Sofía Gisella Brignone · Università di Pisa*	35
O14	Cutaneous delivery of rutin with supramolecular nanosystems *Rita Cortesi · Università di Ferrara*	37
O15	Development and characterization of clay-stabilized Pickering emulsions for dermo-cosmetic application obtained from natural and sustainable ingredients *Samuele Bonafè · Università di Perugia*	39
O16	Spray to film gellan gum-chitosan-sericin hydrogels for the topical delivery of plant-derived bioactives in cosmetic formulations *Maria Rosa Gigliobianco · Università "G. d'Annunzio" Chieti-Pescara*	41
O17	Fermented *Opuntia ficus-indica* cladode extract as a new functional ingredient in a topical cosmetic formulation *Camilla Elena Di Bella · Università "G. d'Annunzio" Chieti-Pescara*	43

Waste to Beauty: integrated nanoformulation strategies for the sustainable valorization of agri-food side streams

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Introduction

The growing demand for sustainable, effective and scientifically validated cosmetic products is driving the development of next-generation skincare products based on renewable natural resources [1]. Among these, agri-food side streams represent a valuable yet largely underutilized source of high-value phytochemicals, including polyphenols, flavonoids and other bioactive compounds with well-documented antioxidant, anti-inflammatory and skin-protective properties. Despite their remarkable biological potential, the practical application of these natural extracts is often limited by poor physicochemical stability and reduced bioavailability [2]. In this context, advanced nanoformulation approaches offer an effective strategy to improve the stability, controlled delivery and biological efficacy of plant-derived bioactives, facilitating their translation into effective and scientifically validated cosmeceutical products [1,2].

Aim of the work

This study presents an integrated research strategy for the sustainable valorisation of agri-food side streams into high-value ingredients through green extraction, comprehensive phytochemical characterization and advanced nanoformulation.

Materials and Methods

Materials: Representative agri-food side streams were selected as renewable sources of bioactive ingredients based on their phytochemical profile and potential cosmeceutical applications.

Extraction, phytochemical characterization and nanoformulation: Bioactive extracts were obtained using sustainable extraction approaches, chemically characterized and incorporated into advanced nanosystems, including liposomes, glycerosomes, penetration enhancer-containing vesicles (PEVs) and nanoemulsions. The resulting formulations were comprehensively characterized in terms of particle size, polydispersity index, ζ-potential, morphology, encapsulation efficiency, storage stability and rheological properties.

Biological evaluation: The biological performance of the developed nanosystems was assessed using validated in vitro models to investigate cytocompatibility, antioxidant, anti-inflammatory and regenerative activities, as well as their protective effects against oxidative stress and other skin-relevant biological responses.

Results and discussion

The integrated research strategy proved effective across diverse agri-food matrices, demonstrating that these renewable resources represent versatile sources of high-value bioactive ingredients for innovative skincare formulations. Despite the chemical diversity of the investigated matrices, the integration of sustainable extraction, comprehensive phytochemical characterization and advanced nanoformulation consistently improved the physicochemical stability, cutaneous delivery and biological efficacy of the resulting bioactive ingredients. This strategy was successfully applied to grape pomace, olive pomace, olive leaves and avocado peel extracts, formulated into liposomes, glycerosomes, penetration enhancer-containing vesicles (PEVs) and nanoemulsions to improve skin protection, preserve skin homeostasis and support healthy skin ageing [1-4]. Furthermore, formulation design was tailored according to the physicochemical characteristics of each extract, highlighting the flexibility of the proposed approach across different agri-food matrices. Recent investigations further expanded this research by integrating technological characterization, including rheological assessment, with biological validation, providing additional insights into formulation performance and cosmetic acceptability. Collectively, these findings demonstrate a multidisciplinary strategy for the sustainable development of scientifically validated, high-value ingredients [3].



Fig. 1. Integrated workflow for the development of sustainable cosmeceuticals from agri-food side streams

Conclusion

This work demonstrates how advanced nanoformulation can unlock the cosmetic potential of agri-food side streams, promoting the development of effective, sustainable and science-based skincare formulations while supporting the transition towards a circular bioeconomy.

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Aspasomal Systems for the Cutaneous Delivery of Depigmenting Oligopeptides

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Introduction

Skin hyperpigmentation is a common cosmetic concern associated with dysregulated melanin production and represents an important target for the development of innovative topical formulations [1]. Decapeptide-12 (DECA-12) and Nonapeptide-1 (NONA-1) are cosmetic oligopeptides investigated for their antimelanogenic activity through distinct mechanisms of action. However, their relatively high molecular weight and hydrophilic nature may limit their ability to cross the skin barrier. Moreover, peptide stability during formulation, storage and topical application may represent an additional challenge [2, 3].

Aspasomes are vesicular nanocarriers composed of ascorbyl palmitate (ASC16), cholesterol (CHOL) and a negatively charged lipid such as dicetyl phosphate (DCP). In addition to their carrier function, the presence of the ascorbyl moiety provides intrinsic antioxidant properties, making these systems particularly attractive for cosmetic applications [4]. Previous studies demonstrated aspasomes ability to form bilayer vesicles and to modulate the release and skin delivery of encapsulated compounds [5, 6, 7].

Aim of the work

The aim of the study was to develop and characterize aspasomal vesicular systems for the topical delivery of DECA-12 and NONA-1. Their physicochemical properties, peptide encapsulation and retention, peptide-bilayer structural interactions, *in vitro* release behaviour and skin permeation and retention were investigated.

Materials and Methods

Materials

DECA-12 and NONA-1 were obtained from RS Synthesis (USA). Ascorbyl palmitate, cholesterol and dicetyl phosphate were purchased from Sigma-Aldrich. HPLC-grade chloroform and methanol were used as organic solvents, whereas phosphate-buffered saline (PBS, pH 7.4) was used for vesicle hydration and dilution. Vivaspin 6 centrifugal ultrafiltration devices (MWCO 5 kDa and 10 kDa PES membrane) were obtained from Sartorius Stedim Biotech, and regenerated cellulose dialysis membranes (MWCO 6–8 kDa, Spectra/Por) were purchased from Cole-Parmer. Porcine ear skin, used as the *ex vivo* tissue model for skin permeation and retention studies, was obtained as a by-product from a local slaughterhouse.

Preparation

Aspasomes were prepared by the thin-film hydration method. ASC16:CHOL:DCP were combined at a molar ratio of 45:45:10, corresponding to a total lipid amount of 200 µmol. The lipid film was hydrated with PBS at room temperature in the presence of the peptide (DECA-12 or NONA-1), followed by probe sonication to reduce vesicle size and improve size homogeneity. Different sonication conditions were evaluated, and the formulations were characterized by dynamic light scattering (DLS) to determine the intensity-weighted mean hydrodynamic diameter (Z-average) and polydispersity index (PDI).

Peptide encapsulation efficiency was determined indirectly by separating free peptide from the vesicular fraction using centrifugal ultrafiltration with Vivaspin 6 devices. The separated peptide was subsequently quantified by fluorescence spectroscopy using excitation and emission wavelengths of 280 and 350 nm, respectively. Formulation stability was evaluated during storage at room temperature protected from light. Differential Scanning Calorimetry (DSC) and ATR-FTIR spectroscopy were employed to investigate changes in the thermal behaviour and spectroscopic profile of the lipid components and peptides following their incorporation into the vesicles.

To further investigate peptide localization and accessibility within the vesicular structure, fluorescence quenching experiments were performed using potassium iodide (KI) as a membrane-impermeant collisional quencher. Fluorescence-quenching data were analysed according to the Stern–Volmer equation, and the behaviour of free peptides was compared with that of aspasome-associated peptides.

In vitro release was evaluated by dynamic dialysis through a regenerated cellulose membrane, using PBS as the receiving medium under controlled temperature and agitation. Samples were collected at predetermined times and the released peptide was quantified fluorimetrically.

Peptides permeation through porcine ear skin and retention within the skin tissue were evaluated after topical application of the formulations using vertical diffusion cells.

Results and Discussion

Optimization of the preparation process demonstrated that sonication strongly influenced vesicle size and size distribution. Under the selected preparation conditions, nanosized vesicular systems with a relatively

homogeneous size distribution were obtained. Peptide incorporation resulted in an increase in hydrodynamic diameter compared with empty aspasomes, suggesting that the presence of the peptides influenced vesicle organization and/or surface hydration. Both peptide-loaded formulations exhibited high encapsulation efficiencies, demonstrating effective incorporation of the actives in the aspasomal systems. The formulations also showed high peptide retention during storage, with only a limited reduction in the encapsulated fraction over the investigated period.

DSC analysis showed that the characteristic thermal transitions of the free peptides were no longer detectable in the peptide-loaded aspasomes. Changes in the thermal transitions of the lipid components were also observed. These findings are consistent with peptide association with the vesicular structure and with interactions between the peptides and the lipid bilayer. ATR-FTIR results further supported this interpretation. The characteristic peptide bands were not clearly distinguishable in the spectra of the loaded formulations, whereas the main lipid-associated bands remained detectable.

The *in vitro* release study highlighted a marked difference between free and aspasome-associated peptides. The free peptide rapidly crossed the dialysis membrane, whereas the peptide-loaded formulation showed very limited release throughout the experimental period. This behaviour indicates a strong retention of the peptides within the vesicular system and suggests that the aspasomes may act as a reservoir capable of limiting its immediate diffusion.

Fluorescence-quenching experiments provided complementary information on peptide accessibility. Free DECA-12 and NONA-1 showed linear Stern–Volmer relationships, consistent with predominantly dynamic collisional quenching. In contrast, the aspasome-associated peptides exhibited a different response to KI, with reduced accessibility of their fluorophoric residues to the external quencher and supporting the shielding effect of the lipid bilayer.

The skin permeation experiments showed no detectable transdermal permeation of the peptides into the receptor phase, while they were retained within the skin. This distribution profile is potentially advantageous for cosmetic applications aimed at producing a local effect while minimizing systemic exposure.

Conclusion

ASC16-based aspasomes were successfully developed as vesicular carriers for the depigmenting oligopeptides DECA-12 and NONA-1. The resulting nanosized formulations showed effective peptide encapsulation and high retention during storage. The combination of a vesicular carrier containing an antioxidant-derived lipid with peptide-based active ingredients and a favourable skin-retention profile supports further investigation of aspasomes as innovative topical delivery systems for cosmetic applications.

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Cutaneous delivery of rutin with supramolecular nanosystems

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Introduction

The use of naturally derived ingredients for the development of cosmetic and dermatological products is a prerogative for companies in the sector. In this context, polyphenolic compounds, such as flavonoids, play a significant role due to their antioxidant and anti-inflammatory activity, after topical application. Among these, rutin (RU) is attracting considerable interest as a therapeutic agent in many pharmaceutical or cosmeceutical formulations. However, RU is scarcely water soluble and bioavailable, so recently, delivery systems have been widely studied to increase the efficacy of the drug and the contact time to improve its absorption and control the release [1].

Aim of the work

The present study aimed to design supramolecular systems for RU delivery, suitable for topical application. Specifically, the β-cyclodextrin inclusion complex containing rutin (Cy/RU) was incorporated into liposomes (L) leading to the formation of RUCyL (Fig.1), and the performances of both Cy/RU and RUCyL were evaluated.

Fig. 1. Schematic representation of systems production.

Materials and Methods

Materials: All materials and solvents were from Sigma-Aldrich (St Louis, MO, USA).

Methods: The supramolecular nanosystems were obtained by *thin film hydration* then characterized in terms of size, morphology, encapsulation efficiency, antioxidant activity and skin safety, by DLS, TEM, UV spectrometry, DPPH radical liberation assay and patch test, respectively. RU release kinetics was determined by Franz cell experiments

Results and discussion

The comparison of the performances of both Cy/RU and RUCyL, showed the maintaining of their physico-chemical stability over time in terms of color, size, RU content, and antioxidant activity. The antioxidant assays demonstrated a greater activity when RU is encapsulated within CyL. Comparative *in vitro* studies (Fig. 2, up) on drug diffusion suggested an improved delivery of RU, possibly controlled by the vesicular system; in contrast, the jellified formulations with carbopol or xanthan gum showed a lower spreadability and greater adhesiveness, ensuring effective skin application. On the other hand, the patch tests (Fig. 2, down) demonstrated an optimal tolerance by the skin of both formulations. Based on these results, the produced Cy/RU and RUCyL formulations may represent an interesting starting point for the topical application of natural antioxidant molecules.

Fig. 1. Schematic representation of a Franz cell and in vitro *release (up) and* in vivo *irritative reactions acquired by patch test (down). Cy/RU (green) and RUCyL (blue) dispersed in xanthan gum (dark color) or carbopol (light color).*

Conclusion

The dual encapsulation of RU through CyL delivery, combined with gelation offers a synergistic approach able to enhance topical release and antioxidant efficacy. This strategy addresses the limitations of RU solubility and stability, ensuring improved skin retention and controlled release, and represents a promising platform for the development of advanced dermal formulations.

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Development and characterization of clay-stabilized Pickering emulsions for dermo-cosmetic application obtained from natural and sustainable ingredients

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Introduction

Nowadays Pickering emulsions are gaining increasing interest for topical applications. These systems are characterized by the presence of solid particles, adsorbed at the O/W interface preventing coalescence [1]. The absence of surfactants, which may cause irritation and compromise the skin barrier, represents a key feature of these systems, making them particularly attractive.
Among solid stabilizers, clay minerals represent an interesting choice due to their multifunctionality [2], acting as rheology modifiers as well. Clay properties such as particle wettability, size, morphology, surface charge, concentration and many others influence the stabilization of Pickering emulsions [3].

Aim of the work

In the present work, Pickering O/W emulgels for skin application were developed and characterized. The composition was optimized taking into account the recent EU Regulation 2023/2055.
Snail secretion filtrate (SSF) was employed as the main component (W phase) due to its interesting properties such as moisturizing, soothing and wound-healing [4]. Two different SSFs obtained from different producers were used (SSF1/SSF2). *Opuntia ficus-indica* seed oil (OFI), which possesses antimicrobial and antioxidant activities [5], was selected as oil phase and bentonite nano-clay (Ben) was used as emulsifying agent. Sodium hyaluronate (NaHy) and chia seed mucilage (CSM) were evaluated as thickeners for the developed emulsions.

Materials and Methods

Materials: snail secretion filtrates (SSF1 and SSF2) were purchased through the appropriate commercial channels; *Opuntia ficus-indica* seed oil was purchased from the Soil Association (Milano, MI, Italy); bentonite nanoclay was purchased from Sigma-Aldrich (Merck KGaA, Darmstadt, Germany); glycerol was purchased from A.C.E.F. S.p.A. (Fiorenzuola d'Arda, PC, Italy); sodium hyaluronate (686 kDa), trisodium citrate dihydrate, and anhydrous citric acid were purchased from Guinama (Valencia, Spain); chia seeds were purchased from a local store. CSM was extracted from chia seeds adapting a published method. [6].
Emulgel preparation: O/W emulsions were prepared as follows: SSF1/2, citrate buffer (pH 5) and glycerol were mixed by high-shear homogenization at 5000 rpm; OFI and Ben were then added in this order and the speed was increased to 15000 rpm and maintained until a total homogenization time of 10 min was reached, obtaining a pre-emulsion. This pre-emulsion was further homogenized by probe ultrasonication in pulsed mode (50% duty cycle) and 60% of the maximum amplitude for 10 min. The emulsions were thickened by adding NaHy or CSM using high-shear homogenization at 15000 rpm for 2 min.
Stability studies*:* the physical stability of the emulsions was evaluated by three different tests by assessing the occurrence of phase separation: i) visual monitoring of formulations stored at 8, 25, and 37 °C (5 weeks); ii) centrifugation test (samples were centrifuged at 25 °C for 20 min at 3000 rpm); iii) visual monitoring of formulations subjected to eight 8/37 °C temperature cycles (2 days at each temperature).
Droplet size analysis: droplet size analysis was performed using a Bettersizer 2600, based on laser diffraction technology. The droplet population was identified by comparison with blank samples containing all components except the O phase. This assignment was further confirmed by optical microscopy observations, which showed droplet size distributions in agreement with the identified peak.
Rheological and textural characterization: the extensibility of the formulations was evaluated by the parallel-plate method according to the Royal Spanish Pharmacopoeia. Rheological measurements were performed using a Discovery HR-3 Rheometer (TA Instruments, New Castle, DE, USA) equipped with a 40 mm aluminum parallel-plate geometry coupled with a Peltier temperature control system. Frequency sweep analyses were carried out at 25 °C within the linear viscoelastic region (LVR) by applying a constant strain of 0.25%. The frequency ranged from 0.01 to 10 Hz. The gap was set at 0.4 mm. Storage modulus (G′) and loss modulus (G″) were recorded as a function of frequency, while tan δ was calculated from the G′ and G″ values. The textural properties of the formulations were characterized using a texture analyzer (TA.XT

Plus, Stable Micro Systems, Godalming, UK) equipped with a 5 kg load cell and a back-extrusion rig. Seventy grams of each formulation were placed into a 50 mm diameter beaker and compressed using a 40 mm diameter plunger. The following parameters were extracted from the force–time curves: firmness, consistency, cohesiveness, and work of cohesion.

Results and discussion

The emulsion composition was optimized to achieve stability against coalescence and a pH value suitable for skin application (approximately 5.0). The selected emulsion had the following composition: SSF1 or SSF2, 50% (w/w); citrate buffer, 23% (w/w); OFI, 10% (w/w); glycerol, 10% (w/w); Ben, 7% (w/w). NaHy and CSM at concentrations of 0.5 and 1.0 % (w/w) were evaluated as thickeners for this optimized emulsion. Stability studies confirmed that the inclusion of a thickener is crucial for effective stabilization of the formulation, as the non-thickened emulsions and the emulgels containing 0.5% (w/w) NaHy failed the centrifugation test. Nevertheless, all formulations remained stable at rest under the tested storage temperatures. Droplet size analyses, coupled with optical microscopy, clearly demonstrated that thickener addition allowed maintenance of smaller droplet sizes (Fig. 1). Moreover, droplet size did not change over 30 days, indicating good stability.

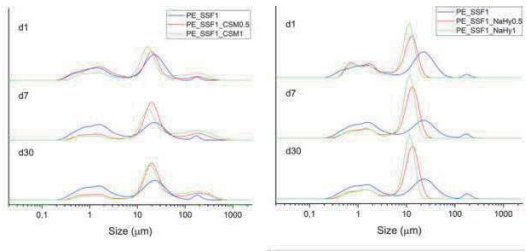


Fig. 1. Droplet size analyses of the non-thickened emulsion (blue line), and the emulsions containing thickeners at 0.5% (w/w) (red line) and 1% (w/w) (green line). From the top, the analyses performed 1, 7 and 30 days after the preparation are shown. For simplicity, only SS1-based formulations are reported.

Based on the extensibility test results, all CSM-based emulgels exhibited the lowest spreading ability, which was inversely proportional to CSM concentration. When NaHy was employed as the thickener, a concentration of 0.5% was not sufficient to reduce spreading ability, as these emulgels showed behavior very similar to that of the non-thickened emulsions. This finding was further confirmed by the back-extrusion test; the non-thickened emulsions and the emulgels containing 0.5% NaHy produced very similar values for all the calculated parameters. In contrast, CSM addition increased the structure and viscosity of the formulations in a concentration-dependent manner. The textural parameters of the formulation containing 1% (w/w) NaHy were very similar to those of the formulation containing 0.5% (w/w) CSM, although the former was slightly tackier (higher negative work of cohesion). Finally, SSF1-based formulations were consistently more structured than SSF2-based ones, suggesting differences in their quali-quantitative compositions. Rheo-textural characterization was completed through frequency sweep. Interestingly, the non-thickened emulsions were characterized by a low loss tangent (≈0.08), indicating a highly elastic but brittle clay "house-of-cards" network. CSM produced a concentration-dependent upward shift of both G′ and G″ over the entire frequency range. In contrast, NaHy showed a less predictable behaviour when comparing SSF1 and SSF2 formulations, exhibiting greater variations in loss tangent at increasing frequencies. Overall, integrating the results obtained from centrifugation, extensibility, back-extrusion, and frequency sweep tests provided a comprehensive interpretation of the rheo-textural behaviour of these complex systems. The baseline emulsion matrix, in the absence of thickeners, is supported by the Ben network and, although highly elastic, is also brittle and lacks structure. The results suggest that 0.5% (w/w) NaHy is insufficient to achieve polymer chain entanglement. Conversely, CSM, a branched heteropolysaccharide, represents a supporting network more efficient than NaHy within the Pickering system, producing a more predictable and structured rheological behavior.

Conclusion

Ben proved to be an effective stabilizer for the emulsions developed in this work. Emulsion composition was optimized to achieve stability and a pH value suitable for skin application. To improve stability and rheological/textural properties, NaHy and CSM were evaluated as thickeners. While NaHy provided greater spreading ability and smaller droplet sizes, CSM was more effective in stabilizing the resulting emulgels. Formulations containing 0.5% CSM and 1% NaHy exhibited very similar textural properties, although the latter showed higher tackiness. The two SSFs employed were shown not to be completely interchangeable, producing slightly different rheological behaviour and textural parameters. This highlights the importance of standardization in the production of naturally derived ingredients. Biological activity studies are still ongoing.

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Spray to film gellan gum-chitosan-sericin hydrogels for the topical delivery of plant-derived bioactives in cosmetic formulations

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Introduction

Cosmetic and dermocosmetic formulations require topical vehicles that combine easy application, uniform distribution, skin compatibility and the protection and retention of labile bioactives.

Gellan gum and chitosan have been investigated in topical films, hydrogels and skin-delivery systems, while sericin has been explored in topical hydrogels and dermocosmetic scaffolds for its moisturising, antioxidant and film-forming properties [1–2]. Liposomal encapsulation can further protect plant extracts and modulate their skin presentation; recent work supports microfluidic production of extract-loaded liposomes for dermal and cosmetic delivery [3].

Aim of the work

The work aims to develop a spray to film gellan gum-chitosan hydrogel containing sericin and microfluidic liposomes, designed to combine easy spray application with rapid post-deposition recovery and local retention of plant-derived bioactives.

Materials and Methods

For hydrogel preparation, two grades of chitosan (CS) were employed: low-molecular-weight CS (Merck Life Science, Milan, Italy; molecular weight 50–190 kDa; degree of deacetylation 75–85%) and high-molecular-weight CS (Merck Life Science, Milan, Italy; molecular weight 310–375 kDa; degree of deacetylation ≥75%). Low-acyl gellan gum (GG) (Kelcogel CG-LA, CP Kelco–Tate & Lyle, London, UK), sodium carboxymethyl cellulose E466 (Acef, Fiorenzuola d'Arda, Italy), hydrochloric acid 37% (density 1.2 g/mL; Merck Life Science, Milan, Italy), and deionized water were also used.

Two procedures were developed to obtain physically crosslinked hydrogels based on low-acyl GG and CS. In both formulations, GG was used at 1% (w/w), whereas CS was dissolved in 0.1 M hydrochloric acid, selected for its high solubilising capacity. Depending on the preparation protocol, CS was used at 1% or 3% (w/w).

In the first procedure, GG was progressively added to the CS solution under continuous stirring at 80 °C, and the resulting mixture was stirred overnight. Several CS ratios were investigated; formulations with ratios of 50:50 and 40:60 were selected for further evaluation based on their mucoadhesive properties. In the second procedure, a more concentrated CS solution (3%, w/w) was prepared. In this case, GG was incorporated during the CS solubilisation step, and the mixture was manually blended at 80 °C for 20 minutes. The formulation was subsequently cooled under continuous stirring. The same CS ratios, namely 50:50 and 40:60, were investigated.

To provide additional bioactive functionality, the hydrogels were loaded with liposomes and sericin. The liposomes were produced using the Sunshine Microfluidic System (Unchained Labs, Pleasanton, CA, USA) and loaded with grape-derived bioactive water extracts at 10 mg/mL.

The resulting hydrogel formulations underwent rheological analysis to assess their response to different mechanical stresses. Measurements were performed using a Haake Mars IQ air rheometer (ThermoFisher Scientific, Munich, Germany) equipped with a parallel-plate geometry with 35 mm diameter plates and a 1 mm gap. We further analysed both unloaded and bioactive-loaded hydrogels by FT-IR and Raman spectroscopy to investigate their structural and compositional characteristics.

FT-IR spectra were recorded using an IR Affinity-1S spectrophotometer (Shimadzu, Milan, Italy) equipped with a sealed and desiccated interferometer, a DTS detector based on deuterated triglycine sulfate doped with L-alanine, and a single-reflection diamond ATR crystal (QATR-10). We collected spectra over 4000–400 cm^{-1} using 45 scans per sample at a resolution of 4 cm^{-1}.

Results and discussion

Rheological analyses showed that all hydrogels exhibited pseudoplastic, shear-thinning behaviour, characterized by a progressive decrease in viscosity with increasing shear rate. Compared with pure GG systems, incorporating CS generally resulted in lower viscosity values. Nevertheless, the 40:60 GG formulation showed an unexpected increase in viscosity at 37 °C compared with room temperature, which could be advantageous for in situ applications requiring enhanced consistency at body temperature. Although this formulation contained a lower proportion of GG than the 50:50 system, it exhibited higher viscosity, likely because of its greater CS content. Although CS alone forms a weaker network than GG,

electrostatic interactions between the two polymers may enhance its contribution to the overall gel structure.
Hydrogels prepared according to method 2, which employed a 3% (w/w) CS stock solution, also showed marked shear-thinning behaviour. Because of their firmer consistency, we analysed these samples using a parallel-plate geometry. The results indicated that higher CS concentration improved gel strength, presumably through stronger, more stable ionic interactions between CS and GG. In particular, the 50:50 formulation prepared using method 2 showed nearly superimposable viscosity profiles at 25 and 37 °C, suggesting a temperature-stable response that could benefit applications requiring consistent performance under physiological conditions. The corresponding 40:60 formulation showed a slight increase in viscosity at 37 °C, similar to the formulation prepared using method 1, supporting its potential use in sprayable systems that can undergo post-application viscosity enhancement. However, the increased stiffness of these systems could negatively affect their sprayability.
FT-IR analysis revealed significant spectral shifts in all tested hydrogels, supporting the formation of a polyelectrolyte complex involving the functional groups of GG and CS. The characteristic signal associated with ammonium–carboxylate interactions was clearly observed in the formulations prepared using method 2 and, unexpectedly, was also detected in the 40:60 GG hydrogel prepared using method 1. This finding was particularly relevant because the latter formulation exhibited lower viscosity, which might otherwise have suggested a reduced degree of polymer interaction.
Among all the investigated samples, the 40:60 GG formulation prepared using method 1 showed the most favourable sprayability under the experimental conditions. Following the incorporation of sericin and the bioactive systems, the gellan gum-chitosan hydrogels maintained their pronounced shear-thinning.

Selected sericin-containing formulations preserved the rheological characteristics required for topical application, while also allowing the evaluation of sericin as a skin-conditioning and film-forming ingredient.

Liposomes prepared using the Sunshine system and loaded with grape-derived extracts were incorporated into the hydrogel matrix without compromising its structural integrity. This approach provides a modular platform for protecting and delivering both hydrophilic and lipophilic plant-derived bioactive compounds.

In vitro assays using normal human dermal fibroblasts indicated favourable cell compatibility, while the release profiles supported topical delivery of the encapsulated bioactive compounds.

Conclusion

This work supports a product-oriented spray to film platform for cosmetic and dermocosmetic skin care. The innovative element is integrating sericin as a skin-conditioning component with microfluidic, Sunshine-produced liposomes carrying plant-derived extracts, within a gellan gum-chitosan gel that sprays easily and recovers viscosity after deposition.

The platform can be tuned to combine film formation, skin retention and controlled presentation of plant bioactives, supporting further optimisation of sensory properties, stability and cosmetic performance.

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Fermented *Opuntia ficus-indica* cladode extract as a new functional ingredient in a topical cosmetic formulation

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Introduction

Opuntia ficus-indica cladodes represent an abundant plant biomass and a valuable source of phenolic compounds, including phenolic acids and flavonoids [1]. Cladode-derived extracts and fractions have shown biological properties relevant to skin applications, including protection against UVA-induced oxidative stress and skin-repairing activity [2,3]. Among the phenolic constituents reported in *O. ficus-indica* cladodes, rutin, gallic acid and catechin have independently shown effects on collagen-related processes in human dermal fibroblasts [4–6], supporting the interest of this matrix as a source of skin-functional ingredients.

The functional properties of plant phenolics are closely related to their chemical structure [7]. In this context, microbial fermentation represents a biotechnological approach for modifying plant phytocomplexes through the release and biotransformation of naturally occurring phenolic constituents [8]. A *Saccharomyces cerevisiae*-mediated fermentation strategy previously developed by our group demonstrated the potential of this approach for obtaining skin-functional ingredients from plant-derived matrices [9].

Although *O. ficus-indica* cladode-derived ingredients have been investigated for topical use, comparatively little attention has been devoted to the formulation of their fermented counterparts. The incorporation of a fermented cladode-derived ingredient into a suitable topical delivery system therefore represents a further step in exploring the potential of this matrix for skin applications.

Aim

The study aimed to develop and characterize a topical formulation designed to deliver a fermented ingredient obtained from *Opuntia ficus-indica* cladodes. To this purpose, a previously developed *Saccharomyces cerevisiae*-mediated fermentation strategy was applied to *O. ficus-indica* cladodes as a new plant matrix, and the effects of fermentation on the chemical composition and skin-related biological properties of the resulting extracts were investigated. The selected fermented extract was subsequently incorporated into an O/W emulsion, whose technological suitability and compatibility with the biotransformed ingredient were evaluated.

Materials and Methods

***O. ficus-indica* cladode extraction and biotransformation**

O. ficus-indica cladode powder was subjected to ultrasound-assisted extraction using water or ethanol/water (60:40, v/v). The resulting extracts were lyophilized and subjected to *Saccharomyces cerevisiae T-58*-mediated fermentation at 37 °C for 24, 48 and 72 h and 14 days, alongside the corresponding non-fermented controls.

Phytochemical characterization

The aqueous and hydroalcoholic extracts were initially screened by HPLC-UV analysis to characterize their phenolic composition, together with spectrophotometric assays for total phenolic content (TPC), total flavonoid content (TFC), ABTS radical-scavenging activity and horseradish peroxidase (HRP) inhibition. Based on the screening results, the aqueous extracts were selected for further investigation, with the 14-day fermented sample (F4) chosen as the representative fermented extract for the subsequent experimental phases.

Biological studies

In vitro biological evaluation was performed in normal human dermal fibroblasts (NHDFs) by comparing F4 with the corresponding non-fermented extract (NF). Cytocompatibility was assessed over the 0.125–1 mg/mL concentration range by MTT assay after 24 and 48 h and by lactate dehydrogenase (LDH) assay after 24 h. The effects on extracellular matrix-related processes were further investigated at 0.5 and 1 mg/mL by evaluating COL1α1 gene expression by real-time RT-PCR after 6 h and the protein expression of Pro-COL1, integrin β1 and MMP9 by Western blotting after 24 h.

Topical formulation development and characterization

For the development of the topical delivery system, an O/W emulsion was optimized for the incorporation of the selected fermented extract (F4). The resulting formulation was subjected to physicochemical and technological characterization, including pH measurement, colorimetric evaluation and accelerated stability testing by centrifugation. Microstructural analysis was performed by optical microscopy, while droplet-size distribution was assessed by laser diffraction. Rheological behavior was evaluated at 25 and 40 °C. The formulation was compared with the corresponding extract-free base and a formulation containing the non-fermented extract (NF).

Results and discussion

Fermentation induced solvent- and time-dependent changes in the phenolic characteristics of the cladode extracts. The aqueous series showed the most pronounced increase in total phenolic and flavonoid contents, with the 14-day fermented extract (F4) reaching the highest TPC (48.36 mg GAE/g dry extract) and TFC (3.81 mg RE/g dry extract) among the investigated samples. Conversely, the hydroalcoholic extract showed lower TPC after prolonged fermentation. These findings supported the selection of the aqueous extracts and the 14-day fermentation time point for subsequent investigations.

HPLC-UV analysis further revealed fermentation-induced changes in the phenolic composition of the aqueous extract. The total concentration of identified phenolic compounds increased from 45.28 µg/mL in NF to 56.11 µg/mL in F4, with pronounced changes in individual constituents. In particular, caftaric acid increased from 2.63 to 9.83 µg/mL, whereas rutin, a glycosylated flavonoid, decreased from 1.78 to 1.29 µg/mL, indicating a substantial remodeling of the phenolic profile following prolonged fermentation.

The fermentation-induced compositional changes were accompanied by a distinct biological response in NHDFs. Both NF and F4 were well tolerated over the investigated concentration range, as indicated by MTT and LDH assays, showing that fermentation did not adversely affect cytocompatibility. More importantly, at 1 mg/mL, F4 significantly upregulated COL1α1 gene expression compared with both NF and untreated cells, whereas NF reduced COL1α1 transcription. At the protein level, both extracts increased Pro-COL1, with F4 inducing significantly higher levels than NF at both 0.5 and 1 mg/mL. These findings indicate that the compositional changes induced by fermentation were associated with a distinct biological response, particularly in collagen-related processes. This is consistent with evidence that microbial fermentation can modify the composition and profile of plant phenolics through their release and bioconversion [8].

F4 also modulated other ECM-related markers, showing lower integrin β1 expression than NF and increased MMP9 expression. Although MMP9 upregulation should not be considered independently as a beneficial effect, its concomitant occurrence with increased COL1α1 transcription and Pro-COL1 levels is consistent with a broader modulation of ECM-related processes involving both matrix synthesis and turnover.

Finally, F4 was successfully incorporated into an O/W emulsion for topical application. The F4-containing formulation maintained a pH of approximately 5.5, showed no macroscopic phase separation and exhibited only minimal color variations. Droplet-size distribution and viscosity remained comparable to those of the extract-free base and NF-containing formulations, while all systems exhibited shear-thinning behavior at both 25 and 40 °C, with no relevant hysteresis. Overall, these findings indicate that incorporation of the fermented extract did not compromise the main physicochemical and rheological properties of the emulsion.

Conclusion

This study demonstrates that a previously developed *S. cerevisiae*-mediated fermentation strategy can be successfully extended to a new plant matrix, *O. ficus-indica* cladodes, supporting their valorization as a source of skin-functional ingredients. The selected fermented extract showed distinct compositional and biological properties compared with its non-fermented counterpart and was successfully incorporated into an O/W emulsion without compromising the main technological properties of the formulation. An in vivo study in healthy volunteers is currently underway to evaluate formulation performance through measurements of skin hydration and elasticity, together with objective Antera 3D analysis of skin surface topography, including wrinkles, texture and roughness.

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Sessione Poster

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Elena Ugazio · Università di Torino

Olive pruning residues as source of bioactive compounds: extraction's valorization for cosmetic and nutraceutical applications

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Introduction

Mediterranean olive growing generates large amounts of pruning residues, mainly leaves and twigs: in Italy alone, 1.65–3.30 million tons of pruning biomass are produced every year [1]. This material is currently shredded in the field, burned or directed to low-value energy uses, although it is rich in phenolic compounds of cosmetic and nutraceutical interest [2]. Available studies, however, focus almost exclusively on leaves separated from the rest of the material and on yield optimization, leaving unexplored the use of the residue as a source of bioactive compounds in the form in which it is actually collected, as well as the two requirements that govern its real application: light stability and cell compatibility.

Aim of the work

The work evaluates unseparated pruning residue of *Olea europaea* L., leaves and twigs together, as a raw material for bioactive extracts, comparing three extraction techniques in terms of yield and phenolic recovery and characterizing the resulting extracts for light stability and cell compatibility.

Materials and Methods

Plant material and compositional characterization: Olive (*Olea europaea* L., cv Frantoio) pruning residues were collected in Perugia (Italy) in April 2026. Leaves and twigs were processed together as a single matrix, with no separation at any stage, so that all determinations refer to the mixed residue. The material was dried at 40 °C to constant weight and ground to a powder. Moisture and dry matter were determined gravimetrically, total carbon by elemental analyser and total nitrogen by the Kjeldahl method, and the C/N ratio was calculated.
Extraction procedures: Microwave-assisted extraction (MAE), ultrasound-assisted extraction (UAE) and maceration were carried out on dry biomass at constant extraction solvent (water/ethanol 50:50) and solid/liquid ratio (1:10), varying only the technique-specific parameters: 800 W, 80 °C, 10 min for MAE; 28 kHz, 240 W, 30 °C, 30 min for UAE; 24 h contact time for maceration. Extracts were filtered, concentrated and dried; yield is expressed as weight percentage of dry biomass.
Extract characterization*:* Total phenolic content (TPC) was determined by the Folin-Ciocalteu method and expressed as mg gallic acid equivalents (GAE) per g of dry extract. Total soluble carbohydrates were determined on the aqueous redissolution of the dry extract by the phenol–sulfuric acid method [3] and expressed as glucose equivalents. Dried extracts were analysed by ATR-FTIR over 4000–400 cm^{-1}.
UV irradiation and cell viability*:* The MAE extract, in dry form, was characterized by FTIR before and after UV irradiation (UV lamp, 396 nm, 3.38 mW/cm^2, total dose 24.3 J/cm^2). The maceration extract was tested on HeLa cells by MTT assay, which assesses cytotoxicity through cell viability, over the 0.625–20 µg/mL range after 24 and 48 h of exposure, viability being referred to the untreated control.

Results and discussion

The starting biomass showed a dry matter content of 81.8 % and a C/N ratio of 39.5, consistent with a predominantly lignocellulosic material. Under the conditions tested, extraction returned 23.6 % w/w of dry extract, with a total phenolic content of 15.47 ± 1.98 mg GAE/g extract and a total soluble carbohydrate content of 311.1 mg glucose equivalents/g extract, corresponding to about 31 % w/w of the extract mass: Co-extracted carbohydrates therefore largely exceed the phenolic fraction. This is consistent with the literature on olive leaves, where microwave- and ultrasound-assisted extraction recover more phenolic compounds, and in considerably shorter times, than conventional procedures [4], and where the technique adopted governs which classes of constituents are preferentially recovered from pruning waste [5]. The systematic comparison of the three extraction techniques, in terms of yield and phenolic characterization, is currently in progress, and will be extended to the light stability and cell compatibility of the resulting extracts.
ATR-FTIR spectra of the MAE extract recorded before and after UVA irradiation at 24.3 J/cm^2 were substantially overlapping, suggesting that the arrangement of functional groups is preserved after exposure. The maceration extract caused no appreciable reduction in cell viability over the concentration range tested, indicating the absence of cytotoxic effects under these conditions.

Conclusion

Unseparated olive pruning residues yielded an extract in which the phenolic fraction is minor compared with the co-extracted soluble carbohydrates. The extract showed no detectable structural change after UVA exposure at the dose tested, and no appreciable reduction of HeLa cell viability over the concentration range examined. These preliminary results support a further characterization of the extracts and of the influence of the extraction technique on their composition.

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Flowing into dermocosmetics: microfluidic-assisted assembly of genipin-crosslinked elastin nanoparticles for ferulic acid delivery

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Introduction

In recent years, the cosmetic industry has shown a growing interest in natural active ingredients for skincare products. One of the most potent natural antioxidants is 4-hydroxy-3-methoxycinnamic acid (ferulic acid, FA). FA exhibits strong free radical scavenging activity, offering protective effects against aging, hyperpigmentation and inflammation. However, its application in cosmetic products is limited by its poor solubility and stability. FA is poorly soluble both in water (being soluble only at alkaline pH) and in oils or solvents suitable for cosmetic applications [1]. Furthermore, FA degrades easily (mainly via decarboxylation), losing its efficacy when exposed to high temperatures, high relative humidity, elevated pH, UV light, and various formulation excipients [2,3]. Therefore, the use of delivery systems that protect the molecule while enhancing its efficacy is of great interest for its practical application.

Aim of the work

Since stability remains a major challenge for the use of FA in cosmetic products, this study focuses on overvcoming this issue through the use of elastin-based protein nanocarriers. Among various approaches, protein nanoparticles stand out due to their biocompatibility, biodegradability, and ability to interact with the skin to deliver antioxidants, peptides, or anti-inflammatory agents with high efficacy and tolerability [4]. Literature predominantly reports the pharmaceutical application of nanoparticles based on recombinant elastin-like polypeptides (ELPs) or, rarely, tropoelastin, both of which are expensive and not readily available on the market. To date, no specific studies have investigated Elastin-derivated Peptides (EDCs), specifically soluble elastin combined with microfluidics, although results on similar proteins such as albumin, zein, and casein are highly promising [5,6].

Therefore, this study aims to launch a feasibility study for the microfluidic production of elastin nanoparticles (ElNPs), the development of which could lead to the application of FA-loaded NPs (FAElNPs) to the scalp. The use of droplet-based microfluidics offers precise control over fluid flows at the micrometer scale, enabling the fabrication of advanced micro- and nanoparticles with highly standardizable and scalable structures and compositions, thus facilitating a faster technology transfer to the cosmetic manufacturing sector.

Materials and Methods

Materials: Cosmetic-grade soluble elastin Matripure® (10-30 KDa) was supplied by Matrihealth GmbH, Germany. Ferulic acid and genipin were purchased from Fisher Scientific. All other chemicals and materials were purchased from Sigma-Aldrich.

Microfluidic set-up:

A syringe pump (IPS14, Inoverso) coupled with a custom 3D-printed toroidal flow microfluidic chip was employed to mix ethanol and an elastine aqueous solution to trigger elastin nucleation and subsequent ElNPs formation. The effect of different variables was expolored: protein concentration (1 and 5 mg/mL); antisolvent (ethanol or isopropanol); FRR (flow rate ratio) ethanol:protein solution (1:1, 2:1 3:1); TFR (total flow rate) 0,025; 0,5; 5 mL/min.

FA loading was achieved by using an ethanolic solution of FA (at 10% w/w relative to elastin) instead pure ethanol, yielding FaElNPs after mixing.

Both ElNPs and FAElNPs were cross-linked with genipin (2% or 5% w/w relative to elastin) by incubation in a thermoshaker at 37 °C and 100 rpm overnight. (Fig.1).

Nanoparticle characterization:

A comprehensive characterization of the NPs was carried out, analyzing: average size, homogeneity (polydispersity index, PDI), and surface charge (DLS; Zetasizer Nano ZS90, Malvern Instruments); morphology (SEM/TEM); variations in protein conformation (micro-Raman LabRAM HR Evolution, Horiba); and degree of cross-linking (FTIR, Bruker Vertex 70, Bruker). Colloidal stability was evaluated under different storage conditions (room temperature, 4 °C, -20 °C).

FA encapsulation was monitored by DSC (Perkin-Elmer Pyris 6 DSC over a temperature range of 30–300 °C) and XRPD. The FAElNPs were ultracentrifuged (16,000 rpm for 20 min), and the encapsulation efficiency was evaluated using UV-Vis plate reader (Varioskan Lux, ThermoFisher).

Results and discussion

The microfluidic formulation protocol for both unloaded (ElNPs) and FA-loaded elastin NPs (FAElNPs) was optimized in terms of instrumental and formulation parameters. In particular, the optimal conditions were found to be: elastin solution concentration 5 mg/mL; ethanol as the organic antisolvent phase; FRR) of 1:1; TFR 5 mL/min; and genipin concentration 5% w/w. Size, PDI, and zeta potential results were used as discriminants to determine the acceptability of the formulations. Among all the ElNPs formulations, only the 3 most promising were selected to prepare FAElNPs. However, FA loading further reduced the number of acceptable formulations, leaving only one with the desired characteristics (Tab.1). All formulations displayed a strong negative surface charge, with zeta potential values hovering around -30 mV, which decreased to approximately -15 mV for the FAElNPs. This highly negative charge suggests excellent colloidal stability due to electrostatic repulsion. Indeed, the unloaded, loaded, and cross-linked NPs remained stable when stored at 4°C for three weeks, showing no changes in physico-chemical characteristics.

Successful cross-linking was monitored colorimetrically by measuring absorbance at 590 nm, detecting the appearance of a blue color associated with the formation of conjugated adducts between genipin and tropoelastin. Spectroscopically (FTIR), bonding was confirmed by the appearance of peaks at 1100 cm^{-1} and 1033 cm^{-1}, attributed respectively to the C-N bond stretching and ether(C-O-C) + hydroxyl (C-O) groups characteristic of the genipin structure integrated into the network.

Crystalline ferulic acid (FA) exhibited a sharp endothermic peak at 173°C (corresponding to its melting temperature), which completely disappeared upon nanoencapsulation. These findings were further confirmed by PXRD analysis. Indeed, the diffraction pattern of pure ferulic acid showed sharp, characteristic crystalline peaks that disappeared when FA was encapsulated within the nanoparticles (NPs) [7].

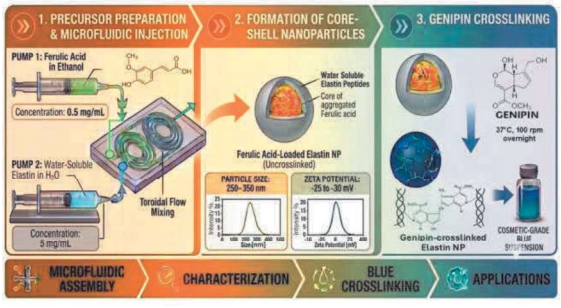


Fig. 1. Schematic of the microfluidic-based approach used for producing elastin-based NPs charging FA (FAElNPs).

Samples (FRR 1:1)	TFR (mL/min)	Z-averege size (nm)	PDI	ζ Potential (mV)
ElNPs1	0.025	240.7 ± 22.4	0.39 ± 0.04	-30.7 ± 1.19
ElNPs2	0.5	280.6 ± 19.2	0.33 ± 0.04	-32.9 ± 1.59
ElNPs3	5	274.4 ± 18.6	0.44 ± 0.01	-29.6 ± 1.66
FAElNPs	5	547,2 ± 20.1	0.50 ± 0.08	-15.5 ± 0.5
FAElNPs crosslinked	5	417.5 ± 6.8	0.40 ± 0.07	-29.3 ± 1.07

Tab.1.

Conclusion and perspectives

FAElNPs, will undergo further characterization to verify their encapsulation efficiency and in vitro effectiveness. Finally, FA release kinetics will be evaluated in physiological saline, simulated sweat at 32°C, and an oily medium composed of isopropyl myristate and isopropyl alcohol to simulate the lipid environment of the hair follicle.

Thanks to the favorable physico-chemical characteristics of these formulations. the potential high throughput production and the precise control offered by the microfluidic technology, the method described in this study for generating protein-based ElNPs and FAElNPs holds great potential for the topical delivery of ferulic acid.

Acknowledgments

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LIPOAMINO ACID ALKYL ESTERS NOT ONLY *SPF BOOSTERS*

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Introduction

It is of great importance to characterize cosmetic raw materials in terms of their complete range of properties, beyond their primary function within a formulation. This highlights the importance of raw material multifunctionality, which allows a single ingredient to contribute to multiple formulation and to skin-related functions.

Lipoamino acids are obtained through the acylation of amino acids with fatty acids. They are amphiphilic molecules exhibiting surfactant properties, good skin affinity, and are of a renewable origin[1] The fatty acid moieties may contain C_4–C_{16} carbon chains, with C_{12} derivatives being predominant, particularly lauroyl and cocoyl derivatives. The main derivatives include glycinates, sarcosinates, alaninates, and glutamates, which are characterized by biodegradability, low toxicity, and cleansing, emulsifying, and foaming properties[1].

Depending on their composition, lipoamino acids may also exhibit anti-ageing, purifying, soothing, and skin-brightening properties. This class includes *Capryloyl Glycine*, which shows antimicrobial and dermopurifying properties; *Dipalmitoyl Hydroxyproline*, known for its antioxidant and anti-wrinkle property; *Undecylenoyl Phenylalanine*, which possesses depigmenting properties and *Cocoyl Proline*, which can act as a natural solubilizing agent.

Aim of the work

Based on previous findings that identified LIPOAMINO ACID ALKYL ESTERS as *SPF booster*, the present study further investigated the mechanisms underlying their booster effect, with particular emphasis on their UV absorption properties and skin affinity, as well as their potential additional functionalities, including the inhibition of oxidative stress and re-epithelialization activity.

Materials and Methods

Materials: Lipoamino acid alkyl esters were synthesized from renewable raw materials, namely olive and hemp oils, and glycine ethyl ester, using a novel green synthesis method based on microwave irradiation (Olive lipoamino acid – OLA; Hemp lipoamino acid – HLA; Olive Oil – OO; Hemp Oil – HO).

To evaluate their spectrophotometric properties, the following UV filters were employed: UVB filters (*Ethylhexyl Methoxycinnamate* - EHMC*, Octocrylene* - OCR and *Ethylhexyl Salicylate* - EHS*)*, UVA filters (*Butyl Methoxydibenzoylmethane*- BMDB and *Diethylamino Hydroxybenzoyl Benzoate* - DHBB*)*, and UVA-UVB filter (*Bis- Ethylhexyloxyphenol Methoxyphenyl Triazine* - BEMT*)*.

An oil-in-water (O/W) emulsion containing either EHMC or BMDM was prepared to evaluate the spectrophotometric properties and photostability of the formulations.

Spectrophotometric analysis*:* The measurements were carried out in solution (ethanol or THF/H_2O (9:1)) using cuvettes with an optical path of 1 cm; on concentrated samples and in emulsion using cuvettes with an optical path of 0.01 mm (thin layer analysis).

Photostability test: The irradiation was performed using a solar simulator with a Xenon arc lamp. The samples were irradiated at 20 MED (600 mJ/cm^2).

In vitro SPF evaluation: The SPF value was determined using a Labsphere UV-2000S Ultraviolet Transmittance Analyzer spectrophotometer, PMMA plates were used as substrate.

Skin affinity evaluation: The test was performed by measuring the contact angle formed between an artificial hydrolipidic film and a drop of sample. An optical contact angle instrument was used applying the sessile drop method[2,3].

Inhibition of oxidative stress: Immortalized human keratinocyte (HaCaT) and human fibroblast (Human Foreskin Fibroblasts, HHF) cell lines were employed to perform this test. The effectiveness of the samples tested (OLA, HLA, OO, HO) to reduce oxidative stress was carried out by stimulating the cells with H_2O_2. The intracellular ROS measurement was assessed by determining the fluorescent adduct, dichlorofluorescein (DCF). The fluorescence signal intensity of DCF is proportional to the amount of ROS produced by the cells.

Scratch wound healing assay: The *in vitro* assessment of re-epithelialization activity was performed according to protocols reported in the literature[4], with appropriate adaptations. The method involves the simulation of a wound in a cellular monolayer by means of a cross-shaped incision, allowing the ability of the cells to repair and close the lesion. The assay was performed using HaCaT cells.

Results and discussion

Evaluation of Spectrophotometric Properties

The spectrophotometric properties were evaluated on the lipoamino acid derivatives (OLA and HLA) and reference oils alone and in the presence of the selected UV filters.

The results showed that the lipoamino acid alkyl esters exhibit a λ_{max} of 270 nm in ethanol and 275 nm in THF/H_2O (9:1), together with higher absorbance values than those observed for the reference oils. These findings highlight that the two SPF boosters do not have UV radiation filtering properties.

In mixtures containing UVB filters, the results showed that the λ_{max} of the filter remains unchanged, whereas absorbance increases in the presence of both lipoamino acid alkyl esters compared with the corresponding mixtures containing the reference oils. The increase in absorbance depends on the specific filter/booster combination. The boosters do not affect the λ_{max} of the UVA filters and present only a limited effect on their absorbance. In the case of the broad-spectrum UVB–UVA filter, both the addition of the two boosters and the reference oils determine a bathochromic shift on the two absorption maxima. In this case, both boosters produce an increase in absorbance compared with the reference oils. Similar results were also obtained through thin-layer analysis.

The emulsions containing both the UVB and UVA filter show a bathochromic shift of the wavelength at the absorption maximum for all tested samples. This is attributable to the influence of the vehicle itself.

In both cases, the addition of the two boosters results in an increase in the absorbance value.

In vitro SPF evaluation confirmed the previously obtained results. An increase of 9/10 points or 3 points was observed in the presence of the two boosters or reference oils respectively.

Evaluation of Photostability

The possible photo-stabilizing properties of the two boosters were evaluated for the EHMC and BMDM filters, known for their photo-instability.

Photostability to irradiation was assessed as the reduction of both the filter absorbance value and the *in vitro* SPF value in the presence of the boosters or reference oils.

For mixtures containing EHMC or BMDM with the lipoamino acid alkyl esters, a lower decrease in absorbance value was observed after irradiation.

The results obtained for the emulsions show a lower loss of absorbance value in the presence of both lipoamino acid derivatives for the UVB filter in comparison with the UVA.

The same trend was observed for the *in vitro* SPF value. After irradiation, a smaller variation was obtained in the presence of the two lipoamino acid derivatives.

These findings highlight an enhancement of the photostability of both UV filters.

Evaluation of Skin Affinity

The results obtained show a lower contact angle for the lipoamino acid derivatives compared to the reference oils, highlighting a greater affinity for the substrate and a greater ability to spread across its surface.

Inhibition of Oxidative Stress

Regarding keratinocytes, no significant effect on the reduction of reactive oxygen species (ROS) production was observed following stimulation with the oxidative agent. However, a slight difference was noted between hemp oil and its corresponding lipoamino acid derivative, with the latter showing a greater ability to inhibit ROS production than the oil used alone.

The most relevant findings were observed in fibroblasts. Treatment with the lipoamino acid mixtures appeared to reduce ROS production, as in both cases ROS levels were lower than those measured in the positive control.

Evaluation of Re-epithelialization Capacity

The results obtained after 24 h of treatment show an improvement in the re-epithelialization process of the simulated in vitro wound. This effect appears to be more pronounced in the lipoamino acid derivatives, suggesting their potential beneficial role in promoting wound closure and tissue regeneration.

Conclusion

The work carried out, partly focused on elucidating the mechanism underlying the booster effect, demonstrated that several factors contribute to the increase in SPF values. These include the lipophilic nature and emulsifying properties of the compounds, which enhance the solubility and dispersion of UV filters, as well as changes in their spectrophotometric properties, such as increased absorbance and the synergistic effect observed in filter/booster mixtures. Furthermore, the formation of systems capable of inducing UV radiation scattering may also be hypothesized.

The study further demonstrated that the lipoamino acid alkyl esters synthesized and initially investigated as SPF boosters possess additional promising properties, including the inhibition of oxidative stress and re-epithelialization capacity.

In conclusion, the results obtained demonstrate that the two boosters have strong potential as novel cosmetic ingredients, as they meet several key requirements of the current cosmetic market, including natural origin, sustainability, and multifunctionality.

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Exploring cosmetic potential of *Spartium junceum* L. pods: extraction, phyto-chemical characterization, ethosomal encapsulation, and biological activity

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Introduction

Annually, the agro-food industry produces 1.3 billion tons of plant-based byproducts including skins, peels, seeds, leaves, and other inedible components [1]. The valorization of aforementioned underexplored plant materials has gained increasing attention as a promising strategy for the development of more sustainable cosmetic ingredients. Among these plant resources, *Spartium junceum* L., commonly known as Spanish broom, has been long recognized as a source of bioactive compounds, especially derived from its yellow flowers [2]; however, the potential of its pods remains poorly explored.

Aim of the work

The aim of the work was to evaluate *S. junceum* L. pods as a source of extract with cosmetic interest and to incorporate it into ethosomal nanocarriers for topical application.

Materials and Methods

Materials: *Spartium junceum* L. pods were collected in September 2024 in Cerchiara di Cal., Cosenza, Italy. After collection, the dried plant material was stored at room temperature in darkness until further processing. Ethanol, methanol, L-α-phosphatidylcholine, sodium carbonate, gallic acid, quercetin, DPPH and ascorbic acid were purchased from Sigma-Aldrich, Milan, Italy. Folin–Ciocalteu reagent and aluminum chloride were purchased from Titolchimica, Pontecchio Polesine, Rovigo, Italy.

Preparation of Spartium junceum L. pods extract:
Pods powder (0.5 g) was dispersed in 50 mL of hydroalcoholic (50:50 *v/v*) solution and macerated for 24 h. After extraction, the macerated was filtered and ethanol was removed using a rotary evaporator (Fig.1). The extract was lyophilized for 48 h using a freeze-dryer (Christ Freeze Dryer ALPHA 1–2, Milan, Italy). Successively, it was stored at 4-8 °C and protected from light until use.



Fig.1 Representative images of Spartium junceum L. pods: (A) intact pods containing seeds, (B) powder obtained after grinding, and (C) hydroalcoholic extract.

Total Phenolic Content (TPC), Total Flavonoid Content (TFC), and Antioxidant Activity (AA%):
The TPC of the extract was determined using the Folin–Ciocalteu method. TPC values were expressed as gallic acid equivalent in mg/g extract (mg GAE/g extract).
The TFC of the extract was determined using the aluminum chloride colorimetric method. TFC values were expressed as mg of quercetin equivalents per gram of extract (mg QE/g extract).
AA% of the extract was evaluated using the DPPH scavenging assay. Results were expressed as percentage of DPPH radical inhibition and calculated according to the following equation: Inhibition (%) = $[(ABS_{control} - ABS_{sample}) / ABS_{control}] \times 100$
where $ABS_{control}$ is the absorbance of the DPPH control and ABS_{sample} is the absorbance of the sample.
All measurements were performed in triplicate.

Antimicrobial activity of pods extract:
The pods extract was tested by well diffusion assay (WDA) against *Staphylococcus aureus* DSM 799, *Escherichia coli* ATCC 10536, *Escherichia coli* NCTC 10538, *Pseudomonas aeruginosa* ATCC 15442, *Enterococcus hirae* ATCC 10541, *Streptococcus pyogenes* ATCC 12344, *Candida albicans* ATCC 10231 and *Aspergillus brasiliensis* ATCC 16404.

Encapsulation of the extract into ethosomes:
Ethosomes were prepared using the ethanol injection method. For the unloaded ethosomal formulation, 75 mg of L-α-phosphatidylcholine were dissolved in 3 mL of ethanol under magnetic stirring to obtain the organic phase. This phase was added dropwise to 7 mL of phosphate-buffered saline (PBS, pH 7.4) under magnetic stirring at 600 rpm. The final dispersion was kept under stirring for 20 min, protected from light, to allow stabilization of the ethosomal system.

Extract-loaded ethosomes were prepared using the same procedure, by dispersing the extract in the PBS phase before addition of the organic phase.

Vesicles characterization:
Vesicle size and PDI were determined by dynamic light scattering (DLS) using a Brookhaven 90-PLUS particle size analyzer (Brookhaven Instruments, New York, USA). Zeta potential was measured by electrophoretic light scattering using a Malvern Zetasizer 3000 HS (Malvern Instruments Ltd., Malvern, UK). The encapsulation efficiency (EE%) was determined using a dialysis method followed by the DPPH radical scavenging assay. The assay was performed in triplicate.

In vitro release studies:
In vitro release studies were performed using Franz-type static glass diffusion cell. A membrane (Spectra/POR, 4 dialysis membrane MWCO 12-14kDa, Cantype, Rancho Dominguez, CA) was placed between the donor and receptor compartments. The receptor compartment was filled with a PBS mixture (70:30, v/v), used as receptor medium, and maintained at 32.0±1.0 °C under stirring throughout the experiment. The donor compartment was loaded with 1 mL of the sample. At predetermined time, aliquots were withdrawn from the receptor compartment and replaced with fresh medium to maintain a constant volume. The collected samples were analyzed by determining TPC.

Ethosomes biocompatibility:
Cell viability was evaluated on WS1 human dermal fibroblasts using the MTT assay.

Results and discussion

In this study, hydroalcoholic *Spartium junceum L.* pod extract was successfully prepared. The TPC and TFC results were 123.55± 5.7 mg GAE/g extract and 9.05 ± 0.5 mg QE/g extract, respectively. DPPH assay showed a prominent antioxidant activity (AA%: 90.85 ± 3.65%) of the prepared extract.

At the concentration tested, the pods extract showed a low antimicrobial activity (0.5-1.0 mm of inhibition halo) on *Staphylococcus aureus* DSM 799, *Escherichia coli* ATCC 10536, *Escherichia coli* NCTC 10538, *Pseudomonas aeruginosa* ATCC 15442 and *Enterococcus hirae* ATCC 10541.

The extract was encapsulated in ethosomes to avoid oxidation and to preserve biological activities for cosmetic use.

Loaded ethosomes showed nanometric size (151.7 ± 7.8 nm), a low polydispersity index (0.178 ± 0.016) and Zeta potential values around -24.51±6.49 mV. EE% based on DPPH radical scavenging activity was 79.5±0.6%.

The release of polyphenols from ethosomes and the free extract solution (1 mg/mL) over time was shown in Figure 2.

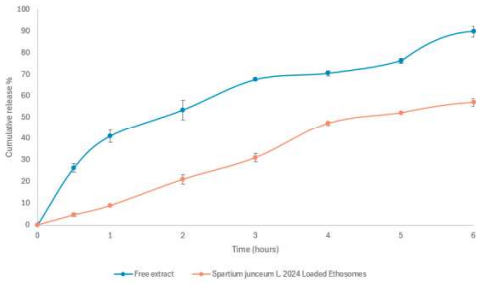


Fig. 2. In vitro release of TPC from hydroalcoholic extract and loaded ethosomes.

The hydroalcoholic extract showed a faster release (89.7 ± 1.2% of TPC released after 6 h) than the extract-loaded ethosomes (74.0 ± 0.4 % after 6 h). This behaviour underlines the capacity of the lipid vesicles to modulate the availability of the phenolic components at the application site.

The biocompatibility of ethosomes was tested on WS1 fibroblasts which are some of the most representative cells of skin tissue and the target of the topical formulation. The MTT assay confirms that WS1 viability is not affected by the extract free or loaded in the ethosomes in the range of concentrations 0.78 to 100 µg/mL.

Conclusions

The results showed that *Spartium junceum* L. pods and its hydroalcoholic extract are a promising source for antioxidant activity in cosmetic products. Future studies should be performed on its chemical composition.

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Cosmetic testers as potential vehicles for the spread of antibiotic-resistant microorganisms

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Introduction

Cosmetic testers represent cosmetics that are frequently handled and used by multiple individuals. They may provide favorable surfaces for the persistence and transmission of microorganisms potentially relevant to consumers' health. There is a lack of research that focuses on microbial resistance in isolated strains coming from makeup testers [1,2].

Aim of the work

This study aimed to assess the microbial contamination of cosmetic testers collected from several perfumeries in the province of Pescara. The antimicrobial susceptibility profiles and biofilm-forming abilities of the isolated microorganisms were also evaluated, as biofilm formation may enhance microbial persistence, antimicrobial tolerance, and transmission among users.

Materials and Methods

Sample collection: 54 cosmetic testers were collected from several anonymized perfumeries in the province of Pescara between February and May 2026.
***Bacterial strain isolation and identification*:** The samples, representing various cosmetic categories, underwent microbiological examination involving culture, morphological observation, Gram staining, and biochemical tests. For each cosmetic tester, direct plating was performed by spreading approximately 20 mg of the product over the entire surface of the culture media. Tryptic Soy Agar (TSA), and Mannitol Salt Agar (MSA), a selective and differential medium for staphylococci, were used.
The plates were incubated aerobically at 37 °C for 48–72 h. The resulting colonies were subsequently subcultured for further identification and antimicrobial susceptibility testing. Bacterial identification was performed in collaboration with the Microbiology Laboratory headed by Prof. Fazii at the Pescara hospital, using the automated VITEK® 2 system (bioMérieux).

Antimicrobial susceptibility test and Biofilm forming assay: The most representative isolates were analyzed for antibiotic susceptibility using the Kirby-Bauer method. Biofilm formation capacity was assessed via a polystyrene microplate assay with safranin staining method [3].

Results and discussion

From cosmetics testers were identified microorganisms belonging primarily to skin and environment, including *Micrococcus luteus, Kocuria rhizophila, Lysinibacillus sphaericus, Peribacillus simplex, Peribacillus muralis,* and *Bacillus subtilis*. Microorganisms of health sanitary significance—such as *Staphylococcus spp., Acinetobacter ursingii,* and *Roseomonas mucosa*—were also isolated. Susceptibility profiles revealed significant variability: alongside susceptible isolates, instances of resistance and multidrug-resistance profiles were observed, particularly among isolates belonging to the *Staphylococcus* genus. Phenotypic assessment further demonstrated the ability of certain microorganisms to adhere to surfaces and form biofilms.

The results confirm that cosmetic testers may serve as potential fomites and highlight the need for standardized sanitization procedures, periodic microbiological monitoring, and increased consumer awareness.

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Valorizing Tunisian Grape Pomace Polyphenols in Cosmetics: From Particle-Size-Driven Extraction to Liposomal Nanoencapsulation and Cream Formulation

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Introduction

Grape pomace, an abundant agro-industrial by-product of wine production, is a rich yet underexploited source of bioactive polyphenols with antioxidant potential [1]. Its valorization in cosmetic formulations is of growing interest, but extraction efficiency and downstream formulation performance remain strongly dependent on the physical properties of the starting material and on the extraction and encapsulation strategy employed [2].

Aim of the work

The aim of this work was to develop a full valorization pathway for Tunisian grape pomace polyphenols for cosmetic application: from particle-size characterization and extraction optimization, through liposomal nanoencapsulation, to incorporation into a finished cream formulation and evaluation of its physicochemical and rheological performance.

Materials and Methods

Plant material and extraction: Grape pomace (Syrah/Carignan, Tunisia) was dried, milled and sieved. Polyphenols were extracted by maceration or ultrasound-assisted sonication in water, 70% ethanol, or absolute ethanol (n = 3), followed by freeze-drying.

Characterization: Particle-size distribution was determined by laser diffraction (Mastersizer, Hydro EV, Mie model) and digital microscopy (20×–100×, 2D/3D). Total phenolic content (TPC) was measured by the Folin–Ciocalteu assay and antioxidant capacity by ABTS, FRAP and DPPH assays.

Liposome preparation: The optimized extract was encapsulated into liposomal nanocarriers by microfluidics (55:45 phospholipid:cholesterol; flow rate ratio 3:1; total flow rate 3000 µL/min; Sunny Trident T490), and characterized by dynamic light scattering (size, PDI, zeta potential) and encapsulation efficiency.

Cream formulation: The liposomal dispersion, the free extract, and an unloaded control were incorporated into an eye-contour cream base. The resulting formulations were characterized by laser diffraction and rotational/oscillatory rheology (flow curves, amplitude sweep, 25 °C).

Results and discussion

The powder showed a broad, polydisperse particle-size distribution (Dv50 = 305 µm), consistent with heterogeneous, partly aggregated particles. Extraction yield was highest for maceration in absolute ethanol (14.3%) but dropped sharply in water (7.15%), whereas sonication gave a more stable yield (10.5–11.8%) and improved water-based extraction by ~62%, consistent with ultrasound-induced disruption of particle aggregates. TPC peaked under maceration with 70% ethanol (3489 µmol GAE/g), while antioxidant capacity did not consistently track TPC: ABTS correlated with TPC, whereas DPPH and FRAP were unexpectedly higher in sonicated extracts despite lower phenolic content, pointing to assay-dependent mechanisms.

Empty liposomes showed a stable hydrodynamic diameter of 144 ± 1 nm (PDI 0.12 ± 0.04; zeta potential −25 ± 1 mV), whereas extract-loaded liposomes displayed a markedly larger diameter (2740 ± 400 nm, PDI 0.73 ± 0.16; zeta potential −32 ± 2 mV), with 10% encapsulation efficiency.

In the cream formulations, laser diffraction showed the extract-loaded cream had the most homogeneous, fine particle distribution (Dx90 = 157.8 µm) versus the base (298.8 µm), while the liposome-loaded cream

displayed a coarser, bimodal distribution (Dx90 = 435.0 µm), consistent with vesicle aggregation. Rheological flow curves confirmed shear-thinning behavior for all formulations, with the extract-loaded cream showing the highest viscosity across the shear-rate range — indicating a more structured, cohesive network — and the liposome-loaded cream the lowest, reflecting partial network disruption by the vesicles despite their greater potential for skin delivery.

Conclusion

Tunisian grape pomace is a structurally heterogeneous, polyphenol-rich by-product whose extraction and antioxidant performance are strongly technique- and solvent-dependent. Its liposomal encapsulation and incorporation into a cosmetic cream are feasible, with the extract-loaded formulation offering the best physical stability and the liposomal formulation offering enhanced potential for skin delivery, pending further optimization of encapsulation efficiency and particle homogeneity.

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Microfluidic-Engineered Liposomes as topical carriers for Bee-Pollen bioactives

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Introduction

Bee pollen (BP) is a natural, inexpensive, and environmentally sustainable bioresource, characterized by a complex phytochemical profile rich in proteins, vitamins, minerals and, notably, phenolic compounds, key contributors to its documented antioxidant and health-promoting properties [1,2].

Despite this promising profile, phenolic-rich natural extracts—including those derived from bee pollen—present intrinsic limitations such as poor aqueous solubility, chemical instability, low bioavailability, and restricted permeation across biological barriers. These features collectively hinder their effective topical delivery and reduce their ability to penetrate and be retained within skin tissues [3-5]. In this context, nanocarrier-based drug-delivery systems have emerged as effective approaches to overcome these limitations. Among them, liposomes represent one of the most established platforms for improving dermal delivery of natural compounds [4,5]. Recent advances in microfluidic technology allow highly controlled and reproducible mixing, yielding liposomes with narrow size distribution, enhanced encapsulation performance, improved batch-to-batch consistency, and scalability toward industrial processes [6,7]. These advantages make microfluidic- engineered liposomes a particularly promising approach for the incorporation of complex natural extracts and for the development of skin-oriented delivery systems with improved functional performance.

Aim

This study aimed to develop and characterize microfluidic-engineered liposomal formulations for the incorporation of bee-pollen derived phenolic extracts designed to overcome their intrinsic limitations and enhancing their suitability for topical applications.

Materials and Methods

Bee-pollen extract preparation and characterization

Ultrasound-assisted extraction (UAE) was performed on multifloral bee pollen waste using 70° ethyl alcohol, varying sonication time (30, 60 and 90 min). Extraction yield was calculated gravimetrically by relating the mass of the obtained dried extract to the initial mass of raw bee pollen material. UV–Vis spectra (200–600 nm) were recorded, and antioxidant properties were determined using the Folin–Ciocalteu total phenolic content (TPC) assay and the DPPH radical-scavenging assay. Based on extraction efficiency and phenolic content, the 30 min extract (BP 30′) was selected for biological and formulation studies.

Biological studies

For the biological evaluation, cytocompatibility was first assessed on normal human dermal fibroblasts (NHDFs) which were exposed to BP 30′ in the concentration range 0.50–0.03 mg/mL. Cell metabolic activity was quantified by MTT assay after 24 and 48 h.

Liposomes preparation and characterization

Microfluidic liposomes were produced using the Sunshine system. The organic phase consisted of L-α-phosphatidylcholine/cholesterol (55:45 mol/mol) in absolute ethanol (≥ 99.8%), while the aqueous phase was either ultrapure water (blank liposomes), or BP 30′ solution at 20 mg/mL for loaded formulations. Liposome assembly was carried out using a Sunny Trident T-490 microchip under a fixed total flow rate (TFR) of 3000 µL/min, while exploring two water:organic flow rate ratios (FRR = 3:1 and 5:1). This yielded four formulations: E3 and E5 (empty liposomes at FRR 3:1 and 5:1) and P3 and P5 (BP-loaded liposomes at the same FRR values).
Following production, the dispersions were purified by dialysis (MWCO 10 kDa) to remove non-encapsulated extract. Entrapment efficiency (EE) was indirectly determined by Folin–Ciocalteu quantification of the non-encapsulated phenolic fraction.
Hydrodynamic diameter (Z-average), polydispersity index (PDI) and ζ-potential were measured by dynamic and electrophoretic light scattering at 25 °C. Based on colloidal characterization, P3 was selected for subsequent studies, lyophilized and reconstituted before use. Entrapment efficiency (EE) was indirectly determined by Folin–Ciocalteu quantification of the non-encapsulated phenolic fraction.

In vitro release study

In vitro release was evaluated over 24 h using automated Franz diffusion cells (Vision® Microette) equipped with synthetic polycarbonate membranes (0.50 µm; diffusion area 1.77 cm^2). PBS containing 0.5% Tween 80 was used as receptor medium at 37 °C. Reconstituted BP-loaded liposomes were compared with free BP extract. Rutin was used as a marker compound and quantified by HPLC-DAD.

Ex vivo skin permeation study

Ex vivo skin permeation was evaluated over 24 h using Franz diffusion cells and porcine dorsal skin (diffusion area 1.86 cm^2), with PBS containing 5% BSA as receptor medium at 37 °C. BP-LPs were compared with free BP extract, using rutin as a marker compound quantified by HPLC-DAD. After 24 h, rutin distribution was evaluated in skin-surface washing, stratum corneum, epidermis, dermis and receptor fluid.

Results and discussion

Ultrasound-assisted extraction of bee pollen waste yielded three extracts (BP 30′, BP 60′ and BP 90′), whose extraction efficiency decreased progressively with increasing sonication time. In all samples, UV–Vis analysis revealed a characteristic absorbance band between 300 and 320 nm attributable to phenolic chromophores; among them, BP 30′ displayed the highest absorbance intensity, consistent with a greater abundance of phenolic constituents. This trend was confirmed by the Folin–Ciocalteu assay, where BP 30′ exhibited the highest total phenolic content, markedly surpassing BP 60′ and BP 90′. DPPH analysis supported these findings, as BP 30′ showed the strongest radical-scavenging activity. Based on this combined spectrophotometric profile, BP 30′ was identified as the most phenolic-rich extract and selected for subsequent evaluations.

The cytocompatibility assessment on NHDFs cells demonstrated that BP 30′ was well tolerated at concentrations ≤0.125 mg/mL, with cell viability remaining ≥80% at both 24 and 48 h. At higher concentrations, a dose-dependent reduction in metabolic activity was observed.

Microfluidic liposomes produced at a TFR of 3000 µL/min displayed excellent colloidal properties across both flow-rate ratios tested (Table 1).

Batch	FRR	TFR (µL/min)	Z-Average (nm)	PDI	ζ (mV)
E3	3:1	3000	150 ± 4	0.14 ± 0.02	-35 ± 1
E5	5:1	3000	145 ± 2	0.12 ± 0.03	-37 ± 1
P3	3:1	3000	174 ± 2	0.14 ±0.01	-42 ± 1
P5	5:1	3000	164 ± 1	0.17 ±0.01	-40 ± 1

Table 1. Colloidal characterization of empty and encapsulated liposomes.

Blank liposomes displayed nanometric size (≈150 nm for E3; ≈145 nm for E5), low polydispersity (PDI ≤0.15) and highly negative ζ-potential values (≈ –35/–37 mV), indicative of good colloidal stability. Incorporation of BP 30′ in P3 and P5 slightly increased vesicle size while maintaining narrow size distribution (PDI ≈0.14–0.17) and further enhancing ζ- potential negativity (up to –42 mV for P3), confirming that the extract did not destabilize the lipid bilayer.

Based on the overall colloidal characterization, P3 was selected for subsequent studies. Following lyophilization and reconstitution, P3 maintained a nanometric size (125 ± 2 nm), a narrow size distribution (PDI 0.17 ± 0.02) and a negative ζ-potential (–31 ± 1 mV). The entrapment efficiency of the phenolic fraction, indirectly determined by Folin–Ciocalteu assay, was 15%.

Both BP-LPs and free BP extract released rutin into the receptor medium over 24 h. The free BP extract showed a faster initial release and reached a higher maximum percentage of released rutin (approximately 73%), whereas BP-LPs exhibited a more gradual initial release profile, reaching a maximum of approximately 52%. Tmax was 3 h for both samples. BP-LPs showed a Cmax of 4.419 µg/mL and an AUC of 22.816, compared with 2.825 µg/mL and 13.490, respectively, for free BP extract.

In the ex vivo permeation study, after 24 h rutin was predominantly recovered in the skin-surface washing fractions, accounting for a mean of 77.3% of the applied dose for BP-LPs and 63.0% for free BP extract. Rutin levels in the stratum corneum were below the quantification limit, while rutin was below the detection limit in the epidermis, dermis and receptor fluid.

Conclusion

This study demonstrated the feasibility of incorporating phenolic-rich bee-pollen extracts into microfluidic-engineered liposomes, combining the favorable antioxidant and cytocompatibility profile of the selected BP extract with the controlled physicochemical properties of the developed nanosystems. Microfluidic processing enabled the production of nanosized, relatively homogeneous and negatively charged vesicles, while the selected BP-loaded formulation showed an encapsulation efficiency of 15%. In vitro release studies revealed a distinct release behavior compared with the free extract, with BP-LPs showing a more gradual initial release of rutin. However, ex vivo studies on porcine dorsal skin showed that rutin remained predominantly at the skin surface, with negligible deposition in the stratum corneum and no detectable penetration into the epidermis, dermis or receptor fluid. Overall, these findings support microfluidic liposomes as a reproducible platform for the incorporation and modulation of the release of bee-pollen-derived bioactives, while highlighting the need for further formulation optimization to enhance their cutaneous delivery.

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Physicochemical Characterisation and In Vivo EEG Evaluation of a Lavandula Essential Oil Cream

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Introduction

Lavender essential oil is well known for its effects on emotional state and brain electrical activity following inhalation [1]. Its incorporation into a topical cosmetic formulation warrants a distinct evaluation, because the experience of application combines olfactory stimulation, cutaneous sensation and expectancy. Neurophysiological research in cosmetics highlights the value of combining objective measurements with self-report, while identifying substantial methodological heterogeneity [2]. EEG studies of cream application further support this combined approach, although neural measures should not be equated directly with emotional benefit [3].

Aim of the work

To characterise the physicochemical stability of an oil-in-water cream containing 1% Lavandula essential oil and investigate responses to its application through an ongoing *in vivo* study combining electroencephalography (EEG), pupillometry and self-reported sensory and emotional assessments, using the base cream as a comparator.

Materials and Methods

Formulation and physicochemical characterisation. A cream containing 1% Lavandula essential oil and its base vehicle were evaluated using the same characterisation framework. Measurements included pH, density and conductivity, droplet-size distribution by laser diffraction (Mastersizer 3000+, Malvern Panalytical), optical and polarised-light microscopy, and rheological assessment using a HAAKE MARS iQ rheometer (Thermo Scientific, Thermo Fisher Scientific). Physical stability was investigated by centrifugation, freeze-thaw cycling, thermal stress and monitoring during storage.

***In vivo* study design.** The ongoing study uses a within-participant design in which both formulations are assessed during a single session, with application order counterbalanced across participants. Before the visit, volunteers provide informed consent and complete screening for cosmetic or lavender allergy, skin conditions, medication affecting the nervous system and impaired olfaction. Cosmetic habits, fragrance preferences and familiarity with lavender are also recorded. Baseline questionnaires assess state and trait anxiety, depressive symptoms, affect, rumination and worry.

EEG and pupillometry protocol. Following placement of the 20-channel EEG system and calibration of pupillometry, a five-minute resting baseline is recorded. The first formulation is applied for one minute, followed by ten minutes of post-application recording. Participants then complete product pleasantness and arousal ratings and the six-item State-Trait Anxiety Inventory (STAI-6). After product removal, the second formulation is applied using the same application, recording and questionnaire sequence. The session concludes with product removal, equipment removal and debriefing.

Subjective assessment. Product liking and skin pleasantness are assessed on five-point scales. Emotional valence and arousal associated with odour, visual appearance and post-application sensation are rated on nine-point scales. Additional questions address perceived well-being during the application ritual, discomfort and willingness to use the product again. The combined recordings and ratings are intended to support within-participant comparisons of responses to the two formulations.

Results and discussion

Physicochemical and stability testing indicated good physicochemical and microbial formulation stability, supporting progression to *in vivo* evaluation. Laser diffraction analysis showed a narrow, unimodal droplet-size distribution for the base cream (5–30 µm) and a broader, more polydisperse distribution for the Lavandula-containing cream (10–80 µm). Rheological assessment at 25 and 35 °C showed shear-thinning behaviour in both formulations, with the Lavandula-containing cream exhibiting slightly lower viscosity than the base cream. EEG, pupillometry and questionnaire assessment is ongoing; neurophysiological and emotional outcomes are not yet available. The protocol integrates formulation characterisation with objective recordings and subjective experience. Differences between formulations will require interpretation in relation to fragrance perception, application order and possible carryover, since the base vehicle does not provide an odour-matched control. Previous studies of cosmetic textures and skincare routines further highlight the need to account for sensory properties and the application experience when interpreting product-related responses [4,5].

Conclusion

The Lavandula-containing cream provides a stable formulation for investigating responses to topical cosmetic application. The ongoing multimodal study will examine whether product-related differences in EEG and pupillometry correspond to reported sensory and emotional experience. Conclusions concerning relaxation or other neurocosmetic benefits await completion and analysis of the in vivo study.

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Formulating with *Opuntia ficus-indica* for acne-prone skin: extract characterization, face gel formulation and evaluation.

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Introduction

Acne vulgaris is a common skin pathology among all age groups and particularly in adolescents and young adults. It affects the pilo-sebaceous unit and is characterized by an increased sebaceous gland activity, follicular hyperkeratinization, imbalances of cutaneous microbiome and the onset of inflammation and oxidative stress[1]. Clinically similar conditions, such as *Malassezia* folliculitis often caused by an imbalance in the skin's microbiome may be misdiagnosed as *acne vulgaris*[2], leading to an inappropriate treatment. Cosmetics are being increasingly recognized as a valuable adjunctive tool to reduce the side effects of acne therapy and improve patient compliance [3,4]. At the same time, growing public demand for sustainable ingredients and formulations is driving research toward renewable and green sources, in line with circular bio-economy principles. By this perspective, *Opuntia ficus-indica* (OFI) cladodes represent an abundant and underutilized mediterranean biomass, characterized by a high phenolic content, with antioxidant and anti-inflammatory activity, which deserves to be deepened and exploited [5,6,7].

Aim of the work

Against this background, the aim of the present study was to obtain and characterize a face gel formulation, suitable for acne-prone skin, incorporating an optimized extract from OFI cladodes.

Materials and Methods

Dried cladodes were submitted to hydroalcoholic extraction using a 30:70 water:ethanol solution under different temperature and time conditions. HPLC-DAD analysis was performed to evaluate the phenolic profile of each extract, while the antioxidant activity was assessed *via* DPPH, ABTS and FRAP assays. To select the most promising extracts, a Principal Component Analysis (PCA), Box Ranking, and Response Surface Methodology (RSM) guided evaluation was conducted. Antimicrobial activity was tested against acne-related microorganisms, such as *Staphylococcus epidermidis* and *Malassezia* clinical isolates and reference strains using broth microdilution method. A selected hydroalcoholic extract was used to develop cosmetic gel formulations for acne-prone skin, testing different polymers as rheology modifiers: xanthan gum (XG), Actigum™ CS 11 (AG) from Caregill, and high molecular weight hyaluronic acid (HA). Each formula also comprised technical ingredients, including a chelating agent, a humectant, and a preservative, other than skin-beneficial ingredients as moisturizing and soothing agents. These formulations were subjected to rheological analysis (*Amplitude sweep*, *Frequency sweep, Flow sweep*) to evaluate their properties. For each formula, the control sample, polysaccharide gel containing all the ingredients except the extract, was also prepared.

Results and discussion

HPLC-DAD analysis identified piscidic acid (21-44 mg/g dry extract) and rutin derivatives (6-11 mg/g dry extract) as characteristic compounds, with optimal recovery at 40–60°C for 6–24 h. DPPH and ABTS assays reported an antiradical activity of 0.20-1 mg GAE/g DE and 60-150 mg TROLOX/g DE respectively and a TPC of 20-60 mg GAE/g DE, while an antioxidant activity of 1-1.8 TROLOX/g DE was determined through FRAP assay. Different extracts were tested for their antimicrobial activity against *Malassezia spp.* and *Staphylococcus epidermidis*, obtaining a minimum inhibitory concentration (MIC) between 10-20 mg/mL. According to preliminary results, the extract (E) obtained with a 24 h extraction at 60°C, was selected and incorporated into gel formulations at the concentration of 1.0 % *w/w*, using XG, AG or HA as gelling agents (Figure 1). These polymers were selected based on preliminary evaluations regarding their natural origin, pH- and ionic strength-independent thickening/gelling capabilities, and compatibility with the extract without loss of viscosity upon incorporation. Furthermore, other aspects such as spreadability and skin feel were evaluated when selecting these polysaccharides over other gelling polymers.

Formulations were then added with glycerol as umectant, phenoxyethanol and ethylhexylglycerin (90:10) as preservative, sodium gluconate as chelating agent, panthenol, glycine, and allantoin as soothing and moisturizing agents, showing excellent compatibility among all the ingredients.

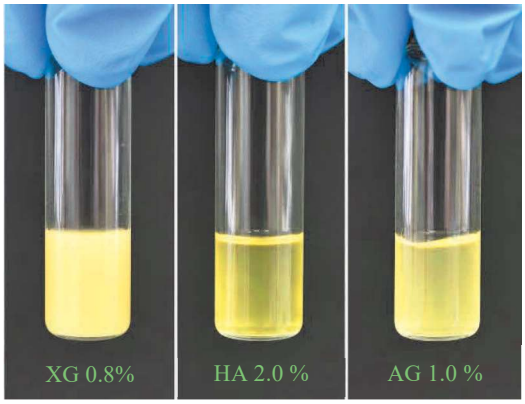


Figure 1. Appearance of the three different gel samples with extract.

Rheological analysis revealed pseudoplastic behavior for all formulations, with slight variations of profiles in extract-containing gels compared to control samples. A high viscosity in the plateau region was recorded for all the samples; HA formulations maintained a high viscosity over a wider shear rate range compared to other formulations.
Mechanical spectra showed weak-gel behavior for the XG- and AG- based gel, suggesting a more structured system, even though XG-based gel showed a certain decrease in G' and G'' when formulated with the extract, while HA formulation exhibited typical viscoelastic behavior, perfectly preserved after the addition of the extract.

Conclusion

The antioxidant and antimicrobial activity against acne-related microorganisms of OFI cladodes extracts suggests their potential application for acne-prone skin. The successful incorporation of the selected extract into gel formulations confirms the technological compatibility of this plant-derived ingredient with different rheology modifiers and the other present cosmetic ingredients, making it suitable for cosmetic formulation. These findings highlight OFI cladodes as a promising, sustainable raw material for the development of dermocosmetic products, in line with circular bio-economy principles.

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QuEChERS-based GC-MS/MS analytical method for a simultaneous determination of potential fragrance allergens in cosmetic complex matrices

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Introduction

The safety evaluation of cosmetic products requires strict compliance with the European Regulation (EU) 2023/1545, which recently amended the EU Cosmetic Regulation (EC) No 1223/2009 by expanding the list of potential fragrance allergens that must be labelled on cosmetic products (from 24 to 80), adding 56 new substances and groups to Annex III [1]. This event leads to a major analytical challenge due to the wide range of compounds to be detected in highly diverse, complex cosmetic matrices. In this context, the preparation of the analytical sample is a crucial step where conventional extraction techniques, such as liquid-liquid extraction [2] or solid-phase microextraction [3], are often time-consuming, solvent-intensive, and prone to matrix effects.

Aim of the work

This work aimed to develop a simple, high-throughput, and robust analytical method using GC-MS/MS for the simultaneous determination of potential fragrance allergens in complex cosmetic matrices as a reliable routine screening tool to comply with labelling requirements. To this end, once developed, the method was verified and applied to the analysis of 5 rinse-off products and 4 leave-on emulsions.

Materials and Methods

Chromatographic analyses were conducted using a gas chromatograph (Model 7890A) coupled with a triple quadrupole mass spectrometer in electron ionization source mode (GC-EI-MS/MS, Model 7000A), equipped with a DB-5MS UI capillary column and an Agilent MassHunter Workstation software (B.06.00) for data acquisition (Agilent Technologies, Palo Alto, CA, USA). Analyte identification was obtained by matching full-scan MS databases, linear retention indices (RI), and selective MRM transitions on a mixture of 62 standards (Table 1, Figure 1). Analytical samples were obtained from the cosmetic complex matrices considered: five body detergents (1-5, Table 2) and four body emulsions (6-9, Table 3) applying a QuEChERS-based extraction profile, by replacing acetonitrile with tert-butyl methyl ether to optimize the partitioning of lipophilic fragrance compounds from complex cosmetic formulations [4]. Absolute Quantification was carried out using Multiple Reaction Monitoring (MRM).

Table 1: Fragrance analytes reported according to Retention Time (RT).

N	Compound	RT (min)	N	Compound	RT (min)
1	α-pinene	14,09	32	vanillin	32,80
2	benzaldehyde	15,56	33	β-damascone	32,20
3	β-pinene	15,99	34	majantol	32,36
4	α-terpinene	17,54	35	β-caryophyllene	32,79
5	limonene	18,06	36	ebanol 1	33,23
6	benzyl alcohol	18,95	37	ebanol 2	33,93
7	salicylaldehyde	18,95	38	coumarin	33,59
8	terpinolene	20,36	39	isoeugenol	33,50
9	linalool	20,84	40	α-isomethyl ionone	34,27
10	camphor	22,99	41	eugenyl acetate	35,41
11	menthol	24,04	42	lilial	35,94
12	methyl salicylate	24,70	43	amyl salicylate	37,39
13	folione	24,69	44	isoeugenol acetate	38,21
14	α-terpineol	24,71	45	phtalide 3-propyldene	39,22
15	citronellol	25,67	46	amyl cinnamaldehyde	39,45
16	geranial	26,19	47	lyral minor	39,85
17	linalyl acetate	26,46	48	Iso E super (a-b)*	40,05
18	geraniol	26,54	49	lyral maior	40,07
19	carvone	26,58	50	α-santalol	40,38
20	neral	27,24	51	amylcinnamyl alcohol	40,52
21	cinnamaldehyde	27,88	52	iso E super (g)*	40,98
22	hydroxycitronellal	27,89	53	farnesol	41,30
23	*p*-anisyl alchol	28,04	54	β-santalol	41,55
24	anethole	28,71	55	hexyl cinamal	42,23
25	cinnamyl alcohol	28,96	56	α-acetyl cedrene	43,02
26	DMBCA	28,97	57	benzyl benzoate	43,05
27	eugenol	30,34	58	galaxolide (1-2)	44,92
28	geranyl acetate	30,92	59	benzyl salicylate	45,89
29	δ-damascone	31,00	60	hexadecanolact-16-one	47,27
30	damascenone	31,21	61	benzyl cinnamate	51,52
31	α-damascone	31,56	62	sclareol	54,01

DMBCA= Dimethyl Benzyl Carbinyl Acetate; *isomeric mixtures

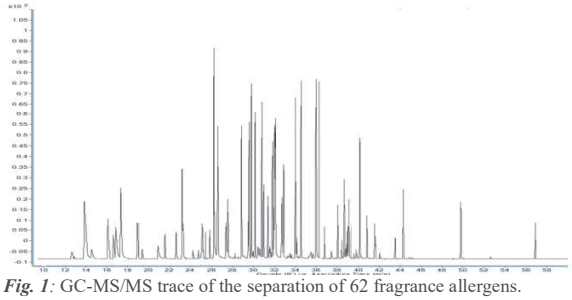


Fig. 1: GC-MS/MS trace of the separation of 62 fragrance allergens.

Two separate calibration curves were obtained, one for each type of matrix, using two blank products without any fragrance (a surfactant aqueous solution, BPF 1, and an O/W emulsion BPF 2), fortified with the analytes of interest.
Linearity and recovery were evaluated after extraction and analysis of BPF 1-2, spiked with internal standards (2-fluorophenylethanol and 1,3-dibromododecyl benzene).

Results and discussion

The analytical method allows the simultaneous detection of 62 fragrance allergens in less than one hour, as reported in Table 1 and Figure 1. The method demonstrated excellent linearity within the range of 1.5 to 18 µg (absolute mass) for all analyzed fragrance allergens, with coefficient of determination values (R^2) ranging from 0.994 to 0.999 for rinse-off products (BF1) and from 0.995 to 0.998 for BF2. Accuracy was thoroughly demonstrated through robust recovery rates. BF1 recovery values ranged from 70.45% to 119.83%, and BF2 recovery values ranged from 81.50% to 117.00%. Despite the potential heavy noise typically encountered in the analysis of leave-on cosmetics, MRM gave clean, unmistakable identification and highly accurate tracking of target allergens by successfully cutting through noise, even in the most complex matrices.
Concerning the commercial products selected, the analyses confirmed the presence, in varying percentages, of all the fragrances listed on the labels, except for some of the newly introduced ones. It should be considered, however, that the deadline for updating the label for non-compliant products (already on the market before 31 July 2026) is 31 July 2028. In Table 2-3, the composition, the labeled fragrances, and the related analytical results for all samples are reported.

Table 2 - Composition, labeled fragrances, and analytical results for samples 1-5 (body detergents).

	INGREDIENTS	Labelled Fragrances	Main Det. (ppm±sd)
1	Aqua sodium laureth sulfate, sodium chloride, cocamidopropyl betaine, laureth-2, parfum, poly-quaternium-7, sodium benzoate, tetrasodium edta, citric acid, CI 19140, CI42090	limonene, linalool	52.6±7.0 78.8±9.2
2	Aqua, sodium laureth sulfate, sodium chloride, cocamidopropyl betaine, parfum, laureth-2, polyquaternium-7, sodium benzoate, tetrasodium edta, citric acid, CI 16035, CI 19140,CI 42090, CI 15985	limonene, hexyl cinamal, linalool	124.3±7.2 80.6±4.7 108.4±1.3
3	Aqua, sodium laureth sulfate, cocamidopropyl betaine, sodium chloride, glycerin, mel extract, parfum, lactic acid, sodium benzoate, potassium sorbate,	benzyl alcohol, vanillin	1469.9±125.2 80.49±9.3
4	Aqua, sodium laureth sulfate, cocamidopropyl betaine, sodium chloride, glycerin, malva sylvestris extract, parfum, lactic acid, sodium benzoate, potassium sorbate,	benzyl alcohol hexamethylindano pyran α-isomethyl ionone, linalool linalyl acetate pentyl salicylate pinene limonene	1302.3±9.8 204.6±9.1 164.7±4.7 302.5±10.9 14.0±12.8 85.2±0.9 26.9±1.1 (α) 10.1±0.9 (β) 48.5±3.15
5	Aqua, sodium laureth sulfate, sodium chloride, parfum, cocamidopropyl betaine, bitter orange flower water, citric acid, tetrasodium edta, sodium benzoate, sodium benzotrazolyl butylphenol sulfonate, buteth 3, tributyl citrate, CI16035 CI17200 CI 19140	geraniol, hexyl cinnamal, limonene, linalool	87.9±21.5 112.1±16.1 129.1±43.7 20.0±5.5

Table 3 - Composition, labeled fragrances and analytical results for samples 6-9 (body emulsions)

	INGREDIENTS	Labelled Fragrances	Main Det. (ppm ±sd)
6	Aqua, ethylhexyl stearate, glycerin, shea butter, cetearyl alcohol, sweet almond oil, pentaerythrityl distearate, hazel seed oil, argan kernel oil, olive fruit oil, ginger root extract, cinnamomum cassia bark extract, tocopherol, tocopheryl acetate, disodium sulfosuccinate, disodium octadecyl sulfosuccinate, xanthan gum, magnesium aluminum silicate, glyceryl stearate, citric acid, ethylhexl glicerin, parfum	benzyl alcohol eugenol coumarin cinnamaldehyde	375.82±18.3 42.76 ±5.51 33.6 ±3.33 26.53±2.64
7	Aqua, ethylhexyl stearate, glycerin, ceteary alcohol, shea butter, sweet almond oil, glyceryl stearate, disodium ceilakil sulfosuccinate, grapefruit extract, peach fruit extract, argani kernel oil, hazel seed oil, olive fruit oil, coconut oil, tocopherol, xanthan gum, magnesium aluminum silicate, citric acid, ethylhexylglycerin, phenoxyethanol, parfum, sodium benzoate, potassium sorbate,	benzyl alcool limonene benzyl salicylate hexyl cinnamal linalool benzyl benzoate coumarin citronellol	1071.8±11.41 41.3±23.1 156.0±17.7 120.2±14.5 65.2±10.2 8.9±2.2 10.28±1.44 8.9±1.7
8	Aqua, Glycerin, Cetearyl Alcohol, Ceteareth-25, Myristyl Alcohol, Shea Butter, Parfum, Cetyl Alcohol, Iris Pallida Root Extract, Dimethicone, Phenoxyethanol, Chlorphenesin, Declene Glycol, Citric Acid, Sodium Benzoate, Potassium Sorbate, Citrus Aurantium Peel Oil,	linalyl acetate benzy salicylate linalool limonene hydroxycitronellal citronellol hexyl cinnamal α-isomethl ionone Iso E super (g) Iso E super (a-b) hexadecanolactone	152.2±24.1 135.7±16.2 162.4±10.5 76.7±9.4 82.7±10.0 65.4±10.7 66.0±12.4 39.8±7.6 1105.6±161.5 474.9±49.9 traces
9	Aqua Glycerin, Cetearyl Alcohol, Ceteareth-25, Myristyl Alcohol, Shea Butter, Parfum, Cetyl Alcool, Cymbidium Great Flower Extract, Dimethicone, Decylene Glycol, Propylene Glycol, Ethylhexyllycerin, Tetrasodium Glutamate Diacetate, Phenoxyethanol, Chlorphenesin,	galaxolide hexyl cinamal acetyl cedrene benzyl benzoate anethole caryophyllene.	206.4±69.8 267.0±88.2 31.0±16.3 46.9±21.7 14.4±5.1 traces

Conclusion

This high-throughput method offers a fast, robust, and reliable solution for day-to-day laboratory screening. This makes the method a perfect fit for meeting the strict safety and labeling standards required by current European regulations.

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Design of Semi-Solid Formulations for Topical Delivery of Palmitoyl-GHK peptide

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Introduction

Biomimetic peptides, such as GHK (Glycine–Histidine–Lysine), are widely used in skin aging treatments to stimulate collagen synthesis and extracellular matrix restoration in the dermis. However, the high molecular weight and hydrophilicity of native peptides restrict their cutaneous penetration [1,2]. Chemical modifications, such as lipophilic conjugation to form Palmitoyl-GHK (Palm-GHK), enhance lipophilicity, permeability, and chemical stability. To overcome delivery barriers and maintain formulation stability, biocompatible lipid- and polymer-based nanocarriers [3] and semi-solid platforms [4] are formulated, including: (i) ethosomes (ETO); (ii) ethosomal gels (ETO-GEL); (iii) organogels (ORG); (iv) poloxamer hydrogels (POL); and (v) poloxamer lecithin organogels (POL-ORG).

Aim of the work

This study aimed to design, formulate, and characterize vesicular and semi-solid delivery systems, specifically ETO, ETO-GEL, ORG, POL, and POL-ORG, for the cutaneous delivery of Palm-GHK. Multiple physicochemical parameters and properties were investigated, such as vesicle size, polidispersity index, spreadability, occlusive factor, in vivo skin hydration capacity, in vitro release kinetics and finally human skin compatibility.

Materials and Methods

Soy phosphatidylcholine (PC), Poloxamer 407 (POL), isopropyl palmitate (IPP), xanthan gum (XG), and synthesized Palm-GHK were utilized as primary materials. ETO were prepared by dropwise addition of water to an ethanolic phosphatidylcholine solution under magnetic stirring, and subsequently thickened with XG 1% w/w to produce the ETO-GEL. ORG were produced by adding water to a 200 mM PC solution in isopropyl palmitate, while the hybrid POL-ORG was obtained by combining the 200 mM PC solution in IPP with a 20% w/w POL hydrogel in a 30:70 w/w ratio. For peptide-loaded formulations, Palm-GHK was solubilized in the respective initial phases prior to complete gelation or vesicle formation. Vesicle diameter (Z-avg) and polydispersity index (PDI) of ethosomes were evaluated via Photon Correlation Spectroscopy (PCS). The spreadability was measured by applying standard weights between Petri dishes, while the occlusive factor was determined by measuring water evaporation loss through filter paper over 48 hours at 32 °C. In vivo corneocyte hydration on the forearms of five healthy volunteers was recorded over two hours using a Moisture MeterSC. Finally, Franz diffusion cells with synthetic nylon membranes were employed over an eight-hour period to evaluate peptide release kinetics via HPLC quantification. Cutaneous compatibility was verified using a 48-hour occlusive patch test on 20 healthy human volunteers.

Results and discussion

Unloaded ETO displayed Z-avg values of 206.5 nm, which decreased to 102.1 nm upon Palm-GHK loading, due to the peptide's surfactant-like interfacial activity, achieving an encapsulation efficiency (EE%) of approximately 98%. Palm-GHK was successfully solubilized in ETO, POL, and POL-ORG; however, it was insoluble in ORG, causing phase separation and leading to the exclusion of ORG-GHK from further studies. Regarding rheological and barrier properties, ETO-GEL showed the highest spreadability at 19.17 $g \cdot cm \cdot s^{-1}$, whereas POL-ORG demonstrated a lower spreadability of 9.17 $g \cdot cm \cdot s^{-1}$ due to its dense interpenetrating network. Conversely, ORG exhibited the highest occlusive factor at 63%, followed by POL-ORG at 39%, POL at 32%, and ETO-GEL at 4%.
Concerning the *in vivo* skin hydration evaluation, ETO-GEL provided a strong initial hydration spike that dissipated rapidly, whereas ORG provided a sustained, progressive moisturizing effect over two hours, due to the presence of isopropyl palmitate content, characterized by emollient properties.
In vitro release testing (Fig. 1) showed that semi-solid formulations were able to control Palm-GHK release, compared with both the free solution and liquid ETO.
ETO-GEL-GHK retained almost all peptide, possibly due to physical entrapment and chemical interactions within the XG network, while POL-ORG-GHK provided a controlled, sustained release governed by its complex supramolecular structure. Crucially, the patch test confirmed that all formulations were completely safe and non-irritating to human skin [1].

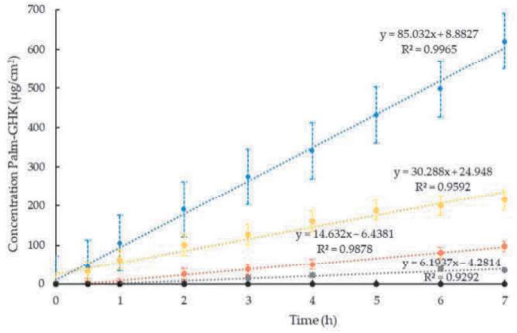


Fig. 1. Palm-GHK release kinetics from the different forms: light blue (SOL-GHK), yellow (ETO-GHK), grey (POL-GHK), orange (POL-ORG-GHK), and black (ETO-GEL-GHK).

Conclusion

This work demonstrated the critical importance of pre-formulation technological design in developing effective cosmetic systems for bioactive peptides like Palmitoyl-GHK. Tailored formulation strategies successfully address key delivery challenges, including peptide solubility, stability, and controlled release. Overall, these advanced semi-solid carriers establish a foundation for high-performance cosmeceuticals that deliver tangible skin benefits while ensuring product safety. Among the evaluated systems, the hybrid Poloxamer lecithin organogel (POL-ORG) represents an optimal, non-irritating, smart semi-solid platform capable of delivering Palm-GHK, assuring a controlled release of the peptide for anti-aging cosmeceutical applications.

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UV-Induced Alterations in Hair: Evaluation of the Photoprotective Activity of Sericin

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Introduction

Although hair lacks essential physiological functions, it significantly influences self-perception and self-esteem. As a result, the haircare industry actively seeks ingredients and formulations that maintain hair appearance and health. Chemical bleaching agents [1], sun exposure, and heat styling are primary contributors to hair damage. Ultraviolet radiation (UV) generates of reactive oxygen species (ROS) within hair fibres, leading to reduced mechanical strength, degradation of lipid and protein components, and oxidation of melanin granules [2].

Aim of the work

This study evaluated *in vitro* the alterations induced in brown Caucasian hair by UV exposure. Identical analyses were conducted on hair tresses previously subjected to chemical stress through treatment with a commercial bleaching formulation.
The project also involved the development of a rinse-off mask functionalized with sericin, a protein derived from silkworm cocoons known for its antioxidant properties [3]. The photoprotective capacity of this formulation was compared to that of a commercial benchmark conditioner containing sericin.

Materials and Methods

Materials: Sericin was extracted using a high-temperature high-pressure degumming process at 120 °C for 15 min with a cocoon-to-water *ratio* of 1:40 w/w. Sericin dispersion was then co-spray-dried with trehalose and methyl-β-cyclodextrin as drying agents [4], and a cationic guar gum derivative, guar hydroxypropyltrimonium chloride, to enhance process yield, reduce residual moisture, and improve the morphology of the sericin powder (SP) [5]. All other materials were of technical grade and used as received.

Formulation development and characterization: A rinse-off mask, an o/w emulsion, was formulated with 1.0% w/w SP. pH and accelerated stability by centrifugation were assessed after 24 hours and after 2 months.

Hair samples: Caucasian natural brown tresses were used. Hair tresses, each approximately 20 cm in length and weighing 2 g, were pre-washed with 10% w/v sodium laureth sulfate.

Hair bleaching procedure: Some brown hair tresses were bleached using a commercial product consisting of a 1:2 mixture of a bleaching powder containing potassium and ammonium persulfates and a 40-volume hydrogen peroxide developer. The mixture was left on for 40 min, after which the tresses were washed.

Hair treatment: The functionalized mask (2 g) was applied to each tress and left on for 10 min before rinsing with water. The tresses were then blow-dried using the cool-air setting. The same application procedure was followed for the benchmark conditioner.

Hair photoirradiation: To simulate light-induced damage, hair tresses were alternately exposed to a UVA lamp (irradiance 1.6 mW/cm²) or a UVB lamp (0.6 mW/cm²) for 168 hours at 10 cm. During the exposure period, samples were periodically turned over and repositioned to ensure uniform photoirradiation on both sides and throughout the entire tress.

In vitro evaluation of photoprotective effect: Hair colour changes were quantified with a colorimetric probe utilizing the CIELab model. The L* (lightness), a* (red-green axis), and b* (yellow-blue axis) parameters were recorded to calculate total colour change (ΔE) according to the formula $\Delta E = \sqrt{[(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2]}$. Lipid degradation was assessed using the malondialdehyde (MDA) assay with thiobarbituric acid (TBA). MDA, generated by oxidation, reacts with TBA to form a pink adduct that absorbs at 535 nm and is detectable by spectrophotometry. Before analysis, lipids were extracted from hair samples in methanol using an ultrasonic bath. The extent of damage was expressed as nmol MDA per gram of hair.

Results and discussion

The INCI composition of the rinse-off mask is reported in Table 1.

Table 1: Composition of rinse-off mask.*

Ingredients	% w/w
Phase A	
Aqua	*q.s. 100.00*
Glycerin	*2.00*
Phase B	
Squalane	*10.00*
Cetyl alcohol	*2.00*
Behentrimonium chloride	*1.00*
Polyquaternium -7	*2.00*
Potassium Olivoyl Hydrolyzed Oat Protein, Cetearyl Alcohol, Glyceryl Stearate, Glyceryl Oleate	*7.00*
Phase C	
Phenoxyethanol	*1.00*
Phase D	
SP	*1.00*

*pH: 4.32±0.1; physical stability after centrifugation test (both at 24 hours and 2 months).

Exposure to solar simulators increased the ΔE value, with a more pronounced effect observed for UVB. UVB, characterized by a shorter wavelength, penetrates the fibre to a lesser extent but damages the cuticle, thereby altering the hair's capacity to reflect incident light. In contrast, UVA radiation influences colour variation primarily by degrading melanin and inducing hypopigmentation.
Colorimetric measurements indicated that the sericin-containing mask exhibited effective photoprotective activity on brown hair, while its efficacy was reduced on bleached hair. In contrast, the benchmark product produced higher ΔE values for all hair samples, likely due to interference from specific components in the formulation (Figures 1,2).

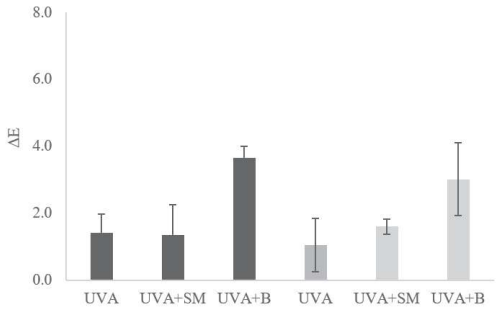


Figure 1. Total color change (ΔE, mean ± SD) in brown (dark) and bleached (grey) hair tresses following UVA exposure, after treatment with sericin-functionalized mask (UVA+SM) or benchmark conditioner (UVA+B) (n=2).

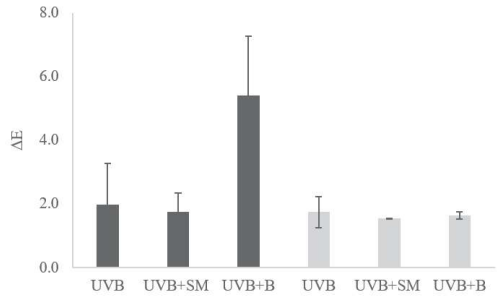


Figure 2. Total color change (ΔE, mean ± SD) in brown (dark) and bleached (grey) hair tresses following UVB exposure, after treatment with sericin-functionalized mask (UVB+SM) or benchmark conditioner (UVB+B) (n=2).

UVA, which penetrates more deeply, caused greater damage to the lipid component of natural brown hair than UVB. Pre-treatment with the experimental mask demonstrated similar protection against both UVA and UVB, as indicated by MDA values measured on tresses irradiated for 168 hours, which were comparable to those of the control group (hair not exposed to the lamps). During exposure to solar simulators, the protein-functionalized preparation exhibited clear photoprotective properties for both brown and bleached hair (Figures 3,4). In contrast, the commercial conditioner was less effective than the laboratory-formulated product.

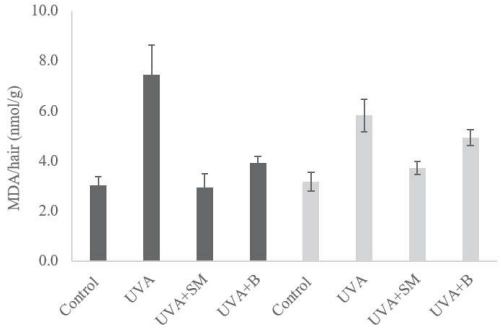


Figure 3. Lipid peroxidation (mean ± SD in brown (black) and bleached (grey) hair tresses before (Control) and following UVA exposure, after treatment with sericin-functionalized mask (UVA+SM) or benchmark conditioner (UVA+B) (n=2).

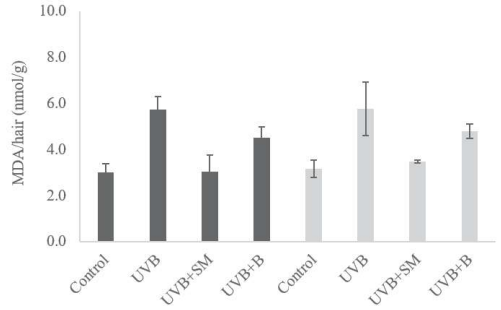


Figure 4. Lipid peroxidation (mean ± SD) in brown (black) and bleached (grey) hair tresses before (Control) and following UVB exposure, after treatment with sericin-functionalized mask (UVB+SM) or benchmark conditioner (UVB+B) (n=2).

Conclusions

UVA and UVB exposure significantly damage hair, altering both structural integrity and mechanical properties. Sericin demonstrates protective effects against physical damage and exhibits notable antioxidant capacity, substantially reducing lipid peroxidation in irradiated hair tresses.

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