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Natural Antioxidants from *Acmella oleracea* Extract as Dermatocosmetic Actives

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Abstract: Compounds from plant extracts make dermatocosmetic products more effective as they avoid the adaptation and resistance of the organism and achieve a synergistic effect of the molecular properties of interest. *Acmella oleracea* extract is considered to have great potential in preventing oxidative damage and improving the appearance of the skin. The purpose of this article is to support the product formulated by preliminary studies of two types of O/W emulsions with 3% and 5% concentrations of *Acmella oleracea* extract. Physico-chemical methods were performed to evaluate the stability, microbiological control, rheological behavior and diffusion through the membrane. Good homogeneity, structural strength and flexibility, adequate skin diffusion, and high physico-chemical and microbiological stability were confirmed. The conclusions lead to the idea that these results require further in vivo studies as well as studies of toxicity and cytotoxicity to obtain the necessary data to place this product on the market.

Keywords: *Acmella oleracea*; dermatocosmetic field; emulsions; skin oxidative stress

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1. Introduction

The current trend among consumers is towards cosmetic products that contain active ingredients of natural origin [1]. The increasing popularity of natural cosmetic products is due to consumers' preference for safer and more effective alternatives to chemically synthesized substances [1].

Consumers are increasingly paying attention to the ingredients used in cosmetic products, so natural ingredients such as vitamins, polyunsaturated fatty acids, oligosaccharides, carotenoids, bioactive peptides and natural compounds are replacing synthetic ingredients as the natural ingredients are considered sustainable and biodegradable [2]. Secondary plant metabolites, polyphenols, flavonoids and terpenes are recognized as valuable active ingredients in dermatocosmetic products [3]. Numerous studies have investigated their biologically active effects in skin care products, antioxidant effect [4], collagen synthesis [5], restructuring of the extracellular matrix [6,7] or UV protection [8]. They can also be used to prevent degenerative skin diseases and provide important micronutrients for maintaining skin homeostasis, with fewer adverse effects [9].

The formulation of cosmetic products with ingredients of natural origin requires a thorough understanding of their properties, and one of the main challenges is to ensure the stability of the active ingredients [10]. It is important to determine the appropriate concentrations to ensure efficacy and determine the pH, as pH can influence the effective penetration of the active ingredients into the skin [11]. In addition, stability is a major challenge in the formulation of cosmetic products with ingredients of natural origin, which often have different pH values, viscosities or solubilities [12,13].

Acmella oleracea (L.) RK Jansen is a plant with great potential that belongs to the *Asteraceae* family and is cultivated as a medicinal plant in South America, especially in Mexico, Peru, Bolivia and Brazil, where it is known by the following names: jambu, paracress or electric plant [14]. The chemical composition of the plant was presented in detail in the specialized literature, where a high content of biologically active compounds of interest in cosmetic products was reported, especially N-alkylamides (such as isobutyl amides [spilanthol], methyl butyl amides, phenyl butyl amides) [15], along with triterpenoids (3-acetylaleurytolic acid, β-sitostenone, stigmasterol), steroid glycosides (stigmasteryl-3-O-β-D-glucopyranoside, β-sitosteryl-3-O-β-D-glucopyranoside), phenolic compounds (vanillic acid, trans-ferulic acid, trans-isoferulic acid), fatty acids (n-hexadecanoic acid, n-tetradecanoic acid), coumarins (scopoletin), and volatile compounds (β-pinene, myrcene, (*E*)-caryophyllene, caryophyllene oxide, germacrene D, β-phellandrene, spilanthol and amelonate) [16–18]. Additionally, the presence of polyphenols in large amounts in leaves and flowers rather than in stems was emphasized, and flavonoids, alkaloids, tannins, saponins and steroid glycosides are distributed in all parts of the plant [19]. The main component, spilanthol, is the primary metabolite responsible for the anesthetic effect [20] and the muscle relaxant effect on the facial muscles and is considered a natural alternative to the botulinum toxin [21].

The biological properties of the plant when used internally or externally (anti-inflammatory, antipyretic, immunostimulant, anti-obesity, antispasmodic, diuretic, anthelmintic, aphrodisiac, sialagogue, analgesic, hepatoprotective) are also of great importance as they do not cause adverse effects. The antimicrobial (antifungal, antiprotozoal) and anesthetic, antioxidant, insecticide, antiseptic) [13,22–25] properties are mainly due to the content of N-alkylamides, a group of molecules found in the genera *Spilanthes* and *Acmella*.

Although it is a tropical plant, it has recently been successfully acclimatized in Romania, a country with a temperate climate, in a small experimental culture in Popricani-Iasi.

The present study focuses on the formulation and characterization of emulsions with ingredients of natural origin and extracts of *Acmella oleracea* (L.) RK Jansen to evaluate the antioxidant capacity of the extract as a biologically active substance in cosmetic compositions.

2. Materials and Methods

2.1. Plant Material

The starting material for the studies was the plant *Acmella oleracea* (*A. oleracea*), which was acclimatized in Iasi County (Romania) (Figure 1a). After harvesting (Figure 1b) and drying (Figure 1c) in a stream of air and without direct exposure to sunlight, the plant was crushed (Figure 1d) and stored in brown containers in a cool place to be characterized and used to obtain an alcoholic extract (Figure 1e) to produce new dermatocosmetic emulsions (Figure 1f). Depending on the characteristics of the soil and growth environment, a number of minerals also occur in addition to the organic compounds.

(**a**) (**b**) (**c**) (**d**) (**e**) (**f**)

Figure 1. The central elements of the study: (**a**) fresh *A.* oleracea plant; (**b**) dry plant; (**c**) grinding the plant; (**d**) plant powder; (**e**) alcoholic extracts obtained from *A. oleracea*; (**f**) emulsion with 5% *A. oleracea* ethanolic extract.

2.2. *A. oleracea* Extract

The *A. oleracea* extract used to obtain the studied emulsion was created through maceration using a liquid–solid extraction method in 30% ethanol solution, for a period of 7 days and using an S/L ratio of 1:5, based on a protocol described in our previous work [26].

2.3. Evaluation of the Mineral Content of the Dried Plant

Before analysis, plant samples were dried to a constant weight and homogenized using a planetary mill Pulverisette 6 (Fritsch, Oberstein, Germany). Next, 0.5 g of each sample was mixed with 5 mL of trace pure HNO_3 (Sigma-Aldrich, Steinheim, Germany) and 2 mL H_2O_2 p.a. (Sigma-Aldrich, Steinheim, Germany) in Teflon vessels. Sample digestion was performed in the Mars 6 microwave digestion system (CEM, Matthews, NC, USA). The obtained solutions were quantitatively transferred into 25 mL flasks and made up to the volume with bi-distilled water. More details about sample preparation can be found in [27].

The content of Al, Ba, Cd, Cr, Co, Cu, Mn, Fe, Ni, Pb, V, Sr, P, S, Ca, Mg, K and Zn was determined using ICP–OES PlasmaQuant 9000 Elite (Analytik Jena, Jena, Germany). The calibration solutions and standards were prepared from IV-STOCK-27 (Inorganic Ventures, Christiansburg, VA, USA) standard solution. All control standards were analyzed after every 10 samples.

2.4. Evaluation of the Main Phytoconstituents of the Extract

The extract used was characterized by the amount of the following main phytoconstituents: polyphenols and flavonoids.

The amount of polyphenols was determined spectrophotometrically using the Foling–Ciocalteu method and the results were expressed in mg of gallic acid equivalent per mL extract (µg GAE/g).

The flavonoid content was determined by reacting with $AlCl_3$ 2% prepared in methanol, following the protocol from our previous works, and the results were expressed in mg of quercitin equivalent per mL extract (mg QE/g) [26].

All analyses were performed in duplicate.

2.5. Evaluation of the Antioxidant Activity of the Extract

For the antioxidant activity, the methods of 2,2-diphenyl-1-picrylhydrazyl radical scavenging assay (DPPH) and 2,2′-azino-bis(3-ethylbenzothiazoline) 6-sulfonic acid radical scavenging assay (ABTS), which had a work protocol detailed in previous works [28] by adapting the information from the literature, were used [29].

2.6. The Methodology of Obtaining the Emulsion Based on the Extract of *A. oleraceea*

An O/W emulsion was prepared with *A. oleracea* extract as an active ingredient with phytochemical properties to protect the skin from oxidative stress. Two types of emulsions

were prepared, with the following extract contents: (i) F1A with 3% extract and (ii) F2A with 5% extract.

The emulsion base (B1A) consists of a lipophilic phase (*Amaranthus Spinosus* Seed Oil, *Psoralea corylifolia* Seed Oil, *Malus domestica* Seed Oil, *Solanum Lycopersicum* Seed Oil, Cetearyl Alcohol [and] Glyceryl Stearate [and] Jojoba Esters [and] *Helianthus Annuus* [Sunflower] Seed Wax [and] Sodium Stearoyl Glutamate [and] Water [and] Polyglycerin-3), a hydrophilic phase (*A. oleracea* Flower Water and Glycerin), additives (Lecithin, Sclerotium Gum, Pullulan, Xanthan Gum) and preservatives (Benzyl Alcohol, Dehydroacetic Acid).

To prepare the emulsions, the protocol presented in our previous work [26] was followed, which assumes that both the hydrophilic and lipophilic phases were heated to 75 °C after the aqueous phase had previously been gelled with the viscosity agent. Under continuous mixing, the lipophilic phase was added to the hydrophilic phase using a Dynamix® DMX combi 160 homogenizer running at 13,000 rpm. Following three 3-min shaking cycles, the emulsion was allowed to cool on a water bath set at 40 °C before the preservative and active ingredients were added. Samples weighing fifteen grams were put into brown glass jars so they could be used in other studies on emulsions.

2.7. Characterization of Emulsion

2.7.1. Microbiological Control

Microbiological control was implemented within 24 h after preparation and at 30 days after storage at room temperature. The methodology previously presented [30] was used to quantify microorganism presence in the analyzed samples.

2.7.2. Rheological Measurements

Rheological tests were performed on a Physica MCR 501 modular rheometer (Anton Paar, Graz, Austria) equipped with a Peltier temperature control system. For the measurements, the geometry of serrated parallel plates with a diameter of 50 mm was used. All isothermal experiments were performed at a constant temperature of 25 °C. Reproducibility was checked for all the rheological tests on three samples from each emulsion. In the amplitude sweep, the frequency is kept constant (10 rad/s), while the oscillation amplitude (γ) varied between 0.01 and 100%. This test is used to determine the linear viscoelastic range. Time sweep tests were performed at constant temperature 25 °C, frequency 1 Hz and amplitude 0.1% (in the linear viscoelastic range). In the dynamic temperature sweep tests, the samples were heated from 10 °C to 50 °C at a 0.5 °C/min rate at a constant frequency of 1 Hz and a constant strain in the linear viscoelastic region (0.1%) [31].

2.7.3. Analysis of Homogeneity of the Emulsion

The homogeneity and stability of the serums during the storage period were studied using the Scanning Electronic Microscopy (SEM) method according to a protocol previously described in our studies [32]. For this determination, the sample was maintained in a vacuum for 24 h. Images were obtained with a Secondary Electrons (Ses) detector (WD 15.5 mm, 30 kV, HV) from a scanning electron microscope (SEM) Vega Tescan-LMHII (Tescan Orsay Holding, Brno-Kohoutovice, South Moravia, Czech Republic).

Also, emulsion images, for checking the homogeneity, were analyzed using a binocular microscope Optika B-159 (OPTIKA S.r.l., Ponteranica [BG], Bergamo, Italy), magnification—1000×, within 24 h of preparation.

2.7.4. In Vitro Evaluation of the Emulsion

To investigate the cutaneous application of emulsions containing *A. oleracea* extract as an active ingredient, preliminary permeation studies were developed using a Franz cell equipped with chicken skin (known to behave similar to human skin). We used the previously developed and tested working protocol in our work [33] as well as those from the literature [34,35].

3. Results

3.1. Determination of Minerals from the Initial Plant Sample

Heavy metals are among the most harmful pollutants, according to the World Health Organization (WHO). These metals are not biodegradable, bioaccumulate in the ecosystem, move up the food chain, and have detrimental impacts on both the environment and human health [36,37]. Heavy metals can undergo species transformations (e.g., valence state changes) and conversions between inorganic and organic forms in the environment due to abiotic and biotic causes. Furthermore, metals come in a variety of sizes in environmental compartments, ranging from tiny particles to massive quantities [38]. Considering these aspects, a possible heavy metal contamination of plants used in cosmetology for specific products could have risks for human health, even in low concentrations, due to their passage through the plant extract. That is why the product standards and the WHO established the non-dangerous limits of these metallic species. In the context of current pollution, the need for research regarding the content of different chemicals such as heavy metals in vegetales intended for utilization in different area is imperative [36].

Along with the polluting metals, beneficial microelements (Ca, K, Mg, Fe, Cu, etc.) can also be present when they are present in certain quantities regulated for the quality of the final cosmetic product. The determination of these quantities was based on the quality control protocol of the starting material in our study. The quality control of the analysis was ensured by analysis of the reference materials 1547 (peach leaves) and 1575a (pine needles). The results of the quality control are presented in Figure 2.

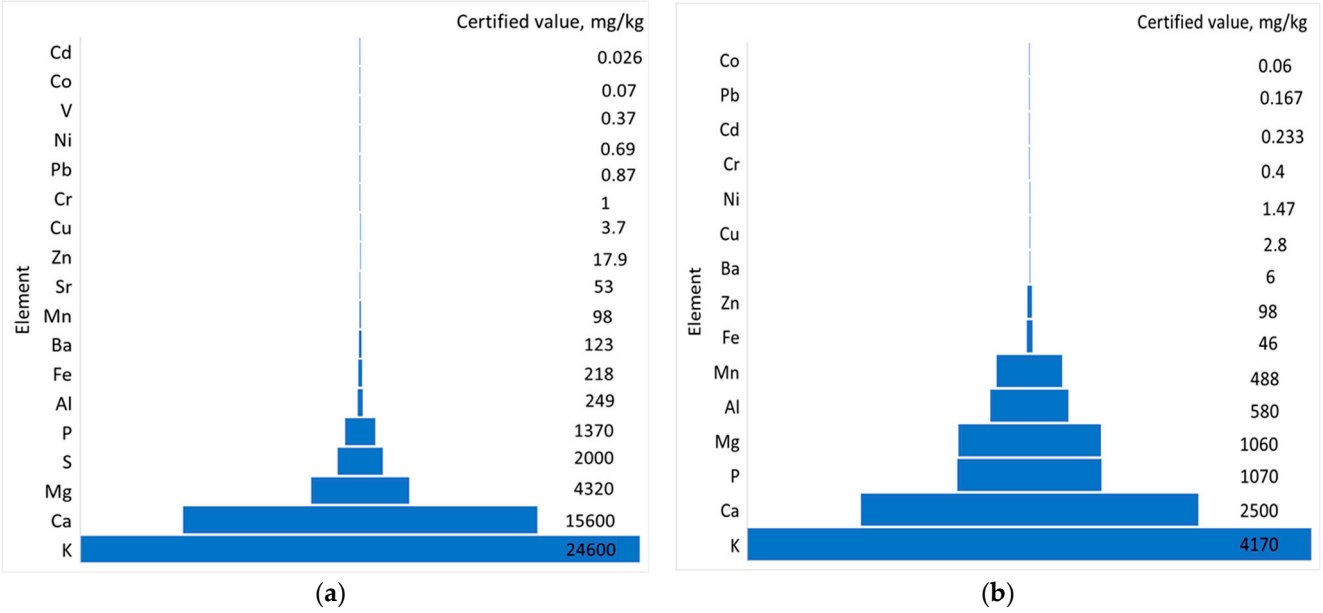


Figure 2. The content of microelements in the dry plant according to the following reference materials: (**a**) 1547 and (**b**) 1575a.

The ISO 21392:2021 standard provides the guidelines for the determination of heavy metals in cosmetic products. It focuses on measuring trace levels of specific metals such as chromium, cobalt, nickel, arsenic, cadmium, antimony and lead using inductively coupled plasma mass spectrometry (ICP–MS). Detection limits for these metals can be as low as 20 µg/kg (0.02 ppm) in the finished product.

The obtained values indicate compliance with the quality standards, namely the fact that the plant extracts will not be contaminated with toxic metal ions.

3.2. Vegetal Extract Characterized

The characteristics of the *A. oleracea* extract used to prepare the studied emulsion are presented in Table 1 [26].

Table 1. Characteristics of *A. oleracea* extract.

Polyphenols Content, mg GAE/mL	Flavonoids Content, mg QE/mL	Antioxidant Activity	
		DPPH (mg TE/mL)	ABTS (mg TE/mL)
3.7986	4.490	0.21 ± 0.05	2.03 ± 0.12

Results are expressed as mean ± standard deviation (SD) of three determinations. Legend: ABTS—2,2′-azino-bis(3-ethylbenzothiazoline) 6-sulfonic acid; DPPH—1,1-diphenyl-2-picrylhydrazyl; TE—Trolox equivalents; GAE—gallic acid; QE—quercitin.

The amount of polyphenols and flavonoids that characterize this alcoholic extract of *A. oleracea* falls within the values reported by other research, regardless of the extraction technique used (Table 2).

Table 2. Example of the amount of polyphenols and flavonoids in *A. oleracea* extract.

Plant	Type of Extract	Polyphenol Content	Flavonoid Content	Ref.
Flowering aerial parts of *A. oleracea*	Solvent Methanol, Soxhlet extract	1.38 GAE mg/g	28.7 QE mg/g	[39]
Leaf of *A. oleracea*	80% ethanol (*v*/*v*), ultrasonic extraction	3.19 mg GAE/g	11.45 mg RE/g	[40]
Flower of *A. oleracea*	80% ethanol (*v*/*v*), Ultrasonic extraction	1.98 mg GAE/g	5.91 mg RE/g	[40]
Dried extract of Flowering aerial parts of *A. oleracea*	Solvent Methanol, Soxhlet extract	7.59 mg GAE/g of dried extract	indefinite	[19]
Fresh leaves of *A. oleracea*	Solvent Methanol, Ultrasonic extraction, 30 min.	588.65 mg GAE/100 g	9.32 mg RE/100 g	[41]
Fresh flower of *A. oleracea*	Solvent Methanol Ultrasonic extraction, 30 min.	292.81 mg GAE/100 g	4.10 mg RE/100 g	[41]
Dried leaves of *A. oleracea*	Solvent Methanol Vortex extraction	10.99 mg GAE/g	11.33 mg RE/g	[42]

Nations: GAE: Gallic Acid Equivalent; RE: Rutin Equivalent; QE: Quercitin Equivalent.

3.3. Characterization the Obtained Emulsion

3.3.1. Microbiological Control

Many types of cosmetics, especially emulsions, can provide suitable growth conditions for bacteria and fungus (molds and yeasts). As a result, preservation is crucial to keep the product from deteriorating and guarantee user safety. Product spoilage over time, as evidenced by the development of unpleasant odors, color or texture changes, and phase separation, frequently points to contamination. The microorganisms in cosmetic products may be saprophyte, meaning there is no risk to the user, or they may be pathogenic or have the potential to become pathogenic under specific circumstances. All microorganisms present in a cosmetic product have the potential to adversely impact product quality throughout the manufacturing process and in product exposure to the environment during regular use. Microorganisms can be introduced in the cosmetic formulation from the following: raw materials, processing equipment, environment, water added, and persons involved in the manufacturing process [30]. Additionally, untreated raw materials are frequently employed in cosmetics to support the "natural" claim, which appeals to consumers more, but if the materials are not treated properly, these products may also contain a high concentration of microorganisms. The goal of creating safe and stable cosmetics may appear intimidating and unachievable given the widespread presence and wide range of microorganism species,

but cosmetic products, if not self-sterilizing, sustain the growth of relatively few organisms, even during normal consumer usage.

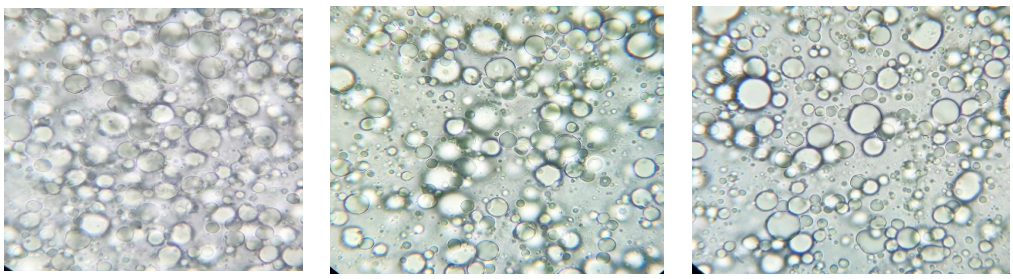
In the notes of guidance for cosmetic products, the Scientific Committee on Consumer Products (SCCP) divided cosmetic products into the following two categories: (a) products for children under three years old or products to be applied to the eye area or mucous membranes, and (b) other cosmetic products. As the tested products are framed in the second category, the maximum number of aerobic mesophilic microorganisms (CFUs; unite formant colonies) that can be found in products is 10^3 CFUs, but the following microorganisms cannot be found in 0.1 mL: *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Candida albicans* [30]. The results are presented in Table 3.

Table 3. Microbiological emulsion test results (plate reading 24/48 h after inoculation).

Sample	Total Viable Microbiological Count, CFU/g	Total Viable Bacteria Count, CFU/g	Total Viable Yeast and Molds Count, CFU/g	Presence of Pathogenic Contaminants
Formulation 1 3% extract *Acmella oleracea* (F1A)	10	10	10	absent
Formulation 2 5% extract *Acmella oleracea* (F2A)	0	0	10	absent
Base (B1A)	0	0	0	absent

3.3.2. Analysis of Homogeneity of the Emulsion

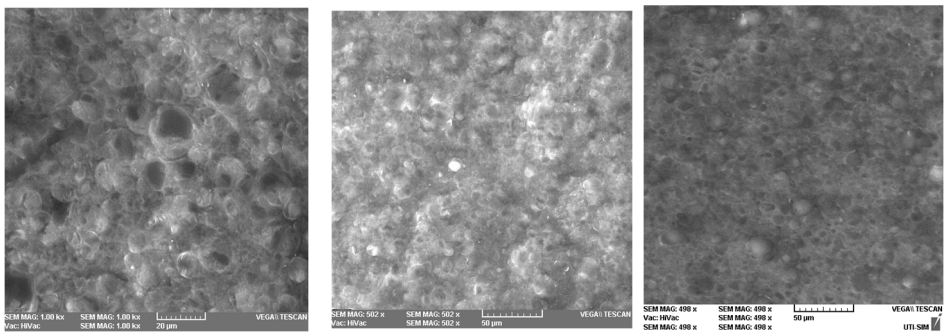
Emulsions are the ideal platform for the inclusion of a wide variety of important components in cosmetics, mainly because they allow the association of hydrophilic and hydrophobic active substances. Cosmetic emulsions are made differently than emulsions used in other sectors because, in addition to performing physicochemical tasks like washing, moisturizing, hydrating, and nourishing at the skin level, one of their main purposes is to provide pleasurable sensory experiences. Since most instability mechanisms and droplets smaller than 100 µm cannot be adequately studied by visual observation, microscopy is used to monitor the droplets that are invisible to the unaided eye and investigate the variables affecting the stability of the emulsion system. Emulsion characteristics at microscopic scale may influence the rheological behavior at macroscopic scale, affecting product stability [43]. The spread of particle-stabilized emulsions is depicted in Figure 3.



(**a**) (**b**) (**c**)

Figure 3. Optical microscopy image of studied emulsions. (**a**) Base (B1A), (**b**) Formulation with 3% extract *A. oleracea* (F1A), (**c**) Formulation with 5% extract *A. oleracea* (F2A).

The surface morphologies were also analyzed through scanning electron microscopy for the characterization of homogeneity and stability of the prepared emulsions with 3% and 5% extract *A. oleracea* together with the base used to obtain these emulsions. Furthermore, micrographs are presented for each emulsion sample, respectively. The images obtained at 10× and 20× magnifications are shown in Figure 4.



(**a**) (**b**) (**c**)

Figure 4. Scanning electron microscopy (SEM) of studied emulsions: (**a**) Base (B1A), (**b**) Formulation with 3% extract *A. oleracea* (F1A), (**c**) Formulation with 5% extract *A. oleracea* (F2A).

3.3.3. Rheological Tests

The Amplitude Sweep

The viscoelastic behavior of emulsions is distinguished by a solid-like character, manifested by a storage modulus G′ that exceeds the loss modulus G″ (Table 4).

Table 4. Dynamic moduli at linear viscoelastic strain.

Samples	Strain (ɣ = 0.1%)	
	G′ (Pa)	G″ (Pa)
B1A	417	86
F1A	671	159
F2A	773	187

The Frequency Sweep

At the linear viscoelastic value of amplitude, a frequency sweep was performed. In the entire frequency range, the storage modulus (G′) is observed to be larger than the loss modulus (G″) (Figure 5).

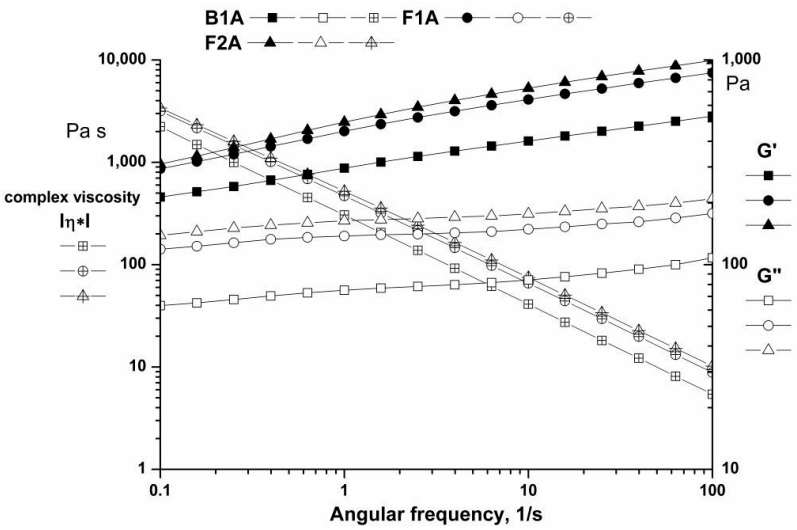


Figure 5. Frequency sweep. Notation: Base (B1A); Formulation with 3% extract *A. oleracea* (F1A); Formulation with 5% extract *A. oleracea* (F2A); η—viscosity.

Dynamic Temperature Sweep Tests

The dynamic moduli of emulsions decrease with an increase in temperature, but the storage modulus is always higher than the loss modulus over the entire temperature range due to the emulsifier agents.

Time Sweep Tests

Oscillatory time sweep tests were performed to evaluate the time stability of the emulsions (Figure 6).

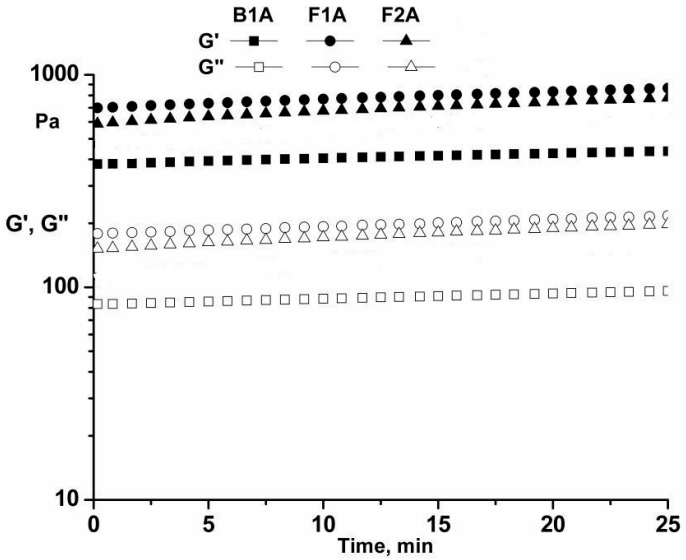


Figure 6. Time test for base (B1A) and emulsion F1A and F2A.

3.3.4. In Vitro Diffusion Test—Frant Cell Test

To investigate the application of emulsions containing 5% *A. oleracea* extract as the active ingredient by cutaneous route and to establish the necessary data for extending this study to real skin (in vivo test), preliminary permeation studies were developed using the diffusion methodology with Franz cells. Two separate methods, each with a specific meaning, are used to show the data (Figure 7):

1. The TPC value for the polyphenols released in the 5 mL receptor chamber is mg/mL mg;
2. The TPC value for the polyphenols' releasing speed is mg/mL/t.

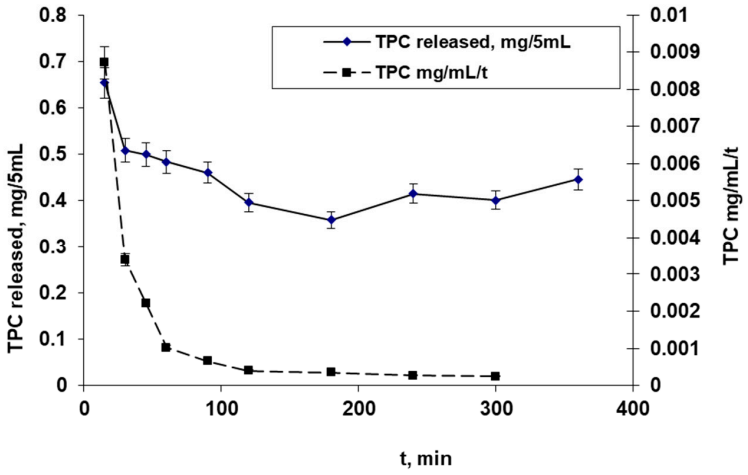


Figure 7. Evidencing the release of polyphenols through the chicken membrane in the case of the Franz cell diffusion test.

4. Discussion

4.1. Microbiological Control of Cosmetic Emulsions

All the tested samples (Table 3) are within the accepted range, proving the preservatives' efficiency. The assessment of all the steps implied in emulsion preparation safety and quality evidenced the emulsion's stability.

4.2. Analysis of Homogeneity of the Emulsion

The microstructure in Figure 3 showed no obvious variations in droplet sizes for the two proposed formulations (structure with slightly non-uniform particles, due to the addition of the extract and manual homogenization, but without cremation, flocculation or sedimentation phenomena). Polydisperse emulsions with droplets suspended in the continuous aqueous phase are observed.

The micrographs from Figure 4 confirm the homogeneity of the emulsions after the incorporation of the *A. oleraceae* extract, which was also observed in the case of the optical microscopy image from Figure 3.

4.3. Rheological Measurements

The Amplitude Sweep

The limit of the linear viscoelastic range (Table 4) was found to be 0.1% for all samples [44–47].

The frequency sweep indicates that the samples exhibit solid-like behavior with enhanced stability [44–46,48–50].

Dynamic temperature sweep tests. The slight decrease in dynamic moduli observed within the temperature range of 30–45 °C can be attributed to the beginning of the emulsion structuring [44].

Time sweep tests showed that the samples are stable over time, exhibiting a constant value for dynamic moduli. However, it should be noted that the absolute value of the dynamic moduli may gradually increases due to solvent evaporation [44].

Rheological tests have been demonstrated to be effective instruments for elucidating the mechanical properties of diverse materials and for the formulation of novel designs. Rheological analysis represents a valuable approach for formulating a time- and cost-saving strategy. The rheological results indicated that the emulsions exhibited solid-like behavior with time stability, which can be posited as the underlying cause of their self-storage stability.

4.4. In Vitro Diffusion Test—Frant Cell Test

The very high release rate can be clearly observed at the beginning of the release process (Figure 7) when the concentration gradient is at its maximum, after which it decreases to the minimum value in about 24 h. This behavior of the emulsion is similar to others even if their structural formula is totally different [33,34,51].

Without being a disadvantage, the overall effect remains low, which supports the aim of preparing a topical effect of the active ingredients without a high percutaneous absorption. The phenomenon of absorption involves overcoming the phase of penetration into the skin tissue, then the complete phase of permeation (the transport of molecules from one layer to another), and the migration of the active ingredients into systemic circulation through their absorption/uptake (lymph and blood). The interest in using a topical product in dermatocosmetics is closely linked to the local action at the level of the skin's own structures in the different areas (epidermis/surface or dermis/deep). The target of the antioxidant defense is aimed at the cellular and extracellular structures in both the epidermis and dermis, but should not bypass these areas by a complete transdermal transport. The effects of systemic absorption of active ingredients are far-reaching and require extensive and complex safety studies regarding the effects of both active ingredients and excipient molecules on the systems in which they are transported to eliminate the risk of a potentially harmful reaction.

The values of the basic characteristics for this new emulsion are based on the alcoholic extract of *A. oleracea* with the role of a natural active ingredient that falls within general product standards and can pass to the third stage of the study involving advanced research (toxicology, cytotoxicology and clinical studies) to determine its effectiveness.

5. Conclusions

Any potentially successful new dermatological formulation is developed based on a need or opportunity so that the end result adds value to either the consumer or the manufacturer. The chances of success increase when the benefits are multiple and reciprocal. The dermatocosmetic formulation at the center of this work, based on the bioactive extract of *A. oleracea*, simultaneously meets the multiple needs of patients through the protective action of the ingredients and through the comfort and pleasure of using the product. The bioactive ingredients have a very convenient source as they are obtained from cultivated plants (*A. oleracea*).

In addition, the active content of the *A. oleracea* extract simultaneously exerts biological effects that are highly valued in dermatocosmetics (antioxidant, wrinkle-reducing, anti-inflammatory) and it is worth every effort to take advantage of them.

In addition to avoiding the body's tendency to adapt and resist, the use of raw plant extracts results in a synergistic effect of the molecular qualities of interest, enhancing novelty and efficiency. In this sense, we applied physicochemical techniques to qualitatively and structurally characterize the dermatocosmetic O/W emulsion formulation with two different concentrations of *A. oleracea* extract (3% and 5%) and to evaluate its stability, microbiological control, rheological behavior and diffusion properties.

Good homogeneity, structural flexibility and strength, adequate skin diffusion, strong physico-chemical stability and microbiological stability were observed. The conclusions suggest that further in vivo research as well as toxicity, cytotoxicity and clinical studies are required to gather the necessary information for the approval of this dermatocosmetic formula.

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