cosmetics

Article

Unlocking Nature's Anti-Aging Secrets: The Potential of Natural Mineral Waters Combined with Plant Extracts in Cosmetics

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Abstract

Skin aging is influenced by intrinsic and extrinsic factors, leading to structural changes in the skin. Current anti-aging cosmetic trends emphasize innovative natural ingredients, including plant extracts, thermal waters, and botanical hydrolats. This work aims to develop six natural anti-aging cosmetics (two serums, two day creams, and two night creams) with innovative and non-irritating profiles. The rational design was guided by market analysis and ingredient properties. Prototype formulations were created with two core mixtures: (1) natural mineral water from Termas de Unhais da Serra and *Thymus* × *citriodorus* hydrolat and (2) natural mineral water from Termas de Chaves and aqueous *Vaccinium myrtillus* (blueberry) extract. The products were evaluated for stability (4 °C and 40 °C, 4 weeks), safety (according to EC Regulation No. 1223/2009), including in vitro testing for skin irritation potential (epiderm model; OECD TG 439). The market study highlighted a gap in anti-aging products combining natural extracts and thermal waters. All prototypes showed desirable textures and remained stable under test conditions. No irritating effects were observed. The results support the development of effective anti-aging cosmetics rooted in natural resources. These innovative products can meet the market demand for natural and sustainable skincare solutions.

Keywords: extracts; hydrolats; natural cosmetics; natural mineral waters; skin-aging

1. Introduction

Skin aging, also known as cutaneous aging, is a natural, multifactorial, and complex biological process influenced by intrinsic and extrinsic factors that lead progressively to a loss of structural integrity and physiological function of the skin [1,2]. Intrinsic aging, also called chronological aging, is driven primarily by genetic and hormonal changes leads to progressive thinning of the epidermis, dermis, and subcutaneous layers, as well as degeneration of collagen and elastin fibers, reduced melanin production, decreased sebum secretion, and structural atrophy. These changes manifest as fine lines, loss of elasticity, drier skin, altered hair growth, and increased skin fragility [3]. In contrast, photoaging—caused by chronic exposure to ultraviolet (UV) radiation and environmental stressors such as wind, cold, and pollution—accelerates the degradation of structural proteins, resulting in irregular thickening of the epidermis, uneven pigmentation, dilated blood vessels (telangiectasis), solar lentigines (age spots), and a higher risk of skin tumors [3]. Lifestyle options (nutrition, tobacco consumption, and sleep habits) also play an important role in the "skin aging exposome" [4]. Clinically, these two forms of aging can be distinguished by comparing sun-protected areas, such as the inner upper arm, which tend to retain a smoother and more youthful appearance, with sun-exposed areas like the back of the hand, where the skin is more wrinkled, pigmented, and less elastic [2,3,5]. Understanding the distinct mechanisms behind these aging processes is essential for developing effective preventative and corrective skincare strategies.

Skin aging is a multifaceted process influenced by oxidative stress, inflammation, and enzymatic degradation of the extracellular matrix [6,7]. Antioxidants play a crucial role in mitigating oxidative stress by neutralizing free radicals and boosting the skin's natural defenses [7,8]. Current cosmetic trends for aging focus on innovative ingredients and technologies aimed at addressing various signs of aging, including wrinkles, loss of elasticity, uneven pigmentation, and dullness [7]. Some key trends include ingredients such as retinoids, peptides, antioxidants, probiotics and/or prebiotics, hyaluronic acid, and plant-derived actives [9]. Plant extracts and botanicals, rich in diverse chemical constituents, have demonstrated promising antioxidant, anti-aging, anti-inflammatory, and whitening activities, making them attractive candidates for anti-aging cosmetic applications [6]. Thus, recent trends in anti-aging research projected the use of natural resources as core ingredients for cosmetics.

A growing awareness of environmental issues is fueling consumer interest in green and sustainable cosmetic products. This escalating demand is pushing the cosmetics industry to embrace sustainable practices, evident in the increasing adoption of bio-efficient ingredients, waste-derived extracts, circular economy models, and natural, biodegradable packaging. This trend reflects a broader shift towards aligning personal care choices with environmental values, as consumers seek products that minimize their ecological footprint [10].

Natural mineral waters (NMW, currently known as "thermal waters") are emerging as promising ingredients in anti-aging cosmetics due to their unique properties [11,12]. Rich in minerals and trace elements, they exhibit antioxidant and anti-inflammatory effects, combating key factors in skin aging [11–13]. Beyond their chemical composition, NMW also harbor unique microbial communities, whose bioactive compounds can potentially improve skin microbiome balance and overall skin health [12]. This combination of mineral-rich composition and beneficial microorganisms position NMW as a valuable asset in the development of innovative and effective anti-aging cosmetic formulations.

This study aimed to develop a range of natural anti-aging cosmetics that combine NMW's and natural extracts. Thus, three base products were developed: a serum, a day cream, and a night cream, in which the following combinations of natural ingredients have been incorporated: thermal water from Unhais da Serra (Portugal) combined with *Thymus × citriodorus* hydrolat and thermal water from Chaves (Portugal) combined with aqueous *Vaccinium myrtillus* (blueberry) extract.

2. Materials and Methods

2.1. Market Study

From November to December 2022, a market study was conducted on anti-aging serums, day creams, and night creams to evaluate the prevalence of natural mineral waters and natural extracts in their INCI compositions. The brands were selected based on a preliminary online search to identify those with anti-aging products available in the Portuguese market. The study was conducted on the brand's websites and in a Portuguese pharmacy. Anti-aging, lifting, hydration, and wrinkles claims were considered as keywords for product selection. In addition, the presence of thermal water and/or natural extracts in the analyzed products was considered an inclusion criterion. A price analysis of the direct competitors' products was also conducted and subsequently compared to the developed products.

2.2. Formulation Development

Before initiating the formulation process, a target product profile (TPP) was defined for each type of product to be developed (Table 1). This approach aimed to guide the rational design of the formulations according to the intended application, galenic form, packaging, use, and desired claims, as well as to ensure alignment with market expectations and regulatory requirements.

Table 1. Target product profile for serum, day cream, and night cream base formulas.

Attribute	Recommended Specification		
	Serum	**Day Cream**	**Night Cream**
Product name	Anti-aging hydrating serum	Anti-aging moisturizing day cream	Anti-aging regenerating night cream
Product application	Face	Face	Face
Galenic form and application	Gel, applied via dropper	Cream with creamy texture, applied by hand with gentle massage	Cream with soft and fluid texture, applied by hand with massage
Packaging	Dropper bottle (glass)	Jar (glass)	Pump bottle (glass)
Use (single or multiple)	Multiple use		
Production method	Cold-process	Emulsion prepared at low temperatures (<60 °C)	
Stability	Greater than 30 months		
Quality specifications	Challenge Test (CT)—Criteria A; Microbiological Quality (QM) according to ISO 17516:2014 [14]; Peroxide value: <10 meq O_2/kg		
Innovation	>90% natural ingredients; use of Portuguese NMW and plant extracts mixtures;		
Intended claims	Hydration, antioxidant, anti-inflammatory, anti-aging (fine lines/wrinkles)	Moisturizing, protective, film forming, antioxidant, anti-aging (fine lines/wrinkles)	Moisturizing, regenerative, protective, antioxidant, anti-aging (fine lines/wrinkles)
Recommended market	European (Natural and sustainable skincare, anti-aging segment)		

The rational design of the products was defined based on the market study and the information obtained from the literature, which covered in vitro studies with natural mineral waters, *Thymus × citriodorus* hydrolat, and aqueous blueberry extract bioactivities, including studies previously developed within our research group [15]. Based on these findings, particularly the ones reported in that study [15], two specific combinations of plant extracts and natural mineral waters were selected and incorporated into the prototype formulations developed in this study. In this study, the authors systematically evaluated the bioactive potential of various Portuguese endemic and native plant species and natural mineral waters, particularly focusing on anti-inflammatory, antioxidant, and anti-aging activities. Among the combinations tested, *Thymus* × *citriodorus* (used as a hydrolat) and *Vaccinium myrtillus* (used as an aqueous extract) were identified as the most promising plant ingredients. These were, respectively, combined with natural mineral waters from Unhais da Serra and Chaves, two thermal mineral waters recognized for their mineral richness and historical dermatological applications. The selection was guided by the demonstrated synergistic effects between the extracts and the mineral compositions of the waters, aiming to maximize efficacy while maintaining biocompatibility and safety. *Thymus* × *citriodorus* is a hybrid species widely cultivated in Portugal for its traditional medicinal uses, while *Vaccinium myrtillus* naturally occurs in high-altitude areas of the Iberian Peninsula, including northern Portugal, and is considered native to the region [15].

The ingredients used in the cosmetic formulations are in Table 2 and were selected based on information provided by the suppliers and material safety data sheets (MSDSs).

Thymus × *citriodorus* (batch: 08TC21) hydrolat was obtained from the Portuguese company Ervitas Catitas™. The aqueous extract was produced from *Vaccinium myrtillus* agroindustrial wastes at the Mountain Research Center (CIMO), Polytechnic Institute of Bragança (IPB). *Vaccinium myrtillus* aerial parts were collected in Baião, Portugal. The plant extracts were produced according to Plasencia et al. [16]. Briefly, 150 mL of ethanol/water 80:20 (v/v) was added to 1 g of freeze-dried sample and subjected to ultra-sound-assisted extraction (Qsonica, Q500, ultrasonic processor sonicator, 20 kHz, Newtown, CT, USA). The extraction was carried out for 10 min, with cycles of 30 s intercalated by pauses of 10 s, at 75% intensity of 500 W. An ice bath was used to avoid sample overheating. Following extraction, the samples were filtered and evaporated under reduced pressure (Büchi R-114, rotary evaporator; Büchi B-480, water bath, and Büchi B-721, vacuum controller system, Flawil, Switzerland) at 37 °C until ethanol was entirely removed. The aqueous extract was frozen, lyophilized, stored, and protected from light until use. The chemical composition of the extracts used in this study was previously characterized and published as part of the same research project [15]. *Thymus* × *citriodorus* is characterized by a high content of 1,8-cineole (43.9%), α-terpineol (21.1%), and borneol (11.5%), compounds known for their antimicrobial and anti-inflammatory properties. *Vaccinium myrtillus* is rich in caffeoylquinic acid derivatives (3-O- and 5-O-caffeoylquinic acid) and procyanidins (dimer and trimer), which exhibit strong antioxidant activity [15].

Two NMWs were used in this study, one from the central region of Portugal (Unhais da Serra) and one from the northern region (Chaves). Each NMW was collected from the boreholes of thermal centers in sterile collection bottles (500 mL; Deltalab, Barcelona), following the previously described procedure [17]. The samples were transported to the laboratory under the recommended conditions of the American Public Health Association (APHA), American Water Works Association (AWWA), and Water Environment Federation (WEF), described in Standard Methods for the Examination of Water and Wastewater (Washington, DC), and kept refrigerated in the dark until use [17].

Table 2. Ingredients used in the formulations (S—serum; DC—day cream; NC—night cream) and core ingredients mixtures (1—NMW from Unhais da Serra + *Thymus* × *citriodorus* hydrolate; 2—NMW from Chaves + aqueous blueberry extract).

INCI Name	Trade Name	Company	Country	Formulas
Natural mineral water from Termas de Unhais da Serra	-	Termas de Unhais da Serra	Portugal	S 1 DC 1 NC 1
Natural mineral water from Termas de Chaves	-	Termas de Chaves	Portugal	S 2 DC 2 NC 2
Aqueous *Vaccinium myrtillus* (blueberry) extract	-	CIMO, IPB	Portugal	S 2 DC 2 NC 2
Thymus × *citriodorus* hydrolat	-	Ervitas Catitas	Portugal	S 1 DC 1 NC 1
Pentylene glycol	A-Leen 5	Unigolden	Spain	S DC NC
Aqua/Water; Propanediol; Asparagopsis Armata extract	ASPAR'AGE	SEPPIC	France	
Xanthan gum	Goma Xantan	Acofarma	Spain	S DC NC
Allantoin	Alantoina	Acofarma	Spain	
Niacinamide	Nicotinamida EP	Escuder	Spain	S NC
Arachidyl alcohol; behenyl alcohol; arachidyl glucoside	Montanov 202	SEPPIC	France	DC NC
Isononyl isononanoate	Lanol 99	SEPPIC	France	NC
C15-19 alkane	Emogreen L15	SEPPIC	France	DC
Vitis Vinifera (Grape) Seed Oil	GSO—Douro Valley Grape Seed Oil	EFP Biotek	Portugal	S DC NC
Caprylic/capric Triglycerides	Caprilic capric trigliceridos EP	Escuder	Spain	
Tocopherol	Tocomix 50	AOM	Spain	
Polyacrylate-13, Polyisobutene, Polysorbate 20	Sepiplus 400	SEPPIC	France	NC
Polyacrylate Crosspolymer-6	Sepimax Zen	SEPPIC	France	DC
Acacia Senegal Gum; Xanthan Gum	Solagum AX	SEPPIC	France	DC NC
Xylitylglucoside; Anhydroxylitol; Xylitol	Aquaxyl	SEPPIC	France	
Glycerin	Glicerina Vegetal EP	Escuder	Spain	S NC
Phenoxyethanol	Sepicide LD	SEPPIC	France	S DC NC
Citric Acid	Acido citrico monohidrato cristal	Acofarma	Spain	

While a formal safety assessment was not performed, the concentrations of the selected cosmetic ingredients were validated through the calculation of the Margin of Safety (MoS), following the methodology recommended by the Scientific Committee on Consumer Safety (SCCS) [18]. The MoS was determined as the ratio between the No Observed Adverse Effect Level (NOAEL) and the Systemic Exposure Dose (SED), with a threshold value of MoS > 100 considered acceptable. This approach, in line with the principles outlined in Regulation (EC) No. 1223/2009, was used to support the safe use of each ingredient in the formulations [19]. Relevant toxicological and regulatory information was retrieved from authoritative sources, including the Cosmetic Ingredient Database (CosIng), Cosmetic Ingredient Review (CIR), and the SCCS Notes of Guidance.

Three base formulas were developed, which included a serum, a day cream, and a night cream to which two different combinations were added as core ingredients, namely NMW from Unhais da Serra and *Thymus* × *citriodorus* hydrolate (mixture 1) and NMW from Chaves and aqueous *Vaccinium myrtillus* (blueberry) extract (mixture 2). The serum was prepared at room temperature; however, and the day cream and night cream were prepared at low temperatures (<60 °C). The three formulas' pH value was adjusted to 5, respecting the recommended range of values for skin balance.

The development of the prototype formulations and the corresponding detailed production method are presented in Appendix A.

2.3. Stability

Preliminary stability was evaluated through the centrifugation test (3000 rpm, 30 min), temperature cycle test (4 °C and 40 °C, alternating for 24 h, overall period of 4 weeks), and by monitoring the organoleptic characteristics and pH over time (4 weeks). Stability was also assessed during 6 months at 4 °C and 40 °C. These tests were performed in cosmetic-grade glass packaging.

2.4. Rheological Characterization

For rheological characterization, properties were measured using a cone–plate viscometer (Brookfield DV3). Measurements were performed under controlled temperature conditions (25 °C ± 2 °C) for 1 min (>5 cone revolutions for each sample) with CP-52 spindle [20]. Two measurements per sample were performed: one at 24 h post-production and another at 4 weeks.

2.5. Skin Irritation Test

2.5.1. Reconstructed Human Epidermis EpiSkin®

Prototypes were tested for skin irritation in vitro through a three-dimensional (3D) reconstructed human epidermis (RHE) model (SkinEthic™) according to OECD test guideline No. 439 and to the supplier's protocol (EpiSkin™) [21]. A total of 6 cosmetic products were tested in triplicate.

2.5.2. Treatment Conditions

At the reception, SkinEthic™ tissues were thoroughly examined for abnormalities or excessive humidity. The tissues were transferred from the transport medium, and the excess agarose was cleaned with sterile gauze. The tissues were transferred to new wells with fresh maintenance culture medium and incubated in the same conditions overnight (18 h–24 h), according to the manufacturer's instructions (37 °C ± 2 °C, 5 ± 1% CO_2, ≥90% RH). After overnight incubation, the tissues were examined and transferred to fresh maintenance culture medium to be topically exposed to 16 ± 2 µL of sample, as well as positive and negative controls (SDS 5% and PBS without Ca^{2+} and Mg^{2+}, respectively).

After application, the exposure of the tissue to the sample and control was maintained for 42 ± 1 min inside the biological safety chamber. Three tissues were used for each condition (sample, negative control, positive control). After exposure, the tissues were rinsed with saline buffer solution (PBS without Ca^{2+} and Mg^{2+}), respecting the order of application, to ensure that all sample/control residues were removed. During the washing process, the plates were kept closed to avoid contamination.

After this step, the tissues were placed in wells containing maintenance culture medium and incubated for 42 h ± 1 h (37 °C ± 2 °C, 5 ± 1% CO_2, ≥90% RH), changing the maintenance culture medium 24 h after the start of incubation.

2.5.3. MTT Assay

The tissues were transferred to MTT medium (0.3 mL of 1 mg/mL of MTT in maintenance culture medium per well). The plates were incubated for 3 h ± 15 min (37 °C ± 2 °C, 5 ± 1%CO_2, ≥90% RH), protected from light. After this incubation, the tissues were transferred and the formazan crystals were extracted, placing the tissues in 0.75 mL of isopropanol and then adding another 0.75 mL of isopropanol to the top of each tissue.

The plates were hermetically sealed, covered with aluminum foil, and incubated at room temperature for at least 2 h with gentle stirring (approximately 120 rpm). After extraction, all tissues were perforated, allowing the mixing of the upper and lower isopropanol extracts placed in the tissue. The extracts were homogenized by pipetting. For each tissue, a duplicate measurement of optical density was performed at 570 nm ± 30 nm (aliquots of 200 μL).

2.5.4. Data Analysis

This method provides an in vitro procedure that can be used to identify the hazards of irritants and mixtures according to UN GHS Category 2. According to the UN GHS classification (UN, 2015), the potential for irritation of substances or mixtures is expected to distinguish between Category 2 substances (irritating to the skin) and unclassified substances (not irritating to the skin).

The potential for irritation of the substances is predicted by the average viability of the tissue exposed to the sample. The sample is considered "Category 2" if the average relative viability after 42 ± 1 min of exposure and 42 ± 1 h of post-incubation is equal to or less than 50% of the negative control and considered unclassified (not irritant to the skin).

2.5.5. Acceptance Criteria

For the test to be accepted, the quality criteria of the results are defined in the procedure and are related to the results obtained for the reference substances used as positive and negative control. These acceptance criteria are those described in DB-ALM Protocol No. 135: SkinEthic^{TM} Skin Irritation Test and according to each test system provider. The test is considered valid if these criteria are met. For the samples, general criteria are established for the observed standard deviation (SD) for the percentage of cell viability. The SD should be less than or equal to 18% for the test to be accepted.

3. Results

3.1. Market Study

In the market study, 20 product brands were considered. To understand the prevalence of thermal waters and natural extracts in the anti-aging skincare market, we analyzed the INCI lists of 92 serums, 117 day creams, and 85 night creams (Table 3). This allowed us to categorize the products based on the presence of thermal water, natural extracts, or both.

Among 92 serums, only 16 products (17.39%) had natural mineral water in their constitution, 63 products (68.48%) had natural extracts in their composition, and 3 products (3.26%) combined a natural mineral water and a natural extract. Furthermore, among 117 day creams, only 24 products (20.51%) had natural mineral water in their constitution, 13 products (11.11%) had natural extracts, and 6 products (5.13%) combined natural mineral water and a natural extract. Lastly, among the 85 night creams, only 19 products (22.35%) had natural mineral water, 13 products (15.29%) had natural extracts, and 5 products (5.88%) combine a thermal water and a natural extract (Figure 1). All brands using NMW were from France and all products combining NMWs and natural extracts belonged to a sole brand (Avène). In total, only 12 products among 294 were formulated with this specific combination of natural ingredients.

The most used ingredients in all products studied were glycerin, hyaluronic acid, niacinamide, retinol, vegetable oils, biomimetic peptides, alpha and beta hydroxy acids, and provitamins E and C. Furthermore, the analysis of the INCI lists allowed us to understand that day creams mostly had light creamy textures, night creams had rich creamy textures and serums had fluid or liquid textures, depending on the product.

The cost analysis for the developed products considered factors like raw material costs, production processes, and packaging. The final retail prices of the developed serums are between EUR 8.00 and EUR 8.21, while for the day and night creams, they range from EUR 13.00 to EUR 13.40. In comparison to market competitors, the developed serums offer a highly competitive price point, with their retail prices significantly lower than the lowest price of EUR 30.90 found in the marketed serums. The day and night creams also present a clear price advantage, with prices ranging from EUR 13.00 to EUR 13.40, compared to the competitors' prices of EUR 27.65 to EUR 44.46. These prices make the developed products highly competitive in the market.

Table 3. Market serums, day creams, and night creams evaluated according to the presence of natural mineral water (NMW) and/or plant extracts in their formulation. The presence in each ingredient category was classified as N or Y (N—No; Y—Yes).

Class of Ingredients		Analyzed Products			Brand
Thermal Water	Natural Extract	Serums (N)	Day Creams (N)	Night Creams (N)	
N	Y	4	3	2	Caudalíe (France)
N	Y	4	8	8	Clarins (France)
N	Y	3	9	4	Eucerin (Germany)
N	Y	9	5	4	Filorga (France)
N	Y	3	3	1	Isdin (Spain)
N	Y	4	6	6	Lierac (France)
N	Y	-	3	3	Noreva (France)
N	Y	5	6	5	Nuxe (France)
N	Y	-	1	1	Rilastil (Italy)
N	Y	5	4	5	Sensilis (Spain)
N	Y	21	11	4	Sesderma (Spain)
N	Y	2	6	5	Shiseido (Japan)
Y	N	2	4	5	Bioderma (France)
Y	N	4	5	3	La Roche Posay (France)
Y	N	1	4	3	Uriage (France)
Y	N	6	5	5	Vichy (France)
N	N	7	9	10	Institut Esthederm (France)
N	N	3	8	6	Martiderm (Spain)
N	N	2	-	-	Noreva (France)
N	N	3	-	-	Rilastil (Italy)
N	N	1	2	2	Topicrem (France)
Y	Y	3	6	3	Avéne (France)
	TOTAL	92	117	85	

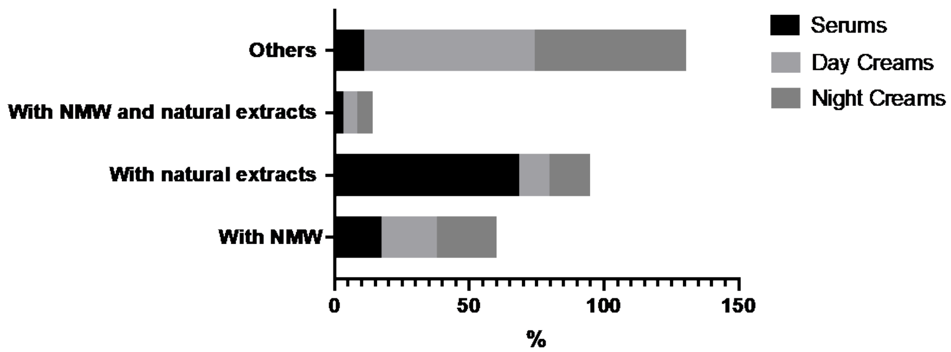

Figure 1. Marketed serums, day creams, and night creams. Percentage of commercial formulations containing natural mineral water (NMW), natural extracts, both, or neither.

3.2. Formulation' Development

A range of products was designed to meet the needs of skin aging and respond to the high demand for natural and sustainable products on the market. A serum, a day cream, and a night cream were developed as base formulations in which the two mixtures of NMW and natural extract were introduced. Two combinations of core ingredients were considered, mixture 1 combined natural mineral water from Termas de Unhais da Serra and *Thymus × citriodorus* hydrolat, while mixture 2 combined natural mineral water from Termas de Chaves and aqueous blueberry extract. The choice of these combinations were based on in vitro tests with these ingredients, which revealed antioxidant and anti-inflammatory properties relevant for their introduction into anti-aging cosmetic products [16,22–24]. In addition to these bioactive ingredients, other humectant ingredients, emollients and vegetable oils were selected that complemented the action of the previous ones and attributed the desired galenical and sensorial characteristics to the product. It is important to note that for all commercial ingredients included, their natural or vegetable origin was privileged. Different formulas were tested to achieve the ideal appearance, texture, and performance. The final prototypes presented a homogeneous appearance, and a slight odor typical of the *Thymus × citriodorus* hydrolat, for mixture 1 versions.

The serum formulation consists of an easy-to-spread gel capable of providing a refreshing effect, with moisturizing and protective action. This action is provided by ingredients that mimic the skin's natural hydration factor, such as niacinamide and glycerin. Furthermore, the serum also has an active ingredient (ASPAR'AGE®), common to all products developed, which minimizes fine lines and wrinkles, helping the skin to regain volume [25]. This serum was designed for dropper packaging, given its highly fluid and spreadable texture. Regarding its organoleptic characteristics, the serum with mixture 1 has a fluid texture, white color, and the characteristic odor of *Thymus × citriodorus* hydrolat, while the serum with mixture 2 has a similar texture and color to the previous one, but without a characteristic odor (Figure 2). The serums have 95% of natural ingredients in their composition (Table 4).

Figure 2. Serum formulations containing mixture 1 (**left**) and mixture 2 (**right**).

Table 4. Serum's ingredients.

Serum (Mixture 1) Ingredients	Serum (Mixture 2) Ingredients
Aqua (Water), Glycerin, Niacinamide, Caprylic/Capric Triglyceride, Propanediol, Asparagopsis Armata Extract, Pentylene Glycol, Vitis Vinifera (Grape) Seed Oil, Phenoxyethanol, Xanthan Gum, *Vaccinium myrtillus* (blueberry) Extract, Citric Acid, Tocopherol.	Aqua (Water), Glycerin, Niacinamide, Propanediol, Asparagopsis Armata Extract, Caprylic/Capric Triglyceride, Thymus Citriodorus (Lemon Thyme) Hydrolat, Pentylene Glycol, Vitis Vinifera (Grape) Seed Oil, Phenoxyethanol, Xanthan Gum, Citric Acid, Tocopherol.

The day cream formulation consists of an easy-to-spread creamy emulsion, capable of providing a refreshing effect, with a moisturizing and protective action. This moisturizing and protective action is provided by ingredients that mimic the skin's natural hydration factor and skin-replenishing lipids, thus restoring balance and improving the skin's barrier function. Furthermore, the day cream also has ASPAR'AGE®. This cream was designed for jar packaging, given the creamy texture it presents. Its texture, combined with the ingredients included, also guarantees a film-forming effect, which will ensure greater protection for the skin during the day when exposed to environmental factors. Mixture 1 day cream has a creamy texture, white color, and characteristic odor of *Thymus × citriodorus* hydrolat. Mixture 2 also has a creamy texture and white color; however, it does not have any characteristic odor (Figure 3). The day creams have 97.5% of natural ingredients in their composition (Table 5).

Figure 3. Day creams formulations containing mixture 1 (**left**) and mixture 2 (**right**).

Table 5. Day creams ingredients.

Day Cream (Mixture 1) Ingredients	Day Cream (Mixture 2) Ingredients
Aqua (Water), C15-19 Alkane, Caprylic/Capric Triglycerides, Xylitylglucoside, Anhydroxylitol, Xylitol, Arachidyl Alcohol, Behenyl Alcohol, Arachidyl Glucoside, Propanediol, Asparagopsis Armata Extract, Vitis Vinifera (Grape) Seed Oil, Thymus Citriodorus (Lemon Thyme) hydrolate, Allantoin, Pentylene Glycol, Polyacrylate Crosspolymer-6, Phenoxyethanol, Acacia Senegal Gum, Xanthan Gum, Citric Acid, Tocopherol.	Aqua (Water), C15-19 Alkane, Caprylic/Capric Triglycerides, Xylitylglucoside, Anhydroxylitol, Xylitol, Propanediol, Asparagopsis Armata Extract, Arachidyl Alcohol, Behenyl Alcohol, Arachidyl Glucoside, Vitis Vinifera (Grape) Seed Oil, Pentylene Glycol, Allantoin, Polyacrylate Crosspolymer-6, Phenoxyethanol, Acacia Senegal Gum, Xanthan Gum, *Vaccinium myrtillus* (blueberry) Extract, Citric Acid, Tocopherol.

The night cream formulation consists of a creamy and fluid emulsion that is easy to spread, capable of providing a refreshing effect, with moisturizing, protective, and regenerating action. These moisturizing and protective actions are provided by ingredients that mimic the skin's natural hydration factor and skin-replenishing lipids, thus restoring balance and improving the skin's barrier function. Furthermore, the night cream also has ASPAR'AGE® that minimizes fine lines and wrinkles, helping the skin to regain volume. This night cream was designed for pump packaging, given its fluid texture, to allow the product to be applied with massage at night and provide users with a moment of self-care. This product in mixture 1 has a fluid texture, white color, and the characteristic odor of *Thymus × citriodorus* hydrolat, while in mixture 2, it does not have a characteristic odor but maintains the texture and color properties (Figure 4). The night creams have 91.3% of natural ingredients in their composition (Table 6).

Figure 4. Night creams formulations containing mixture 1 (**left**) and mixture 2 (**right**).

Table 6. Night creams ingredients.

Night Cream (Mixture 1) Ingredients	Night Cream (Mixture 2) Ingredients
Aqua (Water), Caprylic/Capric Triglycerides, Isononyl Isononanoate, Vitis Vinifera (Grape)Seed Oil, Arachidyl Alcohol, Behenyl Alcohol, Arachidyl Glucoside, Glycerin, Propanediol, Asparagopsis Armata Extract, Niacinamide, Xylitylglucoside, Anhydroxylitol, Xylitol, Thymus Citriodorus (Lemon Thyme) Water, Allantoin, Pentylene Glycol, Phenoxyethanol, Polyacrylate-13, Polyisobutene, Polysorbate 20, Acacia Senegal Gum, Xanthan Gum, Citric Acid, Tocopherol	Aqua (Water), Caprylic/Capric Triglycerides, Isononyl Isononanoate, Vitis Vinifera (Grape)Seed Oil, Arachidyl Alcohol, Behenyl Alcohol, Arachidyl Glucoside, Glycerin, Propanediol, Asparagopsis Armata Extract, Niacinamide, Xylitylglucoside, Anhydroxylitol, Xylitol, Allantoin, Pentylene Glycol, Phenoxyethanol, Polyacrylate-13, Polyisobutene, 9,Polysorbate 20, Acacia Senegal Gum, Xanthan Gum, *Vaccinium myrtillus* (Blueberry) Extract, Citric Acid, Tocopherol

3.3. Stability

All prototypes in their final versions, with mixtures 1 and 2, were stable under centrifugation (30 min at 3000 rpm) and temperature cycles (4 °C and 40 °C, 4 weeks), having also been subjected to prolonged stability (4 °C and 40 °C, 6 months) (Table 7).

Table 7. Stability, organoleptic characteristics, and pH overtime of final formulas.

	Preliminary Stability (Centrifugation 30 min at 3000 rpm)	Temperature Cycles (4 °C and 40 °C, 4 Weeks)	Stability (4 °C and 40 °C, 6 Months)	Organoleptic Characteristics	pH
Serum 1	Stable	No phase separation or color change. Maintain characteristic odor.	Stable Slight odor attenuation; no significant changes.	Texture—fluid Color—transparent Odor—mixture 1 characteristic odor	Initial pH = 5.0 pH (4 weeks) = 5.2 pH (6 months) = 5.2
Serum 2	Stable	No phase separation or color change	Stable No significant changes, maintain organoleptic characteristics.	Texture—fluid Color—transparent Odor—odorless	Initial pH = 5.0 pH (4 weeks) = 5.1 pH (6 months) = 5.1
Day cream 1	Stable	No phase separation or color change. Maintain characteristic odor.	Stable Slight odor attenuation; no significant changes.	Texture—creamy Color—white Odor—mixture 1 characteristic odor	Initial pH = 5.0 pH (4 weeks) = 5.3 pH (6 months) = 5.3
Day cream 2	Stable	No phase separation or color change.	Stable No significant changes, maintain organoleptic characteristics.	Texture—creamy Color—white Odor—odorless	Initial pH = 5.0 pH (4 weeks) = 5.2 pH (6 months) = 5.2
Night cream 1	Stable	No phase separation or color change. Maintain characteristic odor.	Stable Slight odor attenuation; no significant changes.	Texture—fluid cream Color—white Odor—mixture 1 characteristic odor	Initial pH = 5.0 pH (4 weeks) = 5.1 pH (6 months) = 5.2
Night cream 2	Stable	No phase separation or color change.	Stable No significant changes, maintain organoleptic characteristics.	Texture—fluid cream Color—white Odor—odorless	Initial pH = 5.0 pH (4 weeks) = 5.2 pH (6 months) = 5.3

3.4. Rheological Characterization

The viscosity of semisolids is a key parameter for performance. Through rheological characterization, serum was classified as a non-Newtonian, shear-thinning (formerly named pseudoplastic) fluid with non-thixotropic behavior. The day cream was classified as a non-Newtonian, shear-thinning fluid with thixotropic behavior (positive hysteresis area). The night cream was classified as a non-Newtonian, shear-thinning fluid with thixotropic behavior (positive hysteresis area). The rheological characterization of mixture 1 prototypes is shown in Figure 5, and mixture 2 prototypes in Figure 6.

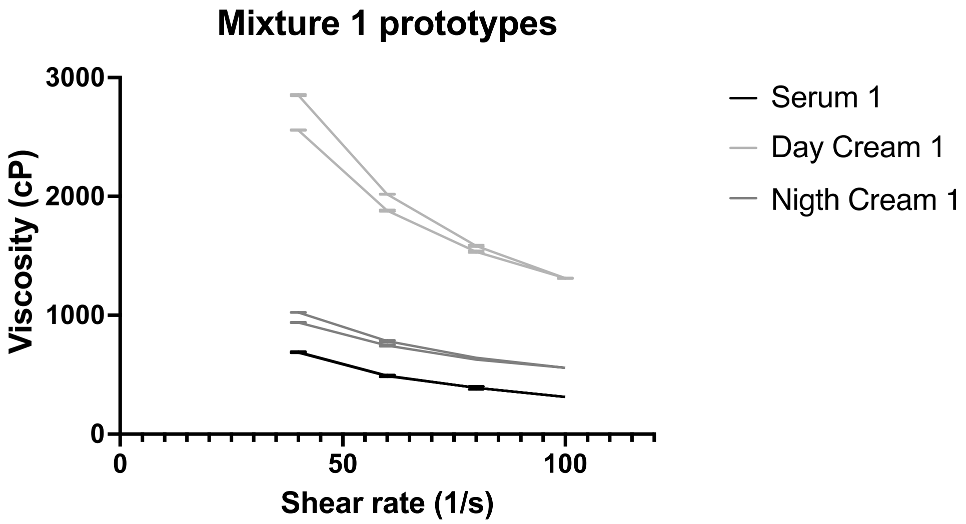

Figure 5. Viscosity (cP) as a function of shear rate (1/s) for the serum, day cream, and night cream corresponding to mixture 1. Each data point represents the mean of two independent measurements ± standard deviation (SD), where the SD corresponds to the difference between the two measurements performed per sample (after production and after 4 weeks).

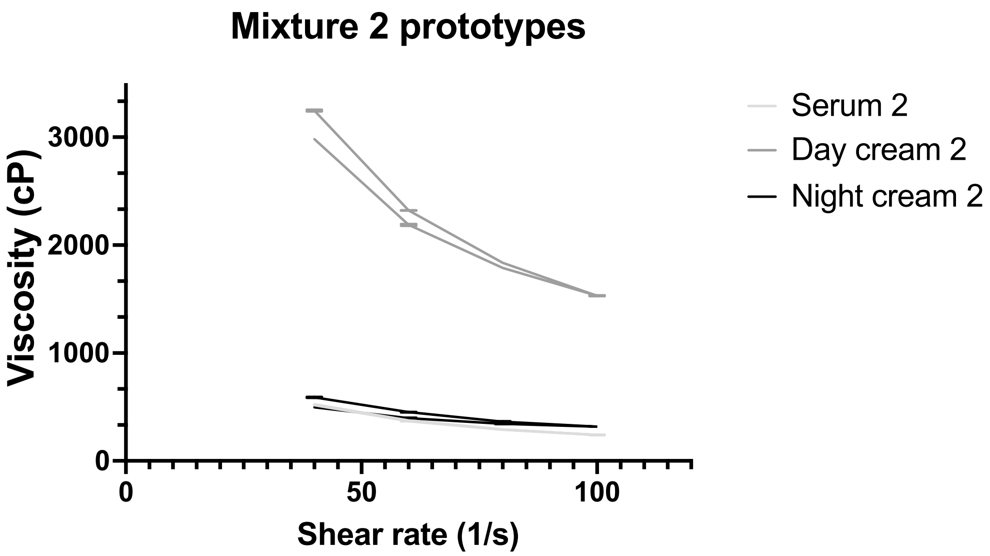

Figure 6. Viscosity (cP) as a function of shear rate (1/s) for the day cream 1 and 2, corresponding to mixture 1 and 2, respectively. Each data point represents the mean of two independent measurements ± standard deviation (SD), where the SD corresponds to the difference between the two measurements performed per sample (after production and after 4 weeks).

As can be seen in Figures 5 and 6 depending on the mixture of core ingredients, the rheological properties of the products are changed. In the case of serums, mixture 1 has the highest viscosity, while in day creams, it is mixture 2, and in night creams, it is mixture 1 that has the highest viscosity values. However, it is worth highlighting that the rheological profile is maintained, with all non-Newtonian, shear-thinning fluids. It is important to highlight that despite the differences in measured viscosity, the perceived viscosity and the sensory characteristics associated with the texture do not undergo relevant changes. Despite the observed variations in rheological properties across different core ingredient mixtures (as seen in Figures 5 and 6), visual inspection of the figures suggests that these differences are not substantial. Moreover, there are no significant apparent changes within products of the same category.

3.5. Skin Irritation Test

The obtained results (Figure 7) for the cosmetics understudy show that the samples are not irritating, since the average percentage of tissue viability is well above 50% in all assays. The standard deviation obtained for the replicas of the samples was below the limit of acceptance of the variation defined as 18%. In conclusion, the developed cosmetics proved to be non-irritating.

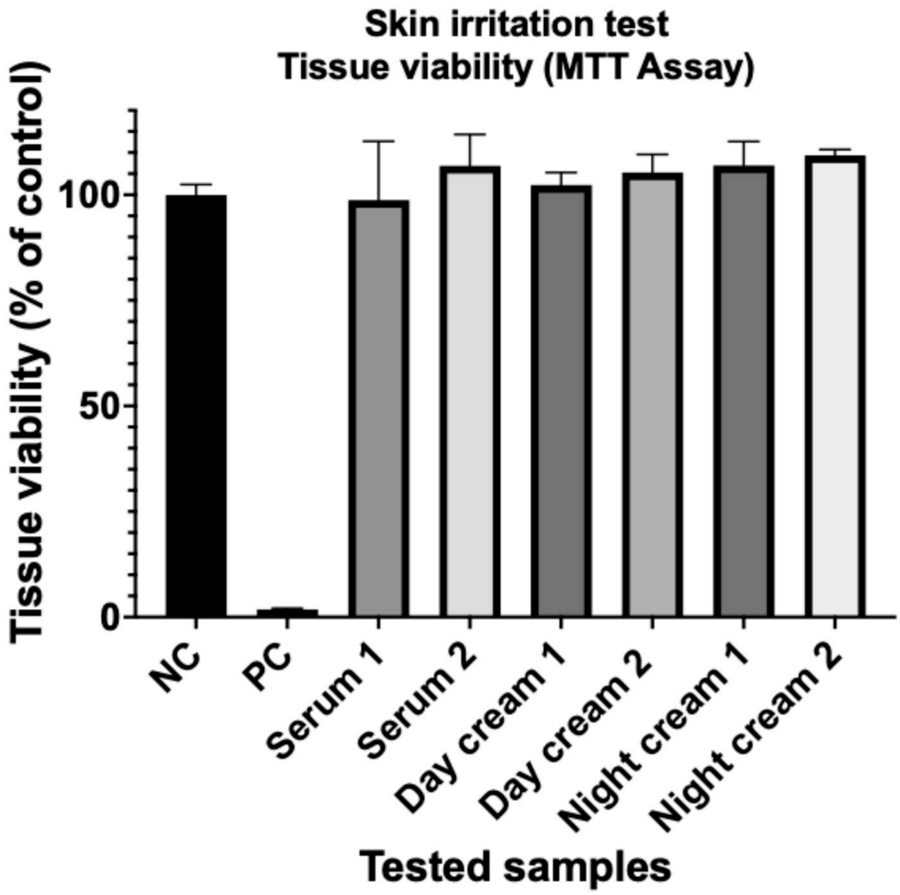

Figure 7. Viability of reconstructed human epidermis (RhE) after exposure to the samples, assessed by MTT assay according to OECD TG 439. Average values obtained for tissue viability of negative control (NC—PBS without Ca^{2+} and Mg^{2+}), positive control (PC—SDS 5%), and samples/prototypes (serum 1; serum 2; day cream 1; day cream 2; night cream 1; night cream 2). The bars represent (n = 3 tissues), and the lines represent the standard deviation (SD).

4. Discussion

When discussing the development of natural creams designed to decrease the severity of skin aging, it is important to consider both the biological and socio-cultural aspects of this process [26,27]. Skin aging involves structural changes like collagen loss and wrinkle formation, coupled with dysregulation of cellular functions. However, perceptions of aging are also heavily influenced by societal beauty standards and generational attitudes [27]. Therefore, successful natural creams should not only address the biological factors, such as oxidative stress and inflammation, but also align with consumer preferences for sensoriality and perceived effectiveness.

Plants and their extracts have been used in cosmetic and pharmaceutical applications since ancient times. Despite the increasing cosmetic innovation in the manufacture of synthetic ingredients, the current trend is the search for green cosmetics with natural ingredients and more sustainable final products [6,28].

The development of natural anti-aging cosmetics is experiencing significant growth, driven by increasing consumer demand for more sustainable products [29]. Plant extracts are central to this trend, offering a wide array of benefits thanks to their inherent bioactive compounds [30]. As highlighted by Xie et al. [29], consumers favor natural extract cosmetics for their safety and efficacy, seeking natural, organic, efficient, and sustainable skincare options. The efficacy of these extracts stems from their ability to address multiple aging pathways, including moisturizing, barrier repair, anti-oxidation, sun screening, whitening, and anti-inflammatory effects [29]. H.C. Siavash and F.A.A. Majid support the incorporation of plant extracts to reduce or delay the process of skin aging, as well as the maintenance of healthy skin by the addition of phytochemicals [28]. This multi-targeted approach is crucial in combating the complex nature of skin aging.

The market reflects this shift, with a growing preference for formulations containing botanicals with DNA-protecting, enzyme-inhibiting, and inflammation-reducing properties [31]. Ferreira et al. note that *Vitis vinifera* (grape seed) oil, *Butyrospermum parkii* butter, and glycine soja are among the most frequently used species in anti-aging products, likely due to their polyphenol content [31]. The use of plant extracts aligns with the broader trend of incorporating natural ingredients, like NMWs, known for their mineral content and potential benefits like anti-inflammatory and antioxidant properties [30].

The variability in plant extract composition poses a major challenge for natural cosmetics due to factors like plant genetics, environment, and processing. To ensure product consistency, manufacturers must implement stringent quality control measures, including standardized sourcing, detailed chemical analysis, and optimized extraction methods. This holistic approach to cosmetic development, which emphasizes safety, sustainability, multifaceted efficacy, and addressing formulation challenges, is poised to shape the future of anti-aging skincare [29].

We began this work by evaluating the market for anti-aging cosmetic products, focusing on the presence of natural ingredients in their composition. This market study highlights the potential for developing innovative skincare products that combine natural extracts and thermal waters, two sectors of natural ingredients, addressing both consumer preferences and sustainability trends. The detailed analysis of textures and ingredients provides a solid foundation for creating formulations that meet market demands while promoting green beauty practices. Considering the small number of marketed products aimed at skin aging that combine natural extracts and thermal waters, it is pertinent to develop new innovative products in this segment, aligned with current sustainability and green cosmetics trends [10,31]. Furthermore, the analysis allowed us to design the three base formulations objectively, having developed a day cream base with a fresh, creamy texture, a night cream with a light creamy texture for nighttime face massage, and a serum with a fluid texture that is quickly absorbed.

Chaves thermal water, sourced from northern Portugal, has been valued for its medicinal properties since Roman times. Its appeal as a cosmetic ingredient lies in its near-neutral pH and low mineralization. The water exhibits anti-inflammatory properties by significantly reducing IL-6 levels in cells exposed to urban pollution [22]. Additionally, it improves skin barrier integrity by reducing transepidermal water loss in human volunteers. Therefore, Chaves' thermal water shows promise as a beneficial cosmetic ingredient for skin health [22].

The mineral natural water from Unhais da Serra, situated in the Beira Interior region of Portugal, possesses a distinctive composition compared to other thermal waters in the region. This uniqueness arises from the mineralogical composition of the geological formations through which the water flows. The geological variability in Portugal contributes to the diversity of thermal waters, each with its own set of properties [24]. The therapeutic indications of thermal waters in the Beira Interior region (where Unhais da Serra is located) for respiratory, rheumatic, and musculoskeletal systems are related to the presence of sulfur and bicarbonate (sulfur in some thermal waters can have an analgesic effect, and bicarbonate ions can help remove inflammation). Unhais da Serra thermal water does not have a specified dermatologic therapeutic effect [24]. However, thermal waters, in general, have demonstrated different effects on the skin, from cellular renewal, skin hydration, recovery of cutaneous barrier, and keratolytic effects to antimicrobial activity, detergent properties, antioxidant capacity, and anti-inflammatory activity [12].

Thymus × *citriodorus* is an emerging aromatic and medicinal hybrid plant with relevant bioactive potential [23]. Historically, *Thymus* species have been employed to address gastrointestinal, respiratory, and skin disorders, highlighting its versatility [32]. The essential oil derived from lemon thyme stands out as a potent antiseptic, boasting deodorant and disinfectant properties, largely attributed to its antimicrobial activity [32]. *Thymus* × *citriodorus* also possesses anti-inflammatory and antioxidant activities [23]. Furthermore, *Thymus* extracts have demonstrated potential as regulators of oily skin and hair, showcasing their potential in anti-acne applications and stimulating effect against the morphological changes in elastic fibers induced by aging [23,32].

Discussing the potential of *Vaccinium myrtillus* (blueberry) extract in various industries reveals its promise as a source of valuable bioactive molecules. Extracts from blueberry aerial parts have demonstrated both antioxidant and anti-tyrosinase capacity, suggesting potential applications in cosmetics for anti-aging and skin-brightening formulations. Furthermore, the antibacterial activity observed in these extracts, comparable to ampicillin, positions them as potential natural preservatives for both the cosmetic and food industries, aligning with the growing demand for natural agents [16].

The inclusion of natural mineral waters from specific sources (Termas de Unhais da Serra and Termas de Chaves) and natural extracts (*Thymus* × *citriodorus* hydrolate and aqueous *Vaccinium myrtillus* (blueberry) extract) reflects an innovative approach to harnessing natural resources with proven benefits. The antioxidant and anti-inflammatory properties of these combinations are particularly relevant for anti-aging products [16,22,24,32]. In addition, the choice to prioritize natural or vegetable-origin ingredients aligns with current trends in sustainability and green cosmetics. This not only caters to consumer demand but also promotes eco-friendly practices in product development. However, although the antioxidant activity of the selected extracts and their combinations has been previously confirmed [15], the lack of direct assessment of oxidative stress biomarkers in cellular or tissue models represents a limitation of the present work. Given the central role of oxidative stress in skin aging—contributing to cellular senescence, collagen degradation, and reduced skin elasticity—future studies should include mechanistic evaluations to better elucidate the anti-aging potential of the formulations [15].

Each product was designed to address specific skincare needs. The properties of the ingredients and the market knowledge in the cosmetic sector led us to design six innovative and sustainable cosmetics (two serums, two day creams, and two night creams). Textural and organoleptic properties are essential attributes in the consumer's choice between cosmetics, which is why we developed three base formulations based on a market study that could make a difference to the consumer [33].

The attention to the sensory properties (texture, appearance, odor, and feel on the skin) ensures that the products are pleasant to use, which is crucial for consumer satisfaction. A positive sensorial experience can significantly enhance a product's appeal, leading to repeat purchases and brand loyalty [33]. The differentiation in the packaging (dropper for serum, jar for day cream, and pump for night cream, all in glass) reflects thoughtful design tailored to the product's use and texture. The high content of natural ingredients (>90%) addresses the actual trends [34].

All prototypes remained stable under centrifugation at 3000 rpm for 30 min. This indicates that the formulations can withstand mechanical stress without phase separation, which is critical for product consistency during manufacturing and transportation. Stability tests at temperature cycles of 4 °C and 40 °C for 4 weeks show that the formulations can endure varying storage conditions. This reflects the robustness of the emulsions, ensuring that they remain effective and visually appealing across different climates and storage environments. Stability over 6 months at both 4 °C and 40 °C is a good indicator for the long-term durability of the products. This long-term stability is essential for shelf-life assurance, giving consumers confidence in the product's efficacy over time. Although this study sustains placement in the market in these early stages, laboratorial confirmation of the shelf life shall be performed through long-term stability studies.

The rheological characterization of serums, day creams, and night creams reveals that while the viscosity and behavior under shear vary with different core ingredient mixtures, the overall rheological profiles remain consistent. All products are classified as non-Newtonian, meaning their viscosity changes with the applied shear rate. This property is desirable in skincare products as it ensures they spread easily under the shear forces of application, providing a smooth and even layer on the skin. Thixotropic fluids exhibit a decrease in viscosity over time under constant shear, followed by a gradual recovery when the shear is removed. Day and night creams show thixotropic behavior, as indicated by the positive hysteresis area in the graphs. This property is beneficial for creams as it allows them to remain stable and thick in the container but become more fluid when applied, aiding in spreading and absorption. Serums, on the other hand, are non-thixotropic, meaning their viscosity returns to its original state immediately after the shear force is removed. This characteristic ensures consistent texture and performance during application. These findings underscore the importance of rheological properties in designing skincare products that perform well and provide a consistent and satisfying user experience [35]. The ability to maintain consistent sensory characteristics despite variations in viscosity is particularly noteworthy, highlighting the careful balance achieved in these formulations.

The skin irritation test results, showing that the cosmetics are non-irritating with tissue viability well above 50%, complement the previous findings on stability, rheological properties, and safety. This comprehensive evaluation confirms that the developed serum, day cream, and night cream are not only stable and effective but also safe for consumers. These attributes make them well-suited for the market, particularly in addressing the demand for natural and non-irritating skincare solutions. The combined insights underscore the robustness of the formulations, ensuring they meet both performance and safety standards, ultimately leading to high consumer trust and satisfaction. Moreover, the use of 3D skin models enables the assessment of the final formulations in conditions that more closely mimic human skin, which is not feasible with traditional monolayer in vitro assays.

The developed products, with up to 97.6% natural ingredients, offer innovative textures and formulations, such as the use of Portuguese natural mineral waters and plant extracts. This, combined with their competitive pricing, positions the products as a promising market alternative, offering effective, sustainable, and affordable skincare solutions.

5. Conclusions

The growing demand for natural, sustainable, and effective anti-aging skincare products, driven by biological needs and evolving socio-cultural perceptions of aging, leads us to this comprehensive approach to developing anti-aging skincare products that meet the demands for natural, effective, and enjoyable skincare solutions.

By combining specific thermal waters and natural extracts with proven skincare ingredients, the formulations provide targeted benefits while promoting sustainability. The use of *Thymus* × *citriodorus* hydrolat and *Vaccinium myrtillus* aqueous extract assures multifunctional properties, including antioxidant, anti-inflammatory, antibacterial, and anti-aging effects. These effects are expected to be synergistically enhanced by the inclusion of Portuguese thermal waters from Chaves and Unhais da Serra, which offer hydration, skin barrier repair, and soothing properties.

The detailed consideration of texture, packaging, and ingredient synergy underscores the thoughtful and innovative approach to product development. The stability and safety findings enhance the earlier insights, confirming that the innovative formulations not only fill a market gap but also meet rigorous quality standards. By integrating thermal waters and natural extracts and ensuring their stability and safety, these products are well-positioned to appeal to consumers seeking effective, sustainable, and reliable skincare solutions. This comprehensive approach underscores the potential for these products to achieve significant market success, catering to the increasing consumer demand for effective, natural, and non-irritating skincare products.

The formulation of cosmetics with these endogenous Portuguese products underscores the feasibility of creating high-performance skincare products rooted in local natural resources while meeting EU regulatory standards.

Future work should focus on validating the safety and efficacy of the formulations through the challenge test, in vivo studies, and exploring interactions with the skin microbiome. Future studies should focus on evaluating oxidative stress markers and assessing the antioxidant capacity of the final formulations, using more appropriate methodologies such as cell-free assays or in vivo models to elucidate the anti-aging potential of these products. Furthermore, assessing the impact of the formulations on skin condition—particularly in terms of hydration, elasticity, and overall health—will be a crucial part of future evaluations. These studies will provide valuable insights into the cosmetic efficacy of the formulations and contribute to a better understanding of their full potential for enhancing skin condition. Sustainability will remain a key focus, with prioritization of new upcycled raw materials and product line expansion to address different needs. Furthermore, integrating consumer feedback in the future could refine sensorial qualities and packaging, ensuring the final products meet both functional and emotional expectations, ultimately leading to more targeted, sustainable, and scientifically grounded skincare solutions.

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Appendix A.

This appendix provides detailed information on the formulation evolution of the six cosmetic products developed in this work. Each table presents the composition of the main development versions, including modifications in ingredients and their concentrations, which reflect the iterative optimization process based on stability.

Appendix A.1. Serum Formulation Evolution, Final Formula, and Productive Method

The serum formulations were developed in two main versions, corresponding to mixture 1 and mixture 2 of core ingredients. Both serums were prepared using a cold process, with sequential addition of phases, and were mixed using a helical stirrer (Heidolph RZR 2041, Heidolph Instruments GmbH & Co., Schwabach, Germany). The third and final version represents the final product, as documented throughout this work.

In Table A1, the three versions developed with mixture 1 are detailed. Modifications were implemented based on observations from temperature cycling tests. Specifically, tocopherol (an antioxidant) was added to address oxidation observed in the initial version. Subsequently, pentylene glycol was incorporated as a preservative booster due to an odor alteration noted during temperature cycling of the second version.

Table A1. Serum 1 formula evolution.

Phase	Ingredient (Name/INCI)	V01	V02	V03
		%	%	%
A	NMW Unhais da Serra	81.55	81.50	80.00
	Thymus × citriodorus hidrolate	3.00	3.00	3.00
	Glycerin	5.00	5.00	5.00
	Pentylene Glycol	-	-	1.50
B	Xantan Gum	0.80	0.80	0.80
	Caprylic/capric Triglycerides	2.50	2,.50	2.50
	Vitis Vinifera (Grape) Seed Oil	1.00	1.00	1.00
C	Aqua/Water; Propanediol; Asparagopsis Armata Extractt	2.00	2.00	2.00
	Niacinamide	3.00	3.00	3.00
	Tocopherol	-	0.05	0.05
	Phenoxyethanol	1.00	1.00	1.00
D	Citric acid	0.15	0.15	0.15

The versions developed with mixture 2 are detailed in Table A2. Based on observations during the development of the prototype with mixture 1, tocopherol was incorporated from the outset in the development of the mixture 2 prototype. Although no oxidation or color alteration was observed during the initial temperature cycling of this prototype, an odor alteration was detected. This led to the incorporation of pentylene glycol in the second and final version of this product.

Table A2. Serum 2 formula evolution.

Phase	Ingredient (Name/INCI)	V01	V02
		%	%
A	NMW Chaves	84.30	82.75%
	Aqueous *Vaccinium myrtillus* (blueberry) extract	0.25	0.25
	Glycerin	5.00	5.00
	Pentylene Glycol	-	1.50
B	Xantan Gum	0.80	0.80
	Caprylic/capric Triglycerides	2.50	2.50
	Vitis Vinifera (Grape) Seed Oil	1.00	1.00
C	Aqua/Water; Propanediol; Asparagopsis Armata Extractt	2.00	2.00
	Niacinamide	3.00	3.00
	Tocopherol	0.05	0.05
	Phenoxyethanol	1.00	1.00
D	Citric Acid	0.10	0.15

Appendix A.2. Day Cream Formulation Evolution, Final Formula, and Productive Method

The day cream formulations were developed in two main versions, corresponding to mixture 1 and mixture 2 of core ingredients. Both day creams were prepared at low temperatures (<60 °C) and mixed using a helical stirrer (Heidolph RZR 2041, Heidolph Instruments GmbH & Co., Schwabach, Germany). Phases A and B were heated (55 °C). Phases C and A were sequentially added to Phase B. Then Phases D, E, and F were added to the previous mixture. Allantoin was incorporated at the end of the production process, once the product had cooled to room temperature.

The day cream versions developed with mixture 1 are detailed in Table A3. An initial version of this formulation was prepared without the ingredient ASPAR'AGE (Aqua/Water; Propanediol; Asparagopsis Armata Extract). Primarily for economic reasons, the stability of the prototype was first assessed using the core ingredients of mixture 1. Following the completion of temperature cycling without observed alterations, ASPAR'AGE was then incorporated.

The day cream versions developed with mixture 2 are detailed in Table A4. For the versions developed with mixture 2 of core ingredients, the same strategy was employed, with the only variations being in the concentrations of NMW and extract relative to the version with mixture 1.

Table A3. Day cream 1 formula evolution.

Phase	Ingredient (Name/INCI)	V01	V02
		%	%
A	NMW Unhais da Serra	71.3	69.15
	Thymus × *citriodorus* hidrolate	3.00	3.00
	Pentylene Glycol	1.00	1.00
	Aqua/Water; Propanediol; Asparagopsis Armata Extractt	-	2.00
B	Arachidyl Alcohol; Behenyl Alcohol; Arachidyl Glucoside	3.00	3.00
	C15-19 Alkane	7.50	7.50
	Vitis Vinifera (Grape) Seed Oil	2.00	2.00
	Caprylic/capric Triglycerides	5.50	5.50
	Tocopherol	0.05	0.05

Table A3. *Cont.*

Phase	Ingredient (Name/INCI)	V01	V02
		%	%
C	Polyacrylate Crosspolymer-6	1.00	1.00
	Acacia Senegal Gum; Xanthan Gum	0.50	0.50
D	Xylitylglucoside; Anhydroxylitol; Xylitol	3.00	3.00
E	Phenoxyethanol	1.00	1.00
F	Citric Acid	0.15	0.30
G	Allantoin	1.00	1.00

Table A4. Day cream 2 formula evolution.

Phase	Ingredient (Name/INCI)	V01	V02
		%	%
A	NMW Chaves	74.05	72.90
	Aqueous *Vaccinium myrtillus* (blueberry) extract	0.25	3.00
	Pentylene Glycol	1.00	1.00
	Aqua/Water; Propanediol; Asparagopsis Armata Extractt	-	2.00
B	Arachidyl Alcohol; Behenyl Alcohol; Arachidyl Glucoside	3.00	3.00
	C15-19 Alkane	7.50	7.50
	Vitis Vinifera (Grape) Seed Oil	2.00	2.00
	Caprylic/capric Triglycerides	5.50	5.50
	Tocopherol	0.05	0.05
C	Polyacrylate Crosspolymer-6	1.00	1.00
	Acacia Senegal Gum; Xanthan Gum	0.50	0.50
D	Xylitylglucoside; Anhydroxylitol; Xylitol	3.00	3.00
E	Phenoxyethanol	1.00	1.00
F	Citric Acid	0.15	0.30
G	Allantoin	1.00	1.00

Appendix A.3. Night Cream Formulation Evolution, Final Formula, and Productive Method

The night cream formulations were developed in two main versions, corresponding to mixture 1 and mixture 2 of core ingredients. Similarly to day creams, night creams were prepared at low temperatures (<60 °C) and mixed using a helical stirrer (Heidolph RZR 2041, Heidolph Instruments GmbH & Co., Schwabach, Germany). Phases A and B were heated (55 °C). Phase C was added to Phase B. Phase A was stirred and subsequently incorporated into the previously prepared mixture under continuous agitation. Then Phases D and E were added to the mixture. Allantoin was incorporated at the end of the production process, once the product had cooled to room temperature.

The night cream versions developed with Mixture 1 are detailed in Table A5. An initial version of this formulation was prepared without the ingredient ASPAR'AGE (Aqua/Water; Propanediol; Asparagopsis Armata Extract) for the same reasons as in day creams. The initial prototype was formulated without Solagum AX (Acacia Senegal Gum; Xanthan Gum). However, owing to insufficient viscosity, Solagum AX was incorporated in the second version to improve the rheological profile of the formulation. Additionally, the glycerin concentration was increased to enhance skin hydration and contribute to the formulation's humectant capacity.

Table A5. Night cream 1 formula evolution.

Phase	Ingredient (Name/INCI)	V01	V02	V03
		%	%	%
A	NMW Unhais da Serra	69.85	64.35	66.35
	Thymus × *citriodorus* hidrolate	3.00	3.00	3.00
	Pentylene Glycol	1.00	1.00	1.00
	Aqua/Water; Propanediol; Asparagopsis Armata Extractt	-	-	2.00
	Niacinamide	1.50	1.50	1.50
B	Arachidyl Alcohol; Behenyl Alcohol; Arachidyl Glucoside	3.00	3.00	3.00
	Isononyl Isononanoate	5.00	5.00	5.00
	Vitis Vinifera (Grape) Seed Oil	3.00	3.00	3.00
	Caprylic/capric Triglycerides	6.00	6.00	6.00
	Tocopherol	0.05	0.05	0.05
C	Polyacrylate-13, Polyisobutene, Polysorbate 20	0.8	0.80	0.80
	Acacia Senegal Gum; Xanthan Gum	-	0.50	0.50
D	Xylitylglucoside; Anhydroxylitol; Xylitol	1.50	1.5	1.5
	Glycerin	3.00	4.00	4.00
	Sepicide LD	1.00	1.00	1.00
E	Citric Acid	0.30	0.30	0.30
F	Allantoin	1.00	1.00	1.00

The night cream versions developed with mixture 2 are detailed in Table A6. The same formulation strategy was adopted for the development of night cream 1, with the only variation being the composition of the core ingredients mixture.

Table A6. Night cream 2 formula evolution.

Phase	Ingredient (Name/INCI)	V01	V02	V03
		%	%	%
A	NMW Chaves	72.60	67.10	69.10
	Aqueous *Vaccinium myrtillus* (blueberry) extract	0.25	0.25	0.25
	Pentylene Glycol	1.00	1.00	1.00
	Aqua/Water; Propanediol; Asparagopsis Armata Extractt	-	-	2.00
	Niacinamide	1.50	1.50	1.50
B	Arachidyl Alcohol; Behenyl Alcohol; Arachidyl Glucoside	3.00	3.00	3.00
	Isononyl Isononanoate	5.00	5.00	5.00
	Vitis Vinifera (Grape) Seed Oil	3.00	3.00	3.00
	Caprylic/capric Triglycerides	6.00	6.00	6.00
	Tocopherol	0.05	0.05	0.05
C	Polyacrylate-13, Polyisobutene, Polysorbate 20	0.8	0.80	0.80
	Acacia Senegal Gum; Xanthan Gum	-	0.50	0.50
D	Xylitylglucoside; Anhydroxylitol; Xylitol	1.50	1.5	1.5
	Glycerin	3.00	4.00	4.00
	Sepicide LD	1.00	1.00	1.00
E	Citric Acid	0.30	0.30	0.30
F	Allantoin	1.00	1.00	1.00

References

1. Tobin, D.J. Introduction to Skin Aging. *J. Tissue Viability* **2017**, *26*, 37–46. [CrossRef] [PubMed]
2. Shai, A. (Ed.) *Handbook of Cosmetic Skin Care*, 2nd ed.; Series in cosmetic and laser therapy; Informa Healthcare: London, UK, 2009.
3. Fisher, G.J.; Kang, S.; Varani, J.; Bata-Csorgo, Z.; Wan, Y.; Datta, S.; Voorhees, J.J. Mechanisms of Photoaging and Chronological Skin Aging. *Arch. Dermatol.* **2002**, *138*, 1462–1470. [CrossRef] [PubMed]

4. Krutmann, J.; Bouloc, A.; Sore, G.; Bernard, B.A.; Passeron, T. The Skin Aging Exposome. *J. Dermatol. Sci.* **2017**, *85*, 152–161. [CrossRef] [PubMed]
5. Gilchrest, B.A.; Krutmann, J. *Skin Aging*; Springer: Berlin, Germany, 2006.
6. Chaiyana, W.; Charoensup, W.; Sriyab, S.; Punyoyai, C.; Neimkhum, W. Herbal Extracts as Potential Antioxidant, Anti-Aging, Anti-Inflammatory, and Whitening Cosmeceutical Ingredients. *Chem. Biodivers.* **2021**, *18*, e2100245. [CrossRef]
7. He, X.; Wan, F.; Su, W.; Xie, W. Research Progress on Skin Aging and Active Ingredients. *Molecules* **2023**, *28*, 5556. [CrossRef]
8. Budzianowska, A.; Banaś, K.; Budzianowski, J.; Kikowska, M. Antioxidants to Defend Healthy and Youthful Skin—Current Trends and Future Directions in Cosmetology. *Appl. Sci.* **2025**, *15*, 2571. [CrossRef]
9. Choi, H.Y.; Lee, Y.J.; Kim, C.M.; Lee, Y.-M. Revolutionizing Cosmetic Ingredients: Harnessing the Power of Antioxidants, Probiotics, Plant Extracts, and Peptides in Personal and Skin Care Products. *Cosmetics* **2024**, *11*, 157. [CrossRef]
10. Amberg, N.; Fogarassy, C. Green Consumer Behavior in the Cosmetics Market. *Resources* **2019**, *8*, 137. [CrossRef]
11. Figueiredo, A.C.; Rodrigues, M.; Mourelle, M.L.; Araujo, A.R.T.S. Thermal Spring Waters as an Active Ingredient in Cosmetic Formulations. *Cosmetics* **2023**, *10*, 27. [CrossRef]
12. Mourelle, M.L.; Gómez, C.P.; Legido, J.L. Hydrobiome of Thermal Waters: Potential Use in Dermocosmetics. *Cosmetics* **2023**, *10*, 94. [CrossRef]
13. Silva, A.; Oliveira, A.S.; Vaz, C.V.; Correia, S.; Ferreira, R.; Breitenfeld, L.; Martinez-de-Oliveira, J.; Palmeira-de-Oliveira, R.; Pereira, C.M.F.; Palmeira-de-Oliveira, A.; et al. Anti-Inflammatory Potential of Portuguese Thermal Waters. *Sci. Rep.* **2020**, *10*, 22313. [CrossRef] [PubMed]
14. *ISO 17516:2014*; Cosmetics—Microbiology—Microbiological Limits. International Organization for Standardization: Geneva, Switzerland, 2014.
15. Gomes, C.P.; Oliveira, A.S.; Rolo, J.; Da Silveira, T.F.F.; Palmeira De Oliveira, R.; Alves, M.J.; Plasencia, P.; Palmeira De Oliveira, A. Natural Mineral Water–Plant Extract Combinations as Potential Anti-Aging Ingredients: An In Vitro Evaluation. *Cosmetics* **2025**, *12*, 113. [CrossRef]
16. Plasencia, P.; Heleno, S.A.; Finimundy, T.; Carocho, M.; Calhelha, R.C.; Añibarro-Ortega, M.; Alves, M.J.; Oludemi, T.; Quidiongo, N.; Barreiro, F.; et al. Recovery of High Valuable Bioactive Molecules from *Vaccinium myrtillus* L. Bioresidues. *Waste Biomass Valorization* **2023**, *14*, 2873–2884. [CrossRef]
17. American Public Health Association; American Water Works Association; Water Environment Federation. *Standard Methods for the Examination of Water and Wastewater*, 20th ed.; APHA: Washington, DC, USA, 1999.
18. Scientific Committee on Consumer Safety (SCCS). *SCCS Notes of Guidance for the Testing of Cosmetic Ingredients and Their Safety Evaluation*; SCCS/1647/22; 12th Revision; European Commission: Brussels, Belgium, 2023; Available online: https://health.ec.europa.eu/publications/sccs-notes-guidance-testing-cosmetic-ingredients-and-their-safety-evaluation-12th-revision_en (accessed on 15 May 2025).
19. Regulation (EC) No 1223/2009 of the European Parliament and of The Council of 30 November 2009 on Cosmetic Products. 2009. Available online: https://health.ec.europa.eu/system/files/2016-11/cosmetic_1223_2009_regulation_en_0.pdf (accessed on 5 May 2021).
20. Machado, A.; Cerca, N. Influence of Biofilm Formation by *Gardnerella vaginalis* and Other Anaerobes on Bacterial Vaginosis. *J. Infect. Dis.* **2015**, *212*, 1856–1861. [CrossRef]
21. OECD. *Test No. 439: In Vitro Skin Irritation: Reconstructed Human Epidermis Test Method*; OECD: Paris, France, 2021. [CrossRef]
22. Pinto-Ribeiro, I.; Castro, C.; Rocha, P.E.; Carvalho, M.J.; Pintado, A.; Mendes, A.; Pedrosa, S.S.; Capeto, P.; Azevedo-Silva, J.; Oliveira, A.L.S.; et al. Chaves Thermal Spring Water Impact on Skin Health: Potential Cosmetic Application. *Appl. Sci.* **2024**, *14*, 7911. [CrossRef]
23. Oliveira, A.S.; Cavaleiro, C.; Martinez-de-Oliveira, J.; Palmeira-de-Oliveira, A. *Thymus* × *citriodorus*: An Emerging Aromatic and Medicinal Hybrid Plant with Relevant Bioactive Potential. *Rev. Bras. Farmacogn.* **2023**, *33*, 1089–1109. [CrossRef]
24. Araujo, A.R.T.S.; Sarraguça, M.C.; Ribeiro, M.P.; Coutinho, P. Physicochemical Fingerprinting of Thermal Waters of Beira Interior Region of Portugal. *Environ. Geochem. Health* **2017**, *39*, 483–496. [CrossRef]
25. Resende, D.I.S.P.; Ferreira, M.; Magalhães, C.; Sousa Lobo, J.M.; Sousa, E.; Almeida, I.F. Trends in the Use of Marine Ingredients in Anti-Aging Cosmetics. *Algal Res.* **2021**, *55*, 102273. [CrossRef]
26. Russell-Goldman, E.; Murphy, G.F. The Pathobiology of Skin Aging. *Am. J. Pathol.* **2020**, *190*, 1356–1369. [CrossRef]
27. McEwen, L.; Déchelette, C.; Fauverghe, S. Anthropological Study on the Perception of Skin Aging and Aesthetic Procedures: An International, Generational Analysis. *Ethics Med. Public Health* **2025**, *33*, 101109. [CrossRef]
28. Chermahini, S.H.; Majid, F.A.A.; Sarmidi, M.R. Cosmeceutical Value of Herbal Extracts as Natural Ingredients and Novel Technologies in Anti-Aging. *J. Med. Plants Res.* **2011**, *5*, 3074–3077.
29. Xie, M.; Jiang, Z.; Lin, X.; Wei, X. Application of Plant Extracts Cosmetics in the Field of Anti-Aging. *J. Dermatol. Sci. Cosmet. Technol.* **2024**, *1*, 100014. [CrossRef]
30. Michalak, M. Plant Extracts as Skin Care and Therapeutic Agents. *Int. J. Mol. Sci.* **2023**, *24*, 15444. [CrossRef] [PubMed]

31. Ferreira, M.; Magalhães, M.; Oliveira, R.; Sousa-Lobo, J.; Almeida, I. Trends in the Use of Botanicals in Anti-Aging Cosmetics. *Molecules* **2021**, *26*, 3584. [CrossRef]
32. Oliveira, A.S.; Rolo, J.; Gaspar, C.; Cavaleiro, C.; Salgueiro, L.; Palmeira-de-Oliveira, R.; Ferraz, C.; Coelho, S.; Pastorinho, M.R.; Sousa, A.C.; et al. Chemical Characterization and Bioactive Potential of *Thymus* × *citriodorus* (Pers.) Schreb. Preparations for Anti-Acne Applications: Antimicrobial, Anti-Biofilm, Anti-Inflammatory and Safety Profiles. *J. Ethnopharmacol.* **2022**, *287*, 114935. [CrossRef]
33. Spence, C.; Zhang, T. Multisensory Contributions to Skin-cosmetic Product Interactions. *Int. J. Cosmet. Sci.* **2024**, *46*, 833–849. [CrossRef]
34. Ahmed, I.A.; Mikail, M.A.; Zamakshshari, N.; Abdullah, A.-S.H. Natural Anti-Aging Skincare: Role and Potential. *Biogerontology* **2020**, *21*, 293–310. [CrossRef]
35. Kim, J.; Jeong, E.H.; Baik, J.H.; Park, J.D. The Role of Rheology in Cosmetics Research: A Review. *Korea-Aust. Rheol. J.* **2024**, *36*, 271–282. [CrossRef]