

# QUALITY ASSESSMENT OF HANDMADE SOAPS, PLANT-BASED OILS, AND SKINCARE CREAMS: PHYSICOCHEMICAL AND MICROBIOLOGICAL ANALYSIS

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**ABSTRACT.** The aim of this study was to evaluate the quality and safety of handmade cosmetic and hygiene products through physicochemical and microbiological determinations, prior to their placement on the market. Three types of samples were analyzed, namely: solid soaps, creams, and cosmetic oils, using standardized methodologies compliant with current European and international regulations. The results demonstrated variations in pH, moisture levels, total alkali and Total Fatty Matter (TFM) content and viscosity, as well as quality indices in the case of oils, alongside the absence of pathogenic microorganisms, with detection occurring in only one sample. Additionally, Gas chromatograph coupled with mass spectrometer (GC-MS) and Fourier transform infrared spectroscopy (FTIR) analyses were performed on representative samples. These findings emphasize the necessity of continuous monitoring and quality assessment of cosmetic products to ensure consumer safety.

**Keywords:** *handmade cosmetic products, microbiological analysis, total alkali, peroxide index, acidity index*

## INTRODUCTION

Cosmetics and personal care products have long been used to cleanse, protect, and hydrate the skin. Soaps, creams, ointments, and vegetable oils remain key categories in topical care. Recently, interest in

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handmade and natural products has increased, driven by a preference for bioactive ingredients and avoidance of synthetic compounds [1,2].

Microbial contamination is a persistent concern, arising from raw material impurities, production conditions, or poor hygiene [3,4]. Microbiological control is essential to ensure safety throughout a product's shelf life [5,6]. Contamination can reduce both efficacy and safety [7], especially for products applied to sensitive skin.

Topical product quality depends on physicochemical properties and microbiological safety. For soaps, important indicators include pH, alkali content, moisture, and Total Fatty Matter (TFM) [8,9]. A pH close to skin's natural value helps maintain the stratum corneum and prevents irritation or atopic conditions [10–12]. Despite this, pH is rarely indicated on packaging.

Creams and ointments are semisolid forms with cosmetic and therapeutic uses. Their pH should match that of healthy skin (5.4–5.9) to preserve the microbiota and barrier function [13–15]. Viscosity also plays a key role in formulation stability and user experience.

Cosmetic oils are evaluated by parameters such as density, acid and peroxide values, which affect their stability and therapeutic potential [16]. Since cosmetics often support microbial growth and are not sterile, contamination with bacteria like *Pseudomonas aeruginosa*, *Staphylococcus aureus*, or filamentous fungi remains a concern [7,18–20].

This study aimed to evaluate the physicochemical and microbiological quality of various cosmetic products, including soaps, creams, ointments, and oils, using standardized methods aligned with current regulations. The antimicrobial activity of selected samples was tested in vitro on Gram-positive bacteria.

The novelty of this research lies in its multidisciplinary evaluation of cosmetic products, combining physicochemical parameters, chromatographic techniques, and microbiological assessments to establish a comprehensive quality profile prior to their market release.

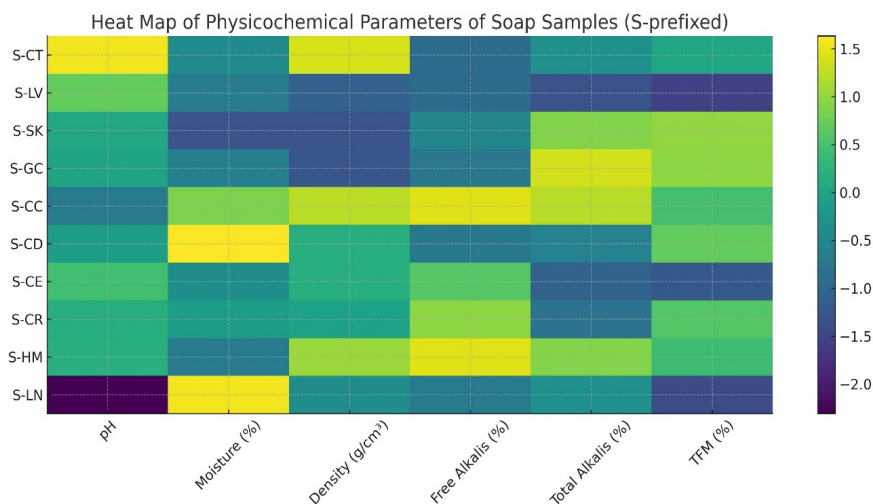
## RESULTS AND DISCUSSION

### 1. *Physicochemical analyses*

#### *Analysis of Handmade Solid Soaps*

The physicochemical analysis of ten handmade solid soap samples is presented in Figures 1–4. The evaluated soap properties: pH, moisture and volatile matter, density, free alkalis, total alkalis, and TFM are presented in the normalized heat map in Figure 1.

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**Figure 1.** Normalised Heat Map for Soap Properties

S-SK, S-GC, S-CD, S-CR, and S-HM show high normalized TFM scores, indicating that they are the richest in fatty matter.

S-CC and S-HM present high normalized alkali values, which may suggest increased skin harshness if not properly buffered.

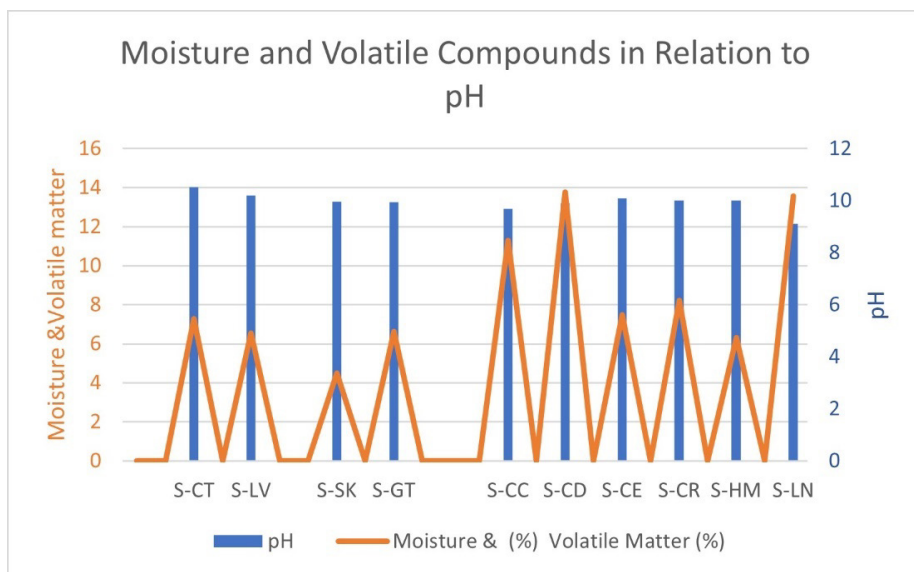
S-LN displays low normalized pH and alkali levels, reflecting a comparatively milder and more skin-friendly formulation.

S-LV, S-CE, and S-LN cluster with lower normalized TFM and alkali scores, suggesting lighter soaps that may be less emollient.

The **pH values** of the analyzed soaps ranged between **9.10 and 10.51**, which is slightly alkaline compared to the natural skin pH (~5.4–5.9). This alkalinity may potentially disrupt the skin's acid mantle if used frequently, increasing the risk of irritation [10,12]. We considered a classification of soaps as follows: pH < 6 acidic, pH= 7 neutral and pH> 8 alkaline. In our study, we have found that all of the samples had a pH value between 9.1 and 10.51, they are alkaline soaps, not very skin-friendly. In Romania [2] have reported pH values for commercial soaps between 9 and 11, consistent with alkaline formulations.

**Moisture** content varied from **4.5% to 13.78%**, reflecting differences in formulation and drying processes. Some values are lower than, while others are similar to, those reported in previous studies in Romania [2]. High moisture content can lead to hydrolysis of unsaponified fats, forming free fatty acids and glycerol, which affects soap hardness and quality.

Considering that moisture and volatile components contribute to the integrity, storage stability, and sensory profile of solid soaps, Figure 2 explores their variation relative to pH across the studied samples.



**Figure 2.** Relationship Between Moisture, Volatile Compounds, and pH

Based on the experimental data, the pH values showed a relatively narrow variation range (9.1–10.51), indicating good alkaline stability across all formulations. In contrast, moisture and volatile matter content exhibited substantial variability (4.5–13.78%), suggesting marked differences in composition and water retention capacity between samples.

A general inverse tendency was observed: formulations with higher pH values tended to present lower moisture and volatile content (e.g., S-SK, S-GT), whereas samples with slightly lower pH showed markedly higher moisture levels (e.g., S-LN, S-CD, S-CC). This suggests a potential negative correlation between pH and moisture/volatile content, indicating that higher alkalinity may be associated with reduced water affinity or volatile retention within the formulations.

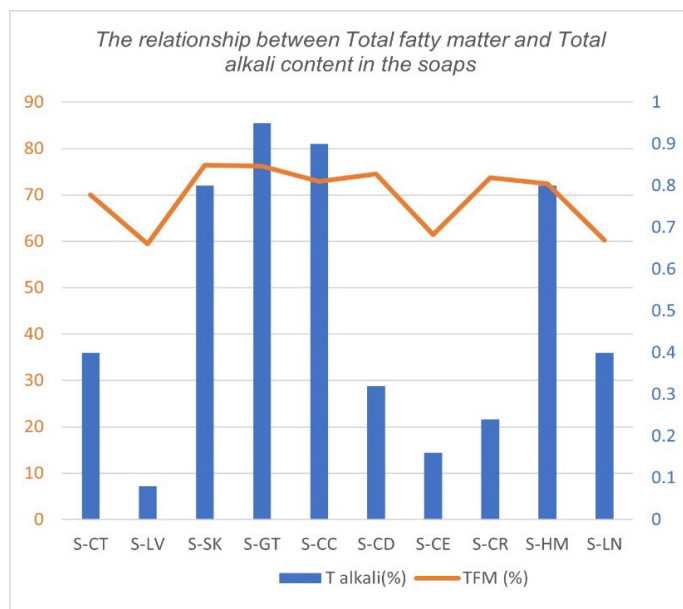
**Density** values ranged from **1.019 to 1.17 g/cm<sup>3</sup>**, consistent with typical handmade soap characteristics.

The **free and total alkali content** remained low in most samples, indicating an adequate saponification process. According to ISO standards, total alkali levels in high-quality soaps should be below 2%, while the Bureau of Indian Standards (BIS) recommends less than 5% for premium soaps [ 21]. High total alkali may indicate excess NaOH, ensuring complete saponification, whereas low total alkali can suggest incomplete reaction or insufficient fatty acids.

**Total Fatty Matter (TFM)**, a key indicator of soap quality, ranged from **59.4% to 76.4%**. Total fatty matter (TFM) reflects the richness and quality of soap. Soaps with higher TFM provide better hardness, lather, and moisturizing properties. Low TFM is associated with poor quality, lower hardness, and reduced consumer satisfaction [22]. In the case of the manufacturers sometimes prefer a small excess of NaOH to ensure complete saponification, obtaining a soap with a high TFM.

Notably, soaps with added natural oils or butters (e.g., green tea and charcoal, calendula, and citronella & rosemary) presented higher TFM values, which aligns with their intended moisturizing and therapeutic effects.

There is an inverse relationship between TFM (Total Fatty Matter) and total alkalinity in a soap. As the fatty matter content increases, total alkalinity decreases, indicating a product richer in fully saponified fats and containing fewer free alkaline salts. Theoretically, this correlation reflects soap quality: high TFM combined with low alkalinity corresponds to a milder, less irritating soap, whereas low TFM with high alkalinity indicates a harsher and more aggressive product, Figure 3.



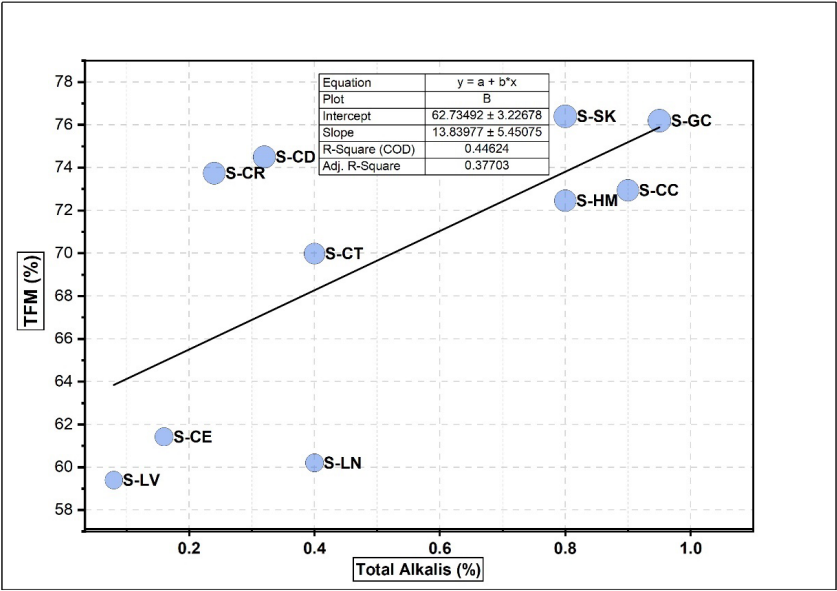
**Figure 3.** The relationship between Total fatty matter and Total alkali content in the soaps

Low alkalinity + low TFM, may indicate incomplete saponification due to lack of base or insufficient raw material in fatty acids.

High total alkalinity + high TFM, may be due to a complete reaction, a lot of bases used, but also a slight excess of free soda.

This general pattern, illustrated in Figure 3 through the combined visualization of TFM and total alkalinity, is further clarified in Figure 4. The scatter plot with linear regression provides a quantitative view of the relationship between these parameters, highlighting the degree of correlation across the soap samples.

The results indicate a moderate positive correlation ( $R^2 = 0.446$ ), which was expected given the diversity of manufacturers and the significant variation in technological processes and product formulations analyzed.



**Figure 4.** Linear Relationship Between Total Alkalinity and TFM in the Soap Samples

Very low total alkalinity values (e.g., S-LV – 0.08%, S-CE – 0.16%) are associated with reduced TFM, which may indicate an incomplete saponification process or excess moisture. In contrast, soaps with high total alkalinity (e.g., S-SK, S-GC, and S-CC at 0.8–0.95%) show elevated TFM levels (~76%), suggesting a well-balanced formulation and an efficient conversion of fatty

acids into soap. This trend is consistent with the principles of saponification, where an adequate amount of base promotes the conversion of fatty acids. Intermediate samples (S-CD, S-CR, S-CT, S-LN, S-HM) reflect the typical variability of commercial products, where TFM levels depend on both the lipid composition and the amount of base used in the manufacturing process. Our results are in agreement with recent studies on commercial soaps, which show that physicochemical parameters such as total alkali and TFM are essential for evaluating product quality [23].

Overall, the physicochemical profiles of the analyzed handmade soaps suggest that while most meet quality criteria, the slightly high pH values may require careful consideration for frequent use on skin, especially for sensitive skin types.

#### *Analysis of creams and ointments samples*

The physicochemical analysis of ten cosmetic creams and ointments samples is presented in Figure 5-6. Properties such as **pH, viscosity, and moisture content** determine texture, absorption, and product stability [24].

The analyzed samples of creams and ointments showed **pH values ranging between 4.95 and 9.1**, indicating a wide variation depending on formulation and intended use. Most samples exhibited pH values within or near the physiological skin range (5.4–5.9), considered optimal for maintaining the balance of the skin microbiome and the integrity of the **stratum corneum** [16]. Most cosmetic creams had slightly acidic to neutral pH values (4.95–7.7), suitable for maintaining the physiological balance of the skin's acid mantle. By contrast, the diaper ointment showed a higher pH (9.1), consistent with its protective and barrier-forming purpose.

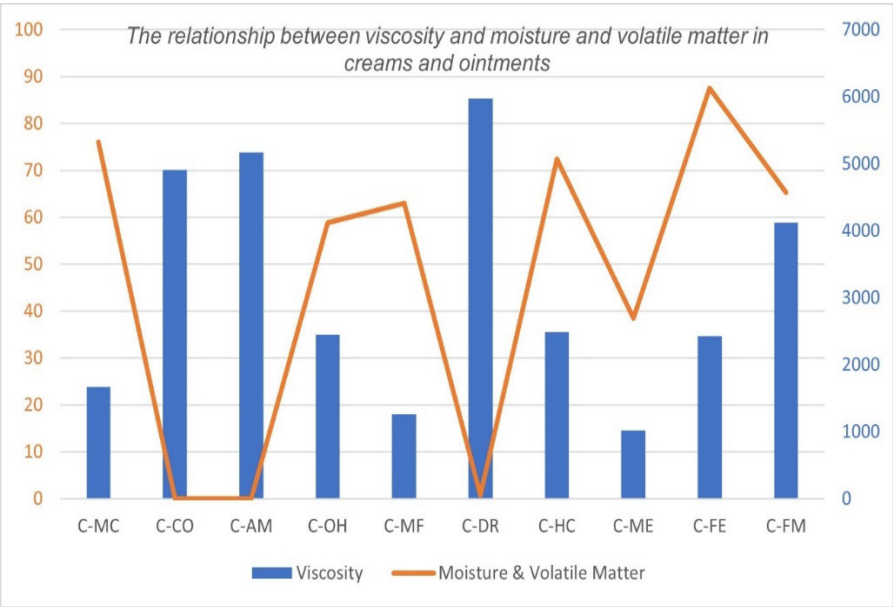
In terms of **density**, the formulations ranged from 0.78 to 1.16 g/cm<sup>3</sup>, reflecting differences in formulation type (oil-in-water vs. water-in-oil emulsions). **Viscosity values** were highly variable (from approximately 1000 to over 5900 mPa·s), confirming differences in texture and rheological behavior among the tested samples. Higher viscosity values were recorded for ointments containing herbal extracts such as *Calendula officinalis* with *Hypericum perforatum*, and *Arnica montana*, likely due to the presence of lipophilic bioactive compounds that increase the consistency of the base.

These plant-derived compounds include **triterpenoids** (faradiol, arnidiol, taraxasterol) from *Calendula officinalis* [25,26], **floroglucinols and phloroglucinol derivatives** such as **hyperforin** and **adhyperforin** from *Hypericum perforatum* [27,28] and **sesquiterpene lactones** like **helenalin** and **dihydrohelenalin** from *Arnica montana* [29,30]. These **lipophilic bioactives**

not only enhance viscosity and formulation stability but also contribute to the **anti-inflammatory**, **wound-healing**, and **antimicrobial** properties of the preparations.

The **moisture and volatile substance content** varied significantly, from 0.01% in ointments to over 80% in certain moisturizing or exfoliating creams, corresponding to the ratio between aqueous and lipidic phases.

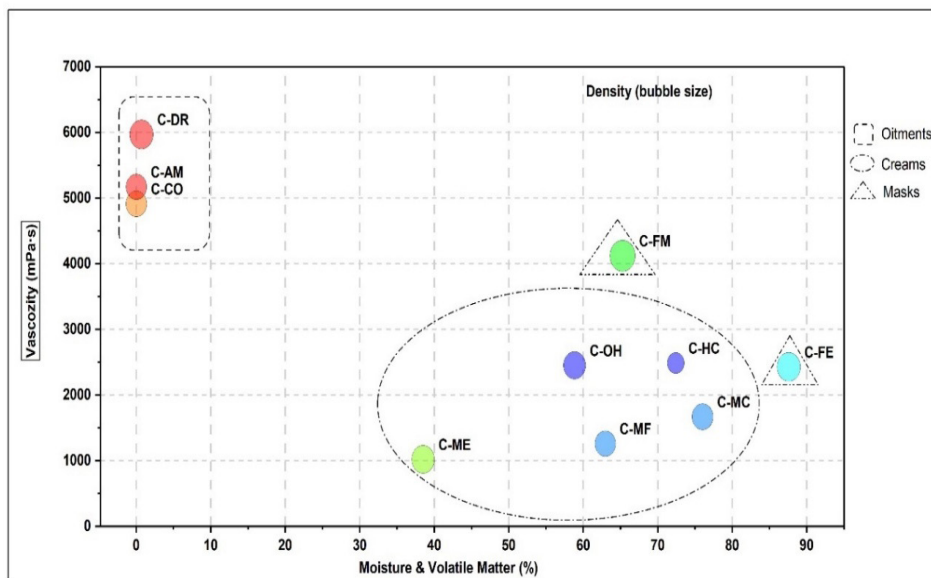
The viscosity of a cream is inversely proportional to its water content (humidity). As humidity increases, the aqueous phase dilutes the network of viscous molecules (fats, polymers, or gelling agents), reducing intermolecular interactions and, consequently, the resistance to flow (Figure 5).



**Figure 5.** Relationship between Moisture Content and Viscosity in the Analyzed Samples

Compared to Figure 5, which illustrates the distribution based solely on moisture and viscosity, Figure 6 provides an extended representation that also includes density as a parameter (represented by bubble size) with clearly highlighting the three product categories: creams, ointments, and facial exfoliants.





**Figure 6.** Dispersion of the Tested Formulations Based on Moisture, Viscosity, and Density

The C-AM, C-CO, and C-DR samples (moisture 0–1%, viscosity 4900–6000 mPa·s) appear clustered in the upper left area of the chart. These are typical for ointments, where the minimal water content leads to a dense structure and very high viscosity values. Their density is moderate and uniform. In contrast, the creams (C-MC, C-MF, C-HC, C-OH, C-ME) exhibit higher moisture levels (40–80%) and moderate viscosities (1000–2500 mPa·s), characteristic of fluid emulsions. The facial exfoliant (C-FE) and the mask (C-FM) show both high moisture content and increased viscosity and density, suggesting a formulation-specific structuring. Overall, the figure highlights the general trend of decreasing viscosity with increasing moisture and the clear separation between the rheological profiles of ointments and creams.

A classification of the quality of creams and ointments according to the main physicochemical is presented below:

#### Moisturizing creams

Results: 1260–1670 mPa·s. Interpretation: low–medium viscosity → Light texture, suitable for normal/oily skin, easily absorbed, ideal for pump-type dispensers; therefore, perfect for a slightly fluid daytime moisturizer. Typical reference range: 1000–5000 mPa·s.

### Hand cream

Result: 2453 mPa·s; Interpretation: medium consistency — pleasant, easy to apply, not very greasy, absorbs quickly. Typical range: 2000–6000 mPa·s.

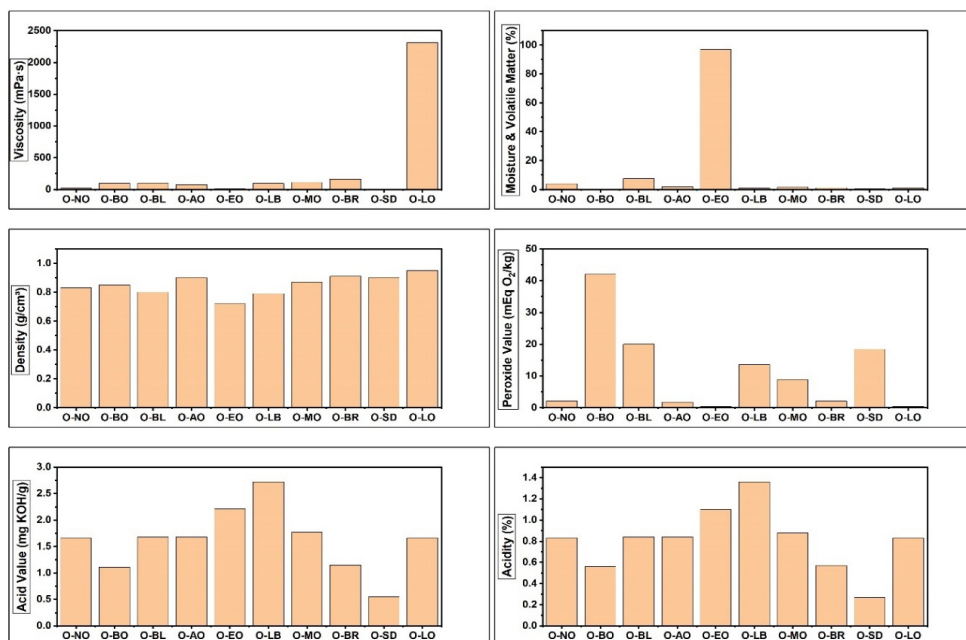
### Ointments

Results: 4912–5171 mPa·s; Interpretation: high viscosity, dense, greasy texture forming a protective film (specific to ointments). These values indicate a W/O type emulsion or a base with a high content of fats/waxes, as expected for ointments. Typical range: 4000–10000 mPa·s.

A viscosity of approximately 1021 mPa·s for moisturizing and emollient face cream is at the lower end of the typical range for moisturizing creams (1000–5000 mPa·s). A cream with a viscosity of ~1020 mPa·s is mainly recommended for normal, combination, and oily skin. For dry skin it may be too light.

### Analysis of oil samples

**Oils**, especially those used in therapeutic or cosmetic applications, are assessed for acidity, peroxide index, viscosity, moisture and density and results are described below and in Figures 7-9.

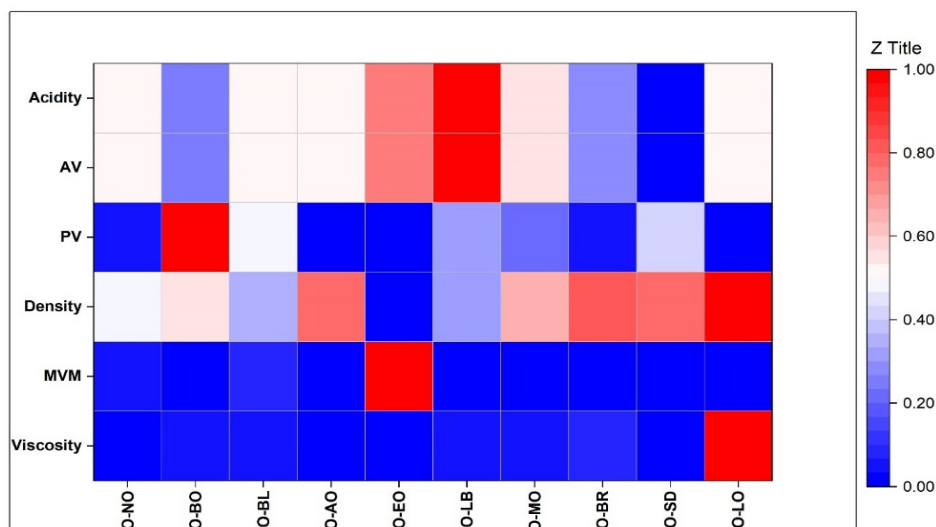


**Figure 7.** Physicochemical analyzes for handmade oils

The cosmetic oil samples showed a **wide variation in viscosity**, ranging from **10 mPa·s** in essential lavender oil to **over 2,300 mPa·s** in lip oil formulations, reflecting differences in oil type, purity, and formulation viscosity modifiers. Lower viscosity values were characteristic of **essential and volatile oils**, while higher viscosities were observed in complex formulations such as **lip oils** or **massage blends**, which include heavier triglycerides or waxy emollients.

The **moisture and volatile content** ranged between **0.16% and 3.89%** for most oils, except for the **lavender essential oil (97%)**, where the value reflects its highly volatile nature. **Density values** varied between **0.728 and 0.953 g/cm<sup>3</sup>**, consistent with typical cosmetic oil formulations composed of esters and natural triglycerides.

Regarding **oxidative stability**, the **peroxide values** ranged from **0.38 to 42.0 mEq O<sub>2</sub>/kg**, indicating different degrees of lipid oxidation. The highest peroxide value was found in the **body oil containing a blend of grape seed, almond, and jojoba oils**, which could be attributed to the unsaturated fatty acid profile of these ingredients, making them more prone to peroxidation. Lower peroxide values (below 2 mEq O<sub>2</sub>/kg) were characteristic of **pure essential or refined oils**, demonstrating good oxidative stability. Fresh oils typically have low acidity and peroxide values, while higher values indicate oxidation, hydrolysis, or degradation, often due to improper storage [24].



**Figure 8.** Heat Map of PhysicoChemical Profiles – 10 Types of Handmade Oils

The **acid value** ranged from **0.55 to 2.72 mg KOH/g**, corresponding to **acidities between 0.27% and 1.36%**, within the limits recommended by cosmetic quality standards (generally <2 mg KOH/g for cold-pressed cosmetic oils). Higher acid values, as observed in some lavender-based oils, may indicate partial hydrolysis of triglycerides or prolonged storage before testing.

A heat map representation was selected because it allows a simultaneous visualization of all measured parameters, highlighting patterns, correlations, and sample groupings that are not easily identifiable through individual plots.

The two indices, **Peroxide Value (PV)** and **Acid Value (AV)**, measure different degradation processes: oxidation in the case of PV and hydrolysis in the case of AV. Together, they provide complementary information: one index may indicate oil deterioration over time, while the other can demonstrate product compliance [31].

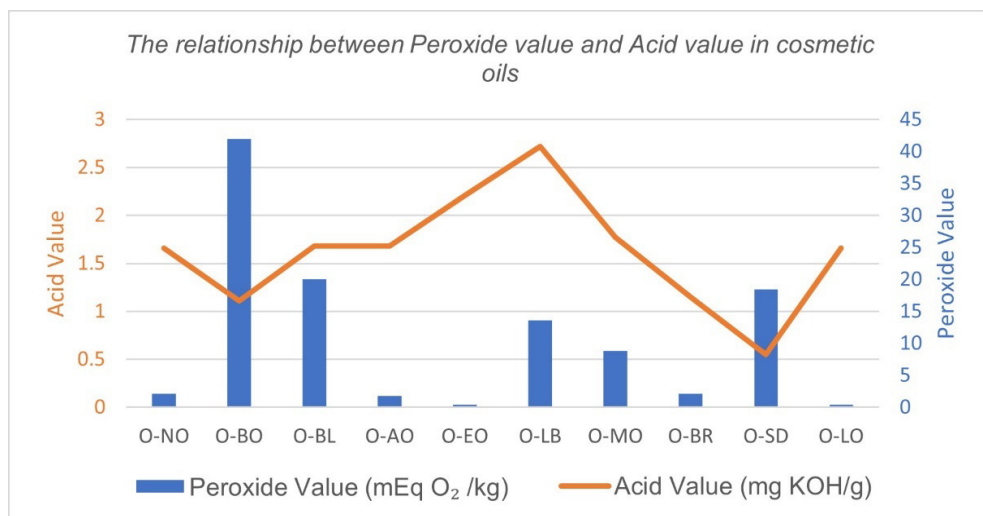
**Cosmetic oils** showed acceptable density, PV, and AV /acidity, indicating good oxidative stability and overall quality. The combination of PV and AV allowed the assessment of both oxidative and hydrolytic degradation, while acidity values confirmed that all oils fell within the **excellent to good range**. Microbiological analyses confirmed high safety, with no pathogenic microorganisms detected in any of the samples.

Overall, the results highlight the importance of continuous monitoring of handmade cosmetic products to ensure physicochemical quality, microbiological safety, and product stability. The study demonstrates that careful formulation, use of high-quality raw materials, and proper handling can produce handmade cosmetics that meet both safety and performance standards, making them suitable for consumer use.

The comparative analysis of the cosmetic oils shows that most products display high chemical stability and excellent microbiological safety, confirming an overall strong quality profile. Products such as Nutritive Oil, Anticellulite Massage Oil, Beard Oil, and Lip Oil stand out through a well-balanced combination of moisturizing properties and stability, making them suitable for regular use. Formulas with a heavier texture (e.g., *Beard Oil*, *Lip Oil*) prioritize emollience over lightness, which is appropriate for their intended function.

Lavender-based oils and body oil blends demonstrate good moisturizing performance but may benefit from improved oxidative stability, as certain botanical components are more sensitive to degradation. Overall, the product selection offers a diverse range of options tailored to different skin needs — from very light, fast-absorbing textures to richer, more protective formulations.

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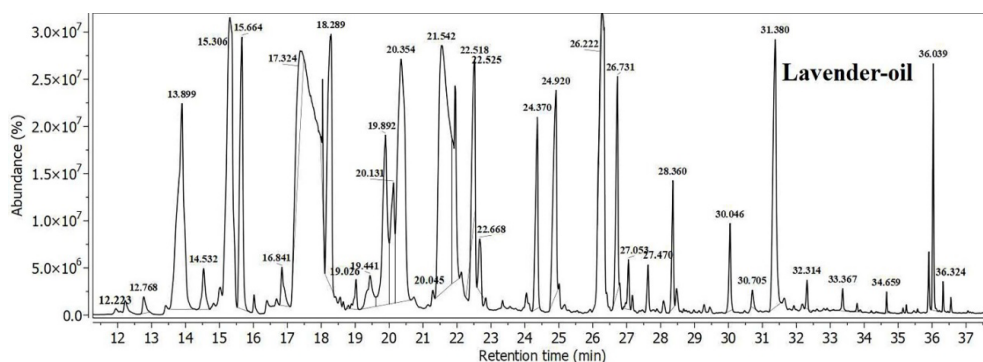


**Figure 9.** Relationship between Peroxide Value and Acid value in cosmetic oils

### *GC-MS analysis Lavender Essential Oil*

To extend the physicochemical analyses with an accurate molecular compositional profile, the *Lavender oil* sample was selected for GC-MS investigation due to single-origin essential oil, unlike the other commercial, non-essential oil products, included in the study, which may contain additives or blends.

The Figure 10 shows the total ion chromatogram (TIC) of the sample, with major constituents identified based on mass spectral matching (NIST library) and retention indices. The main compounds detected were: beta-myrcene (RT = 13.95 min; 6.19%), trans-beta-ocimene (RT = 15.30 min;

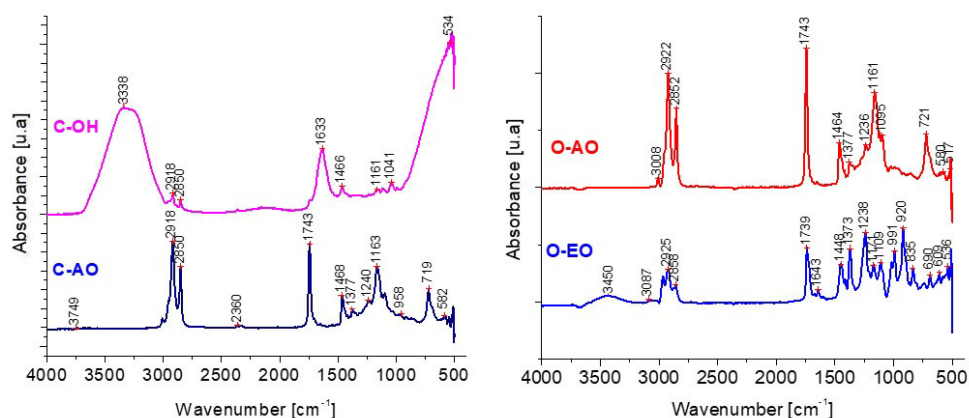


**Figure 10.** Chemical Profile of Lavender Essential Oil by GC-MS

8.11%), linalool (RT = 17.38 min; 21.91%), allo - ocimene (RT = 18.15 min; 5.00%), alpha-terpineol (RT = 20.35 min; 6.12%), linalyl acetate (RT = 21.513 min; 14.54%), lavandulyl acetate (RT = 22.52 min; 3.64%), and caryophyllene (RT = 26.24 min; 7.31%). Minor constituents (each <5%) included a series of monoterpenes and sesquiterpenes such as  $\alpha$ -pinene, cis-beta-ocimene, terpinen-4-ol, geranyl acetate, beta-farnasene, and tau-cadinol. The presence of characteristic lavender markers such as linalool, linalyl acetate, terpinen-4-ol, ocimene isomers, lavandulyl acetate, and  $\beta$ -caryophyllene confirms the authenticity of the essential oil and is consistent with published profiles of *Lavandula angustifolia* [32,33].

### FTIR-Based Physicochemical Structural Characterization

FTIR spectroscopy was selected for the analysis of creams and oil products due to its ability to rapidly characterize formulation matrices at a molecular level, based on the intrinsic physicochemical behavior of chemical bonds and infrared absorption of functional groups. This technique enables reliable identification of major compositional features such as esters, alcohols, carbonyls, and hydrocarbon chains, which are defining structural elements in semisolid and lipidic formulations. FTIR also provides valuable insight into molecular interactions within complex mixtures, without requiring prior separation or chemical modification of the sample. The obtained spectra showed well-defined characteristic bands consistent with the expected functional group profiles reported in the literature, supporting both sample integrity and the suitability of FTIR for comparative physicochemical screening and structural validation in this study. Figure 11 illustrates the FTIR spectra of the oat-based cream and the arnica ointment formulation and spectral profiles of lavender essential oil and the antifungal oil.



**Figure 11.** The FTIR-ATR spectra of colloidal Oat meal and Orange hand cream, ointment with Arnica, antifungal oil and Lavender oil

Lavender oil was chosen for its single-origin traceability and consistent spectral fingerprint, whereas the antifungal oil blend was selected to identify characteristic absorption bands of its bioactive compounds, ensuring IR-based evaluation of functional composition over botanical origin.

Because the antifungal oil (O-AO) is a mixture of six essential oils (eucalyptus oil, clove oil, frankincense oil, almond oil, lavender oil) and vitamin E, its FTIR spectrum is marked by the absorption bands of the major component. As can be seen in that spectrum, the strong absorption peak of C–H stretching appears at  $2852\text{ cm}^{-1}$  and  $2922\text{ cm}^{-1}$ , methyl asymmetric and symmetric bending peaks appear at  $1464\text{ cm}^{-1}$  and  $1377\text{ cm}^{-1}$  respectively; C–O stretching of the phenol appears at  $1236\text{ cm}^{-1}$ ; ether group appears at  $1161\text{ cm}^{-1}$  belonging to vitamin-E [34]. The band at  $3450\text{--}3087\text{ cm}^{-1}$  represents the stretching vibration of the -OH groups of the linalool substances, and another band at  $2925\text{ cm}^{-1}$  is correlated with the antisymmetric stretching vibration of the -CH<sub>2</sub> groups in lavender oil (O-EO) [35,36]. Also, the strong band at  $1739\text{ cm}^{-1}$  corresponds to the C=O stretching vibration of the ester compositions in lavender oil [35]. The linalyl acetate ester in this oil shows the C=CH<sub>2</sub> in-plane deformation vibration that occurs near the value of  $1443\text{ cm}^{-1}$  [37]. Frankincense essential oil contains majority diterpene alcohols, which show two strong bands, in the  $1377\text{--}1464\text{ cm}^{-1}$  region, while the second band, assigned to the C-O stretching vibration, are shown at  $1161\text{ cm}^{-1}$ , respectively. [37] The FTIR spectrum of clove oil shows several distinct peaks at  $1095\text{ cm}^{-1}$ , attributed to the C-O stretching vibration of the eugenol compound. Other peaks should be present at  $841.1\text{ cm}^{-1}$ , due to the C-H bending vibration, a peak at  $2920.4\text{ cm}^{-1}$  due to the C-H stretching vibration of the methylene group, and a peak at  $1279\text{ cm}^{-1}$ , due to the C-H rotational vibration of the isoeugenol compound present in clove oil. These peaks are either too small due to dilution or overlap with the peaks of the majority compounds.

Regarding hand creams, the colloidal oatmeal(C-OH) presents a large peak at  $3338\text{ cm}^{-1}$ , which can be attributed to N–H stretching vibration due to the presence of protein content. The peaks at  $2918$  and  $2860\text{ cm}^{-1}$  are attributed to C–H stretching vibrations. The peak at  $1633\text{ cm}^{-1}$  is attributed to the N–H bending, and peak at  $1406\text{ cm}^{-1}$  belong to the C–N bending [38].

For hand cream with arnica(C-AO): the FTIR band at  $2918\text{ cm}^{-1}$  in oils corresponds to the asymmetric C-H stretching vibration of aliphatic methylene -CH groups. This peak is typically seen alongside a symmetric -CH stretching vibration at a lower wavenumber, at  $2850\text{ cm}^{-1}$ . The arnica oil presents a strong band at  $1743\text{ cm}^{-1}$ , which is characteristic of carbonyl C=O groups, and band at  $1163\text{ cm}^{-1}$ , corresponds to C-O stretching vibrations such as sesquiterpene lactones from this plant [37].

## 2. Microbiological Analysis

### *Microbiological analyses for creams and ointments*

In the case of creams and ointments, microbiological safety is essential; the presence of pathogenic bacteria or fungi can cause infections or adverse reactions (Table 1).

Regarding **microbiological safety**, all products except the facial mask sample were free of pathogenic bacteria, fungi, and aerobic microorganisms. The **facial mask** showed contamination with potentially pathogenic microorganisms, with microbial colonies visible on most culture media in counts of several hundred (Figure 12).

The **microbiological contamination** or biological load of a cosmetic product is a very important component in product safety. Microbial growth can change physicochemical and organoleptic properties that affect marketing and consumer satisfaction. In addition to skin problems, microbial growth can change the color of a cosmetic product, or affect the viscosity, which gives the product a lumpy appearance or makes it more liquid, and also alter the shape of the packaging [39,40].

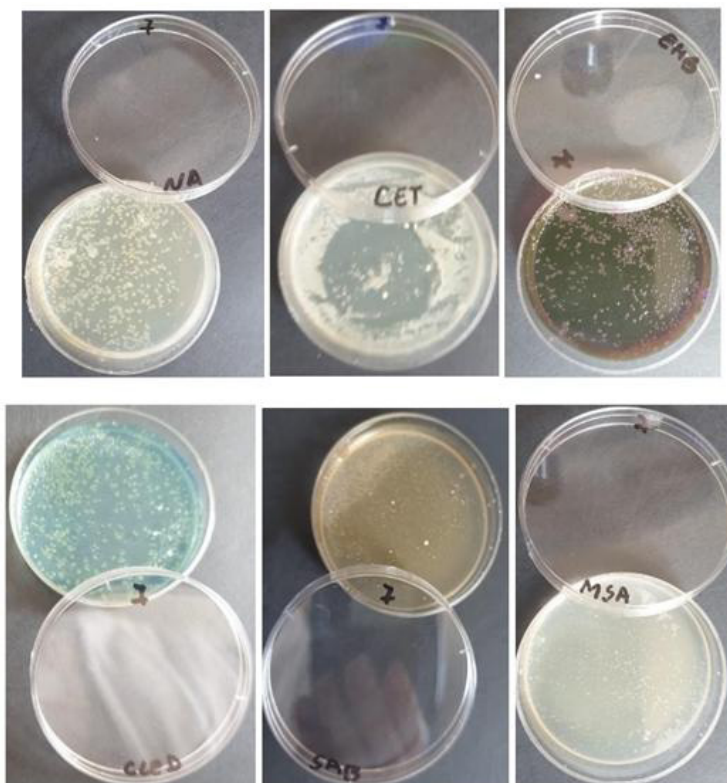
Cosmetic companies are not required to create sterile products. But they are responsible for guaranteeing the product's safety to the prospective buyer [41]. The determination of the microorganisms present in cosmetic products consisted of isolating and identifying possible microorganisms that could contaminate cosmetic products either during their formulation or during storage.

**Table 1.** Microbiological analyses for creams and ointments

No.	Sample	Microbiological Analysis
1	C-MC	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
2	C-CO	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
3	C-AM	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
4	C-OH	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
5	C-MF	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
6	C-DR	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
7	C-HC	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
8	C-ME	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
9	C-FE	Pathogenic bacteria, pathogenic fungi, and aerobic bacteria – <b>ABSENT</b>
10	C-FM	<b>PRESENCE of potentially pathogenic microorganisms</b> observed on all tested media; microbial colonies in the range of hundreds were detected on most culture plates.



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**Figure 12.** Microbial colonies developed on selective and non-selective test media developed for sample10; (NA = Nutrient Agar; Cet = Cetrimide Agar; EMB = Eosin Methylene Blue Agar; CLED = Cystine-Lactose-Electrolyte-Deficient medium; MSA = Mannitol Salt Agar; Sab = Sabouraud Chloramphenicol Agar)

This result indicates either inadequate preservation or contamination during the manufacturing process, emphasizing the need for strict hygienic practices and continuous microbiological monitoring of handmade cosmetic products.

Overall, the analyzed creams and ointments met the major physicochemical quality parameters, with the exception of one contaminated sample. These findings underline the importance of monitoring both formulation parameters (pH, viscosity, moisture) and microbiological safety to ensure consumer protection and product efficacy.

### *Microbiological Analyses of Handmade Oils*

The results are detailed in Table 2.

**Table 2.** Microbiological Analyses of Handmade Oils

No.	Sample	Microbiological Analysis
1	O-NO	Negative for <i>E. coli</i> , <i>P. aeruginosa</i> , <i>Enterobacter</i> sp., <i>S. aureus</i> , <i>C. albicans</i> ; total aerobic bacteria absent
2	O-BO	Negative for pathogenic microorganisms; total aerobic bacteria absent
3	O-BL	Negative for pathogenic microorganisms; total aerobic bacteria absent
4	O-AO	Negative for pathogenic microorganisms; total aerobic bacteria absent
5	O-EO	Negative for pathogenic microorganisms; total aerobic bacteria absent
6	O-LB	Negative for pathogenic microorganisms; total aerobic bacteria absent
7	O-MO	Negative for pathogenic microorganisms; total aerobic bacteria absent
8	O-BR	Negative for pathogenic microorganisms; total aerobic bacteria absent
9	O-SD	Negative for pathogenic microorganisms; total aerobic bacteria absent
10	O-LO	Negative for pathogenic microorganisms; total aerobic bacteria absent

From a microbiological perspective, all analyzed oil samples were negative for pathogenic microorganisms, including *Escherichia coli*, *Pseudomonas aeruginosa*, *Enterobacter* spp., *Staphylococcus aureus*, and *Candida albicans*. Total aerobic bacterial counts were also negative, confirming that the oils met the microbiological purity requirements for cosmetic products.

Overall, the results indicate that the analyzed handmade cosmetic oils were microbiologically safe, exhibited physicochemical parameters within acceptable cosmetic standards, demonstrated how PV and AV can be used together to monitor product stability and quality.

Continuous monitoring of peroxide and acid values is recommended to ensure product stability and maintain quality throughout storage and distribution.

## CONCLUSIONS

This study evaluated the physicochemical and microbiological quality of 30 handmade cosmetic products including creams, ointments, oils, and soaps, using standardized methods prior to market release. The novelty of the research lies in the pre-market assessment of these formulations, enabling early identification of potential risks related to skin compatibility and microbiological safety, while also supporting regulatory compliance and consumer protection.

The analyses demonstrated that:

The analyzed **solid soaps** exhibited a wide range of physicochemical properties, including pH, total and free alkalis, and total fatty matter, reflecting variability in formulation and composition. While most samples met quality standards, the slightly elevated pH values may require attention for frequent use on sensitive skin. Functional profiles varied from mild, moisturizing formulations suitable for daily cleansing to more abrasive or aromatic variants designed for intensive use.

The evaluated **creams and ointments** exhibited pH values within the physiological range, appropriate viscosity and density for topical application, and satisfactory microbiological safety. Viscosity measurements confirmed consistency with their intended uses — from light, fast-absorbing moisturizing creams suitable for normal to oily skin, to denser ointments with occlusive properties ideal for protective and reparative purposes. Plant extracts such as *Hypericum perforatum*, *Calendula officinalis*, and *Arnica montana* contributed to emulsion stability and potential therapeutic benefits through their lipophilic bioactive compounds.

The analyzed **cosmetic oils** demonstrated good chemical stability and excellent microbiological safety, with peroxide and acid values confirming minimal oxidative and hydrolytic degradation. Products such as Nutritive Oil, Anticellulite Massage Oil, and Beard Oil presented a balanced profile of moisturization and stability, supporting regular use. Higher-viscosity formulations (e.g., Beard Oil, Lip Oil) emphasized emollience and protection, while lavender-based blends showed good hydration but may benefit from enhanced oxidative stability.

## EXPERIMENTAL SECTION

### **Materials and Methods**

#### **1. Materials**

A total of 30 handmade cosmetic and hygiene products were analyzed, consisting of 10 samples from each category divided into three main categories:

**Creams and Ointments (10 samples):** 2 Hand creams, 1 Moisturizing face cream, 2 Ointments with medicinal plant extracts, 5 additional samples of various creams and ointments: Moisturizing cream (**C-MC**), *Calendula officinalis* and *Hypericum perforatum* Ointment (**C-CO**), *Arnica montana* Ointment (**C-AM**), Oat Hand Cream (**C-OH**), Moisturizing Face Cream (**C-MF**), Diaper Rash Ointment (**C-DR**), Hand Cream with *Calendula officinalis* and Glycerin (**C-HC**), Moisturizing and Emollient Face Cream (**C-ME**), Facial Exfoliant (**C-FE**), Facial Mask (**C-FM**). (*Semisolid formulations intended for topical application*).

**Cosmetic Oils (10 samples):** 3 Body oils, 1 Antimicrobial oil, 1 Essential oil, 5 additional oils for body or massage, including plant extract-enriched formulations: Nutritive Oil (**O-NO**), Body Oil (Grape Seed + Almond + Jojoba) (**O-BO**), Body Oil with Lavender (**O-BL**), Antifungal Oil (**O-AO**), Lavender Essential Oil (**O-EO**), Lavender Body Oil (**O-LB**), Anticellulite Massage Oil with Plants (**O-MO**), Beard Oil (**O-BR**), Body Oil “Silky Drops” (**O-SD**), Lip Oil (**O-LO**). (*Liquid formulations intended for skin application.*)

**Solid Soaps (10 samples):** 10 Handmade toilet soaps, including scented, exfoliating, and plant-enriched formulations: Citrus Scented Solid Soap (**S-CT**), Lavender and Violet Solid Soap (**S-LV**), Handmade “SKIN” Solid Soap (**S-SK**), Green Tea and Charcoal Solid Soap (**S-GC**), Coffee, Honey, and Cinnamon Soap (**S-CC**), Calendula Soap (**S-CD**), Coffee Exfoliating Soap (**S-CE**), Citronella and Rosemary Soap (**S-CR**), Honey and Oat Milk Soap (**S-HM**), Luxury Natural Soap (**S-LN**). (*Solid cleansing products produced by classical alkaline saponification or semi-synthetic methods.*)

## 2. Physicochemical Analyses

### 2.1 pH Determination:

For solid soaps, pH was measured in a 10% w/w aqueous solution using a Hanna Instruments pH meter, according to STAS SR EN 1262/2004 [42].

For aqueous solutions of creams, pH was determined according to STAS 8619/3-90 [43] and measured with a HALO 2 Wireless pH Meter.

### 2.2 Dynamic Viscosity:

Measured with a B-One Viscometer (LAMY RHEOLOGY) at 250 rpm for 60 s. For creams: RV4 spindle; for oils: RV1 spindle.

### 2.3 Moisture & Volatile Matter:

Determined according to SR:ISO 672:1996 [44] using a BIOBASE oven and RADWAG analytical balance.

### 2.4 Appearance and Stability:

Organoleptic evaluation and accelerated stability tests were performed, involving alternating exposure to low and high temperatures ( $4\text{ }^{\circ}\text{C} \leftrightarrow 40\text{ }^{\circ}\text{C}$ , 24 h per cycle, for 4–6 cycles).

### 2.5 Quality Factors for Oils:

**2.5.1 Peroxide Value (PV):** Determined by iodometric titration according to ISO 3960/2017 [45], representing the amount of iodine released from potassium iodide by peroxides in the sample. The peroxide value for all determinations is calculated by the equation:

$$PV = \frac{1000(V_0 - V_1) \times c}{m}$$

where:  $V_1$  is the volume in mL of thiosulphate solution used for titration,  $V_0$  is the volume in mL of thiosulphate solution used for titration of a control,  $c$  is the mol L<sup>-1</sup> thiosulphate concentration and  $m$  the amount of sample.

**2.5.2 Acid Value (AV):** Determined according to SR EN ISO 660:2020 [46] representing the amount of KOH (mg) required to neutralize the free fatty acids in 1 g of oil. It indicates the degree to which the triglycerides in the oil have decomposed to release free fatty acids.

The acid value was calculated by the equation:

$$AV = \frac{M \times C \times V}{m}$$

where:  $AV$  - the acid value expressed as a mass fraction,  $M$  is the molar mass, in grams per mole, for KOH,  $C$  is the L<sup>-1</sup> mole concentration of the standard volumetric solution of potassium hydroxide KOH used,  $V$  is the volume, in milliliters, of KOH and  $m$  is the mass, in grams, of the oil sample.

## 2.6 Determination of Alkalis and Fats in Soaps:

### 2.6.1 Free alkalis were determined according to SR:ISO 684:1996 [47]

The principle of the method consists of dissolving the soap in an ethanolic solution and neutralizing the free alkalis with a sulfuric acid solution whose known excess is back titrated with an ethanolic solution of potassium hydroxide. The total content of free alkali, expressed as sodium hydroxide (NaOH) in mass percent, is given by the formula:

$$\%NaOH = 0.040 \times \frac{V_0 T_0 - V_1 T_1}{m} \times 100 ,$$

where:  $m$  is the mass, in grams, of the test portion;  $V_0$  is the volume, in milliliters, of the standard volumetric acid solution used;  $V_1$  is the volume, in milliliters, of the standard volumetric potassium hydroxide solution used;  $T_0$  is the exact normality of the standard volumetric acid solution;  $T_1$  is the exact normality of the standard volumetric potassium hydroxide solution.

**2.6.2 Total alkalis, and total fatty matter (TFM)** were determined according to SR:ISO 685:2020 [48]. The principle of the method consists in decomposition of the soap by a known volume of standard volumetric mineral acid solution, extraction and separation of the liberated fatty matter with light petroleum and determination of the total alkali content by titration of the excess of acid contained in the aqueous phase with a volumetric

standard sodium hydroxide solution. After evaporation of the light petroleum from the extract, dissolution of the residue in ethanol and neutralization of the fatty acids with a standard volumetric potassium hydroxide solution. Evaporation of the ethanol and weighing of the soap formed to determine the total fatty matter content. Calculate *the total alkali content*, expressed as a percentage mass fraction of sodium hydroxide (NaOH) for sodium soaps,  $w$  NaOH, using formula:

$$w \text{ NaOH} = 0.040 \times (V_0 T_0 - V_1 T_1) \times \frac{100}{m},$$

where:  $m$  is the mass, in grams, of the test portion;  $V_0$  is the volume, in milliliters, of the standard volumetric acid solution used;  $V_1$  is the volume, in milliliters, of the standard volumetric sodium hydroxide solution used;  $T_0$  is the exact normality of the standard volumetric acid solution  $T_1$  is the exact normality of the standard volumetric sodium hydroxide solution.

Calculate *the total fatty matter content*, expressed as a percentage mass fraction,  $w$ , using formula:

$$w = [m1 - (V \times T \times 0,038)] \frac{100}{mn},$$

where:  $m$  is the mass, in grams, of the test portion;  $m1$  is the mass, in grams, of the dried potassium soap;  $V$  is the volume, in milliliters, of the standard volumetric ethanolic potassium hydroxide solution used for the neutralization;  $T$  is the exact normality of the standard volumetric ethanolic potassium hydroxide solution.

## 2.7 GC-MS Analysis:

The lavender essential oil was analyzed using on gas chromatograph coupled with mass spectrometer instrument Model Agilent 7890 & 5975 Series MSD, equipped with a HP-5MS (5% phenyl)-methyl polysiloxane fused silica column Agilent (30 m x 0.25 mm x 0.25  $\mu$ M). The oil sample (0,1g) diluted in hexan (1 mL) and a volume of 1 $\mu$ l was injected into the GC device. GC-MS data was obtained under the following conditions: carrier gas helium (He 6.0), flow rate 1ml/min, injector temperature was 260°C, splitless mode. The temperature program was the following: Oven temperature was programmed as 40°C for 1 min and an increase by 5 °C /min to 200 °C. From 200 °C to 240 °C, increase with 20 °C /min. It is maintained at 240 °C for 5 minutes. Mass spectra: electron impact (EI+) mode, 70 eV and ion source temperature, 230°C. Mass spectra were recorded over 50-500 a.m.u. range, scan mode. All analyses were carried out in duplicate. Data

acquisition and processing were performed using MSD ChemStation software. NIST library was used for identification/confirmation of the structure components. In addition, a C8-C20 standards alkanes (Alkane Standard Solution C8-C20, Sigma Aldrich) was used for calculation of the linear retention index (LRI), and matching the experimental values with those reported in the literature for similar chromatographic columns, in the same condition. The qualitative analysis was based on the percent area of each peak of the sample compounds [49].

### 2.8 FTIR analysis:

An analysis was performed using a Fourier-transform infrared spectrophotometer (FTIR) (Jasco FTIR-610) (Jasco® International Co., Ltd., Tokyo, Japan) equipped with an attenuated total reflectance (ATR) accessory with a horizontal ZnSe crystal (Jasco PRO400S). The samples were placed in direct contact with the ZnSe crystal and then the spectra were recorded at a resolution of 4 cm<sup>-1</sup>. The scans were repeated 100 times.

## 3. Microbiological Analyses

### Detection of microbial contamination:

- Manitol Salt Agar (MSA) medium for detection of *Staphylococcus aureus*
- Cetrimide Agar for detection of *Pseudomonas aeruginosa*;
- Sabouraud Chloramphenicol Agar for detection of *Candida albicans*;
- Cystine-Lactose-Electrolyte-Deficient (CLED) medium, for detection of *Enterobacter* sp.;
- Eosin Methylene Blue (EMB) agar for detection of *Escherichia coli*;
- Nutrient Agar for detection of total aerobic bacteria [50,51].

Sample preparation involved 1:10 dilution: 1 g of sample added to 9 mL of Eugon LT 100 Broth.

Dilutions were inoculated on selective media for detection of potential pathogenic microorganisms. Plates were incubated at 37 °C for 24 h (for bacterial strains) and 48 h (for fungal strains), after which microbial growth was evaluated. All microbiological analyses were performed according to ISO 17516:2014 [52].

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