CHITOSAN-BASED FILMS WITH SELECTED MOISTURIZING ADDITIVES

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Abstract

This study presents the development and characterization of thin films based on chitosan, modified with selected moisturizing additives, including hyaluronic acid, snail mucus extract, natural moisturizing factor (NMF), and gluconolactone, chosen for their common application in skin care products. Chitosan was dissolved in 0.1 M acetic acid to prepare a 1% w/v solution, and each additive was incorporated at 10% w/w relative to the chitosan content, both individually and in combination. The resulting mixtures were cast and dried under ambient conditions to form flexible films. Comprehensive characterization was performed using Fourier Transform Infrared Spectroscopy (FTIR), atomic force microscopy (AFM), contact angle measurements, and mechanical testing. The results revealed significant differences in chemical interactions, surface morphology, wettability, and mechanical behavior depending on the additive used. Notably, the formulation combining all moisturizing agents exhibited the most favorable balance of mechanical strength, flexibility, and hydration-related surface characteristics. FTIR spectra indicated molecular-level interactions between chitosan and the additives, particularly through hydrogen bonding. AFM analysis showed variations in surface roughness, while contact angle measurements highlighted changes in surface energy and hydrophilicity. These findings indicate that the modified chitosan films are promising candidates for innovative, sustainable dermatological and cosmetic applications, especially for effective skin hydration and use in skin care products.

Keywords: *chitosan, hyaluronic acid, snail mucus extract, natural moisturizing factor , gluconolactone, films*

Introduction

In recent years, increasing attention has been devoted to the ecological impact of everyday products, particularly in the cosmetics industry, where the use of synthetic and non-biodegradable materials remains widespread [1]. Consumers are becoming more conscious not only of product ingredients but also of the environmental footprint left by packaging and disposable cosmetic items. This has led to the rising popularity of sustainable, zero-waste cosmetic solutions that emphasize the use of biodegradable, renewable, and skin-compatible materials [2]. Among these, natural biomaterials—especially polysaccharide-based polymers such as chitosan, hyaluronic acid, and starch derivatives—have gained prominence due to their excellent biocompatibility, non-toxicity, and ability to retain moisture [3]. These materials not only align with environmental sustainability goals but also offer functional benefits for skin care, such as forming protective films, enhancing hydration, and improving skin elasticity. Their film-forming capabilities and interaction with the skin's natural moisture barrier make them ideal candidates for the development of modern skin care products, including facial masks, topical gel and emollient creams [4].

Among natural polymers, chitosan has emerged as a particularly promising biopolymer due to its high biocompatibility, biodegradability, antimicrobial activity, and ability to form flexible films [5]. Derived primarily from the deacetylation of chitin obtained from crustacean shells, chitosan can be processed into a variety of structures, such as membranes and hydrogels, which are advantageous for cosmetic and biomedical applications. Its inherent cationic nature allows for electrostatic interactions with anionic molecules and bioactive compounds, expanding its utility in multifunctional formulations [6]. Despite concerns regarding its potential to cause allergic reactions in individuals with shellfish allergies, chitosan's use in cosmetics is generally considered safe due to its high purification standards and low residual protein content. Compared to other polysaccharides, chitosan offers a unique combination of properties—including antimicrobial efficacy, film-forming ability, and bioadhesiveness—that justify its selection in applications where both performance and environmental sustainability are critical.

Hyaluronic acid (HA), a naturally occurring polysaccharide present in the human body, is widely known for its exceptional moisture-retaining capacity and skin-replenishing properties. Its incorporation into topical formulations enhances skin hydration, elasticity, and protection against environmental stressors [7]. From a materials science perspective, HA contributes to the formation of viscoelastic, hydrophilic matrices that support water retention at the skin surface and within the stratum corneum. Due to its high molecular weight and polyanionic nature, HA forms stable networks within polymeric films, improving their mechanical flexibility and ability to adhere to the skin [8]. This not only prolongs the contact time of active ingredients but also creates a semi-occlusive layer that reduces transepidermal water loss (TEWL). Additionally, HA's compatibility with other biopolymers, such as chitosan, facilitates the formation of composite materials with synergistic moisturizing and protective effects, making it an essential component in advanced skin care applications [9].

This study focuses on the preparation and characterization of chitosan-based films enriched with moisturizing agents such as hyaluronic acid, snail mucin extract, natural moisturizing factor (NMF), and gluconolactone. The goal is to evaluate the mechanical, surface, and spectroscopic properties of these thin films and to determine the most effective composition in terms of elasticity and material performance. The results are expected to contribute to the development of sustainable and efficient products in line with contemporary environmental and consumer health standards.

Materials and Methods

Chemicals

Chitosan (CTS, M_v=1.4 × 10^6 g/mol, DD=78%) was purchased from Sigma-Aldrich. Acetic acid waspurchased from Pol-Aura. Hyaualuronic acid (M_v=1.8 × 10^6 g/mol), snail mucus extract, natural moisturizingfactor (NMF) and gluconolactonewere purchased from Zrób Sobie Krem company.

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Films preparation

Chitosan was dissolved in 0.1 M acetic acid to reach a concentration of 1% w/v. The CTS solution was mixed with a hyaluronic acid (HA), snail mucus extract (SME), natural moisturizing factor (NMF) and gluconolactone (GL) separately, which were added 10 w/w% based on the chitosan content. Chitosan-based films were also modified using all mentioned additives mixed in equal ratio and added in the same content. The mixtures were stirred with a magnetic stirrer for 1 hour and then placed on a plastic holder (40 mL per 10 cm × 10 cm). Thin films were obtained by evaporating the solvent at room conditions (thickness 0.135 ± 0.024 µm). Pure chitosan-based films were utilized as controls for comparison (TABLE 1). Films are presented in FIG. 1.

TABLE 1. Nomenclature and composition of the different films

Abbreviation	Sample
CTS	Film based on chitosan
CTS + HA	Film based on chitosan with the 10% hyaluronic acid
CTS + SME	Film based on chitosan with the 10% snail mucus extract
CTS + NMF	Film based on chitosan with the 10% natural moisturizing factor
CTS + GL	Film based on chitosan with the 10% gluconolactone
CTS + MIX	Film based on chitosan with the 10% combination of all listed humectants

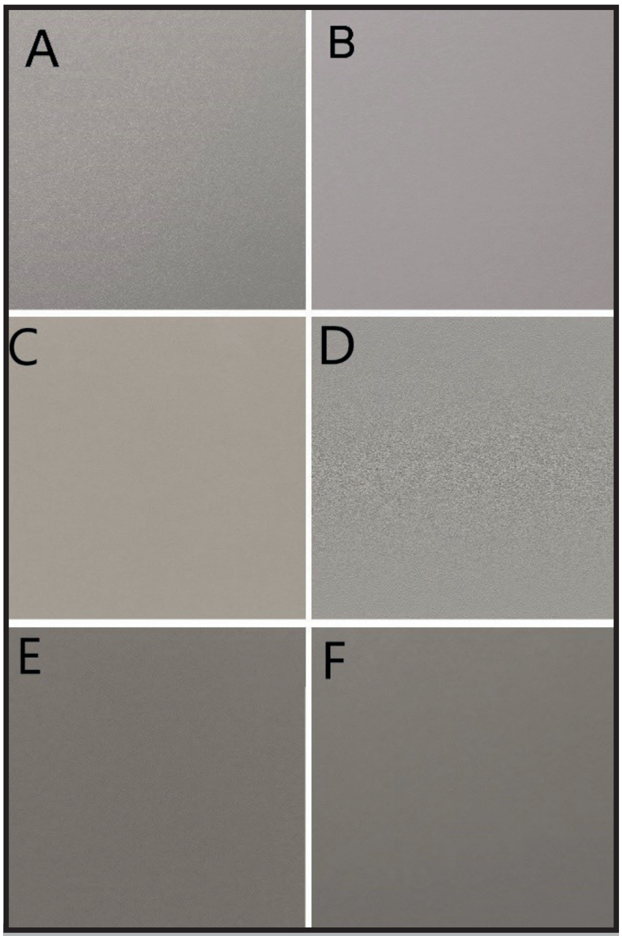

FIG. 1. The images of the obtained films A) CTS B) CTS+HA C) CTS+SME D) CTS+NMF E) CTS+GL F) CTS+MIX (magnification 5x)

Fourier Transform Infrared Spectroscopy—Attenuated Total Reflectance (FTIR–ATR)

Structural characterization of the samples was performed using FTIR–ATR spectroscopy. Spectral data were collected within the 4000–500 cm^{-1} range, employing a resolution of 4 cm^{-1} and averaging 64 scans per sample. Measurements were carried out using a Nicolet iS10 spectrometer (Thermo Fisher Scientific Inc., Waltham, MA, USA), equipped with a germanium ATR crystal, operating in absorbance mode.

Atomic Force Microscopy (AFM)

Surface morphology and roughness of the samples were assessed under ambient conditions using a NanoScope IIIa MultiMode scanning probe microscope (Veeco Metrology, Inc., Santa Barbara, CA, USA) in tapping mode. Quantitative surface roughness parameters, including root mean square roughness (Rq) and arithmetic mean roughness (Ra), were calculated with the use of Nanoscope Analysis software v6.11 (Bruker Optoc GmbH, Ettlingen, Germany).

Mechanical Testing

Mechanical behavior of the film samples was examined using a Shimadzu EZ-Test EZ-SX universal testing machine (Kyoto, Japan). Specimens were subjected to uniaxial tensile testing at a crosshead speed of 5 mm/min. Young's modulus (E_{mod}), ultimate tensile strength (σ_{max}), and elongation at break (dl) were determined from the linear portion of the stress-strain curve using Trapezium X Texture analysis software (Kyoto, Japan).

Contact Angle Measurement

Wettability of the film surfaces was evaluated by measuring the static contact angle of liquid droplets deposited on the sample surface. Surface free energy, including polar and dispersive components, was estimated via the Owens–Wendt approach, utilizing two probe liquids—water and diiodomethane [10]. Measurements were taken at constant temperature using a goniometer integrated with a drop shape analysis system (DSA 10 Control Unit, Krüss, Germany).

Statistical analysis

Quantitative data were processed using SigmaPlot 14.0 software (Systat Software, San Jose, CA, USA). The Shapiro–Wilk test was applied to assess normality of data distribution. Results are presented as mean ± standard deviation (SD). Statistical differences between groups were evaluated using one-way analysis of variance (ANOVA), followed by Bonferroni's post hoc test for multiple comparisons, with significance considered at $p < 0.05$.

Results and Discussions

Fourier Transform Infrared Spectroscopy—Attenuated Total Reflectance (FTIR–ATR)

The FTIR spectra of all analyzed chitosan-based films exhibited characteristic absorption bands associated with the chemical structure of chitosan (TABLE 2). Prominent peaks observed in the region of 3000–3500 cm^{-1} correspond to the stretching vibrations of hydroxyl (O–H) and amine (N–H) groups, indicative of the polysaccharide nature of chitosan [11]. Additionally, a distinct band near 1650 cm^{-1} is attributed to the C=O stretching vibrations in the amide I region, while signals around 1550 cm^{-1} are assigned to amide II bands, reflecting N–H bending and C–N stretching vibrations [12].

The incorporation of each tested moisturizing agent induced subtle yet distinct spectral shifts, particularly in the 1600–1700 cm^{-1} region and between 1200–1400 cm^{-1}. These alterations suggest molecular-level interactions between the hydrophilic functional groups of the added components and the primary chitosan matrix. Such interactions are likely governed by hydrogen bonding or electrostatic attractions involving hydroxyl, carboxyl, amine, or amide groups.

In the case of hyaluronic acid, a shift in the carbonyl stretching region indicates potential hydrogen bonding between its carboxyl and hydroxyl groups and the amino functionalities of chitosan [13]. A similar spectral behavior was observed with the addition of snail mucus, known to contain glycoproteins and polysaccharides capable of engaging in intermolecular interactions [14]. Gluconolactone appeared to induce broader changes across the spectra, including the emergence and attenuation of certain bands, suggesting a more pronounced interaction with the chitosan backbone, possibly due to its polyhydroxy structure.

The most complex spectral profile was observed for the MIX sample, which combines all moisturizing agents. This sample exhibited multiple overlapping bands and notable shifts, indicating synergistic or cumulative interactions among the components and the chitosan matrix. The presence of diverse functional groups within the mixture likely contributed to the formation of a more intricate hydrogen bonding network, influencing the chemical environment of the film.

TABLE 2. Summary of wavenumber and functional group results of all studied samples

CTS		CTS + HA		CTS + SME	
Wavenumber [cm $^{-1}$]	functional groups	Wavenumber [cm $^{-1}$]	functional groups	Wavenumber [cm $^{-1}$]	functional groups
3118.21	N-H, O-H	3118.45	N-H, O-H	2929.79	N-H, O-H
2917.19	C-H	2916.36	C-H	2891.18	C-H
2871.89	C-H	2872.13	C-H	1730.29	C=O, N-H
1641.83	C=O, N-H	1646.83	C=O, N-H	1588.37	C-N, N-H
1551.13	C-N, N-H	1554.51	C-N, N-H	1409.63	CH-OH
1376.81	CH_2	1376.26	CH_2	1378.21	CH_2-OH
1317.37	C-N	1316.28	C-N	1326.07	CH_2
1151.78	C-O-C	1151.85	C-O-C	1298.44	C-N
1060.51	C-OH	1060.96	C-OH	1215.22	C-O-C
1025.21	CH_2-OH	1026.09	CH_2-OH	1070.82	C-OH
896.37	C-H	896.18	C-H	1026.43	CH_2-OH

CTS + NMF		CTS + GL		CTS + MIX	
Wavenumber [cm $^{-1}$]	functional groups	Wavenumber [cm $^{-1}$]	functional groups	Wavenumber [cm $^{-1}$]	functional groups
3210.86	N-H, O-H	3118.23	N-H, O-H	3273.30	N-H, OH
2923.29	C-H	2915.10	C-H	2929.75	C-H
2875.05	C-H	2871.94	C-H	2887.77	C-H
1660.01	C=O, N-H	1646.88	C=O, N-H	1658.30	C=O, N-H
1581.52	C-N, N-H	1553.09	C-N, N-H	1588.69	C-N, N-H
1379.69	CH_2	1376.10	CH_2	1378.46	CH_2
1295.18	C-N	1315.89	C-N	1326.43	C-N
1149.35	C-O-C	1151.92	C-O-C	1125.57	C-O-C
1064.76	C-OH	1061.13	C-OH	1071.42	C-OH
1029.23	CH_2-OH	1025.32	CH_2-OH	1028.84	CH_2-OH
895.89	C-H	896.04	C-H	877.86	C-H

Atomic Force Microscopy

Atomic Force Microscopy (AFM) analysis was conducted to examine the surface topography of chitosan-based films and to assess the potential influence of moisturizing additives on their microstructure (Figure 2). The control chitosan film exhibited a relatively smooth and uniform surface, which is typical for pristine chitosan matrices [15]. Quantitative roughness parameters, including average roughness (Ra) and root-mean-square roughness (Rq), confirmed this observation (TABLE 3).

The incorporation of moisturizing agents such as hyaluronic acid (HA), snail mucus extract (SME), natural moisturizing factor (NMF), gluconolactone (GL), and their combination (MIX) led to only minor changes in surface roughness compared to the control. While some formulations exhibited slightly increased Ra and Rq values—particularly those containing SME and the MIX—the differences were not pronounced. These findings suggest that the moisturizing additives had a negligible overall effect on the surface topography of the films.

Slight roughness variations may be attributed to the molecular characteristics of the additives and their interactions with the chitosan matrix. For instance, HA may promote minor surface smoothing through hydrogen bonding [16], while SME and NMF could introduce mild heterogeneity due to their complex biochemical compositions. Similarly, the modest increase observed in GL-containing films may reflect its partial compatibility with the chitosan network [17], leading to only minimal disruption of surface uniformity. Nevertheless, the general uniformity of roughness values across all samples indicates that the additives did not significantly alter the microstructure of the chitosan films.

TABLE 3. The roughness parameters for all studied films (n=5; no significantly different from 100CTS (p<0.05)).

Specimen	Ra [nm]	Rq [nm]
CTS	1.65 ± 0.67	2.05 ± 0.83
CTS + HA	1.26 ± 0.01	1.61 ± 0.04
CTS + SME	2.09 ± 0.63	2.62 ± 0.86
CTS + NMF	2.70 ± 0.16	2.10 ± 0.15
CTS + GL	1.76 ± 0.46	1.44 ± 0.38
CTS + MIX	2.20 ± 0.52	1.79 ± 0.38

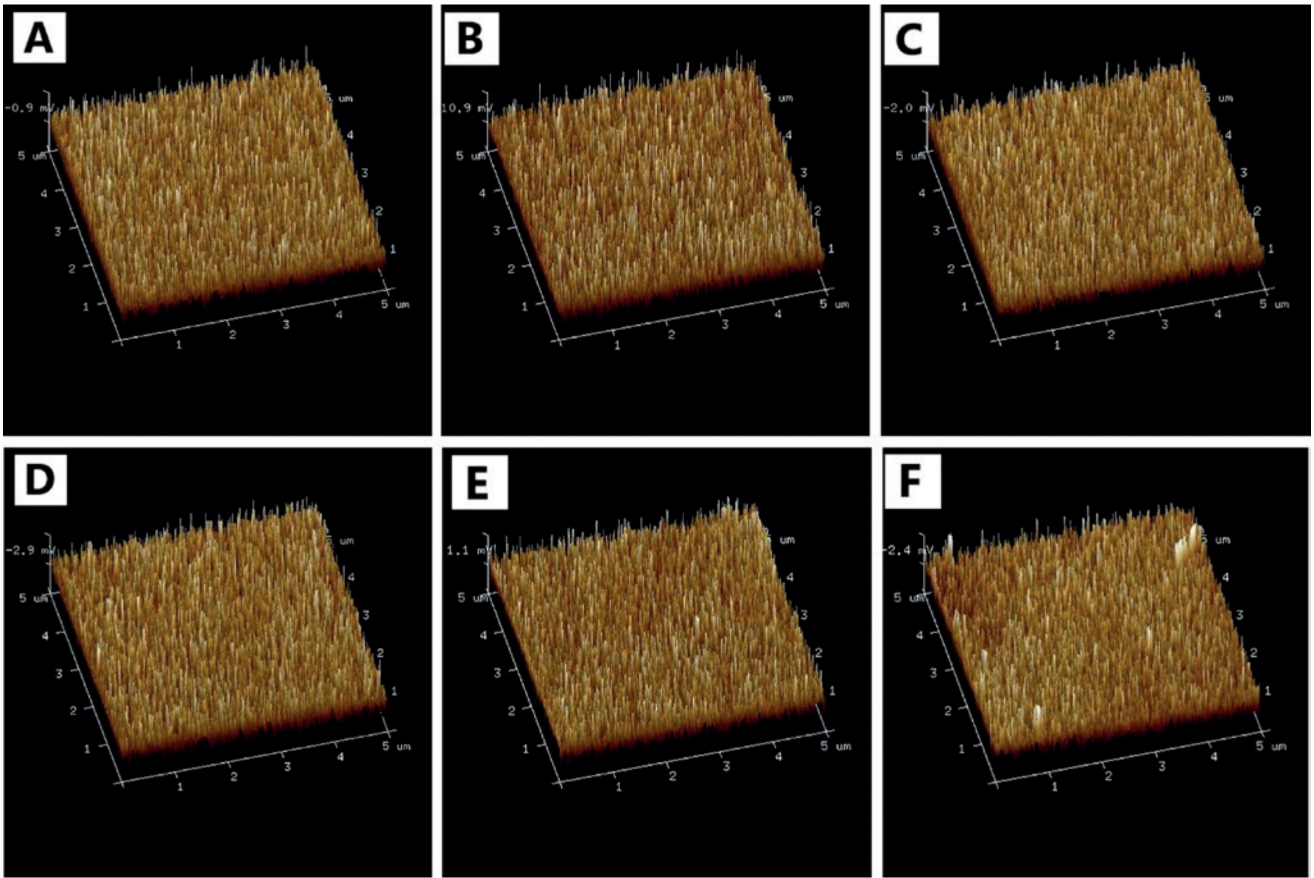

FIG. 2. The 3D images of surfaces of films.
A) CTS; B) CTS + HA; C) CTS + SME; D) CTS + NMF; E) CTS + GL, F) CTS + MIX.

Mechanical Testing

The stiffness of the chitosan-based films, as indicated by Young's modulus, varied noticeably depending on the type of moisturizing agent incorporated (FIG. 3). Films containing hyaluronic acid or snail mucus exhibited increased stiffness compared to the pure chitosan film, suggesting that these components enhance the structural integrity of the polymer matrix. Particularly, snail mucus appeared to significantly reinforce the film structure. In contrast, films containing NMF, gluconolactone, or a combination of all moisturizing agents showed a marked decrease in stiffness, indicating a more flexible and less rigid material. This reduction suggests a plasticizing effect or a disruption of the polymeric network, leading to a softer structure [18].

Tensile strength remained relatively stable or improved slightly in films enriched with hyaluronic acid or snail mucus, pointing to enhanced resistance to mechanical stress. However, the addition of NMF, gluconolactone, and especially their combination resulted in a substantial weakening of the film structure, with significantly reduced tensile strength. This indicates that although these additives may benefit flexibility, they can compromise the mechanical stability of the material.

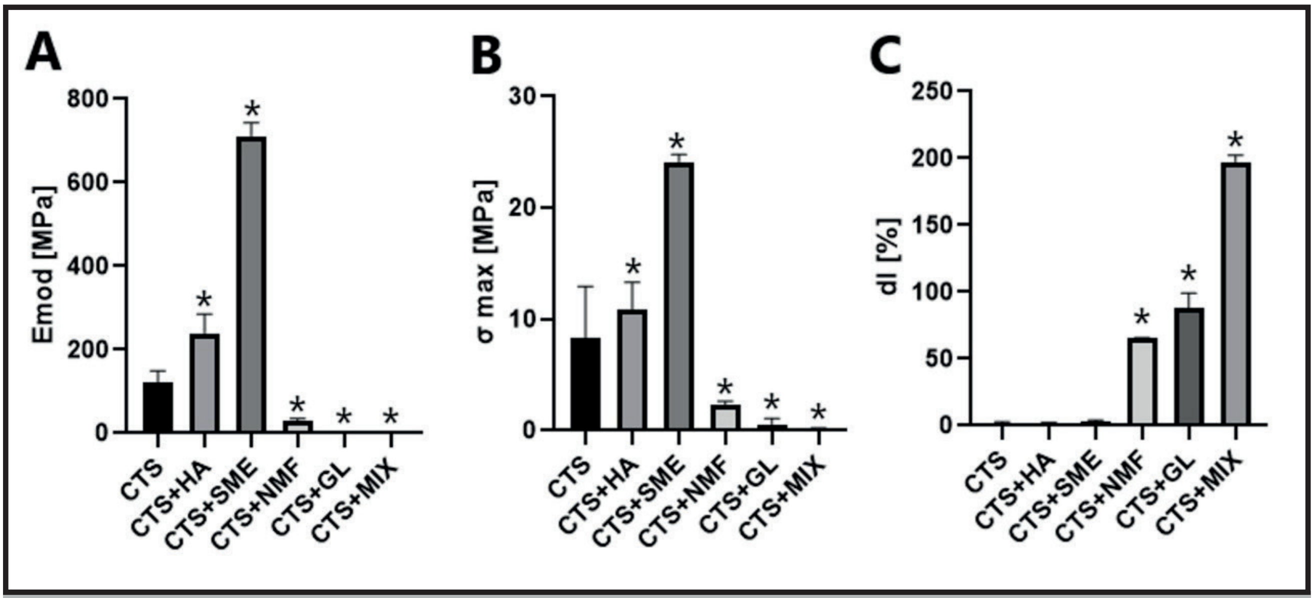

FIG. 3. Mechanical parameters A) the Young Modulus (E_{mod}), (B) maximum tensile strength (σ_{max}), and (C) elongation at break (dl) determined for films based on chitosan (n = 5; *significant differences from 100CTS ($p<0.05$)

The most substantial changes were observed in the films' ability to stretch before breaking. While the control chitosan film and those with hyaluronic acid or snail mucus exhibited limited elongation, films containing NMF and gluconolactone showed significantly higher extensibility. The most flexible formulation was the one combining all moisturizing agents, which demonstrated the highest elongation, indicating superior ductility. This trend implies that although these additives reduce stiffness and strength, they greatly enhance the film's elasticity [19].

Contact Angle Measurement

The wettability and surface characteristics of the chitosan-based films were investigated using contact angle measurements and surface free energy calculations based on the Owens–Wendt approach (TABLE 4). These analyses provided insights into how the incorporation of various moisturizing agents, such as hyaluronic acid, snail mucus, natural moisturizing factor, gluconolactone, and a combined mix, influenced the interfacial properties of the films.

Films composed solely of chitosan or with the addition of hyaluronic acid, snail mucus, or NMF exhibited relatively similar contact angle values. This consistency is likely due to the dominant presence of chitosan in each formulation, whose hydrophilic nature is attributed to hydroxyl and amine functional groups in its structure [20]. The surface free energy values for these samples also remained in a close range, particularly within the dispersive component, suggesting that the addition of small quantities of these moisturizing agents does not significantly alter the dominant physicochemical behavior of the chitosan matrix.

More pronounced effects were observed upon the incorporation of gluconolactone and the combined moisturizing agent mix. These films displayed notably lower contact angles, indicating reduced wettability and suggesting a decrease in surface hydrophilicity. However, despite lower overall wettability, the gluconolactone-containing film exhibited the highest polar contribution to the surface free energy, implying a strong polar character at the interface. This may be due to the unique molecular interactions introduced by gluconolactone, potentially involving its hydroxyl and carbonyl functionalities, which can rearrange the surface chemistry and enhance polarity. The film containing snail mucus demonstrated the highest contact angle among all variants, indicating a relatively more hydrophilic surface. At the same time, it exhibited the lowest polar contribution to surface free energy, suggesting that while the surface appears highly wettable, it may be governed more by dispersive interactions. This discrepancy highlights the complex interplay between topography, surface roughness, and molecular composition in defining film wettability.

In addition to the analytical data, the practical implications of these findings for cosmetic applications are noteworthy. The physicochemical and mechanical properties of the developed chitosan-based films suggest their high potential for real-world cosmetic and dermatological applications. From a practical perspective, the observed balance between flexibility and mechanical integrity is particularly important for products intended for direct application to the skin, such as sheet masks, under-eye patches, or wound dressings. Films that are both soft and elastic are more likely to conform closely to the skin's surface without

TABLE 4. The surface free energy (IFT(s)), its polar (IFT (s,P)) and dispersive (IFT (s,D)) components of chitosan based films (n=5; * significantly different from CTS- p<0.05)

Specimen	θ G [°]	θ D [°]	IFTS(s) [mJ/m^2]	IFTS(s,D) [mJ/m^2]	IFTS(s,P) [mJ/m^2]
CTS	73.10 ± 6.22	61.15 ± 2.73	30.60 ± 1.17	22.05 ±0.58	8.56 ± 0.60
CTS + HA	68.68 ± 3.33	58.17 ± 2.80	33.27 ± 0.99*	22.98 ± 0.57*	10.29 ± 0.42*
CTS + SME	76.64 ± 2.94	60.68 ± 1.00	29.67 ± 0.46	23.13 ±0.22*	6.54 ± 0.24*
CTS + NMF	72.03 ± 3.25	59.50 ± 1.33	31.59 ±0.60	22.88 ± 0.29*	8.71 ± 0.31
CTS + GL	44.88 ± 1.72*	49.70 ± 2.45*	47.00 ± 0.94*	23.46 ± 0.50*	23.54 ± 0.44*
CTS + MIX	46.23 ±1.44*	45.75 ± 2.35*	47.02 ± 0.88*	25.86 ± 0.50*	21.16 ± 0.38*

wrinkling or detaching prematurely, thus enhancing comfort and prolonging contact with active ingredients. In particular, the MIX formulation—containing all moisturizing additives—demonstrated superior extensibility and elasticity, indicating a high level of structural adaptability. This feature is essential for ensuring that the film remains in intimate contact with facial contours during application, allowing for even distribution of hydration and actives. Moreover, the relatively low contact angle and increased polar surface energy observed in the MIX and GL films imply enhanced wettability, which can improve adherence to moist skin and facilitate the diffusion of hydrophilic compounds. Surface roughness, while often considered a structural characteristic, may also affect user experience. Smooth surfaces are generally perceived as more pleasant in cosmetic applications, and the data confirm that none of the additives introduced excessive surface irregularities. In fact, the negligible variation in roughness across samples supports the conclusion that the films retain a cosmetically acceptable texture regardless of formulation.

Although peel adhesion testing was not conducted, the material's film-forming ability and surface energy characteristics suggest that adhesion would be sufficient for temporary skin application. Nevertheless, this aspect warrants further quantitative assessment in follow-up studies to fully validate the usability of these formulations in a commercial setting.

Finally, the natural origin and biodegradability of all ingredients make these films suitable for single-use applications with minimal environmental impact. This aligns with current consumer preferences for eco-conscious, zero-waste cosmetics and supports their potential as sustainable alternatives to synthetic polymer-based products.

Conclusion

This study demonstrated the successful development and characterization (FIG. 4) of chitosan-based films enriched with selected moisturizing additives, including hyaluronic acid, snail mucus extract, natural moisturizing factor, and gluconolactone. The incorporation of these agents significantly influenced the physicochemical, mechanical, and surface properties of the films. FTIR analysis confirmed interactions between chitosan and moisturizing additives, primarily through hydrogen bonding, leading to structural changes in the polymer matrix. AFM showed slight changes in roughness. Mechanical tests showed that hyaluronic acid and snail mucus enhanced strength, while NMF and gluconolactone increased elasticity. Films with all additives were the most flexible. Contact angle analysis indicated changes in surface wettability, particularly in gluconolactone and mixed formulations.. Among all tested variants, the film containing the MIX of all moisturizing agents demonstrated

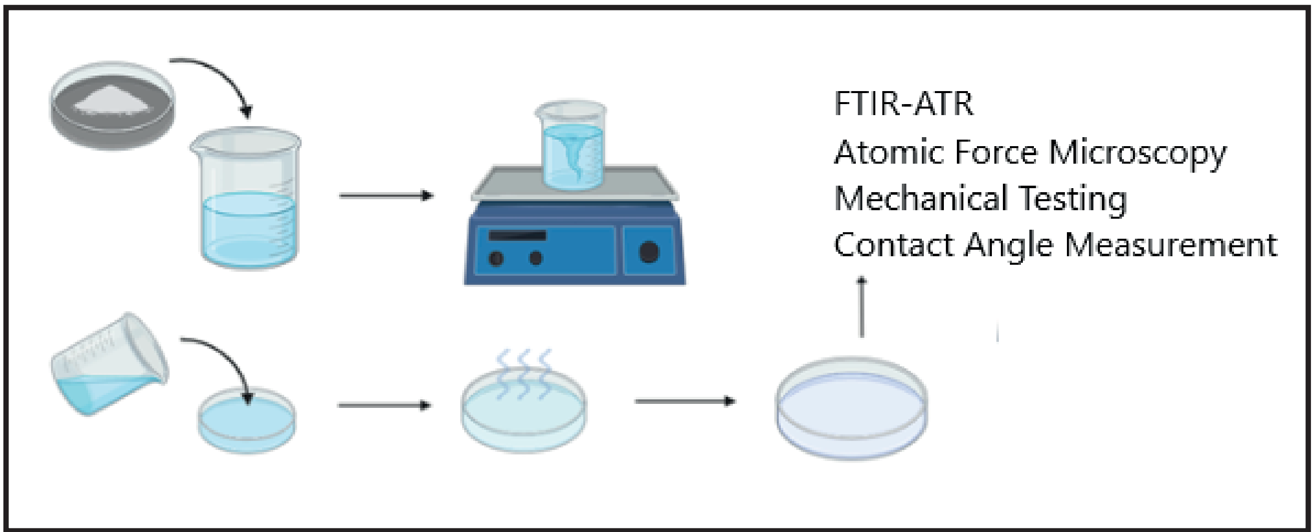

FIG. 4. A graphical summary of the testing workflow.

the most balanced profile in terms of elasticity, extensibility, hydrophilicity, and surface energy, making it the most promising candidate for cosmetic skin applications.

These findings suggest that the modified chitosan films represent promising and sustainable materials for skin-related applications. Given their favorable flexibility, hydration properties, and biocompatibility, such films may serve as innovative components in cosmetic facial masks, transdermal patches, or biodegradable wound dressings. The versatility of the system also opens possibilities for incorporating active pharmaceutical or cosmetic agents. Future studies may explore the long-term skin compatibility, controlled release behavior, and potential scale-up of these formulations in industrial applications.

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