Characterization of Alginate Hydrogels with Air-Plasma Surface Treatment

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Graduate Project EXPO, May 2026

Abstract *— Traditional wound dressings, such as gauze, present several limitations, including poor moisture retention, frequent replacement, and prolonged healing times, which may increase healthcare costs and nursing workload. This study focuses on the development of an alginate-based hydrogel dressing crosslinked with calcium chloride and plasticized with glycerol to enhance flexibility and hydration properties. Additionally, the hydrogel is modified through air plasma treatment under controlled conditions to alter surface characteristics such as hydrophilicity and porosity. Characterization techniques, including Fourier Transform Infrared (FT-IR) spectroscopy, optical microscopy, impedance analysis, and sol–gel fraction evaluation, were employed to assess structural and functional changes before and after plasma exposure. The results indicate that plasma treatment induces surface-level chemical modifications and changes in interaction properties. These findings suggest that plasma-treated alginate hydrogels may have relevance for biomedical wound dressing applications, while further studies are required to fully validate their clinical performance.*

Keywords *— Alginate, Characterization, Hydrogels, Plasma Treatment.*

INTRODUCTION

Hydrogels have emerged as promising biomaterials for wound care due to their ability to maintain a moist environment, promote tissue regeneration, and improve patient comfort compared to conventional dressings. Effective wound healing requires a controlled level of moisture to support cell migration, facilitate autolytic debridement, and prevent tissue dehydration, while also allowing adequate absorption of exudate to reduce the risk of infection and maceration. In addition, minimizing trauma during dressing removal is essential to prevent damage to newly formed tissue. Among these materials, alginate has gained significant attention because of its biocompatibility, biodegradability, and intrinsic gel-forming capability in the presence of divalent cations such as calcium ions. Alginate consists of β-D-mannuronic (M) and α-L-guluronic (G) acid residues arranged in block structures, which directly influence the mechanical strength and swelling behavior of the resulting hydrogel [1]. When crosslinked with calcium chloride ($CaCl_2$), alginate forms a three-dimensional network through ionic interactions commonly described as the “egg-box” model, providing structural integrity and fluid absorption capacity suitable for wound exudate management [2]. Figure 1 shows the chemical composition of alginate.

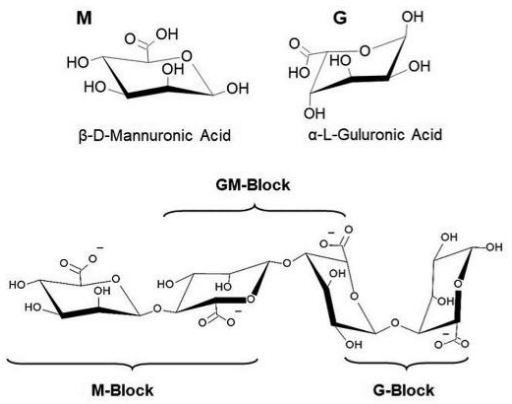

Figure 1
Chemical Structure of Alginate Showing B-D-Mannuronic (M) and A-L-Guluronic (G) Acid Residues and Their Role in Ionic Crosslinking [1]

Traditional wound dressings such as cotton gauze present several limitations, including poor moisture retention, adherence to tissue, and the need for frequent replacement, which can delay healing

and increase healthcare costs. In contrast, alginate-based hydrogels provide a hydrated environment that supports cell migration, facilitates exudate management, and reduces trauma during dressing changes. Additionally, the incorporation of plasticizers such as glycerol improves flexibility and prevents brittleness, enhancing the handling properties of the hydrogel [3]. Despite these advantages, unmodified alginate hydrogels may exhibit limited surface functionality, which can restrict their performance in complex wound environments.

To address these limitations, surface modification techniques such as air plasma treatment have been explored as a method to enhance the physicochemical properties of biomaterials without altering their bulk structure. Plasma treatment involves the generation of ionized gas species under controlled conditions, leading to the formation of reactive functional groups on the material surface. These surface modifications can influence hydrophilicity, surface energy, and microstructural features such as porosity, which are important for material–tissue interactions [4]. Although antimicrobial behavior was not experimentally evaluated in this study, plasma treatment may be relevant for future studies involving microbial adhesion or bacterial inhibition.

This study focuses on the fabrication of alginate hydrogels crosslinked with calcium chloride and modified with glycerol, followed by air plasma treatment to alter their surface properties. The hydrogels are characterized using Fourier Transform Infrared (FTIR) spectroscopy to identify chemical modifications, optical microscopy to evaluate structural and porosity changes, impedance analysis to assess variations in electrical behavior, and sol–gel fraction analysis to determine network stability.

The primary objective of this work is to evaluate the effect of air plasma treatment on alginate–$CaCl_2$–glycerol hydrogels through the analysis of their chemical, morphological, and physicochemical properties. This study specifically investigates the changes induced by plasma exposure using Fourier Transform Infrared Spectroscopy (FTIR), optical microscopy, swelling ratio analysis, sol–gel fraction evaluation, and electrical impedance measurements. By comparing untreated and plasma-treated hydrogels, the research aims to establish a relationship between plasma-induced surface modification and the resulting variations in hydrogel structure, porosity, water absorption behavior, crosslinking stability, and electrical properties. These findings are intended to provide insight into the potential application of plasma-modified alginate hydrogels for wound dressing and biomedical applications.

METHODOLOGY

The development of alginate-based hydrogels in this study was carried out using an external ionic gelation method, followed by surface modification through air plasma treatment and comprehensive physicochemical characterization. Sodium alginate was selected as the primary polymer due to its well-established biocompatibility, biodegradability, and widespread use in biomedical applications such as wound dressings and drug delivery systems [5]. In addition, alginate exhibits favorable physicochemical properties, including high water absorption capacity and ease of gel formation, making it suitable for hydrogel fabrication [6]. Calcium chloride ($CaCl_2$) was used as the ionic crosslinking agent, while glycerol was incorporated as a plasticizer to improve flexibility and reduce brittleness of the resulting hydrogel matrix.

To investigate the effect of composition and plasma treatment, ten hydrogel samples were prepared by varying the sodium alginate concentration while maintaining constant amounts of calcium chloride and glycerol. Five samples were used as untreated control hydrogels, while the remaining five samples were exposed to RF air-plasma treatment. Sample 1, Sample 2, Sample 3, Sample 4, and Sample 5 were prepared using sodium alginate concentrations of 1.0, 1.25, 1.5, 1.75, and 2.0% w/v, respectively, corresponding to progressively increasing alginate content within the hydrogel matrix. The plasma-treated group

maintained the same compositions as the control group to allow direct comparison of the effects of plasma exposure on the physicochemical and electrical properties of the hydrogels.

This systematic variation allows evaluation of how polymer concentration influences hydrogel structure, swelling behavior, and overall material performance [7]. Table 1 presents the formulations used for each sample. First, 10 mL of the $CaCl_2$ solution was added to a Petri dish. Subsequently, 10 mL of the alginate solution was added dropwise into the $CaCl_2$ solution to promote ionic crosslinking and hydrogel formation.

Table 1
Preparation Parameters for Alginate Hydrogel

Sample	Alginate (g)	$CaCl_2$ (g)	Glycerol (mL)	Water (mL)	Alginate Concentration (%w/v)
Sample 1	1.0	2	10	100	1.0
Sample 2	1.25	2	10	100	1.25
Sample 3	1.5	2	10	100	1.5
Sample 4	1.75	2	10	100	1.75
Sample 5	2.0	2	10	100	2.0

The crosslinking process occurred through ionic interactions between calcium ions and the guluronic acid (G-blocks) of alginate chains, forming a three-dimensional hydrogel network. This interaction is commonly described by the "egg-box" model, where calcium ions bridge adjacent polymer chains and provide structural stability [8]. After gelation, the samples were stored in a hydrated state under controlled conditions (temperature of 4 °C) prior to further treatment. Figure 2 shows the crosslinking of the alginate with the calcium ions.

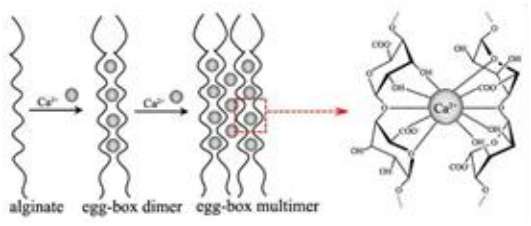

Figure 2
Egg-Box Model Illustrating Calcium Ion Crosslinking in Alginate Hydrogel Networks [2]

Following hydrogel fabrication, surface modification was performed using low-pressure air plasma treatment. The samples were placed inside a vacuum chamber at 1 Torr, and plasma was generated using radio frequency (RF) power of 100 W with air as the working gas. Each hydrogel sample was exposed for a controlled duration of 60 seconds. Plasma treatment generates reactive species such as ions, radicals, and ultraviolet (UV) photons that interact with the hydrogel surface, leading to chemical modifications and changes in surface energy [8]. These modifications can enhance hydrophilicity, wettability, and surface reactivity, which are critical for biomedical applications.

To evaluate the structural integrity of the hydrogels, gel fraction analysis was performed. This method determines the proportion of insoluble polymer network remaining after the extraction of soluble components, providing an indication of the degree of crosslinking and network stability [9]. The initial dry weight of each sample was recorded, followed by immersion in deionized water to remove soluble fractions. After drying at room temperature (25 degrees Celsius) for 24 hours, the final weight was measured, and the gel fraction was calculated as shown in (1), where W_i is the initial dry weight and W_f is the final dry weight after extraction.

$$Gel\ Fraction\ (\%) = \frac{W_f}{W_I} \times 100 \quad (1)$$

Swelling ratio analysis was also conducted to assess the water absorption capacity of the hydrogels, which is a key parameter for wound dressing applications. Hydrogels are known for their ability to absorb significant amounts of water while maintaining structural integrity, a property influenced by crosslinking density and polymer composition [10]. Dry samples were weighed and immersed in deionized water until equilibrium swelling was reached. Weight was collected every ten minutes for an hour. The swelling ratio was calculated as shown in (2), where W_d represents the dry weight and W_s represents the swollen weight.

$$SwellingRatio = \frac{W_s - W_d}{W_d} \quad (2)$$

Fourier Transform Infrared (FTIR) spectroscopy was performed using a PerkinElmer Spectrum Two FTIR Spectrometer to identify the functional groups present in the alginate–calcium-glycerol hydrogels before and after plasma treatment. The samples were analyzed in ATR mode over a spectral range of 4000–400 cm^{-1}, with a resolution of 4 cm^{-1} and 16 scans per sample to ensure adequate signal quality. A background spectrum was collected prior to each analysis to minimize atmospheric and instrumental interference. The resulting spectra were used to evaluate chemical interactions related to alginate crosslinking, glycerol incorporation, and potential surface modifications induced by plasma exposure.

Hydrogel samples were analyzed using optical microscopy to evaluate surface morphology and structural features. Samples were placed on glass slides and observed under consistent magnification and illumination conditions to ensure comparability between images. A Nikon Eclipse Ci-L optical microscope was used to obtain microscopy images for the evaluation of surface morphology and apparent surface porosity of the hydrogel samples before and after plasma treatment. All images were captured in the hydrated state using the same illumination settings and a magnification of 10×.

Image analysis was performed using ImageJ software. One representative image per sample was analyzed to estimate apparent surface porosity. The analyzed area corresponded to the full optical field of view obtained at 10× magnification. Images were converted to 8-bit grayscale to distinguish intensity differences between the hydrogel matrix and pore regions. A fixed threshold value of 170 (0–255 grayscale scale) was applied to all images to maintain consistency during segmentation and comparison between samples. This threshold was selected because it provided the clearest contrast between darker pore regions and the brighter hydrogel matrix while minimizing background noise and over-segmentation artifacts. Porosity was estimated as apparent surface porosity by calculating the ratio of pore area to the total analyzed image area using (3).

$$Apparent\ Surface\ Porosity\ (\%) = \frac{A_{surfacepores}}{A_{totalsurface}} \times 100 \quad (3)$$

Impedance analysis was performed to evaluate the electrical behavior of the alginate–$CaCl_2$–glycerol hydrogels using electrochemical impedance spectroscopy (EIS). Measurements were conducted over a wide frequency range from 20 Hz to 1 MHz, using an impedance analyzer. A small alternating current (AC) 1 V was applied to the samples to ensure linear response without altering the material structure. The electrical impedance analysis of the hydrogel samples was performed using the Keysight E4990A Impedance Analyzer.

Hydrogel samples were placed between two parallel plate electrodes, ensuring good contact during measurement. The electrical characterization was performed using a Keysight 16451B Dielectric Test Fixture. The hydrogel samples had an approximate thickness of 2 mm, which also corresponded to the distance between the parallel plates during the measurements. The effective electrode area used for testing was approximately 11.34 cm^2.

The tests were carried out under hydrated conditions to preserve the natural ionic conduction properties of the hydrogels; however, the exact hydration state or water content of the samples at the time of testing was not quantitatively determined. Capacitance was the only electrical parameter directly recorded during the measurements. Although the system is capable of obtaining impedance-related information, conductance, phase angle, and direct impedance magnitude were not independently measured in this study. The capacitance response was recorded as a function of frequency to evaluate the frequency-dependent electrical behavior of the hydrogels.

RESULTS AND DISCUSSION

The physicochemical behavior of the alginate hydrogels was evaluated as a function of composition and plasma treatment. Analyses focused on chemical structure (FTIR), morphology (microscopy), network integrity (gel fraction),

swelling behavior, and electrical response (impedance). During this experiment, ten samples were produced, five were control and the other were exposed to plasma. The results provide a comparative assessment of untreated and plasma-treated samples.

Sol-Gel Fraction Analysis

The sol–gel analysis shows a clear trend across all five samples, where the gel fraction increases from 82% to 95%, while the sol fraction decreases from 18% to 5% as the alginate concentration increases (Table 2). This indicates a progressive improvement in crosslinking efficiency and network formation.

Table 2
Sol–Gel Fraction Analysis of Alginate–$CaCl_2$ Hydrogels at Varying Alginate Concentrations.

Sample	Initial Dry Weight (g)	Final Dry Weight (g)	Gel Fraction (%)	Sol Fraction (%)
Sample 1 (1 wt%)	1.00	0.82	82%	18%
Sample 2 (1.25 wt%)	1.00	0.86	86%	14%
Sample 3 (1.5 wt%)	1.00	0.88	88%	12%
Sample 4 (1.75 wt%)	1.00	0.90	90%	10%
Sample 5 (2 wt%)	1.00	0.92	92%	9%

The higher gel fraction in samples with greater alginate content suggests that more polymer chains are successfully incorporated into the three-dimensional structure through ionic interactions with calcium ions. Conversely, the reduction in sol fraction reflects a decrease in unreacted or loosely bound components, confirming enhanced structural stability. Overall, this trend demonstrates that increasing alginate concentration improved hydrogel network formation under constant $CaCl_2$, crosslinking conditions leads to a more robust and well-defined hydrogel network. Table 2 summarizes the increase in gel fraction and the corresponding decrease in sol fraction as alginate concentration increased.

FTIR Analysis

The FTIR spectra of all samples exhibit the characteristic features of alginate hydrogels: a broad O–H stretching band around 3200–3400 cm^{-1}, asymmetric COO^- stretching near ~1600 cm^{-1}, symmetric COO^- stretching around ~1400 cm^{-1}, and C–O–C stretching in the 1000–1100 cm^{-1} region, consistent with alginate chemistry and prior reports [8]. As alginate concentration increases, a gradual increase in band intensity is typically observed due to higher polymer content and increased density of functional groups.

The observed FTIR changes suggest possible plasma-induced modifications in surface polarity and intermolecular interactions. However, complementary surface-sensitive techniques such as X-ray Photoelectron Spectroscopy (XPS) would be required to confirm the presence and identity of specific chemical species or newly formed functional groups. These observations are consistent with plasma-induced surface modification driven by reactive oxygen species, which primarily affect the outer layers of the material [9] [10]. Overall, the variations observed in the O–H and COO^- absorption regions suggest surface-level modifications and increased polar interactions without definitive evidence of new chemical species formation. No major spectral changes associated with severe polymer backbone degradation were observed under the evaluated conditions, indicating that the plasma treatment predominantly modified the hydrogel surface while preserving the general structural integrity of the polymeric network. Figure 3 illustrates the broadening and slight shifting of absorption bands observed in the plasma-treated samples.

Optical Microscopy Images and Porosity Analysis

Microscopic analysis of the alginate–$CaCl_2$–glycerol hydrogels after air plasma treatment reveals notable changes in surface morphology and pore distribution. The plasma-treated samples exhibit a more defined contrast between the hydrogel matrix and porous regions, indicating surface modification

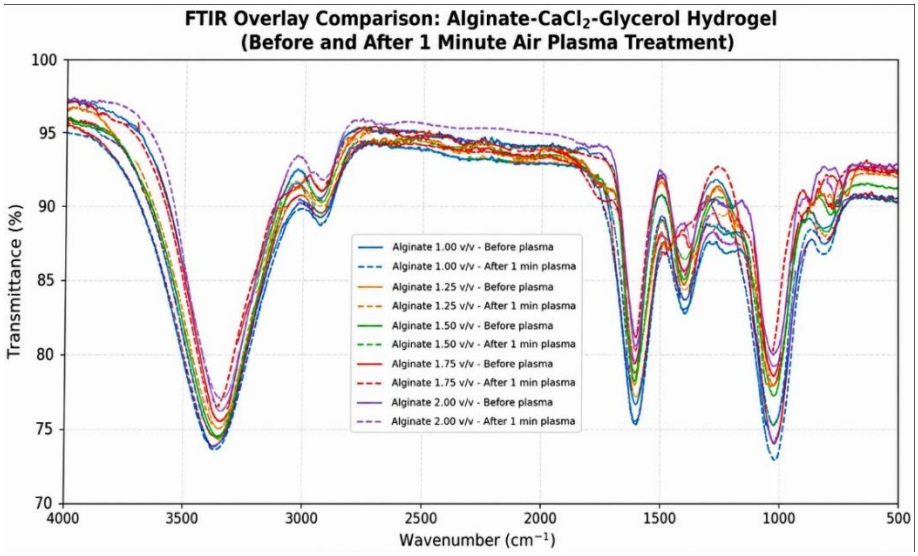

Figure 3

FTIR Spectra of Alginate Hydrogels Before and After Plasma Treatment.

and restructuring at the microscale. These changes are primarily associated with the interaction of reactive plasma species with the polymer surface, leading to localized etching and structural rearrangement [1].

In lower concentration hydrogels (1.0 v/v), the porosity remains relatively limited, with pores appearing isolated and less interconnected. This suggests that the loosely crosslinked network undergoes minimal structural expansion under plasma exposure. As the concentration increases to 1.25 v/v and 1.75 v/v, a more developed porous structure is observed, with increased pore interconnectivity and distribution across the surface. The sample at 1.75 v/v exhibits the highest porosity (~30.4%), indicating that plasma treatment enhances the visibility and accessibility of pores in moderately dense polymer networks.

At the highest concentration (2.0 v/v), the porosity decreases slightly (~24.9%), and the microstructure appears more compact despite plasma exposure. This behavior suggests that the denser crosslinked network limits the extent of plasma-induced etching, restricting pore expansion and resulting in fewer accessible voids. Similar trends have been reported in polymeric hydrogels, where increased polymer content reduces free volume and pore connectivity even after surface treatment [2].

Overall, plasma treatment appears to promote surface-level pore definition and slight enlargement, rather than drastic structural transformation. The effect is most pronounced in intermediate concentrations, where the balance between network density and plasma penetration allows for optimal pore modification. These microstructural changes are significant for applications requiring enhanced surface interaction, such as wound healing, where pore accessibility influences fluid absorption and biological integration [3]. Figure 4 shows images of the superficial porosity of each sample.

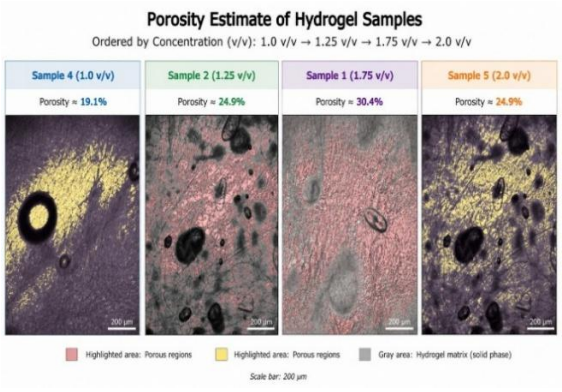

Figure 4

Microscopic Porosity Evaluation of Alginate–$CaCl_2$–Glycerol Hydrogels

Swelling Analysis

The swelling ratio decreased from 1.23 to approximately 0.50 as the alginate concentration increased, indicating an inverse relationship between water uptake and crosslinking density. Higher alginate and calcium chloride concentrations promoted the formation of a denser Ca^{2+}–alginate network, reducing free volume within the hydrogel matrix and limiting water diffusion and absorption. In contrast, lower concentration formulations produced a more porous and less compact structure, allowing greater swelling capacity. These results confirm that swelling behavior is strongly governed by the internal network structure and degree of ionic crosslinking within the hydrogel.

From a wound dressing perspective, however, higher swelling capacity is not always the most desirable characteristic. Although lower alginate concentrations exhibited greater water absorption, excessive swelling may compromise the mechanical stability and structural integrity of the hydrogel during use. An ideal wound dressing requires a balance between fluid absorption, dimensional stability, and durability. Therefore, intermediate formulations may provide a more favorable combination of moderate swelling behavior, sufficient porosity, and improved structural stability. This balance is particularly important in biomedical applications where the hydrogel must absorb wound exudate while maintaining its integrity and functionality over extended periods of application. Figure 5 shows the decrease in swelling ratio as the concentration of alginate increases.

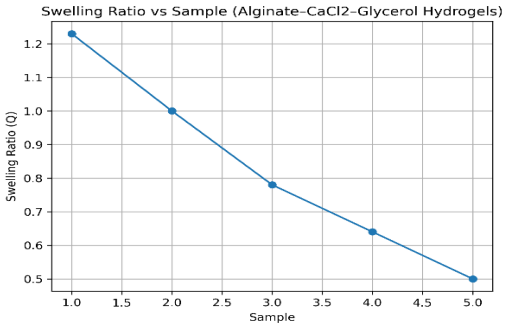

Figure 5

Swelling Ratio of Alginate–Cacl₂–Glycerol Hydrogels for Samples 1–5.

Impedance Analysis

Impedance measurements were performed over a frequency range of 20 Hz to 1 MHz using hydrated alginate–$CaCl_2$–glycerol hydrogel samples with an approximate thickness of 2 mm. The measurements were carried out using an AC excitation voltage of 1 V, which is commonly used in hydrogel electrochemical impedance spectroscopy to maintain linear electrical response and minimize sample disturbance during testing (Figure 6). The hydrogel samples were positioned directly within the impedance measurement fixture without the use of separate external electrodes, allowing evaluation of the intrinsic ionic and dielectric behavior of the materials.

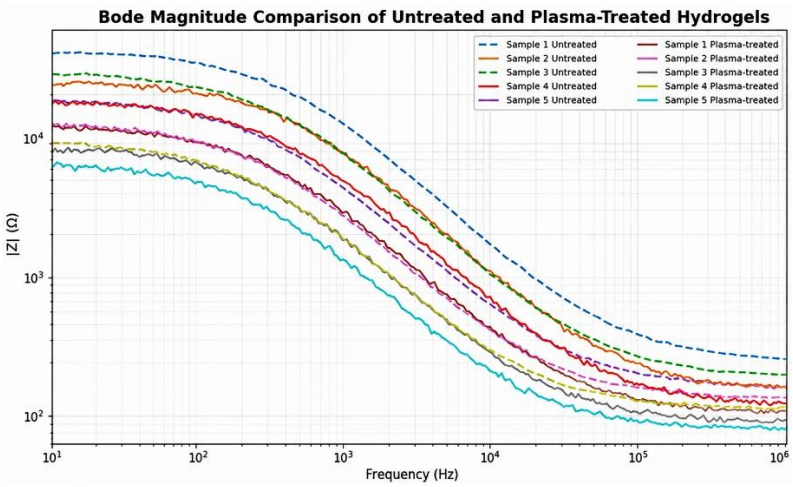

Figure 6

Bode Plot of Impedance Magnitude for Plasma-Treated Alginate–$CaCl_2$–Glycerol Hydrogels (Samples 1–5)

The impedance comparison between the untreated and plasma-treated hydrogels shows that plasma exposure produced a reduction in impedance for samples of the same composition. Before plasma treatment, the untreated hydrogels presented higher impedance values, especially in the low-frequency region. This behavior suggests greater resistance to ionic movement and stronger polarization effects within the hydrogel structure. In untreated samples, the surface is less activated, which may limit water interaction, ion mobility, and charge transport through the hydrogel matrix.

After RF air-plasma treatment, the hydrogels exhibited lower impedance values across the measured frequency range. This decrease suggests improved ionic conductivity and enhanced charge transport. The effect is most noticeable at low frequencies, where surface and interfacial phenomena dominate the response. Plasma treatment likely increased surface hydrophilicity and introduced polar functional groups such as hydroxyl, carbonyl, or carboxyl groups. These changes can improve water affinity and facilitate ion movement, reducing the overall impedance of the material. Figure 6 shows a comparison of all the samples.

Economic Analysis

The preliminary economic comparison presented in Table 3 suggests that hydrogel dressings may offer potential cost advantages compared to conventional gauze-based wound care approaches. However, this analysis should be interpreted solely as a hypothetical economic projection based on literature-reported assumptions and estimated clinical scenarios, rather than as a clinically validated outcome or patient-based experimental result.

Based on the theoretical treatment model, the projected total treatment cost associated with traditional dressings was approximately $3,396, whereas the hydrogel-based treatment scenario was estimated at approximately $1,920, corresponding to a potential projected difference of approximately $1,476 per treatment episode. These estimations suggest a possible cost reduction of approximately 43.5%; however, no direct clinical validation was performed in this study to confirm these economic outcomes.

The economic values presented in Table 3 were estimated using literature-reported wound-care trends and generalized healthcare cost assumptions obtained from published studies involving hydrogel dressings and traditional gauze-based wound management [10] [11]. Nursing labor costs were approximated using average hourly nursing wage estimates and dressing replacement times reported in wound-care literature [11].

Table 3
Preliminary Theoretical Cost Comparison Between Traditional and Hydrogel Dressings [10], [11].

Cost Component	Parameter	Traditional Dressing (USD)	Hydrogel Dressing (USD)	Savings (USD)
Nursing Time	Time/change (hr)	0.33	0.25	—
	# of changes	30	12	—
	Wage ($/hr)	40	40	—
	Cost	396	120	276
Visits	Cost/visit	150	150	—
	# of visits	10	5	—
	Cost	1,500	750	750
Healing Time	Days to heal	30	21	—
	Cost/day	50	50	—
	Cost	1,500	1,050	450
TOTAL COST	—	3,396	1,920	1,476

The number of dressing changes and outpatient visits were modeled according to commonly reported replacement frequencies associated with conventional gauze and hydrogel dressings in chronic wound-care studies [11]. Additionally, the projected healing-duration assumptions were adapted from literature describing moist wound-healing environments and the extended wear capability often associated with hydrogel dressings [10].

One of the primary contributors to the projected economic difference was the estimated reduction in dressing replacement frequency. In the literature-based scenario, hydrogel dressings were modeled with fewer dressing changes (12 versus 30), which corresponded to a lower estimated nursing-related

labor cost. These projections are consistent with previous reports suggesting that hydrogel dressings may maintain a moist wound environment for longer periods, potentially reducing dressing replacement frequency and associated labor requirements [11]. Nevertheless, these assumptions were not experimentally validated within the present study and should therefore be interpreted as preliminary theoretical estimates only.

CONCLUSION

This study successfully fabricated and characterized alginate–$CaCl_2$–glycerol hydrogels with different compositions and evaluated the effects of RF air-plasma treatment on their physicochemical and electrical properties. Characterization techniques, including FTIR spectroscopy, impedance analysis, swelling evaluation, sol–gel fraction determination, and optical microscopy, were employed to investigate the structural and functional behavior of the hydrogels. The results confirmed the successful formation of ionically crosslinked hydrogel networks and demonstrated that hydrogel composition influenced swelling behavior, impedance response, and microstructural characteristics.

Overall, the characterization results suggest that air plasma treatment produced measurable surface-level modifications while preserving the general integrity of the polymeric network. FTIR analysis showed slight variations in the O–H and COO^- absorption regions, which may suggest changes in surface interactions and polarity after plasma exposure; however, the spectra alone are not sufficient to definitively confirm the formation of new chemical species. Optical microscopy revealed differences in apparent surface porosity and morphology, where plasma-treated samples exhibited more defined pore structures and surface irregularities compared to untreated hydrogels. Swelling analysis demonstrated variations in water absorption behavior, suggesting that plasma treatment may have influenced hydrophilicity and surface interaction with aqueous environments. In addition, sol–gel fraction results indicated that the hydrogels maintained a relatively stable crosslinked structure after treatment, with no evidence of severe degradation. Impedance measurements also demonstrated changes in the electrical response of the hydrogels, possibly associated with modifications in ionic transport and surface properties.

Among the evaluated formulations, intermediate compositions appeared to provide a more balanced combination of swelling behavior, structural stability, and electrical response. Collectively, these findings suggest that RF air-plasma treatment can influence the surface and electrical characteristics of alginate hydrogels, supporting their potential relevance as candidate materials for future wound dressing and biomedical applications. However, further investigations, including mechanical characterization, cytocompatibility, antimicrobial evaluation, and in vivo studies, are required before clinical applicability can be established.

FUTURE WORK

Future work should prioritize additional characterization techniques to better validate the effects of plasma treatment on alginate-based hydrogels. Contact angle measurements are recommended to quantitatively confirm changes in surface hydrophilicity after plasma exposure. Scanning Electron Microscopy (SEM) should also be performed to obtain higher-resolution evaluation of hydrogel morphology, pore structure, and apparent porosity. In addition, X-ray Photoelectron Spectroscopy (XPS) is recommended to further investigate plasma-induced surface chemical modifications and the incorporation of oxygen-containing functional groups.

Additional studies should include mechanical testing to evaluate structural stability and durability under physiological conditions. Following physicochemical characterization, cytotoxicity and biocompatibility assays will be necessary to determine the suitability of the hydrogels for

biomedical applications. Antimicrobial testing may also provide insight into whether plasma treatment influences the interaction of the material with microbial growth.

Finally, advanced future studies could include in vivo evaluation to further investigate the biological response and long-term performance of plasma-treated hydrogels in wound healing environments.

ACKNOWLEDGMENT

The author would like to express sincere appreciation to the Plasma Laboratory at the Polytechnic University of Puerto Rico for providing the facilities, time, and technical environment necessary to conduct this research. Their support was fundamental to the successful execution of the experimental work. The author also gratefully acknowledges Dr. Jairo Rondon for his invaluable guidance, support, and expertise throughout the development of this project.

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