Hindawi

Research Article

Effect of Collagen/Ibuprofen Hydrogel in Wound Healing: An In Vivo Study

Kamyar Abbasi [1] **Reza Eftekhar Ashtiani** [1] **Mitra Abdolahi,**[2] **Maryam Hosseini,**[3]
Reza Sayyad Soufdoost [4] **Mostafa Alam** [5] **and Sadaf Fani-Hanifeh** [6]

[1]*Department of Prosthodontics, School of Dentistry, Shahid Beheshti University of Medical Science, Tehran, Iran*
[2]*Department of Pathology, School of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran*
[3]*School of Dentistry, Shahid Beheshti University of Medical Sciences, Tehran, Iran*
[4]*Dentistry Research Institute, School of Dentistry, Shahed University, Tehran, Iran*
[5]*Department of Oral and Maxillofacial Surgery, School of Dentistry, Shahid Beheshti University of Medical Science, Tehran, Iran*
[6]*Rajaie Cardiovascular Medical and Research Center, Iran University of Medical Sciences, Tehran, Iran*

Correspondence should be addressed to Mostafa Alam; aalaammostaafaa@gmail.com and Sadaf Fani-Hanifeh; sadaf.fn77@
gmail.com

Background and Aim. Wound healing is a complicated physiological process to preserve skin integrity after injuries and includes the proliferative phase, hemostasis/inflammatory phase, and remodeling through extracellular, intercellular, and intracellular components synchronization. The study aimed to examine human placental collagen/ibuprofen's effect on wound healing in an animal model. *Materials and Methods*. The cell viability test was performed by MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide) assay. In this animal study, three circular excisions were made in the back of each of the 12 rats and injected with the following groups: collagen/ibuprofen, group 1; collagen, group 2; and unfilled as the control group, group 3 for two times. The healing procedures were explored via calculating wound contraction percentage after taking photographs on days 7 and 14. Also, histopathological assessments were conducted on days 7 and 14. *Results*. The MTT results showed no significant cytotoxicity, and macroscopic results demonstrated that the percentage of wound contraction was 85%, 79%, and 41% in the collagen/ibuprofen, collagen, and control groups on 14th day, respectively. Histopathological assessments showed that hair follicles number, fibroblasts content, and angiogenesis in the collagen/ibuprofen were remarkably more significant than collagen and control groups. *Conclusion*. The collagen/ibuprofen group had significant outcomes compared with collagen and the control groups in wound healing and wound contraction. Collagen/ibuprofen hydrogel can be a potential hydrogel in accelerating wound healing.

1. Introduction

Skin wounds are prevalent injuries that generally occur via different causes involving metabolic diseases such as diabetes mellitus and thermal or physicochemical damages. Skin wounds have the potential to induce morbidity along with imbalance of function, disability, and elevated pain [1]. The statistical analysis estimated that about 5.6 million people in the United States are impacted via wound lesions,

and treatment costs over 50 billion dollars each year. Also, skin wounds can be categorized into chronic and acute wounds. Acute wounds usually heal themselves sufficiently and smoothly [2]. Contrarily, circumstances such as related disorders, inadequate nutrition, the extensive extension of the wound, age, and impairment of the skin regeneration process lead to chronic wounds [3]. Wound treatment concluded with wound debridement, pressure off-loading, and infection management with topical antibiotics and

topical antiseptics also revascularization, which are the main principles of therapeutic wound approaches [4]. There are different advances in debridement such as surgical processes, biosurgery, wet to dry dressing, preparations of enzymes, polysaccharide beads or dextranomer polysaccharide paste, and hydrogels. When wound dressing alters the wound from moist to dry, they provide nonselective debridement, hence cleaning the wound with necrotic tissue removal. It is supposed that wounds be investigated at each dressing change. Moisture-retentive dressing concluding films, hydrocolloids, hydrofibres, alginates, and hydrogels are helpful in diverse clinical surroundings [5]. One of the critical clinical problems is wound care therapy, which possesses several procedures containing inflammation, hemostasis, proliferation, angiogenesis, connective tissue remodeling, and wound strength recovery. Regeneration and spontaneous growth generally restore the integrity of damaged tissue in the wound healing process. The endogenous release of chemokines, cytokines, and growth factors is crucial for restoring wound tissue integrity that mediates the interaction between fibroblasts, keratinocytes, neutrophils, endothelial cells, and macrophages [6]. Pain management is a significant burden with chronic wounds in patients. Also, cells release pain neuropeptides responding to a broad range of stimuli, and responses to pain stress can be detrimental to amelioration. Neuropeptides generally induce monocytes, leukocytes, and other immunoreactive cells to discharge proinflammatory cytokines [7]. An imbalance of proinflammatory cytokines impacts wound healing negatively, subscribing to related inflammatory disorders and leading to the destruction of tissues. Multiple advances to prevent acute pain and inflammation in wound care have been assessed. Also, ibuprofen-loaded materials with controlled-release actions have been utilized to prevent immoderate inflammation through the early phases of healing, attenuate pain, and stimulate the repairing of tissues in the duration of later phases [8]. Ibuprofen has been classified in a nonsteroidal anti-inflammatory drug (NSAID) class to treat acute pain, inflammatory, and degenerative diseases by prohibiting cyclooxygenases (COX) pathways. Ibuprofen is biotransformed rapidly due to its short half-life and must be used rapidly to preserve effective plasma concentration. Nevertheless, multiple doses of ibuprofen can hinder compliance and decrease clinical efficacy [9]. Moreover, ibuprofen's sustained and controlled release systems can improve the conditions of therapeutic burdens by stimuli-responsive carriers providing demanding drug release [10]. Similarly, with injectable hydrogels, temperature-inducible, localized drug delivery systems may modulate drug release and alleviate nonspecific drug distribution to healthy organs and tissues. Likewise, they can support inhibition of multiple drug-using and increase patients' compliance [11]. Collagen provides integrity of tissue structures and cells that generally recognize the most presented protein in the skin. Arranged myofibroblasts and fibroblasts in the fibril forms of tissues synthesize skin collagen regularly under shear stresses and tensile [12]. One of the forms of collagen is type I collagen, which exists in the fasciae, tendons, dermis, and components of essential scar tissue. Collagen is used to administer in the synthesization of skin substitutes that are also delivered in transmembrane proteins composition, fibril surfaces, or beaded structures [13]. Therefore, the purpose of the study was to investigate the effect of collagen and ibuprofen hydrogel on wound healing in the in vivo test and also assess the histopathological changes and the percentage of wound contraction.

2. Materials and Methods

2.1. Synthesis of Hydrogel. Collagen was extracted according to the protocol [14]. In brief, the human placenta was derived from the abdomen via a general surgeon. Afterward, the placenta was washed (distilled water, 4°C) and then chopped 1-2 cm on ice. Sodium hydroxide was used for removing the noncollagenous proteins (1N, refreshed every 2 h, stirred for 6 h). Then, extra fat was eliminated by butyl alcohol (10%, 24 h, filtered with cheesecloth). The precipitate of 20% collagen was acquired using NaCl (pH = 7, 3M, centrifuged 5000 g for 30 min). The remaining was dissolved in 2% acetic acid, transferred to dialysis tubing, stirred in distilled water (5 days), and freeze-dried [14]. The harvested collagen was immersed in diluted hydrochloric acid (pH = 2, to obtain 1% (w/v) solution) [15]. Then, ibuprofen (-2-(4-isobutylphenyl) propionic acid; Arora Pharmaceuticals, Delhi, India) (25 wt%) was added to the collagen solution while stirring. The obtained homogeneous suspension was stored at 4°C.

2.2. Cell Viability Test. The MTT (methyl thiazolyl tetrazolium) test was done directly according to the ISO 10993–5 standards. The cell culture plates maintain the negative control without cytotoxicity. MTT assay was conducted to assess the cytotoxicity of the hydrogel. Hence, 20000 adipose-derived stem cells (ADSCs) were added to the culture medium (50 μL), and 20000 fibroblasts were added to the culture medium (50 μL) as the negative control without cytotoxicity, separately which are containing serum and cultured on the hydrogel. The culture medium was added to each well after 3 hours and was removed. Afterward, MTT solution (100 μL, 0.5 mg/mL) was added to each well. The supernatant solution was eliminated after 4 hours. Then, DMSO (100 μl) was added to dissolve the formed purple formazan crystals. The solution optical density was detected at 570 nanometers wavelength in an ELISA reader (Convergent ELReader 96X). Findings were shown as a percentage (control value = 100.00%). Also, the assays were displayed in triplicate to acquire accurate outcomes. The cell viability was investigated via the following equation:

$$\textbf{Cell viability}\,(\%) = \frac{\text{Value of therapy sample}}{\text{Value of sample control}} \times 100. \qquad (1)$$

2.3. In Vivo Test. Twelve male Wistar rats (180–200 g and 3-4 weeks) were allocated in standard conditions (20–25°C, 65–75% humidity) under the supervision of a veterinarian. General anesthesia was induced by intramuscular injection of 2% xylazine (10 mg/kg) and 10% ketamine (50 mg/kg).

Afterward, three circular excisions (8 mm in diameter) in the back of each of the 12 rats were made using a punch on day 7 and day 14. The defective sites were injected as follows: collagen/ibuprofen (group 1), collagen (group 2), and un-filled control group (group 3). The extracted collagen (3 mg/ml) was mixed with 40 mg/ml of ibuprofen, and then, the mix was injected over the wound.

2.4. Macroscopic Analysis. In macroscopic analysis, wound contraction pictures were obtained of the excision wounds in all three groups using a digital camera 7 and 14 days after the intervention. The Image J software investigated the wound contraction pictures concerning the wound contraction percentage. The wound contraction percentage was calculated using the following equation .

$$\text{Wound contraction}\,(\%) = \frac{\text{Initial wound size} - \text{specific day wound size}}{\text{Initial wound size}} \times 100. \tag{2}$$

2.5. Microscopic Analysis. The rats were sacrificed after 7 and 14 days. The samples (each group with 1 cm area) were fixed in 10% formalin for 48 h. Then, the samples were dehydrated in a graded series of 80%–100% ethanol solution. All the specimens were located in paraffin, cut into a 5 μm section, and then stained with hematoxylin and eosin staining. The samples were evaluated and photographed using a light microscope. The histological investigations, including the numerical density of angiogenesis, hair follicles numbers, and fibroblasts, comprise a morphological description of at least three sections under a light microscope.

2.6. Statistical Analysis. The nonparametric Friedman test was used to compare the groups at $P < 0.05$ level of significance.

3. Results

3.1. Cytotoxicity Test. This cell viability investigation was performed at 24 and 48 hours. In this test, cells cultured on conventional culture dishes were considered control. According to Figure 1, the results showed that the survival of cells was less than the control group in 24 hours and overtime; an increasing trend of survival is observed, which may indicate that cells resumed their reproduction after tolerating a shock on the first day and went through an upward trend. Furthermore, the cell survival rate in 48 hours reached about 98% in both groups (Figure 1).

3.2. Macroscopic Results. The rats were sacrificed and photographed at 7 and 14 days, and wound contraction was evaluated. Figure 2 shows the macroscopic perspective of the wounds. The outcomes showed that the wound healing rate was higher in the collagen/ibuprofen group than in other groups. Wound contraction and healing in the collagen/ibuprofen group were more significant than in collagen and control groups. Wound contraction was 40%, 25%, and 17% in the collagen/ibuprofen, collagen, and control groups on day 7, respectively. On day 14, wound healing and contraction were more remarkable than day 7, and collagen/ibuprofen could significantly enhance wound healing. Wound contraction was higher in the collagen/ibuprofen group than in collagen and control groups. Wound

contraction was 85%, 79%, and 41% in the collagen/ibuprofen, collagen, and control groups on day 14, respectively (Figures 2 and 3).

3.3. Microscopic Results. Histopathological investigations reported proper wound healing in the collagen/ibuprofen group on day 7. The fibroblasts relocated into the wound site, and hair follicles were detected on day 14, which showed the crucial role of synthetic hydrogel in the improvement of wound healing and stimulation of other factors migration that possess practical functions in wound healing progression on day 14 (Figures 4 and 5). Hair follicles, blood vessels, and fibroblasts were discovered in the collagen/ibuprofen group in 7 days compared with other groups, indicating that this synthetic compound (collagen/ibuprofen) can play a fundamental role in the enhancement of wound healing. The procedure remained and expanded by day 14, which implied the positive response of cutaneous tissue to the synthetic hydrogel and its crucial part in the increment of wound healing (Figures 4 and 5). The rate of wound healing was higher in the collagen/ibuprofen group during 14 days compared to 7 days. The cells were transferred to the wound site and promoted angiogenesis on day 14 more than day 7. Wound healing was observed with collagen sheets and slight migration of cells into the wound site in the collagen/ibuprofen group; however, higher migration of cells and further angiogenesis were detected on day 14 in the collagen/ibuprofen group. Both hair follicles and angiogenesis were considerable in collagen/ibuprofen and collagen groups, but the number of fibroblasts in the collagen/ibuprofen group was higher than the collagen group. Wound healing in the collagen group began from the peripheral areas of the wound during 7 days and was enhanced by day 14, based on slight migration of cells to the wound area and increased angiogenesis.

On the contrary, wound healing was unremarkable in the control group, and negligible healing was detected under the layer of connective tissue. The wound healing rate was significantly higher on day 14 than on day 7 in the control group. The number of blood vessels, hair follicles, and fibroblasts (vital factors in wound restoration) was drastically lower in the collagen and control groups on days 7 and 14 compared to the collagen/ibuprofen group.
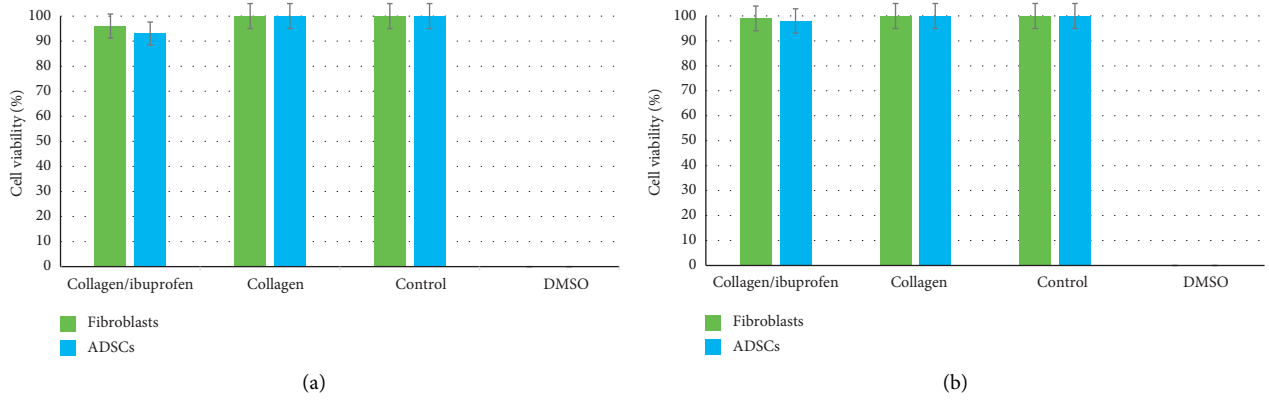
(a)

(b)

FIGURE 1: Cell viability of fibroblasts and ADSCs in control, collagen, and collagen/ibuprofen groups at 24 (a) and 48 (b) hours ($P < 0.05$).

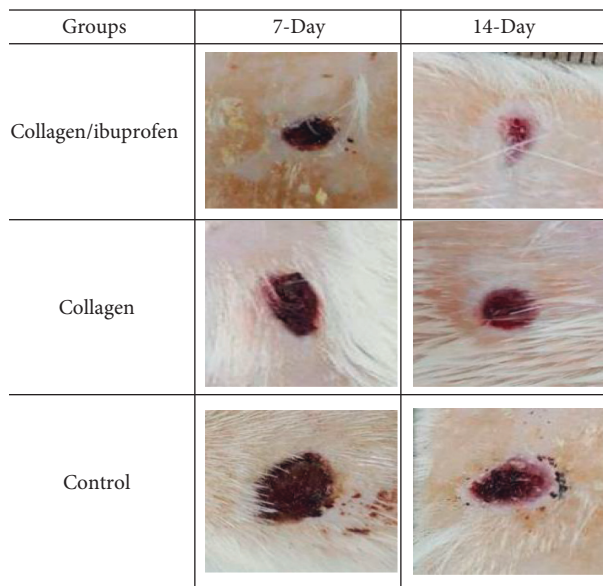| Groups	7-Day	14-Day
Collagen/ibuprofen		
Collagen		
Control		

FIGURE 2: Macroscopic investigations of control, collagen, and collagen/ibuprofen groups on days 7 and 14.
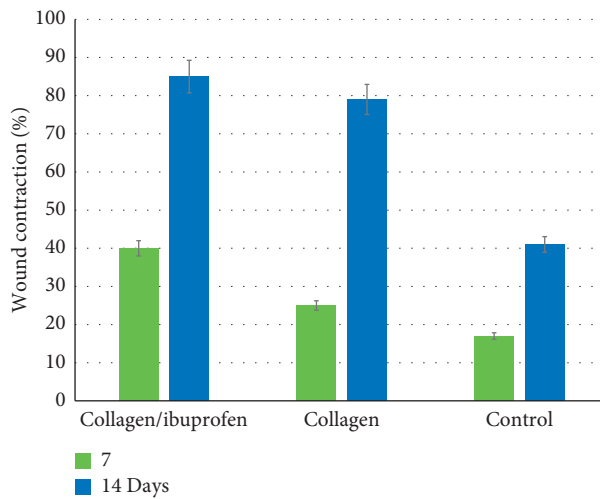
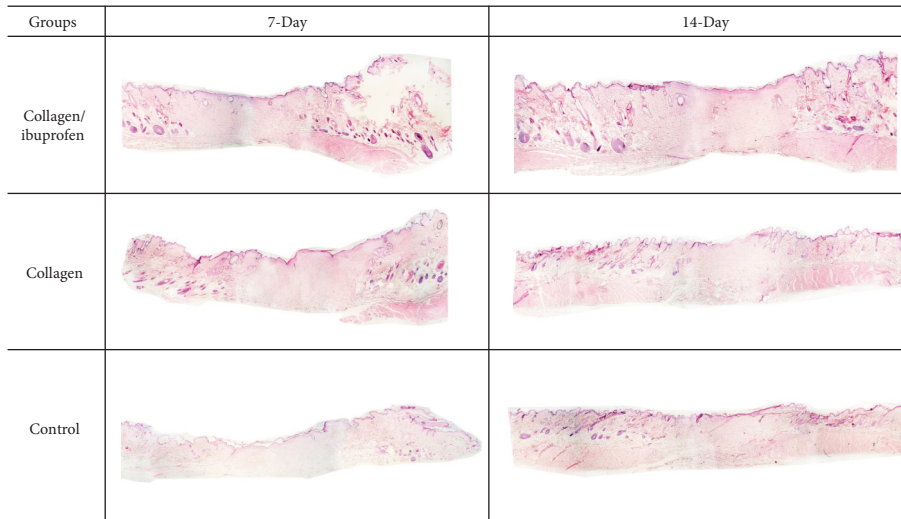FIGURE 3: Wound contraction of collagen/ibuprofen, collagen, and control groups on days 7 and 14 ($P < 0.05$).

Groups	7-Day	14-Day
Collagen/ ibuprofen		
Collagen		
Control		

FIGURE 4: Histopathological analysis of collagen/ibuprofen, collagen, and control groups on days 7 and 14 with H&E staining (×40).
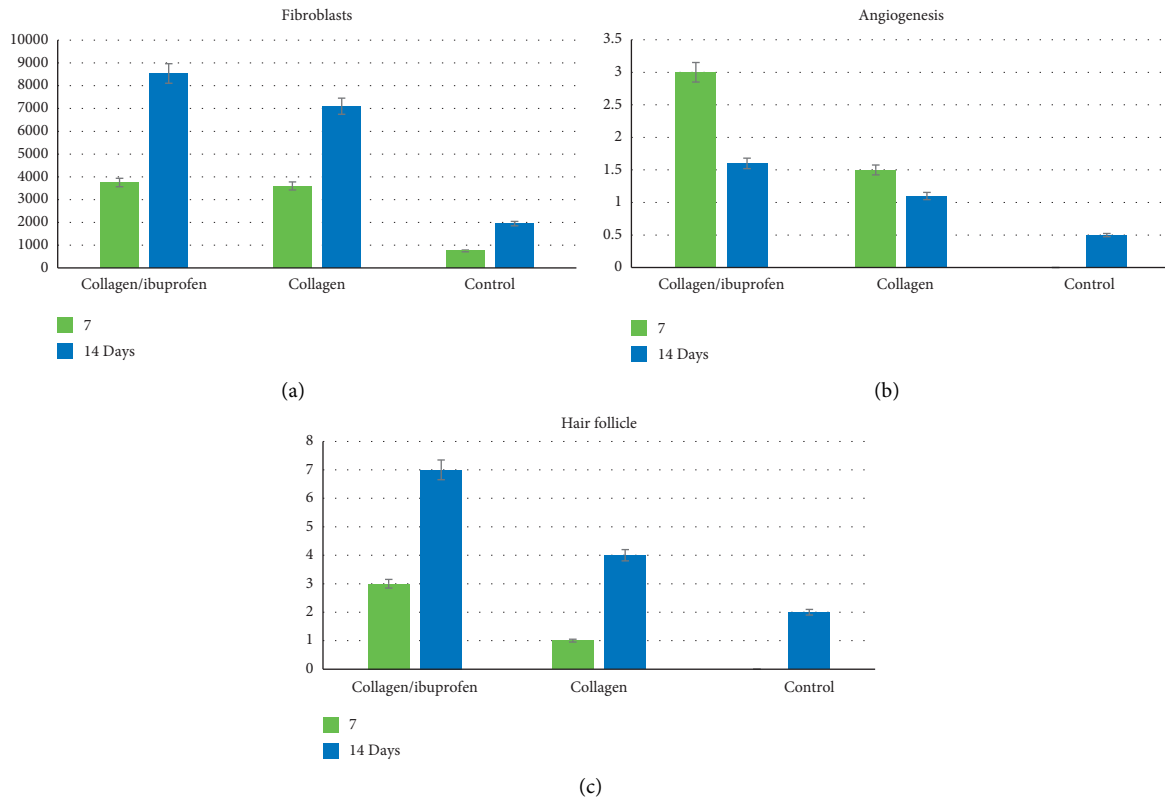

(a)

(b)

(c)

FIGURE 5: Fibroblasts (a), angiogenesis (b), and hair follicles (c) of collagen/ibuprofen, collagen, and control groups on days 7 and 14 ($P < 0.05$).

4. Discussion

The progression of functional therapy for increasing angiogenesis and wound healing, especially for complicated or nonhealing wounds, demands the ability of the compound to work in the challenging environment of insufficient wound healing. Additionally, several components have shown significant promise in increasing angiogenesis and wound healing in animal wound healing models. Previously, various wound healing therapeutic approaches that have been prosperous in preclinical investigations eventually deteriorate to present advantages to patients when examined in clinical trials [16, 17]. Hydrogels are broadly administered for repairing and regenerating damaged tissues, principally because of their characteristics, such as permeability, viscoelastic properties, porosity, biocompatibility, and

biodegradability. Wound healing experiments reported that hydrogels showed induced dermal regeneration and anti-inflammatory characteristics. For example, microspheres of gelatin integrating ginsenoside Rg1 (a compound derived from *Panax ginseng*) were loaded in chitosan/collagen scaffolds. Therefore, the dressing was usually administered to regenerate the damaged tissues of the skin because of the effect of the bioactive compounds [18, 19]. The inflammatory reactions are a series of well-coordinated cellular occurrences causing wound repair that can perform both deleterious and beneficial roles in repairing cutaneous layers. NSAIDs are abundantly used to manage inflammation because of their antipyretic, analgesic, anti-inflammatory, and thrombotic properties because of inhibition of cyclooxygenases 1 and 2 [20, 21]. Also, NSAIDs such as ibuprofen are the first-line drugs for treating osteoarthritis, soft tissue injuries, gout, pain, and inflammatory diseases with an estimated administration of more than 30 million per day. Recent findings have revealed that the combination of anti-inflammatory factors such as diclofenac and dexamethasone can decrease levels of prostaglandin E, edema, and histopathological damage when used after the manifestations of first clinical symptoms. Diclofenac presents to affect via inhibition of cyclooxygenases 1 and 2, hence inhibiting prostaglandin synthesis. Likewise, clobetasol inhibits phospholipase A2 and cyclooxygenase 2 activity by suppressing the glucocorticoid receptor and inhibiting cytokines production and altering the population of cells at the inflammation area. Also, the effectiveness of combination therapy improved due to different mechanisms of diclofenac and clobetasol [22]. Ibuprofen would have had the most significant impact on healing in the early alleviation period in the duration of acute inflammation reactions to defect and injury [23]. Connizo et al. have explored that the early use of ibuprofen in the postoperative duration devastated tendon healing [24]. In contrast, delayed use did not affect the improvement of the tendon. The findings of Connizo et al. also supposed that ibuprofen administration has the most significant restorative effect in early postoperative duration (between 2 and 4 weeks) [24, 25]. Korat and Kapupara showed that local infiltration of the surgical wounds with low doses of ibuprofen, levobupivacaine, and epinephrine affects the sutured muscle wound in postoperative pain. The surgical wound was infiltrated with $50 \, \mu\text{L}$ solution comprised of 8 mg/mL epinephrine, 0.3% w/v levobupivacaine, and 2 mg/mL ibuprofen and in contrast to infiltration of water over the sutured muscle wound before closing the skin. The related outcomes to the study revealed that local infiltration of the surgical wound with epinephrine, levobupivacaine, and ibuprofen combination has efficient functions in the surgical wound healing and postoperative pain through reducing pain, improvement of angiogenesis, and strength of tensile in the rat model study [26]. Li et al. studied that the chitosan/collagen gel combination supplemented with a cell-penetrating peptide gel had a fantastic rate of healing and acceleration in treatment via increasing granulation tissue formation, stimulating angiogenesis, and enhancing deposition of collagen in wound tissue. In contrast, negligible cytotoxicity of gel was reported in the histopathological analysis [27].

This study demonstrated that wound healing was significantly enhanced in the collagen/ibuprofen group than collagen and control groups. Wound contraction was 40% in collagen/ibuprofen groups, while this rate was 25% in the collagen and 17% in the control group on day 7. Contrarily, the rate of wound contraction was more significant on day 14. Wound healing in collagen/ibuprofen groups was more remarkable than in the collagen and control groups. The collagen group showed higher wound healing and contraction than the control group. On day 7, wound healing in the collagen/ibuprofen group was highly considerable; migration of fibroblasts to the wound site and observation of hair follicles at the healing site on day 14 presented that the synthetic hydrogel significantly enhanced the wound healing and promoted the migration of other factors involved in this process to the wound site. The rate of healing in the collagen/ibuprofen group on day 14 was higher than that on day 7 and characterized by the formation of collagen layers and migration of cells and angiogenesis. On day 14, the wound margins' closure was more excellent in the collagen/ibuprofen group. The healing in the control group was negligible on day 7 and was initiated beneath the connective tissue layer. The healing rate on day 14 was much higher than that on day 7 in the control group. Based on the outcomes, the collagen/ibuprofen synthetic hydrogel may be concluded to enhance wound healing during 14 days effectively. Microscopically, the collagen/ibuprofen synthetic hydrogel increased wound healing and induced the migration of factors responsible for the healing process, which presented the promising capacity of the synthetic hydrogel comprised of collagen and ibuprofen in acute wound healing.

Data Availability

The data used to support the findings of this study are included within the article and are available from the corresponding author upon request.

Conflicts of Interest

The authors declare that there are no conflicts of interest.

References

[1] E. M. Tottoli and R. Dorati, "Skin wound healing process and new emerging technologies for skin wound care and regeneration," *Pharmaceutics*, vol. 12, no. 8, p. 735, 2020.

[2] C. K. Sen and G. Gordillo, "Human skin wounds: a major and snowballing threat to public health and the economy," *Wound Repair and Regeneration*, vol. 17, no. 6, pp. 763–771, 2009.

[3] T. A. T. Araujo and M. C. I. J. A. F. Y. A. Almeida, "Collagen membranes for skin wound repair: a systematic review," *Journal of Biomaterials Applications*, vol. 36, no. 1, pp. 95–112, 2021.

[4] J. G. Powers and C. K. T. J. Higham, "Wound healing and treating wounds," *Journal of the American Academy of Dermatology*, vol. 74, no. 4, pp. 607–625, 2016.

[5] P. G. Bowler, B. I. Duerden, and D. G. Armstrong, "Wound microbiology and associated approaches to wound management," *Clinical Microbiology Reviews*, vol. 14, no. 2, pp. 244–269, 2001.

[6] A. T. Andrgie and H. Darge, "Ibuprofen-loaded heparin modified thermosensitive hydrogel for inhibiting excessive inflammation and promoting wound healing," *Polymers*, vol. 12, no. 11, p. 2619, 2020.

[7] K. Y. Woo, "Exploring the effects of pain and stress on wound healing," *Advances in Skin & Wound Care*, vol. 25, no. 1, pp. 38–44, 2012.

[8] Z. Yuan and J. Zhao, "Ibuprofen-loaded electrospun fibrous scaffold doped with sodium bicarbonate for responsively inhibiting inflammation and promoting muscle wound healing in vivo," *Biomater. Sci.*vol. 2, no. 4, pp. 502–511, 2014.

[9] R. Bushra and N. Aslam, "An overview of clinical pharmacology of Ibuprofen," *Oman Medical Journal*, vol. 25, no. 3, pp. 155–161, 2010.

[10] S. Kralj and T. P. S. D. Potrc, "Design and fabrication of magnetically responsive nanocarriers for drug delivery," *Current Medicinal Chemistry*, vol. 24, no. 5, pp. 454–469, 2017.

[11] P. I. Morgado and S. Miguel, "Ibuprofen loaded PVA/chitosan membranes: a highly efficient strategy towards an improved skin wound healing," *Carbohydrate Polymers*, vol. 159, pp. 136–145, 2017.

[12] M. Sawant, B. Hinz, and K. Schönborn, "A story of fibers and stress: m," *Wound Repair and Regeneration*, vol. 29, no. 4, 530 pages, 2021.

[13] S. Sharma and V. K. R. K. T. S. Rai, "Collagen-based formulations for wound healing: a literature review," *Life Sciences*, vol. 290, Article ID 120096, 2022.

[14] A. Karami, H. Tebyanian, R. Sayyad Soufdoost, E. Motavallian, A. Barkhordari, and M. R. Nourani, "Extraction and characterization of collagen with cost-effective method from human placenta for biomedical applications," *World Journal of Plastic Surgery*, vol. 8, no. 3, pp. 352–358, 2019.

[15] W. J. Irwin, F. D. Sanderson, and Li, "Percutaneous absorption of ibuprofen: vehicle effects on transport through rat skin," *International Journal of Pharmaceutics*, vol. 66, no. 1-3, pp. 193–200, 1990.

[16] A. P. Veith and K. A. A. D. A. B. Henderson, "Therapeutic strategies for enhancing angiogenesis in wound healing," *Advanced Drug Delivery Reviews*, vol. 146, pp. 97–125, 2019.

[17] E. Hajmohammadi, T. Molaei, S. H. Mowlaei et al., "Sonodynamic therapy and common head and neck cancers: in vitro and in vivo studies," *European Review for Medical and Pharmacological Sciences*, vol. 25, no. 16, pp. 5113–5121, 2021.

[18] A. Gaspar-Pintiliescu, A.-M. Stanciuc, and O. Craciunescu, "Natural composite dressings based on collagen, gelatin and plant bioactive compounds for wound healing: a review," *International Journal of Biological Macromolecules*, vol. 138, pp. 854–865, 2019.

[19] L. K. Hakim and M. Yazdanian, "Biocompatible and biomaterials application in drug delivery system in oral cavity," *Evidence-based Complementary and Alternative Medicine*, vol. 202112 pages, 2021.

[20] S. M. Razavi and D. Khayatan, "Licofelone, a potent COX/5-LOX inhibitor and a novel option for treatment of neurological disorders," *Prostaglandins & Other Lipid Mediators*, vol. 157, Article ID 106587, 2021.

[21] S. K. Shukla and A. Sharma, "Pharmacological control of inflammation in wound healing," *Journal of Tissue Viability*, vol. 28, no. 4, pp. 218–222, 2019.

[22] J. L. Plahovinsak and M. Buccellato, "Selection of non-steroidal anti-inflammatory drug and treatment regimen for sulfur mustard-induced cutaneous lesions," *Cutaneous and Ocular Toxicology*, vol. 35, no. 3, pp. 208–217, 2016.

[23] B. Jørgensen, G. J. Friis, and F. Gottrup, "Pain and quality of life for patients with venous leg ulcers: proof of concept of the efficacy of Biatain-Ibu, a new pain reducing wound dressing," *Wound Repair and Regeneration*, vol. 14, no. 3, pp. 233–239, 2006.

[24] B. K. Connizzo and S. Yannascoli, "The detrimental effects of systemic Ibuprofen delivery on tendon healing are time-dependent," *Clinical Orthopaedics and Related Research*, vol. 472, no. 8, pp. 2433–2439, 2014.

[25] J. D. Packer and A. Varthi, "Ibuprofen impairs capsulolabral healing in a rat model of anterior glenohumeral instability," *Journal of Shoulder and Elbow Surgery*, vol. 25, no. 10, pp. e315–324, 2016.

[26] P. S. Korat and P. P. Kapupara, "Local infiltration of the surgical wound with levobupivacaine, ibuprofen, and epinephrine in postoperative pain: an experimental study," *Biomedicine & Pharmacotherapy*, vol. 96, pp. 104–111, 2017.

[27] M. Li and M. Han, "Oligoarginine mediated collagen/chitosan gel composite for cutaneous wound healing," *International Journal of Biological Macromolecules*, vol. 122, pp. 1120–1127, 2019.