




## BIOPAD & BIOSPRAY

The safe and fast way  
to heal Wounds

**EURORESEARCH**   
natural growing, natural healing



## About us

Euroresearch is an Italian company that was founded in 1983. It operates in various pharmaceutical fields, with particular focus on research and development, marketing, licensing and trading. With over 30 years of experience, Euroresearch has developed high quality products and positioned itself as a leading company in the collagen market.

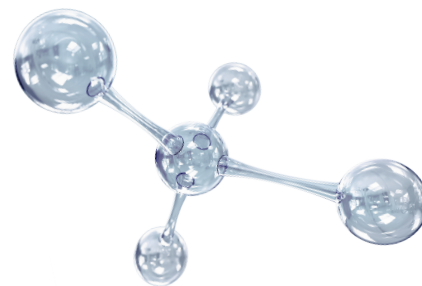
Because of its well-established manufacturing processes, Euroresearch products typically have a high degree of purity and safety, thus ensuring optimum quality. The company's mission began with research and development of highly effective, innovative solutions for the treatment of chronic injuries. Based on this experience, Euroresearch developed the idea of investing in new technological solutions in the following areas: aesthetic medicine, traumatology and dentistry.



## A market leader in high quality, innovative products

Euroresearch's mission started with research and development of innovative solutions for wound management. With its innovative and highly effective wound healing products, Euroresearch adds value to the everyday job healthcare professionals in wound management: the peculiarity of Euroresearch's approach could be summarized in "not to cover but to care". The company's commitment in developing new and cost-effective wound healing products continues day by day, to provide patients the best and fastest solutions to their needs. Innovation is at the core of Euroresearch's daily work, with the aim of striking a balance between the needs of patients and those of medical treatments. The experience achieved in wound management enables Euroresearch to offer new and valid products in a number of different areas.

The dynamism and professionalism characterizing Euroresearch's staff ensures a high-quality service to medical practitioners and - eventually - to patients. Euroresearch's goal in health care is to expand its range of products and services to better meet the needs of both physicians and patients, either in hospitals or at home, while assuring the necessary resources to provide educational programs and assistance to doctors and nurses.





BIO  
SPRAYZS

SHAKE WELL BEFORE USE

100% NATURAL



## What is healing?

Healing is a complex, dynamic biological process aimed at repairing damaged organs or tissues. This physiological process is conventionally divided into four stages, each of them closely interconnected and dependent upon the others.

Hemostasis, inflammation, proliferation and remodeling make up a (seemingly simple) pathway that leads to healing through physical, chemical and biological mechanisms. In most cases the perfection of the human body guarantees a successful outcome; however, failures must not be overlooked. These are often linked to either overall conditions or local imbalances: as far as the latter are concerned, collagen surely fits in and plays a paramount role. Collagen is the protein most commonly found in the extracellular matrix: this component represents 25% to 80% of the proteins that make up the skin, and it is indeed considered the most crucial structure to achieve good tissue reconstruction results.





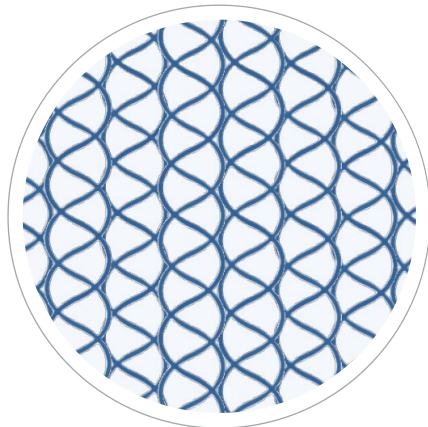
# What is collagen, and what role does it play in the healing process?

From a structural point of view, collagen is made of a triple helix of diversely composed filaments, differentiated by polypeptide amino-acids.

Collagen molecules are predominantly synthesized by fibroblasts, but also by chondroblasts, osteoblasts and epithelial cells. These molecules are then stored in the extracellular matrix (ECM), where the triple helix structure, composed of three alpha-chains, connects with other more complex fibrils of a reticular and elastic nature, thus reinforcing its characteristics.

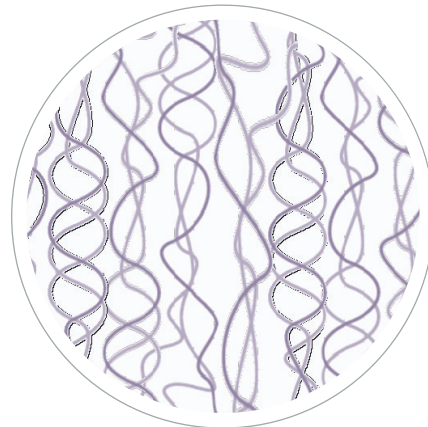
This assembly can be considered a real tissue matrix, capable of taking on different degrees of elasticity and flexibility (*figure 1*).

## Native Collagen



Collagen dressing with native structure

## Hydrolyzed Collagen



Collagen dressing with denatured structure

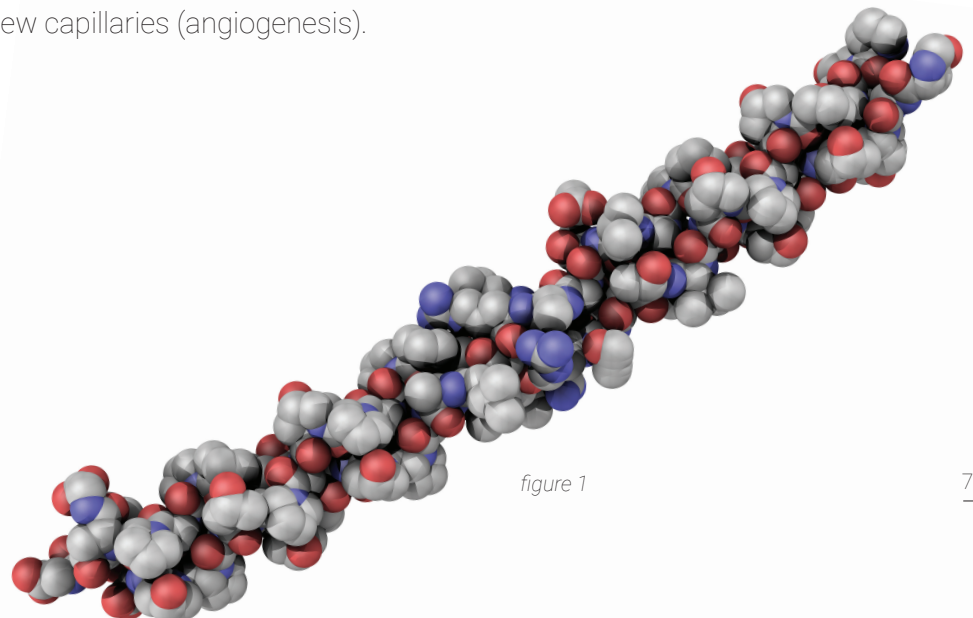
Ushiki T. Collagen fibers, Reticular fibers and Elastic fibers, a comprehensive understanding from a morphological viewpoint.



Collagen is a fibrous protein found in the ECM of human tissues such as skin, tendons, ligaments, cartilage, bones, and teeth; it makes up approximately 25% of all proteins in the human body. Collagen plays an important role in the biological process of cicatrization, in that it is the driver of tissue repair processes.

Its mechanical action provides structural support and contributes to:

- offering a load-bearing structure (scaffold) to recover substance loss
- controlling differentiation, migration and synthesis of several cell proteins
- fostering physiological production of fibroblasts, granulation tissue and native collagen
- facilitating contact between platelets and coagulation factors (hemostasis)
- favoring the formation of new capillaries (angiogenesis).





# What happens in chronic wound healing?

1

Deposition of new collagen is delayed<sup>1</sup>

2

Recruitment of fibroblasts (that synthesize collagen) is delayed<sup>1</sup>

3

High levels of MMPs and elastase are responsible for the proteolytic degradation of collagen<sup>1</sup>

4

Age-related delayed wound healing is caused by impaired collagen synthesis and increased degradation<sup>2</sup>

5

In diabetic patients, hyperglycemia reduces collagen production and induces the formation of abnormal rigid collagen<sup>2</sup>







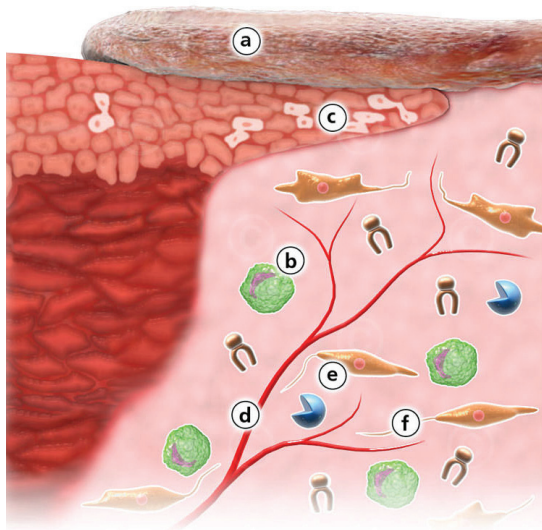
Chronic wounds are characterized by a destroyed or damaged ECM incapable of contributing to wound healing.

The most common types of chronic wounds are:

- pressure sores
- venous leg ulcers
- diabetic foot ulcers
- ulcers of different etiology.

In chronic wounds, growth factor activity is reduced and matrix metalloproteinases (MMPs) are superabundant. The combination of these two features leads to ECM degradation.

### Normal healing process in acute wounds



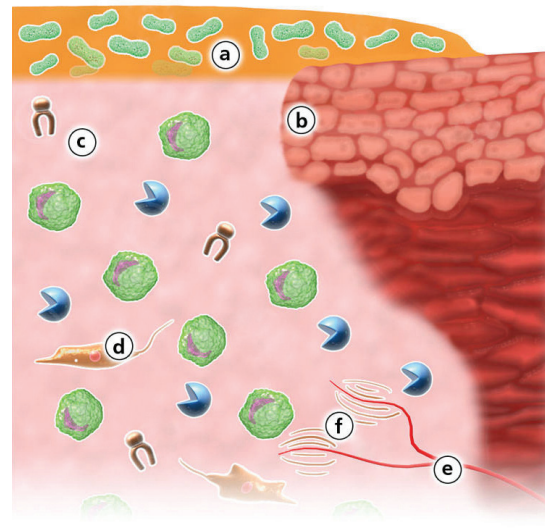
#### Initial phase

- (a) Scab formation
- (b) Immune cell infiltration

#### Healing phase

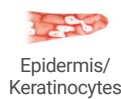
- (c) Re-epithelialisation
- (d) Angiogenesis
- (e) Fibroblast migration
- (f) Collagen deposition

### Impaired healing process in chronic wounds



#### Chronic wound abnormalities

- (a) Colonization and infection
- (b) Hyperproliferative epidermis
- (c) Inflammation, exudate
- (d) Fibroblast senescence
- (e) Impaired angiogenesis
- (f) Fibrin cuffs (barrier to oxygen)





## What are BIOPAD's advantages?

- it speeds up the wound healing processes
- it prevents traumas to the granulation tissue
- it has a hemostatic effect
- It is free from BSE-related risks
- It is not affected by chemical cross-linking processes
- It is well tolerated and easy to use
- It is available in different formulations



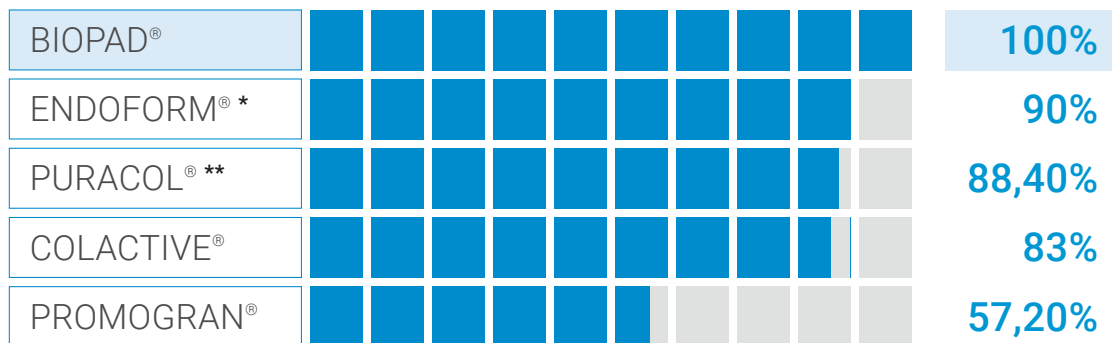


## Our solution

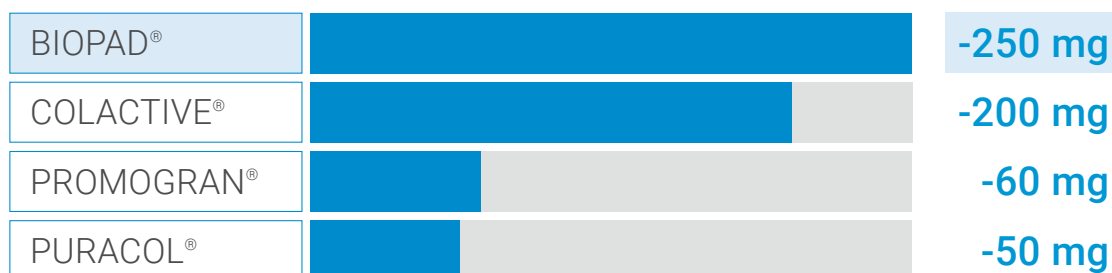
**BIOPAD®** is an ivory-white soft sponge made exclusively of type I collagen of equine origin that keeps its native structure

**BIOPAD®** has the highest collagen content currently on the market (250 mg of collagen)<sup>3</sup>

### Collagen content (%)<sup>3</sup>



### Collagen content (mg)<sup>3</sup>



\* Manufacturer's statement

\*\* Puracol collagen content determined under this method was 88.4%, which differs from the manufacturer's statement of 100% collagen content

Collagen content (%)<sup>3</sup>

Collagen content (mg)<sup>3</sup>

(3) JC. Karr et al (2011): Adv Skin Wound Care 24:208-216



## BIOPAD® indications

- pressure sores
- donor sites and other bleeding surfaces
- dehisced surgical incisions
- draining wounds
- lacerations
- venous stasis ulcers
- diabetic ulcers
- partial and full thickness wounds
- post-laser surgery
- podiatry
- surgical and traumatic wounds

## Preparations

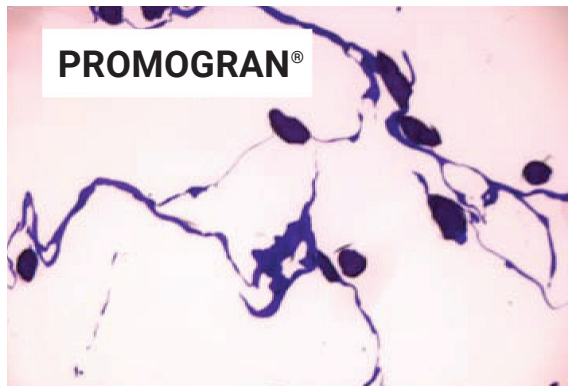
- 1 Prepare the wound bed as per the facility's wound care protocol and debride the wound when necessary.
- 2 Cut BIOPAD®, if necessary, to fit the size of the wound  
Wound with limited exudate: hydrate BIOPAD® with sterile saline solution  
Wound with heavy exudate: do not hydrate BIOPAD®. Rinse out the wound bed with saline solution prior to application.

## Application

- 3 Apply it directly onto the wound bed, covering the entire wound; do not overlap the edges of the wound. The gel that is formed with the wound exudate is biodegradable and does not need to be removed.
- 4 Apply a secondary dressing as per the physician's protocol to cover BIOPAD®. Depending on the amount of exudate, BIOPAD® can be reapplied every day or as per the physician's protocol. If the wound has a small amount of exudate, BIOPAD® can be reapplied every 48 hours or as per the physician's protocol.



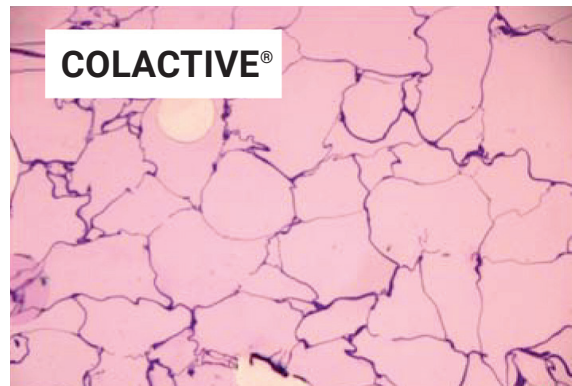
## Competitive analysis. Why is BIOPAD<sup>®</sup> better than others?



**PROMOGRAN<sup>®</sup>**

The high value for the mesh area indicates that there are too few collagen strands ( $8\ \mu\text{M} - 12\ \mu\text{M}$ )

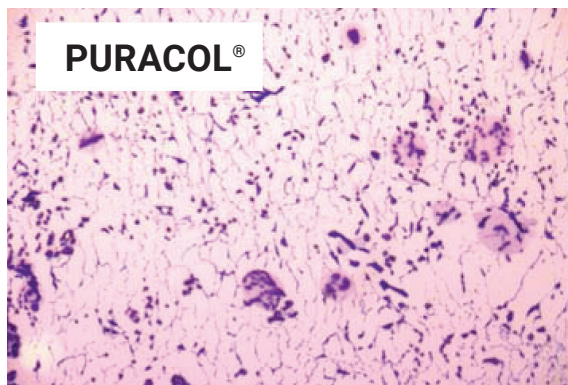
**Fibroblasts do not interact very easily with PROMOGRAN<sup>®</sup>**



**COLACTIVE<sup>®</sup>**

COLACTIVE<sup>®</sup> has a reasonable value for the mesh area, but this value is associated with thin collagen strands ( $< 2\ \mu\text{M}$ )

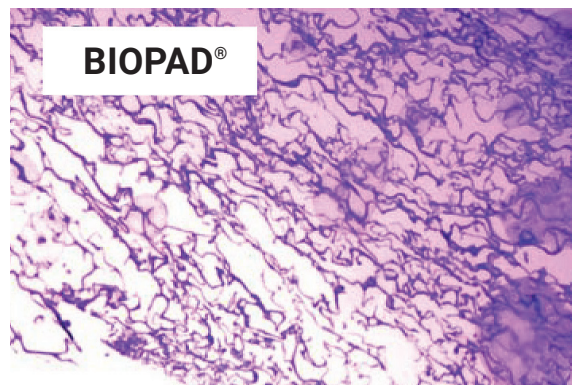
**Fibroblasts do not interact very easily with COLACTIVE<sup>®</sup>**



**PURACOL<sup>®</sup>**

Collagen strands are densely distributed, but this value is associated with thinner collagen strands ( $0.2\ \mu\text{M}$ )

**Fibroblasts do not interact very easily with PURACOL<sup>®</sup>**



**BIOPAD<sup>®</sup>**

BIOPAD<sup>®</sup> has a reasonable value for the mesh area and this value is associated with thicker collagen strands ( $4\ \mu\text{M}$ )

**Fibroblasts have an "active behavior"**



# Ultrastructural investigation on fibroblast interaction with collagen scaffold



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**Abstract:** Collagen-based scaffolds are used as temporary or permanent coverings to help wound healing. Under natural conditions, wound healing is affected by such factors as cell types, growth factors and several components of the extracellular matrix. Due to the complexity of the cell-to-matrix interaction, many cell based mechanisms regulating wound healing *in vivo* are not yet properly understood. However, the whole process can be partially simulated *in vitro* to determine how cells interact with the collagen scaffold in relation to such features as physico-chemical properties, matrix architecture and fiber stability. Under these conditions, cell migration into the collagen matrix can be easily assessed and causally correlated with these features. In this study, we aimed at providing a structural analysis of how NIH3T3 fibroblasts migrate and proliferate *in vitro* when seeded on a native type-I collagen scaffold. To this end, samples were collected at regular time intervals and analyzed by light micro-

scopy (LM), scanning electron microscopy (SEM) and transmission electron microscopy (TEM). Through this experimental approach we demonstrate that collagen is gradually frayed into progressively thinner fibrils as fibroblasts migrate into the matrix, embrace the collagen fibers with long filopodia and form large intracellular vacuoles. A key role in this process is also played by microvesicles shed from the fibroblast plasma membrane and spread over long distances inside the collagen matrix. These observations indicate that a native type-I equine collagen provides favorable conditions for simulating collagen processing *in vitro* and eventually for unraveling the mechanisms controlling cell uptake and intracellular degradation. © 2015 Wiley Periodicals, Inc. J Biomed Mater Res Part A: 00A:000–000, 2015.

**Key Words:** fibroblasts, collagen matrix, electron microscopy, *in vitro* culture, microvesicles

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## INTRODUCTION

Wound healing is a physiological process naturally addressed to repairing skin injury. It consists of a complex cascade of molecular events, from removal of damaged tissues to restoration of normal morphogenesis.<sup>1,2</sup> To this end, the extracellular matrix provides a structural scaffold for cells to migrate through the wound area and differentiate into specific functional types.<sup>3</sup> As a major constituent of the extracellular matrix, the collagen plays a key role in the regenerative process of the wound by acting as an anchoring support for the migrating cells.<sup>4,5</sup> Because of its relative abundance in bodily proteins, low immunogenicity and high stability, the collagen is *par excellence* the best biomaterial employed as a prosthetic medical device to treat dermal injury.<sup>6,7</sup> However, to satisfy these requirements, the collagen scaffold should be endowed with such physico-chemical properties as mechanical integrity, structural uniformity and have a matrix porosity suitable for sustaining cell migration.<sup>8</sup> In addition, the matrix itself should be such to be

retained on the wound bed for time intervals sufficient to complete tissue remodelling.<sup>9</sup>

Prosthetic collagen matrices differ in absorption and stability depending on the type and concentration of chemical substances added to modulate these capacities.<sup>10–12</sup> Therefore, a wide variety of collagen-based dressings differing in structural and biochemical properties have been developed with the intent to simulate the overall function of the extracellular matrix and improve the regenerative process of the injured area in normal or chronic situations.<sup>13,14</sup> Amongst the physiological variables that characterize the collagen matrix, the overall three-dimensional architecture and porosity are the most relevant since they condition such cell behaviours as proliferation, migration and, ultimately, signalling and differential gene expression.<sup>15–17</sup> Matrices with high porosity values provide healthier healing environments, especially in case of chronic wounds, due to their capacity to absorb more efficiently blood and lymphatic fluids from the wound area.<sup>9,18</sup>

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# Scientific literature

## BIOPAD®

- 1 - Baj A, Beltrami GA, Romano M, Lauritano D, Gaudio RM, Palmieri A, Cura F, Gianni AB. - Genetic effects of BIOPAD® on fibroblast primary culture. - *J Biol Regul Homeost Agents*. 2017 Apr-Jun;31(2 Suppl 1):209-214
- 2 - Frangez I, Kasnik T, Cimerman M, Smrke DM - Guided tissue regeneration with heterologous materials in primary subtalar arthrodesis: a case report. *J Med Case Rep*. 2016 May 3;10(1):108
- 3 - Bertone M, Dini V, Romanelli P, Rizzello F, Romanelli M. - Objective analysis of heterologous collagen efficacy in hard-to-heal venous leg ulcers. - *Wounds*. 2008 Sep;20(9):245-9.
- 4 - Torra i Bou JE, Casaroli-Marano RP, Martínez Cuervo F, Reina M, Soldevilla Agreda JJ, Vilaró S. - The use of collagen in the cicatrization of wounds. - *Rev Enferm*. 2000 Oct;23(10):715-22.
- 5 - Abramo AC, Viola JC - Heterologous collagen matrix sponge: histologic and clinical response to its implantation in third-degree burn injuries. - *Br J Plast Surg*. 1992 Feb-Mar;45(2):117-22.
- 6 - Mian M, Aloisi R, Benetti D, Rosini S, Fantozzi R. - Potential role of heterologous collagen in promoting cutaneous wound repair in rats. - *Int J Tissue React*. 1992;14 Suppl:43-52
- 7 - Palmieri B - Heterologous collagen in wound healing: a clinical study. - *Int J Tissue React*. 1992;14 Suppl:21-5.
- 8 - Beghé F, Menicagli C, Neggiani P, Zampieri A, Trallori L, Teta E, Rosini S. - Lyophilized non-denatured type-I collagen (Condress) extracted from bovine Achilles' tendon and suitable for clinical use. - *Int J Tissue React*. 1992;14 Suppl:11-9.
- 9 - Niebauer GW, Oz MC, Goldschmidt M, Lemole G. - Simultaneous use of microfibrillar collagen hemostat and blood saving devices in a canine kidney perfusion model. - *Ann Thorac Surg*. 1989 Oct;48(4):523-7
- 10 - Lazaridis N, Merten HA, Luhr HG. - Experimental study of bone deficits' healing in animals, following their filling with gluing fibrous substance and heterologous collagen. - *Hell Period Stomat Gnathopathoprosopike Cheir*. 1986 Mar;1(1):15-20. Greek, Modern.
- 11 - Jakob H, Campbell CD, Qiu ZK, Pick R, Replogle RL. - Evaluation of fibrin sealing for cardiovascular surgery. - *Circulation*. 1984 Sep;70(3 Pt 2):1138-46
- 12 - Motta G, Ratto GB, De Barbieri A, Corte G, Zardi L, Sacco A, Castagnola M Can heterologous collagen enhance the granulation tissue growth? An experimental study. - *Ital J Surg Sci*. 1983;13(2):101-8.
- 13 - Benfer J, Struck H. - Experimental studies on fracture healing. 3. Accelerated fracture healing through soluble heterologous collagen. - *Arch Surg*. 1973 Jun;106(6):838-42. No abstract available

## BIOSPRAY®

- 1 - Kallis G, Friedman AJ - Collagen powder in wound healing. - *J Drugs Dermatol*. 2018 Apr 1;17(4):403-408 PMID: 29601617
- 2 - Sreelesh LS, Laxminarayan Bhandari P. - An Easy Technique for Negative-Pressure Wound Therapy for Extremities Using Collagen Powder and Sterile Gloves. - *Indian J Surg*. 2017 Feb;79(1):81-83. Epub 2017 Jan 7. PMID: 28331276
- 3 - Zhou K, Krug K, Stachura J, Niewczyk P, Ross M, Tutuska J, Ford G - Silver-Collagen Dressing and High-voltage, Pulsed-current Therapy for the Treatment of Chronic Full-thickness Wounds: A Case Series. - *Ostomy Wound Manage*. 2016 Mar;62(3):36-44
- 4 - Cavanagh MH, Burrell RE, Nadworny PL - Evaluating antimicrobial efficacy of new commercially available silver dressings. *Int Wound J*. 2010 Oct;7(5):394-405. PMID: 20626470
- 5 - Giannini S, Buda R, Vannini F, Cavallo M, Grigolo B. - One-step bone marrow-derived cell transplantation in talar osteochondral lesions. - *Clin Orthop Relat Res*. 2009 Dec;467(12):3307-20. Epub 2009 May 16. PMID:19449082
- 6 - Garrido Villoldo A, Compañ Aguilar MJ. - Treatment of a long-standing lesion with early stimulation of wound healing. *Enferm Clin*. 2007 Nov-Dec;17(6):322-5. PMID: 18039441
- 7 - Akay AF, Sahin H, Nergiz Y, Aflay U, Bircan MK. - Reconstruction techniques in renal injury. - *Saudi Med J*. 2005 Nov;26(11):1750-4. PMID: 16311660
- 8 - [No authors listed] - New collagen powder for wound healing + cost control. - *Krankenpfl J*. 2004;42(7-10):260. German. No abstract available. PMID: 15675418
- 9 - Cuadrillero Martín F, Sandoval Cuadrado P, Rodríguez Hernández F. - Collagen powder: hydrocolloidal dressings. Their effectiveness in radiodermatitis. - *Rev Enferm*. 2004 May;27(5):17-22. PMID: 15239494
- 10 - Ballesté Torralba J, Blanco Blanco J. - Collagen powder dressings in the healing of secondary intention wounds. Report of a case - *Rev Enferm*. 2002 Dec;25(12):50-4. PMID: 14502896
- 11 - Bou Torra JE, Soldevilla Agreda JJ, Martínez Cuervo F, Rueda López J. - Collagen powder dressing in the treatment of pressure ulcer. Multicenter comparative study assessing effectiveness and cost. - *Rev Enferm*. 2002 Sep;25(9):50-7. PMID: 13677749
- 12 - Dunn CJ, Goa KL. - Fibrin sealant: a review of its use in surgery and endoscopy - *Drugs*. 1999 Nov;58(5):863-86. Review. PMID: 10595866
- 13 - Rutherford RB, Wahle J, Tucker M, Rueger D, Charette M - Induction of reparative dentine formation in monkeys by recombinant human osteogenic protein-1 - *Arch Oral Biol*. 1993 Jul;38(7):571-6. PMID: 8368953



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