Use of Collagen and Fibroblasts in Modern Medicine (Review of Literature)

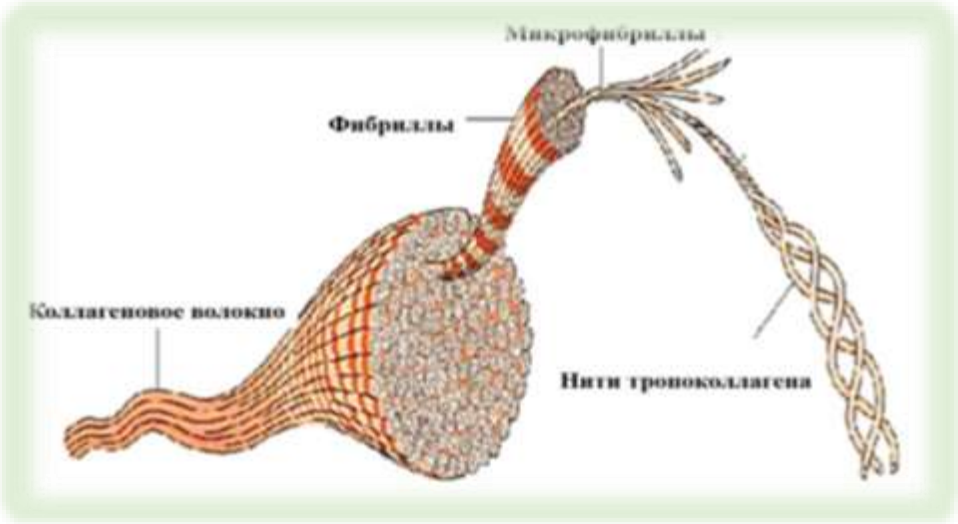
U.Y.Ergashev	Department of General Surgery №2 Tashkent Medical Academy
N.M.Malikov	Department of General Surgery №2 Tashkent Medical Academy
D.R.Yakubov	Department of General Surgery №2 Tashkent Medical Academy
B.A.Abdusalomov	Department of General Surgery №2 Tashkent Medical Academy
B.T.Gafurov	Department of General Surgery №2 Tashkent Medical Academy

ABSTRACT

The work is devoted to a review of the options for the use of collagen and materials based on collagen in medicine and the pharmaceutical industry. The problem of effective impact on the local manifestation of the wound process and at the present stage of development of medicine remains unresolved, despite the emergence of various drugs and techniques. Among the many types of cells that can have a clinical effect, of particular interest are dermal fibroblasts, which are a heterogeneous population of mesenchymal cells and play a key role in the regulation of cell interactions and maintaining skin homeostasis. Currently, there are more than 60 modern cellular or tissue drugs for the treatment of wounds, which makes it difficult to choose an appropriate, safe and effective adjuvant therapy. A review of the world literature on the role of collagen in the wound healing process is presented. The problems of the epidemiology of chronic wounds and ulcers of various origins, the physiology and pathophysiology of the phases of wound healing are considered. The pathogenetic role of different types of collagens, as well as the mechanisms of functioning of collagen, macrophages, fibroblasts, matrix metalloproteinases, and other cytokines during ulcer healing, are discussed.

Keywords:	Collagen, "collost", biomaterial, allogeneic fibroblasts, human fibroblasts as a skin substitute, repair of damaged tissues, chronic wounds.

Introduction. Collagen is a fibrillar protein that forms the basis of the connective tissue of the body and ensures its strength and elasticity. Connective tissue contains from 1 to 9% collagen. Collagen belongs to a class of proteins called scleroproteins. A feature of proteins of this class is their phylogenetic relationship in different species of animals and humans. The term "collagen" refers to specific monomeric protein molecules and aggregates of these molecules, which form fibrillar structures in the extracellular matrix of the connective tissue (Fig. 1.). In the collagen molecule, every third amino acid is a glycine. Collagen is also characterized by amino acids that are not found in other proteins, such as hydroxyproline and oxylysin, the content of which is 23% of the total amino acid composition of the collagen molecule [1].



Picture 1 - The structure of the collagen fiber

In the pharmaceutical and medical industries, collagen has found wide application. On its basis, various dosage forms (soft and liquid), special patches and sponges (hemostatic collagen sponge, collagen sponge with methyluracil, collagen sponge with sanguirythrin, etc.), as well as a variety of means for quickly stopping bleeding (means of local hemostasis) have been developed medical devices for the treatment of wounds, burns, trophic ulcers, bedsores and other soft tissue defects of various origins [2]. A complex three-helix molecule is ordered in such a way that the free glycine side chains of each polypeptide chain are inside the common helix, and the proline, hydroxyproline and side groups of amino acids protrude outwards (Fig. 2). The unique physicochemical, physicomechanical and biological properties of collagen make it possible to widely use it as an auxiliary substance in the production of prolonged dosage forms, as a hemostatic agent and a matrix for guided tissue regeneration in the form of membranes, sponges, coatings, and also as a component of complex combined drug systems, including a collagen-based depot matrix, a drug substance (or a combination of substances) and a release modulator. In addition, collagen itself, due to its “typical” specificity as a macromolecular compound, has a high potential for pharmaceutical development [2,3]. To date, scientists have identified more than 40 genes that collectively code for 28 types of collagen. They are designated by Roman numerals I-XXVIII. Such a pronounced diversity of collagen types is necessary to ensure different physiological roles in various tissues and organs [9].

Figure 2 - Trinity-helical model of the collagen molecule

Biomaterial "Collost" was implanted starting from the 2nd phase of the wound process. Collost is a bioplastic material based on native non-reconstructed bovine collagen with a completely preserved structure. To close planar wounds and ulcerative defects, the biomaterial "Collost" was used in the form of membranes. A membrane pre-soaked in warm (38°C) sterile saline for 15 min was applied to the wound treated with ultrasound. When the wound defect was localized on the plantar surface, the membrane was fixed with 3–4 Tisorb 00 ligatures. After the manipulation was completed, a gel dressing (Hydrosorb, Gelepran) was applied [4]. "Collost" is a sterile bioplastic collagen material with a completely preserved fibrous structure, which provides tissue regeneration. The mechanism of action of Collost is due to the fact that its basis is type I collagen, which acts as an extracellular matrix and provides guided contact between epithelial cells and fibroblasts, creating their optimal migration and orientation, as well as binding cells to form new tissue. The use of ultrasonic cavitation and bioplastic material "Collost" accelerates the processes of wound cleansing from devitalized tissues and microbial contamination of wounds, and also prevents secondary infection. The use of ultrasound and bioplastic material "Collost" significantly improves the cytological picture of wounds, which affects the acceleration of reparative processes and the timing of epithelialization. [5]. Fibroblasts are one of the main secretory cells of the body involved in the formation of the extracellular matrix, repair of skin lesions, and stimulation of the growth of keratinocytes and blood vessels. In accordance with their location in tissues and their functions, fibroblasts are able to produce procollagen, fibronectin, glycosaminoglycans, proelastin, nidogen, laminin, chondroitin-4-sulfate, tenascin [6]. Transplantation of cultured allogeneic fibroblasts improves the clinical indicators of the course of the wound process, significantly reducing the time of wound healing by an average of 7-8 days compared to traditional methods of treatment, which significantly reduces the time of treatment, mortality, and costs for the victims. For the current level of

care for seriously ill patients, the most acceptable is the transplantation of allogeneic fibroblasts cultured in vitro. Dermal fibroblasts are a heterogeneous population of mesenchymal cells and play a key role in the regulation of cell interactions and maintenance of skin homeostasis [7, 10]. The connective tissue frame of the heart, lungs, gastrointestinal tract, muscles and other organs contain fibroblasts that perform specialized functions. Differences in gene expression between fibroblasts of the dermis and skin derivatives have been shown; fibroblasts obtained from different anatomical sites have tissue-specific cytophysiological differences [19].

The following are involved in the regeneration of periodontal tissues: transforming growth factor alpha (affects angiogenesis), growth factor beta (stimulates the synthesis of type 1 collagen, fironectin and osteonectin) and basic fibroblast growth factor (affects the growth of all types of cells in tissues). In experimental studies, a pronounced effect of GFGF (general fibroblast growth factor) on the acceleration of angiogenesis processes was also established (it enhances the proliferation of capillary endotheliocytes, smooth muscle cells and pericytes, which play an important role in the formation of blood vessels). OFGF is also a powerful mitogenic factor for cells of mesenchymal origin; it significantly reduces their doubling time [8]. An experimental model of the wound process was formed in male Wistar rats on the background of alloxan diabetes. Studies of the cellular structures of wounds demonstrate the positive dynamics of the course of reparative processes in experimental animals with alloxan diabetes with local administration of the sorbent, Beta-Beta. This was manifested in the improvement of angiogenesis processes, the development of granulation tissue and epithelialization of wound surfaces. An experimental model of alloxan diabetes has been formed to study the therapeutic reparative effects of Beta-Beta when applied topically to purulent-necrotic wounds of soft tissues in rats [12]. The treatment of chronic wounds is a continuously evolving area. Problems of excessive mechanical forces, infection, inflammation, decreased production of growth factors, and of course lack of collagen will affect treatment outcomes. Numerous studies have shown that collagen preparations are bioactivators and promote their own tissue regeneration, integrating into the surrounding natural tissues. Their main advantages are regulation of the biochemical environment of the wound, stimulation of chemotaxis and angiogenesis. They have the properties of a thin layer of natural skin, but are devoid of the disadvantages inherent in foreign cellular elements that contribute to the rejection of the skin graft [9]. Foot ulcers in diabetic patients are more common and often result in lower limb amputation unless a prompt, rational, multidisciplinary approach to therapy is adopted. The main components of treatment that can ensure successful and rapid healing of diabetic foot ulcers include education, blood sugar control, wound debridement, extended dressing, unloading, surgery, and advanced therapies that are used in clinical practice. These approaches should be used whenever possible to reduce the high morbidity and risk of serious complications resulting from foot ulcers [18]. Tissue damage is accompanied by trauma to microvessels and activation of the coagulation cascade aimed at stopping bleeding and forming a platelet clot. The latter contains fibrin, fibronectin, vitronectin, von Willebrand factor, thrombospondin, which form a matrix for cell migration. Immediately after injury, collagen begins to contact the wound surface with blood, which promotes platelet aggregation and activation of a number of chemical factors [17].

The problem of treatment of long-term non-healing wounds is one of the most urgent in medicine due to the wide variety of possible causes of their occurrence, the difficulties in choosing a treatment. The article presents the results of a study of possible causes of impaired wound healing, among which one of the most significant is the violation of the synthetic function of fibroblasts. In this case, there is a change in the spectrum of expressed cytokines and growth factors, including an increase in the expression of pro-inflammatory cytokines. These factors lead to the impossibility of the formation of a full-fledged extracellular matrix,

and hence the impossibility of fibroblast migration, disruption of cell differentiation and wound healing. Thus, long-term non-healing wounds are characterized by stereotypical changes regardless of their etiology and localization [13]. Currently, a rapidly developing area of medicine is regenerative medicine, where cellular technologies are used using cultured human cells. The proposed innovative method of treatment is the complete closure of the ulcer after implantation of the cell product. As a result, the frequency and duration of medical examinations are significantly reduced, the quality of life and the patient's social activity improve. At least 100 patients who received this treatment between 2000 and 2015 in clinical trials had no follow-up visits. The use of this method is also extremely important for the part of patients in whom the existing open ulcer of a small area does not allow performing a surgical operation for the underlying disease. According to some reports, at least 2,000 patients a year in St. Petersburg need treatment for trophic ulcers [11]. In addition, high activity in long-term non-healing wounds of proteolytic enzymes, primarily serine, and matrix metalloproteinases, combined with a lack of their inhibitors, can cause increased utilization of various cytokines and growth factors, thereby causing their deficiency and, in combination with other factors, leading to the development of a long-term pathological process functioning on the principle of a vicious circle. Based on the conducted morphological and immunohistochemical studies, it can be said that long-term non-healing wounds are characterized by the presence of chronic inflammation, a feature of which is the predominance of monocyte-macrophage cells in the infiltrate in combination with increased expression of pro-inflammatory cytokines and a violation of their normal ratio. In addition, there is an increase in proteolytic activity in the wound area, combined with a decrease in the activity of protease inhibitors. Decreased expression of fibrogenic growth factors. Violation of the formation of normal connective tissue is manifested by a decrease in the content of type III collagen, the accumulation of tenascin and the plasma form of fibronectin in the stroma, the redistribution of laminin, and a decrease in the number of myofibroblasts in the wound. These changes are stereotyped for most long-term non-healing wounds, regardless of their etiology and localization [13,14,15].

In patients with trophic ulcers of venous etiology, skin fibroblasts retain the ability to form collagen under the stimulating effect of biological growth factors. Autologous platelet-rich plasma stimulates the synthesis of collagens by the culture of skin fibroblasts in patients with trophic ulcers of venous etiology, which may be an experimental justification for the use of platelet concentrates in clinical practice [16]. Thus, in recent decades, significant progress has been made in understanding the molecular mechanisms of the main phases of the normal and complicated wound process. Fibroblasts represent a universal biological model for studying in vitro dynamic molecular regulatory processes underlying cell growth and proliferation, metabolism and transduction of intra and extracellular signals. A new step towards an effective solution of the socially significant problem of closing extensive wound defects may be further study of the processes of direct intercellular interaction and the choice of connexin proteins as indicators of the state of the healing process and a target for pathogenetic influence. Given the above data, the treatment of chronic ulcers and other ulcers complicated by diabetes mellitus remains unresolved and requires further study.

Bibliography

1. Бегма А.Н., Бегма И.В. Оценка эффективности применения коллагеновых губок в лечении хронических ран различной этиологии // Главная медицинская сестра. 2014. №3. С. 56-64.
2. Применение биопластического материала Коллост для лечения раневых дефектов у пациентов с осложненными формами синдрома диабетической стопы // Под ред. проф. Б.С. Брискина / МЗСР РФ ГОУ ВПО «Московский государственный

медико-стоматологический университет». – М., 2014.
3. Нестеренко В.Г., Сафоян А.А., Суслов А.П. Коллост – биологическая коллагеновая матрица для репарации поврежденной кожи // Тез. 2 Всерос. конгр. дерматовенерологов. – СПб., 2007.
4. М.Д. Дибиров, Р.У. Гаджимурадов, Ф.Ф. Хамитов, М.В. Полянский, С.А. Терещенко, М.Р. Какубава, К.А. Корейба. Применение современных технологий в лечении гнойно некротических осложнений синдрома диабетической стопы. //Журнал имени академика Б.В. Петровского №2/2016 60-68 ст.
5. А.В.Прошин, Р.А.Сулиманов, И.П.Завалий, А.В.Ребинок. Процессы репаративной регенерации в гнойных ранах при местном сочетанном лечении физическими методами и биологически активными материалами. //Вестник новгородского государственного университета №3(101) 2018 61 ст.
6. В.Л. Зорин, А.И. Зорина, О.С. Петракова, В.Р. Черкасов. Дермальные фибробласты для лечения дефектов кожи. //Клеточная трансплантология и тканевая инженерия. Том IV, № 4, 2014.
7. А.Н. Маркова, А.В. Лизунов, Т.Э. Хамидулин. Использование фибробластов в комбустиологии. Вестник совета молодых учёных и специалистов Челябинской области №3 (14) Т. 3 2016 87-88 ст.
8. Кяримова Р.Р., Лахонина К.А., Фаркашди Ш. Применение фибробластов в стоматологии. Электронный научно-образовательный вестник "Здоровье и образование в XXI веке" № 12, 2008 г. (Т. 10).
9. Силина Е.В., Ступин В.А., Габитов Р.Б. Роль коллагена в механизмах заживления хронических ран при синдроме диабетической стопы. Клиническая медицина. 2018; 96(2).
10. А.В. Мелешина., А.С. Быстрова., О.С. Роговая., А.В. Васильев., Е.В. Загайнова. Тканеинженерные конструкты кожи и использование стволовых клеток для создания кожных эквивалентов. //СТМ ∫ 2017 — том 9, №1 198-219.
11. Блинова М.И., Юдинцева Н.М., Александрова О.И., Баллюзек М.Ф., Хабарова И.Г., Маркин С.М., Чагунава О.Л. Клинический опыт заживления трофических язв с использованием клеточного продукта «эквивалент дермальный эд». //Хирургические болезни 690-695 ст.
12. И.П.Завалий. Экспериментально-клиническое исследование состояния и лечение гнойных ран при сахарном диабете. //Вестник новгородского государственного университета №1(92) 32-36 ст.
13. Е.М. Фоминых. Межклеточные и клеточно-матриксные взаимодействия при репарации длительно незаживающих ран. //Вестник медицинского института непрерывного образования / Выпуск № 1, 2022 66-70 ст.
14. Суханов Ю.В., Воротеляк Е.А., Лядова И.В., Васильев А.В. Терапия мезенхимальными стволовыми клетками – сосуд наполовину полон или наполовину пуст? Онтогенез. 2020; 51 (4): 316–320 ст.
15. Васильев А.В., Батин М. «Дорожная карта» регенеративной медицины. Клеточная трансплантология и тканевая инженерия. 2010; 5 (2): 89–90 ст.
16. Богдан В.Г., Толстов Д.А., Зафранская М.М. Оценка стимулирующего влияния обогащенной тромбоцитами плазмы в экспериментальной модели культур фибробластов пациентов с трофическими язвами венозной этиологии.

//Медицинские новости №9• 2014 87-89 ст.
17. Ю.С. Винник, А.Б. Салмина, А.И. Дробушевская, О.В. Теплякова, Е.А. Пожиленкова, А.Р. Котиков. Особенности патогенеза длительно незаживающих ран. //Лекции, обзоры 2011.
18. Эргашев У.Ю., Моминов А.Т., Маликов Н.М., Якубов Д.Р. Нарушение целостьности стопы у пациентов с синдромом диабетической стопы (обзор литературы). // Volume–7 17_Issue-1_November_2022 7-17 ст.
19. Paquet-Fifield S, Schlüter H, Li A, et al. A role for pericytes as microenvironmental regulators of human skin tissue regeneration. Journal of Clinical Investigation. March 2009. doi: 10.1172/jci38535.
20. Karimov X. Y., Ergashev U. Y., Yakubov D. R. Complex treatment in severe forms of acute paraproctitis //Web of Scientist: International Scientific Research Journal. – 2022. – Т. 3. – №. 9. – С. 199-203.
21. Моминов А. Т. и др. Проблемы обезболивания в амбулаторной хирургии //European Journal of Interdisciplinary Research and Development. – 2022. – Т. 10. – С. 81-89.5.
22. Эргашев У. Ю., Якубов Д. Р., Моминов А. Т. ЎТКИР ПЕЛВИОРЕКТАЛ ПАРАПРОКТИТ БЎЛГАН БЕМОРНИ КОМПЛЕКС ДАВОЛАШ (КЛИНИК ХОЛАТ) //Development of pedagogical technologies in modern sciences. – 2022. – Т. 1. – №. 2. – С. 63-64.
23. Эргашев У. Ю., Маликов Н. М., Якубов Д. Р. КЛИНИЧЕСКИЙ ОПЫТ ХИРУРГИЧЕСКОГО ЛЕЧЕНИЯ ВЕНТРАЛЬНЫХ ГРЫЖ У БОЛЬНЫХ С ИЗБЫТОЧНОЙ МАССОЙ ТЕЛА И ОЖИРЕНИЕМ //Proceedings of Scientific Conference on Multidisciplinary Studies. – 2022. – Т. 1. – №. 3. – С. 1-2.