



## ANALYSIS OF THE RESULTS OF APPLYING THE METHOD PLATELET-RICH PLASMA (PRP) FOR THE TREATMENT OF PROBLEMATIC SKIN WOUNDS

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### ABSTRACT

**OBJECTIVE:** To show analysis of the results of applying the method platelet-rich plasma (PRP) for the treatment of problematic skin wounds. The paper's objective is to prove its reliability and relevance, to evaluate its efficiency and applicability to Bulgarian patients.

**MATERIAL AND METHODS:** Out of a total of 154 hospitalized patients with problematic skin wounds 83 have been treated with platelet-rich plasma, comprising the Experimental group (EG), and 71 patients with similar wounds have been treated by using traditional methods for the respective pathology at the Department of Orthopedics and Traumatology, Vessel and Plastic Surgery Department, comprising the Control Group (CG). Data on the condition of monitored symptoms of researched cases was collected for the research. A variable is defined for every symptom and its value and weight is correspondingly introduced. Scores introduced by Cancela AM are used for the assessment of the respective wound. Each of these scores is used for assessing specific wound parameters: Total Wound Score (TWS); total anatomic score (TAS) of wound and Total Score of Wound data (TSWD).

**RESULTS:** Analysis of variance was performed to achieve the objectives and solve the research tasks; variables containing information on initial and final data from each series of experiments have been analyzed in order to determine basic numerical characteristics of variables, and by comparative analysis to check how they tend to vary in experimental (EG) and control (CG) group. Statistical survey shows that the platelet-rich plasma method gives significant results when treating problematic skin wounds leading to full recovery as compared to traditional methods typical for the pathology. The graphical interpretation allows to identify forecast relations between assessed aspects of problematic wounds and weeks of treatment.

**CONCLUSION:** By analyzing the results of our study we can conclude that the use of platelet-rich plasma to treat problematic skin wounds is a safe and effective treatment method. It is not universal for every wound, but follows the principles of biological wound treatment and leads to full recovery of high percentage of problematic

skin wounds. We support proponents of the application of platelet-rich plasma to treat problematic skin wounds.

**Keywords:** problematic skin wounds, platelet rich plasma, functional scoring scales

Skin ulcerations are a clinical problem with an incidence of 0,78%. Annual costs for treating such wounds are immense. For instance, 2% of healthcare budget in the European Union, 40 million pounds in the UK, and \$ 1.3 billion in the United States, are spent annually for treatment of patients with pressure ulcers [1]. Problematic skin wounds are a serious treatment challenge in Bulgaria as well. Approximately 100 000 patients annually have various problematic wounds in Bulgaria [2].

A number of concomitant conditions might be a prerequisite for getting such wounds. Chronic venous insufficiency is most often the reason for chronic wounds of lower extremities as it is believed that it comprises 60-80% of cases [1]. Diabetes Mellitus is a worldwide public health problem. Its high incidence places this condition among the major causes of chronic wounds, especially with respect to lower extremities [3,4]. Pressure wounds, also known as decubitus ulcers are localized injuries of the skin and /or underlying tissue, usually occurring over bony prominences resulting from pressure in combination with shearing and/or friction. Most often it damages the skin covering the sacrum, coccyx, heel or thighs but other areas might be affected as well: elbows, knees, ankles or the back of the skull [5].

Chronic and hard-to-heal skin wounds are highly prevalent and hard to treat due to loss of growth factors, necessary for the healing process and are often complicated by superinfections [6]. The term *regenerative medicine* was introduced in the late 1990s. A number of research papers on stem cells, growth factors and extracellular matrix give the opportunity for development of a new treatment philosophy, different from the concept for the classic tissue engineering with a striving to regenerate fully functioning tissue [7, 8].

The platelet-rich plasma method gives an opportu-

nity for a modern, biological treatment of problematic skin wounds. This method for treatment of problematic skin wounds has not been researched in Bulgaria until now. This paper's objective is to prove its reliability and significance, to evaluate its efficiency and applicability to Bulgarian patients.

**MATERIAL AND METHODS:**

The research was carried out at the Department of Orthopedics and Traumatology, MHAT Ruse, AD for a period of 84 months: from February 2009 to September 2016. Out of a total of 154 hospitalized patients with problematic skin wounds 83 have been treated with platelet-rich plasma, comprising the Experimental group (EG), and 71 patients with similar wounds have been treated by using traditional methods for the respective pathology at the Department of Orthopedics and Traumatology, Vessel and Plastic Surgery Department, comprising the Control Group (CG).

This is the first longitudinal research on treating patients with problematic skin wounds by applying the platelet-rich plasma method in Bulgaria.

Data on the condition of monitored symptoms of researched cases was collected for the research. A variable is defined for every symptom and its value and weight is correspondingly introduced. Scores introduced by Cancela AM are used for the assessment of the respective wound [1]. Each of these scores is used for assessing specific wound parameters: Total Wound Score (TWS); total anatomic score (TAS) of wound and Total Score of Wound data (TSWD).

All patients, included in the research have had problematic skin wounds for various periods, accompanied by a psychological trauma, pain and difficulty in normal everyday tasks.

The formation of samples meets all requirements for sample formation in medical research for statistical process-

ing: reviewed sample is representative; sample's size is enough; Researched groups (CG and EG) are selected randomly and the requirement for homogeneity at the beginning of the trial is met, i.e. both groups have relatively equal opportunity, and the requirement for CG and EG to be similar in size with roughly the same number of patients in them is met; coefficients of variation are calculated to check the homogeneity of the groups, in order to determine the degree of uniformity of the CG and EG. An analysis of variance of trial results based on researched symptoms and their respective coefficients of variation have been calculated.

Statistical analyzes have been performed by using the software SPSS (Statistical Package for the Social Sciences). During the research the following three nonparametric tests have been used: Kolmogorov-Smirnov, Mann-Whitney and Wilcoxon to verify the significance of relations between features.

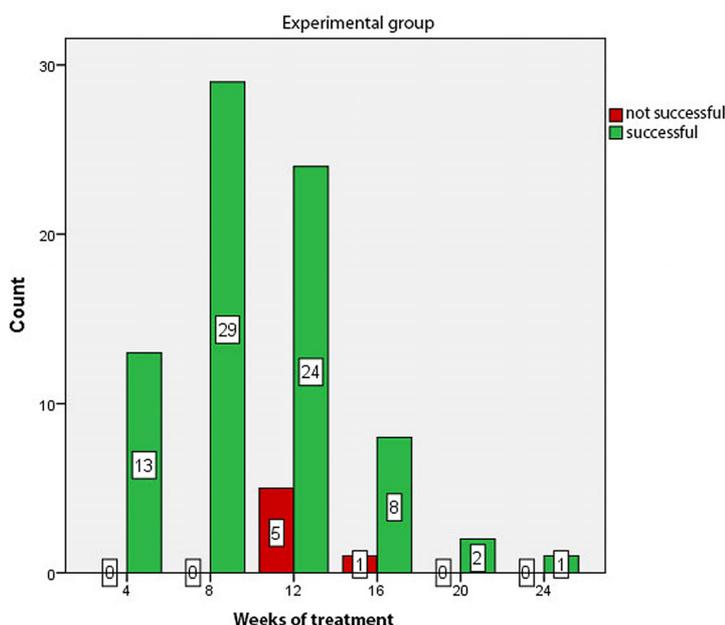
**RESULTS:**

*Analysis of variance*

Analysis of variance was performed to achieve the objectives and solve the research tasks; variables containing information on initial and final data from each series of experiments have been analyzed in order to determine basic numerical characteristics of variables, and by comparative analysis to check how they tend to vary in experimental (EG) and control (CG) group.

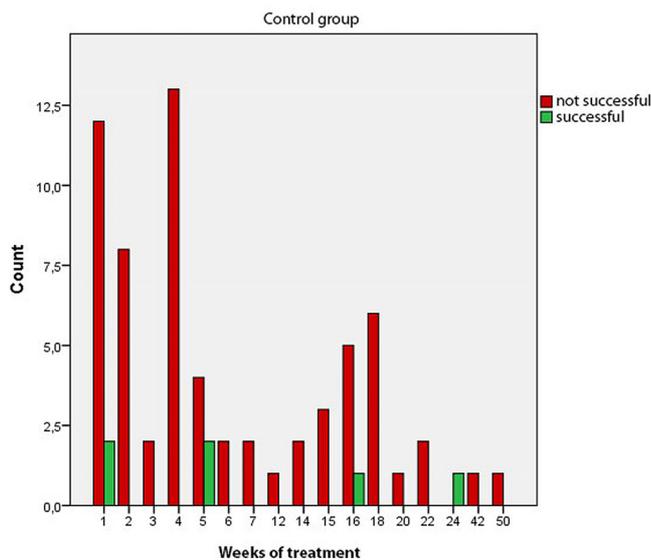
The analysis of variance shown on fig. 1 compares treatment outcome by week for the experimental group and shows that most patients are completely cured within 8 and 12 weeks, respectively 34.94% and 28.91%, which in total is 63.85% successfully treated patients after application of the platelet-rich plasma method. Accordingly, the platelet-rich plasma provides the best results for a treatment period with duration from 8 to 12 weeks.

**Fig. 1.** Analysis of variance by „Treatment outcome“ compared to „Weeks of treatment“ for EG



Only 7.22% of platelet-rich plasma treated patients have a bad outcome due to comorbidities in an advanced stage. The analysis of variance shown on fig. 2 compares treatment outcome by week for the control group and shows that only 8.45% of patients have been cured completely by using the traditional method for the pathology and for the remaining 91.55% no positive outcome was achieved. Therefore, the traditional method fails to bring good results when treating problematic skin wounds.

**Fig. 2.** Analysis of variance Treatment outcome“ compared to „Weeks of treatment“ for CG



Breakdown of patients by week of treatment shows that the largest number of patients (69,88% of all patients in EG) have been treated from 8 to 12 week and 91,38% of them have positive treatment outcome (complete wound healing). This result allows drawing the conclusion that optimum results are achieved between the 8 and 12 week of treatment of problematic skin wounds.

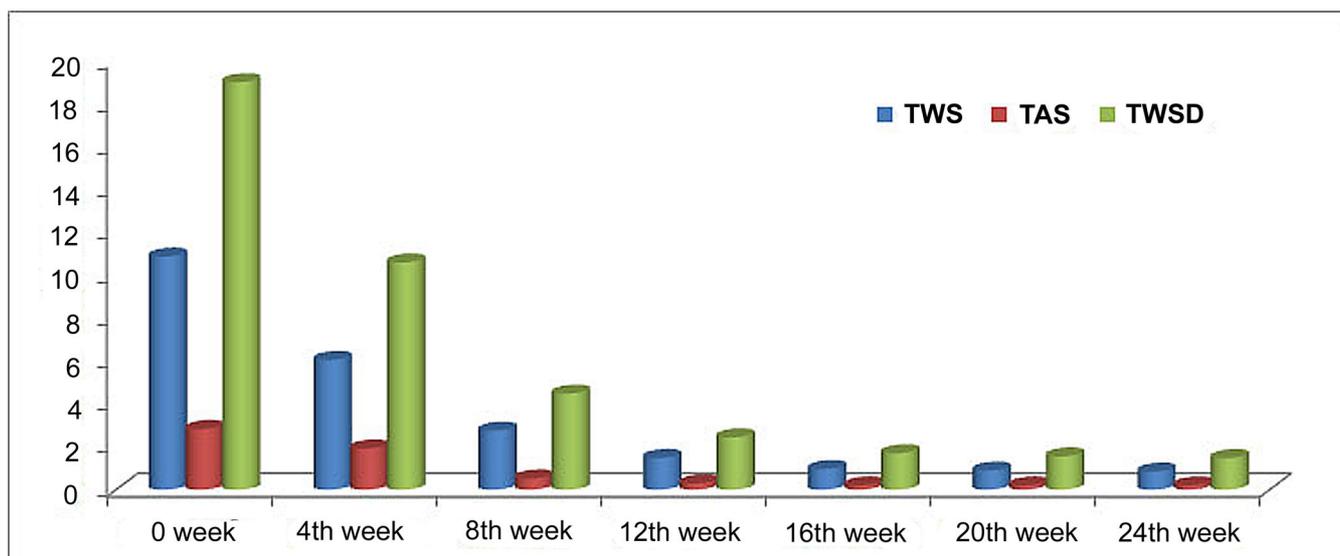
Statistical survey shows that the platelet-rich plasma method gives significant results when treating problematic skin wounds leading to full recovery as compared to traditional methods typical for the pathology.

**Comparison of outcome based on criteria scores**

TWS criterion stands for total wound score and involves the following parameters: edema around the wound, erythema around the wound, puss discharge, fibrin, granulation, edema at the bottom of the wound and ocher edema. At baseline (week 0) 84% of all patients have a score between 10 and 12 points. During week 4, only 17% of them have the same high scores. The majority of patients after week 4 have scores from 0 to 8, and the number of patients with 0 score increases by increasing the weeks of treatment, which indicates the successful conclusion of the treatment.

TAS criterion stands for total anatomic score where wounds are assessed based on open, tibia bone, open tendons, a.dorsalis pedis pulse and a.tibialis posterior pulse. At baseline (week 0) 30% of all patients have a score between 7 and 10 points tabl.4.2. During Week 8, only 7% of them have the same high score but the score is lower than 10. By Week 8 successful treatment outcome is achieved by applying the platelet- rich plasma method.

**Fig. 3.** Visualization of summarized data of platelet –rich plasma treatment method based on the three criteria: TWS, TAS, TSWD

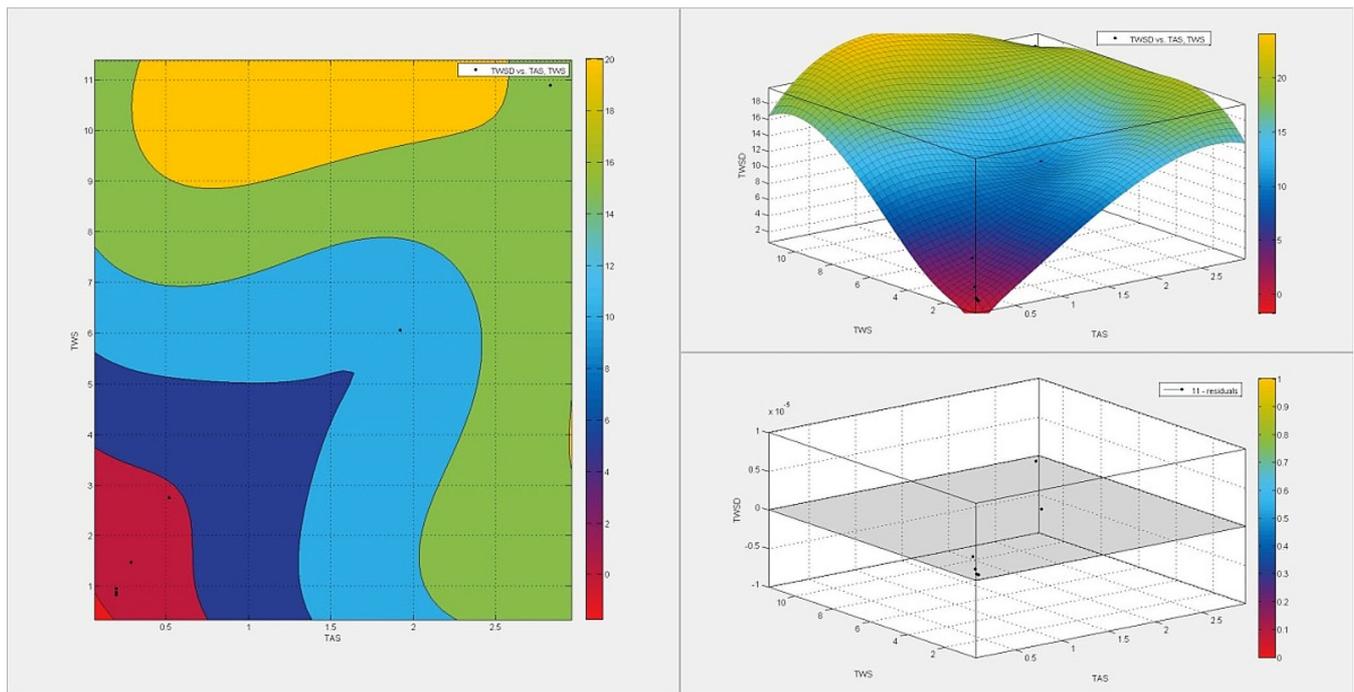


TSWD criterion stands for Total Score of Wound data where wounds are assessed based on: size in sq. mm, depth in mm, erosion in mm and period of existence of the wound. The results represent the treatment dynamics and scores vary widely– from 0 to 20. At baseline (week 0) 36,6% of all

patients have scores of 18 points. It is evident that in the 4th week only 6.7% of patients have high scores, but those comprise 1.2% of all patients. After Week 8, the treatment outcome is successful for 92.7% of all patients treated by the platelet-rich plasma method.

Data derived from the summarized results obtained from treatment of problematic skin wounds by applying the platelet-rich plasma method, assessed by the three criteria TWS, TAS, TSWD, are presented in Fig. 3. The graphic interpretation gives varying rate of convergence of the results of the three criteria, which shows the significance of the assessment of treatment outcomes after applying the method. Fig. 4 represents the average variation of the TWS, TAS, TSWD according to the summarized outcome. The graphical interpretation allows to identify forecast relations between assessed aspects of problematic wounds and weeks of treatment.

**Fig.4.** Graphical interpretation of average variation of TWS, TAS, TSWD criteria



**DISCUSSION:**

This paper examines the reliability and relevance of the platelet-rich plasma method, evaluates its effectiveness and applicability when treating problematic skin wounds on Bulgarian patients. In this paper we review acute, chronic and hard-to heal -skin wounds by their etiology, physiology, pathophysiology and treatment method and refer to them as the so called problematic skin wounds, which is new for Bulgaria.

Hard-to-heal wounds and chronic wounds are problematic by themselves. Acute skin wounds heal primarily in the normal way of wound healing, but when they are accompanied by intrinsic and extrinsic factors (age, comorbidities, taking various medications, presence of pathogenic infectious agents, wound location, etc.) they may become chronic [9, 10]. About 15% of acute wounds become chronic and fall into the group of problematic skin wounds [11]. The main reason for the difficult and delayed healing is the lack of growth factors in the wound bed due to various factors negatively affecting normal wound healing.

Activated platelets in the platelet-rich plasma supply the necessary growth factors to the wound bed and the

peripheral wound edges. Growth factors stimulate normal wound healing and lead to recovery of problematic skin wounds. Presently in Bulgaria there are no data on treating problematic skin wounds by applying platelet-rich plasma, which encourages us to study and research method in this paper. The results of our research indicate that 92.78% of patients enjoy full recovery, while 7.22% of patients have no positive wound healing outcome (unhealed wound).

By analyzing results one can discuss the contribution of our work to the researched field. We use platelet-rich plasma prepared by one time centrifugation of patient's own blood. The plasma is removed without leukocytes. We support authors who use plasma obtained without leukocytes. We confirm published evidence that platelet-rich plasma inhibits the development of certain infectious agents without the need for antibiotic treatment and full wound healing is achieved by the use of our method.

Our research and analysis is the first in Bulgaria to consider in detail the possibility to digitize the assessment criteria for condition of problematic skin wounds upon first encounter with the wound, as well as the weekly follow-

up until completion of skin tissue regeneration and a forecast is provided.

In terms of the interaction between treatment outcome and comorbidities, the most common concomitant disease is diabetes (as the only concomitant disease) and the use of platelet-rich plasma has led to 100% positive outcome and complete wound healing. This method is successful for patients with a single concomitant disease. The following meta-analysis of studies gives similar studies, but most of them have lower rates of successful complete wound healing.

In terms of the interaction between treatment outcome and etiologic groups our study proved that, the treatment outcome for all patients with problematic skin wounds where the platelet-rich plasma method is applied is successful for 93.9% of them with traumatic wounds, 88.8% with pressure sores and 100% of patients with wounds of inflammatory origin. The extremely high rate of successful treatment of pressure sores focuses our attention on comparison of this particular kind of wounds in order to demonstrate the effectiveness of the method for them.

In reporting our results on the interaction between treatment outcome and weeks of treatment depending on the agent it is found that for patients with *S.aureus* treatment should be extended to 24 weeks due to the difficulty in its response to medication. When the other agent *E.coli* is present, treatment should be extended to 16 weeks, in case of *Kl.pneumoniae* -treatment should reach 20 weeks. For our patients, where *S.aureus* and *E.coli* have been isolated we used only platelet-rich plasma to the full recovery of the problematic skin wounds. Our data supports the above-mentioned conclusions also drawn by other authors [12]. Patients with *Kl. pneumonia*, *Enterococcus faecalis* and *Ps. aeruginosa*, in addition to platelet-rich plasma, we used the appropriate antibiotics in accordance with the microbiological test. Successfully treated patients with *S.aureus* comprise 37.21% of our patients, distributed in all 4 week periods. According to the results obtained, the type of agent causing the wound affects the duration of treatment. Our research has shown that the platelet-rich plasma method gives significant results for treating problematic skin wounds in spite of the presence of infectious agents. Furthermore, in terms of the interaction between cause and comorbidities we found a strong correlation, and we believe the agent has a strong impact on treatment of problematic skin wounds by applying platelet-rich plasma. The most common concomitant disease is diabetes (as the

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only concomitant disease), which is combined with all the agents and prolongs the treatment of problematic skin wounds.

In terms of the interaction between anatomic location and weeks of treatment it is evident that anatomic location of problematic skin wounds impacts treatment duration. Our research has shown that the vast percentage of wounds are located in the area of lower limbs.

Statistical survey shows that the platelet-rich plasma method gives significant results when treating problematic skin wounds in spite of aggravating factors such as causing agent, comorbidities and localization, and for 92,77% of the experimental group patients a positive outcome of full recovery has been achieved.

A review of the results of our study makes it evident that 7.22% (6/83) of are patients without a positive outcome in the process of wound healing, i.e. patients with poor results, no recovery and deterioration of wound parameters. These patients also have a number of additional factors that aggravate the process of wound healing. These are advanced age, comorbidities, wound localization and etiologies. Their percentage is significantly low thus the platelet -rich plasma is considered successful for treatment of problematic skin wounds.

#### **CONCLUSION**

By analyzing the results of our study we can conclude that the use of platelet-rich plasma to treat problematic skin wounds is a safe and effective treatment method. It is not universal for every wound, but follows the principles of biological wound treatment and leads to full recovery of high percentage of problematic skin wounds. We support proponents of the application of platelet-rich plasma to treat problematic skin wounds.

An important quality of platelet-rich plasma is the opportunity to use it for prophylactic treatment of acute skin wounds that could become problematic. Furthermore, the platelet rich plasma method is cost-effective and could be of used in outpatient practices.

Using the applied TWS, TAS and TSWD criteria for wound assessment allows accurate, prompt and easy evaluation of the entire wound healing process, at the treatment onset as well as throughout the whole period of tissue regeneration of the problematic skin wound and last but not least it allows estimation of specific wound treatment duration. The same results are obtained statistically and by assessing wounds based on the used criteria, which proves the adequacy of the methodology.

## REFERENCES:

1. Cancela AM, Lana JF, Annichino-Bizzachi JM, Belangero WD, Malheiros Luzo AC. Use of Platelet-Rich Plasma (PRP) in Treating Chronic Wounds. In: Lana JF, Shimojo AA, Belangero WD, Malheiros Luzo AC. (eds.), Platelet-Rich Plasma. Lecture Notes in Bioengineering. Springer-Verlag Berlin, Heidelberg. 2014; p. 281-288. [CrossRef].
2. Hristov V. Over 100 000 Bulgarians suffer from wounds that do not heal! 2014 Jan. [Internet]
3. Anitua E, Aguirre JJ, Algorta J, Ayerdi E, Cabezas AI, Orive G, et al. Effectiveness of autologous preparation rich in growth factors for the treatment of chronic cutaneous ulcers. *J Biomed Mater Res B Appl Biomater*. 2008 Feb;84(2):415-21. [PubMed] [CrossRef]
4. Papanas N, Maltezos E. Growth factors in the treatment of diabetic foot ulcers: new technologies, any promises? *Int J Low Extrem Wounds*. 2007 Mar;6(1):37-53. [PubMed] [CrossRef]
5. Agrawal K, Chauhan N. Pressure ulcers: Back to the basics. *Indian J Plast Surg*. 2012 May;45(2):244-54. [PubMed] [CrossRef].
6. Baharestani M. The clinical relevance of debridement. In: Baharestani M, Gottrup F, Holstein P, Vanscheidt W, editors. The Clinical Relevance of Debridement. Springer-Verlag Berlin. 1999; p.1-13.
7. Cugat R. Front Matter: Foreword I. In: Lana JF, Shimojo AA, Belangero WD, Malheiros Luzo AC. (eds.), Platelet-Rich Plasma. Regenerative Medicine: Sports Medicine, Orthopedic, and Recovery of Musculoskeletal Injuries. Springer-Verlag Berlin, Heidelberg. 2014; p v-vi. [CrossRef]
8. Lana JF, Weglein A, Vicente E, Perez AG, Rodrigues A, Luzo AC, et al. Platelet Rich Plasma and Its Growth Factors: The State of the Art. In: Lana JF, et al. (eds.), Platelet-Rich Plasma. Lecture Notes in Bioengineering. Springer-Verlag Berlin, Heidelberg. 2014; p1-59. [CrossRef].
9. Eppley BL, Pietrzak WS, Blanton M. Platelet-rich plasma: a review of biology and applications in plastic surgery. *Plast Reconstr Surg*. 2006 Nov;118(6):147e-159e. [PubMed] [CrossRef]
10. Snyder RJ. Treatment of nonhealing ulcers with allografts. *Clin Dermatol*. 2005 Jul-Aug;23(4):388-95. [PubMed] [CrossRef]
11. Lindholm C, Searle R. Wound management for the 21st century: combining effectiveness and efficiency. *Int Wound J*. 2016 Jul;13 Suppl 2:5-15. [PubMed] [CrossRef]
12. Anitua E, Alonso R, Girbau C, Aguirre JJ, Muruzabal F, Orive G. Antibacterial effect of plasma rich in growth factors (PRGF®-Endoret®) against Staphylococcus aureus and Staphylococcus epidermidis strains. *Clin Exp Dermatol*. 2012 Aug;37(6):652-7. [PubMed] [CrossRef]

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