MATERIALE HEMOSTATICE INOVATIVE - EXPERIENȚĂ CLINICĂ UTILIZÂND ADEZIVII BAZAȚI PE FIBRINĂ ÎN ARTROPLASTIILE DE ȘOLD
NOVEL HAEMOSTATIC MATERIALS - CLINICAL EXPERIENCE WITH FIBRIN BASED SEALANT IN HIP ARTHROPLASTIES

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Fibrin-based sealants are mainly based on fibrinogen and thrombin; in the presence of even small amounts of Factor XIII and calcium, fibrinogen is polymerised by thrombin into insoluble fibrin, which organises as a network with multiple functions: it has a major role into the coagulation cascade and it also enhance wound healing, thus playing a key role in human body reaction to any type of aggression, including the surgical procedures. Due to their properties, fibrin-derived materials have been intensively studied and enhanced, so that they nowadays represent a group of great interest for many medical fields. One of the most frequently performed orthopaedic procedures, hip arthroplasty is, in the same time, considerably invasive upon homeostasis, especially due to bleeding, which can lead to dramatic outcome of the patients, as well as upon the soft tissues, with different healing properties. Intensive studies have been thus performed in order to find materials able to diminish these effects, fibrin-based sealants proving themselves as able to fulfil these requirements. This paper reflects the clinical experience of using a Fibrin-based sealant in major orthopaedic procedures, revealing the advantages of using this type of materials for the outcome of the patients.

Adezivii pe bază de fibrină conțin mai ales fibrinogen și trombină; în prezența unor cantități chiar mici de Factor XIII și calciu, fibrinogenul este polimerizat de către trombină în fibrină insolubilă, care formează o rețea insolubilă cu mai multe funcții: are un rol major în cascada coagulării, și de asemenea stimulează procesele de vindecare a plăgilor, jucând astfel un rol cheie în reactivitatea post-lezională, inclusiv cea după intervenții chirurgicale. Datorită proprietăților lor, materialele bazate pe fibrină au fost continuu studiate și dezvoltate, astfel încât astăzi constituie un grup de interes major pentru multe domenii medicale. Una dintre cele mai frecvente intervenții ortopedice, protezarea șoldului, este, în același timp invazivă, afectând semnificativ homeostazia organismului, în special datorită sângerării care poate complica dramatic evoluția pacienților, dar în același timp sunt afectate și țesuturile moi, cu capacitate diferită de vindecare. Din aceste motive au fost efectuate studii amănunțite pentru a crea materiale capabile să antagonizeze aceste efecte negative, adezivii bazați pe fibrină dovedindu-se capabili să îndeplinească aceste cerințe. Această lucrare reflectă experiența clinică în utilizarea unui adeziv bazat pe fibrină (FBS) într-o procedură chirurgicală ortopedică majoră, prezentând beneficiile pe care aceste materiale, corect folosite, le pot determina în evoluția pacienților .

Keywords: *fibrin sealants, hip arthroplasty, haemostatic, fibrin glue*

1. Introduction

An impressive number of materials have been created for medical use, thus appearing the category of biomaterials, with different properties resulting in different indications. Stopping bleeding and enhancing tissue regeneration are particularly important for all surgical specialities, for which materials with haemostatic properties, as well as those increasing tissular adherence are of great interest. Due to large incisions and massive soft tissue involvement, arthroplastic surgery represents one of the most invasive types of orthopaedic procedures, especially when it refers to the hip. High severity of the osteoarthritis (for which arthroplasties are indicated) and increased bone mass increase the bleeding risk, while decreasing the process of tissue regeneration; quite frequent, general conditions (diabetes being by far the most frequent) impair healing, while bleeding- stimulating circumstances (such as therapeutic anticoagulation for cardiac dysfunctions) increase surgical risk, the systemic modalities of controlling them being limited. That is why medical research focused on finding local possibilities to solve these problems, to find materials able to decrease bleeding and to stimulate tissular bonding and healing, resulting into three categories of materials: adhesives, haemostatics and sealants.

Adhesives represent materials able to glue tissues together; they become active in a dry field and are usually self-polymerising, so do the

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sealants, which are materials able to create a barrier preventing leakage of a gas or liquid from a cavity or a structure. A haemostatic agent is effective in the presence of blood, on which it acts, producing primarily blood clotting. By adhering to the leaking blood vessels, thus closing them, both adhesives and the sealants diminish bleeding, without initiating clot formation [1].

Several types of haemostats have been approved, their mechanisms of action being: mechanic (like gelatine, collagen, cellulose), active (based on thrombin), flowable (combining gelatine and thrombin) and fibrin-based, extracted from plasma, enriched with thrombin or collagen.

FDA approved as adhesives the cyanoacrylate group, the albumin &glutaraldehyde group and fibrin-based (human pooled plasma liquid) and as sealants, the PEG group (Polyethylene glycol polymer), the albumin& glutaraldehyde group, the cyanoacrylate group and the fibrin-based sealants [2-3].

As it can be seen, there is only one group performing simultaneously all the three actions, the fibrin-based sealants (FBS) which are two-component materials: fibrinogen and thrombin. Fibrinogen is converted into insoluble fibrin by thrombin in the presence of small quantities of calcium and factor XIII, thus resulting the final stable form [4].

Since 1909, when Bergel first used fibrin as a haemostatic agent, then used for the first time in 1915 by Lippincott for haemostatic purposes during cerebral surgery [5], research progressed focused not only on the haemostatic properties, but also on the healing ones, so that fibrinogen and thrombin where used as a biologic glue for the purposes of skin grafting in 1944 by Cronkite and in the 1980s, Gestring and Lerner, Siedentop and colleagues, Spotnitz and colleagues used chemical and cryoprecipitation methods for producing concentrated fibrinogen used in fibrin sealants [6]. In 1998, FDA approved the fibrin sealant, thus opening the way for clinical use and enhancing research in this fold.

Due to its complex mechanism of action (Figure 1), FBS has a double haemostatic action: besides the primary one, through the fibrin scaffold, it is also a sealant and an adhesive, closing the discontinuities in the blood vessels, thus having a secondary haemostatic effect, since it does not involve clot formation. [7, 8]

Among different fibrin-based sealants, those derived from autologous plasma have both the advantages of extremely low antigenicity and maximal activity. This paper describes clinical experience using a FBS containing a mixture of cryoprecipitate and thrombin prepared by a special device from autologous plasma in a closed, sterile fluid path. Usually, each unit of cryoprecipitate (10 - 15 mL) usually contains fibrinogen (150–250 mg), Factor VIII (80–150 U), combined with von

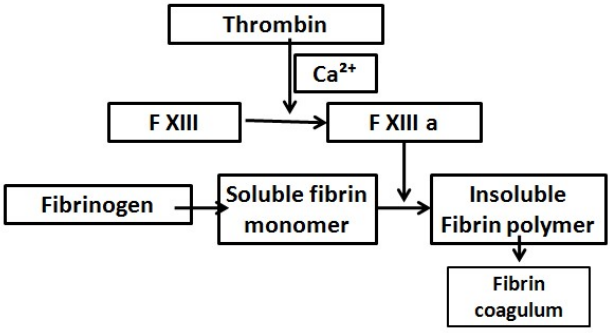


Fig. 1- Mechanism of action for FBS / *Mecanismul de acțiune al FBS.*

Willebrand factor (100-150 U), Factor XIII (50–75 U) and fibronectine (20-40 mg) [9, 10].

Practically, the solution of concentrated fibrinogen and factor XIII is combined with a solution of thrombin and calcium to form (within seconds, or slower if a more dilute form of thrombin is used) a fibrin clot, similar to the coagulum from the final stages of the clotting cascade.

Also, the local application diminishes the risk of adverse effects reported with the systemic administration of cryoprecipitate such as: haemolytic post-transfusion reactions, allergic reactions, from urticaria to anaphylaxis, cardiac insufficiency due to overload, transfusion related acute lung injury, post-transfusion rejection reaction or purpura [11].

The second effect of the FBS is enhancing the wound healing, due to the fibrin which acts like an autologous biocompatible, biodegradable scaffold which can bind cells and substances essential for tissular healing. Regardless of the type of the injury and its' site, cells start the coagulation cascade and normally form a fibrin scaffold as an initial step. This scaffold degrades in a balanced rate with tissue regeneration, it is highly adhesive and biocompatible, since it does not induce any toxic, allergenic or inflammatory reactions [12-14].

Using the electronic microscopy scanning it has been described that the structure of the fibrin network depends on the fibrinogen concentrations: lower concentrations of fibrinogen (5 mg/ml) result in thick fibres and a dense structures, whereas looser gel and thinner fibres result from higher fibrinogen concentrations (20 mg/ml); no significant differences occur when the concentration of thrombin increases from 0.5 U/ml to 5 U/ml [15]

Due to their properties of adhesion, as well as their ability to bind and then deliver cells and substances, fibrin scaffolds have been intensively studied for their possible carrier function for cells (for example mesenchymal cells), growth factors (fibroblast growth factor, neurotrophic 3, transforming growth factor beta 1, transforming growth factor beta 2, nerve growth factor, brain

derived neurotrophic factor)[16-19].

Indicatons for FS in orthopaedic surgery refers both to elective surgery (hip and knee prosthesis) and trauma: closed and open fractures, muscular injuries, skin defects (it improves the binding between the graft and the receptor). Figure 2 shows a case where FBS was used in an open fracture stabilised with an external fixator, with a bone defect.

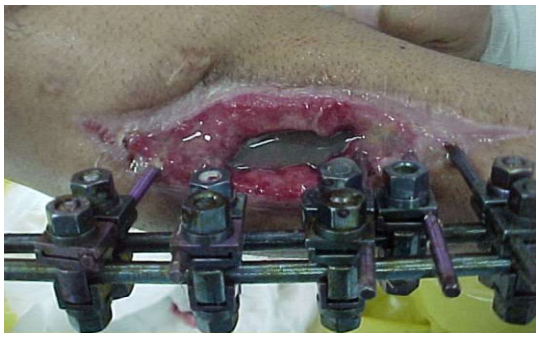


Fig. 2- FBS used in a bone defect after an open fracture/ *FBS utilizat intr-n cazdedefect osos post fractură deschisă.*

2. Matherial and method

This paper retrospectively evaluates 50 patients operated in the Orthopaedic and Trauma Clinic, Emergency Hospital Bucharest for hip osteoarthritis (HOA); 28 of them received FBS (group A) and 22 of them did not (group B). In order to evaluate the efficacy of FBS, a major orthopaedic procedure (total hip arthroplasty) was selected, because these type of surgery has high bleeding risk not form a unique blood vessel, but from a surface with small blood vessels, on which FBS may be active. Therefore, bleeding was assessed separately for the two groups, as to find out wether FBS are efficient or not, using two criteria: the post-operative drainage and the decrease of haemoglobin.

For all the patients of the two groups, the diagnosis of HOA was established following the same protocol; pre-operative evaluations showed physiological levels of haemoglobin and coagulation, with no bleeding abnormalities.

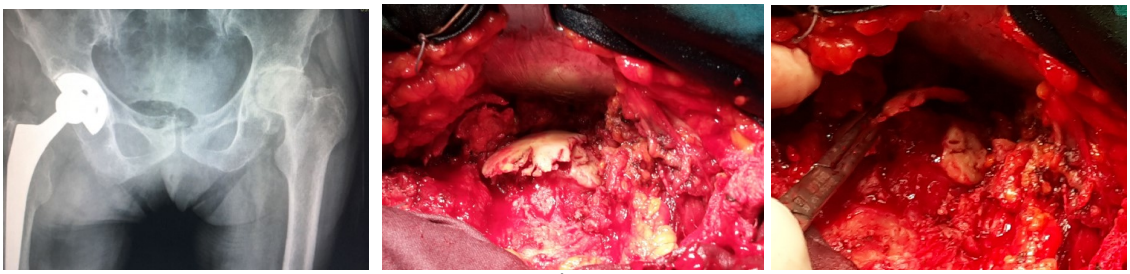
Arthroplasty was performed using the same approach (external) by the same two teams of experienced surgeons all the prosthesis had the same producer; 16 patients in group A and 13 patients in group B (57%) had cementless prosthesis , while for the rest, cemented arthroplasty was performed.

FBS was prepared according to the recommendations of the producer and was applied (after the insertion of the prosthesis, prior to suture) in the immediate peri-prosthetic area and in the subfascial space; since the FBS becomes active after prosthesis is inserted when practically the two types of prosthesis have the same behaviour form the point of view of bleeding, no distinction was made between patients with cementless and cemented prosthesis in groups A and B.

The human-derived FBS Crystal results from donor autologous plasma, thus minimising the risk of infections associated with pooled products and that of allergic reactions, as well as those aprotinin (especially immune) related (it does not contain aprotinin) Due to its strictly local action, no immune mediated coagulopathies have been reported and no risk of transmissible spongiform encephalopathies (prion disease) or bovine spongiform encephalopathy [20].

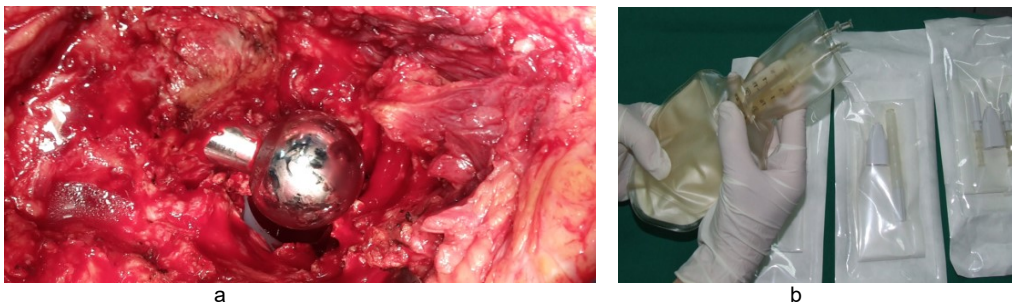
Each set contains 2 syringes filled with cryoprecipitate and thrombin in equal amounts, with 6-16 ml of sealant produced from each unit of plasma [20].

The components may be stored at -18 Celsius grd for 1 year. Once thawed, it can be kept active up to 6 hours at 34-37 Celsius grd or on ice for up to 4 hours and 2 additional hours at 34-37 Celsius grd. The components are pre-warmed at least 10 minutes prior to use. The FBS application system is specially designed so as to insure the proper mixture (due to a single cannula) and the focused application (as spray or drops) of the resulted product on the desired surface. The producer recommendations should be followed in order to ensure the desired effect. Figure 3 presents a case where FBS was used : pre-



a b c

Fig. 3 - Pre-operative Xray (a), intra-operative aspect with HOA (b) and complete avulsion of the femoral head cartilage(c) / *Radiografia pre-operatorie (a), aspect intra-operator cu coxartroză (b) cu avulsia cartilajului capului femoral (c).*



a b

Fig. 4- Reduction of the prosthesis (a) and FBS handling (b) / *Reducerea protezei (a) și prepararea pentru aplicare a FBS (b).*

operative Xray (a)shows marked deformity of the left hip (previously operated on the right) ; intra-operative aspect with HOA (b) and complete avulsion of the cartilage(c) , typical for severe HOA.

A total hip prosthesis was inserted, and, after the insertion of the prosthesis (c), FBS was prepared and introduced (Figure 4).

It must be underlined that the suture wires were put in place, then the FBS applied and then the sores tightened, so that the flaps were the sealant were applied were not at all disturbed by potential movements necessary for wire insertion. This rule was followed for all the layers were the FBS was spread so as to ensure maximum adhesivity and prevent seroma formation.

The patients of the two groups received the same thrombo-prophylaxis regimen according to their comorbidities and pre-op evaluation, followed the same rehabilitation protocol and were evaluated regarding: age and gender, duration of surgery and bleeding landmarks , consisting of :

- Post-operative active drainage – it was kept for 48 hrs.
- Haemoglobin levels- were evaluated 6 hours after surgery was finished, provided that haemostasis was performed, and then 24, 48 and 72 hrs after surgery.

The two groups were also compared regarding the incidence of local and general complications which could be connected with FBS during admission: haematoma, seroma or infection at the surgical site, morbidity and mortality bleeding related.

All the patients signed the inform consents for the treatment protocol; the study was performed according to the European Communities Council Directive of 24 November 1986 (86/609/EEC) and the treatment of the patients was performed following the local Ethical Regulations.

3. Results

As shown in Figure 5, the two groups demographic characteristics compatible with the onset and outcome of HOA : the men age was 73 yrs old in group A and 71 in group B, with no significant differences regarding gender.

The duration of surgery was analysed considering that it might be connected with the bleeding and soft tissue damage, so that differences between groups regarding bleeding could be explained by differences between the extents of surgical procedures. As shown in Figure 6, most surgical procedures lasted between 90 and 120 minutes (from the moment of incision to the moment when suture was completed), with an average interval of 130.9 min (group A) and 126.8 min (group B); it is to be underlined that the application of sealant takes less than 1 minute for each vial, so the differences between the two groups cannot be related to sealant application. In the same time, differences regarding the post-op bleeding cannot be due to the operative time, since the structure of the two groups does not show any relevant data for that.

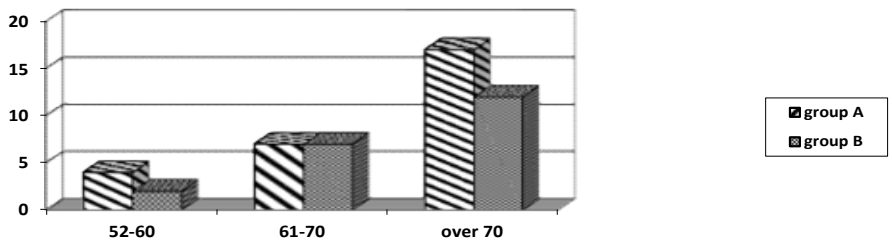


Fig. 5- The age and gender of the patients (yrs) / *Analiza demografică- vârsta și sexul pacienților*.

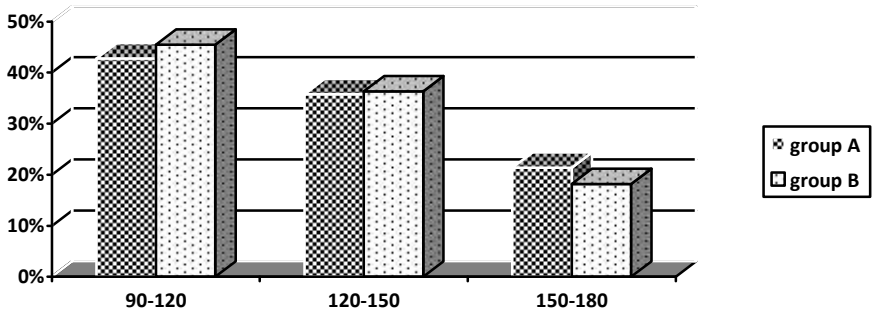


Fig. 6- The duration of the surgical procedure (min) / *Durata intervenției chirurgicale-min.*

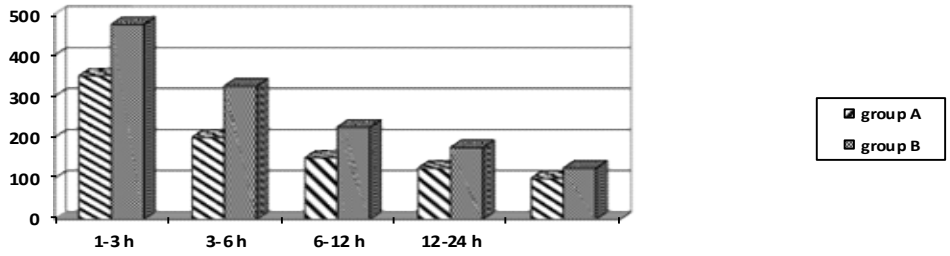


Fig.7- Post-operative drainage (ml) / *Drenajul post-operator ; ml.*

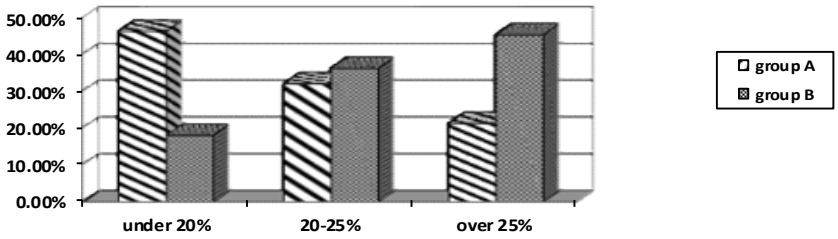


Fig.8- Post-operative decrease of haemoglobin / *Scăderea post-operatorie a hemoglobinei -%.*

The post-operative drainage, as shown in Figure 7, was decreased by the FS, but it is interesting to underline the fact that the difference was significant in the first 3 hours (the application of FS decreased with 20% the post-operative bleeding), then the difference decreased until 6 hrs (11.6%) and then becomes even less after that time.

As for the decrease of haemoglobin, this is somehow parallel to the previously presented data, and higher for the group who did not receive FBS. as shown in Figure 8, most of the patients who had a variation of more than 20% requiring blood transfusions.

As it can be seen, 53.5% of the patients in group A had haemoglobin decrease over 20%, compared to 81.81% in group B, so the necessity of transfusions was considerably higher when FBS were not used. More than that, only 21.4% of the patients in group A had a haemoglobin decrease over 25%, while in group B the percentage is double (45.45%), all these demonstrating the efficacy of the sealant. There was no bleeding-related mortality in neither of the two groups.

As for the local complications, 2 patients in group B developed a seroma 2 weeks after surgery requiring surgical revision; the microbiological exam was negative and the outcome after secondary surgery was favourable. No local septic complications were reported for the patients of the two groups 2 years after surgery.

4. Discussions

Orthopaedic surgery represents one of the most impressive fields from the point of view of bleeding, which can be produced by two types of

vessels: those which can be ligated or cauterised, and those generating diffuse bleeding, such as capillaries, where the two methods previously described are useless. Form the point of view of the tissue generating it, both soft tissue and bones can bleed, either due to trauma or to surgical cuts. Although they do not have an immediate homeostatic effect, but a late one, if they persist, diffuse bleeding are difficult to manage, especially when they result from bony fragments, since especially cancellous bone is well vascularised.

Under these circumstances, stopping these diffuse bleeding focused medical research on discovering local therapeutic means, as the systemic ones have limited effect, especially that large blood loss generates anaemia, requires transfusions (with all the contamination and post-transfusional risks) and impairs recovery, thus prolonging hospitalisation.

In our study, all the parameters monitoring bleeding showed that the FBS was effective in decreasing bleeding, thus having, besides the local effect, a positive systemic one, diminishing the impact of a major surgical procedure, otherwise potentially generating anaemia and hypoxic consequences

This study confirms the validity of the considerable interest which have been demonstrated for the development of FBS, arthroplastic surgery having definite indications for using them, due to the following reasons:

- It requires considerable incisions, followed by muscular splitting, depending on the articular damages, so as to be able to properly see them.
- The surfaces where bones are sectioned bleed with no possibility to stop bleeding by ligation and low effectiveness of cauterization.

FBS have multiple actions: perform haemostasis by direct stimulation of clotting (due to its content) and by closing the small vessels and enhance healing due to its gluing properties; since the product is obtained from human autologous plasma, an issue worthing discussion is the potential variability of the concentration of factor, and thus, of the activity of the product depending on the manufacturing protocol.

For the product used in this study, fresh frozen apheresis plasma is used as a source of allogeneic single donor FBS; the method involves cryoprecipitation to derive fibrinogen and precipitation by ethanol and dicalcium chloride to get thrombin. Laboratory studies were performed in order to assess the concentrations of fibrinogen and thrombin depending on the temperature of initial plasma. Significant differences regarding the concentrations of thrombin were detected when the initial temperature was 2-8 Celsius grd (27.5±17.5UI/ml thrombin concentration) vs 34-37 Celsius grd (41.2±13.2 UI/ml), P=0.041; the volume of product / unit of plasma (in a group of 41 units of plasma) was 12.4±3.3 ml FBS, starting from the initial volume of plasma of 281 ±23 ml. The concentrations of main components were relatively homogenic: 19.9 ±4.6 mg/ml for fibrinogen and 25.7 ±11.1 IU/ml for thrombin [4, 21].

The concentration of fibrinogen also influences the influence upon healing, as Ib studies shown that lower levels of fibrinogen (<20 mg/ml, compared with >80 mg/ml) stimulate matrix degradation and increase vascular growth [22]

An aspect which must be discussed regarding this study is related to seroma prevention after hip arthroplasties, as the surgical technique is invasive resulting in massive flaps which can result, in the absence of a proper adherence, in cavities where seroma can occur. The gravity of this fact consists of the high infection risk of any liquid-filled space, so seroma prevention is extremely important for the pathology described in this study. Therefore, the absence of this complication in group A (with 2 cases in group B) shows not only the efficacy of the sealant, but also the fact that it was properly applied.

The connection between the application technique and the effectiveness of FBS adhesion has been discussed in literature. Several authors (Baily, Cha, Llewelyn-Bennett, Jones, Spotnitz) revealed that depending on the application technique, sealants can work as adhesives or anti-adhesives; if the sealant is applied immediately prior to suture, it will glue the tissues secured by suture; if the FBS is applied with the flaps distracted, it will form a layer which practically isolates each flap from the surrounding tissues, so the effect will be to prevent adhesion. Therefore, prevent seroma formation requires not only the proper action of the glue, but also a proper handling of the product.[4], [23-26]

The results of this study not only confirmed the efficacy of FBS in orthopaedic surgery but also demonstrated the importance of research in the field of materials with medical indications, as FBS can significantly improve the post-operative outcome of patients with total hip arthroplasties. The importance of this fact refers not only to the increasing number of these procedures due to increased life expectancy , but also to identifying topical means to improve haemostasis in order to decrease diffuse post-operative bleeding. As post-operative bleeding requires transfusions, impairs the post-operative capacity of healing and enhances the risk of infections , any mean able to control it will reduce the necessity of transfusions (and the associated risks) and the risk of infections and will help healing, thus resulting in a shorter hospital stay (which is itself a circumstance increasing the probability of nosocomial infections, due to prolonged exposure to hospital bacteria) and an earlier rehabilitation of the patient. Therefore, not only that the expenses will be lower, but the most important thing is that the quality of life of our patients will be significantly improved by

using FBS in orthopaedic surgery

Although our study is limited by the fact that it was a retrospective study it confirmed previous data, for example those of Sabatini and Notarnicola [27, 28], especially that most of the studies were performed on patients undergoing knee surgery. Therefore, as it refers to hip arthroplasty, this study offered data confirming the efficacy of this group of materials in a relatively new field of research.

5. Conclusions

Major orthopaedic surgery is followed by significant bleeding which can have severe systemic consequences, especially that some of the sources of this bleeding cannot be antagonised by surgical method solely. Therefore, biomedical research focused on discovering materials which can act as active haemostatic agents, thus improving the outcome of the patients and decreasing hospital stay and the global costs of recovery. FBS proved to act not only as haemostatics, but also as healing-enhancing factors, especially due to their fibrin scaffold. The human-derived FBS described in this paper is biocompatible and with practically no allergic and infectious risk, thus effective and safe as well. Further prospective randomised studies are necessary in order to complete the existing data, but this paper confirms the data from literature and underlines the importance of the interdisciplinary approach, bringing together medicine and bio-engineering to enhance the success of medical procures and improve the patients' quality of life .

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