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A NEW FIBRIN SEALANT FROM *Crotalus durissus terrificus* VENOM: APPLICATIONS IN MEDICINE

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Fibrin sealant, a widely available tissue adhesive, has been used since 1940 in a variety of clinical applications. Commercially available fibrin sealant products are synthesized from bovine thrombin and human fibrinogen, which may transmit infectious diseases, and recipients may also develop antibodies against bovine thrombin. Bearing these disadvantages in mind, a new fibrin sealant was developed in 1989 by a group of researchers from the Center for the Study of Venoms and Venomous Animals, in Sao Paulo State, Brazil. The main purpose was to produce an adhesive fibrin without using human blood, to avoid transmitting infectious diseases. The components of this novel sealant were extracted from large animals and a serine proteinase extracted from *Crotalus durissus terrificus* snake venom. The applicability of this sealant was tested in animals and humans with beneficial results. The new fibrin sealant can be a useful tool clinically due to its flexibility and diversity of applications. This sealant is a biological and biodegradable product that (1) does not produce adverse reactions, (1) contains no human blood, (3) has a good adhesive capacity, (4) gives no transmission of infectious diseases, and (5) may be used as an adjuvant in conventional suture procedures. The effectiveness of this new fibrin sealant is reviewed and its development and employment are described.

Suturing materials and techniques have presented problems in conditions such as fistulas, granulomas, lacerations of inflamed areas, and tissue ischemia, which prompted research into adhesive materials that could bind tissue and wounds without producing trauma (Matras, 1985). A substance that could bind tissues more rapidly is an important development, since it could promote hemostasis and solidify tissue adhesion with no carcinogenic effects (Morandini and Ortiz, 1992).

The concept of using fibrin sealants to approximate the skin edges of wounds or produce adherence to other tissues is relatively novel. In essence, there are three basic types of fibrin sealants: (1) autologous, (2) homologous (both obtained from cryoprecipitate), and (3) synthetic/commercial (Fattahi et al., 2004).

As a hemostatic agent, fibrin sealant achieves hemostasis resulting in decreased blood loss and potentially improved outcomes following bleeding conditions. A hemostatic compound may also seal tissues to prevent fluid loss and facilitate tissue adherence, thus eliminating potential openings. As a result, it may also reduce operation time and complications, thereby improving effectiveness.

The potential economic impact of fibrin sealants include (1) reduced stay in critical care unit; (2) decreased duration of hospital stay; (3) reduced costs associated with complications of transfusion; (4) potential to increase patient recovery time due to the effects already listed; (5) decreased operating room time; and (6) less claims for medical negligence resulting from surgical complications.

MECHANISM OF ACTION AND BIOCHEMESTRY

Many fibrin sealants, composed of concentrated fibrinogen and thrombin, have been studied and developed (Sierra, 1993). The activity of fibrin sealants is based upon the natural coagulation

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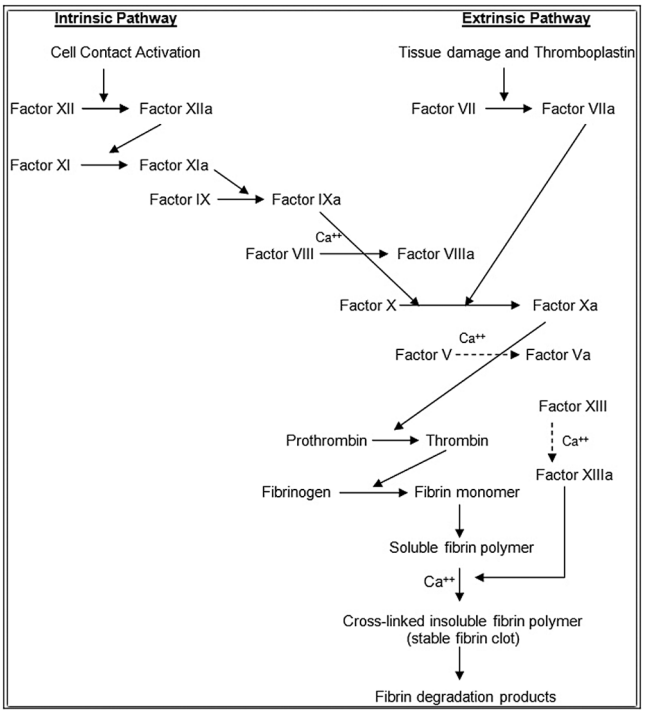

FIGURE 1. The coagulation cascade pathway, showing the intrinsic pathway started by cell contact activation and the extrinsic pathway induced by tissue damage and thromboplastin activation resulting in stable fibrin clot.

cascade, in which the final step involves the conversion of fibrinogen to fibrin by thrombin and the cross-linking of fibrin monomers into an insoluble complex (Figure 1). Thus, these substances mediate the coagulation cascade that leads to clot formation (Lerner & Binur, 1990). The final step in this biochemical process has been investigated and illustrated in Figure 2. This process continues to be studied in order to determine the influence of confounding factors and interrelationships (Mosensson, 2003).

In this cascade, thrombin cleaves fibrinogen into fibrin monomers. The fibrin monomers spontaneously polymerize within seconds into soluble fibrin polymers, which form a relatively weak clot. Thrombin also converts factor XIII (fibrin stabilizing factor) into active factor XIII (factor XIIIa), which, in the presence of calcium (Ca^{2+}), catalyzes the formation of covalent bonds between adjacent soluble fibrin polymers, creating insoluble fibrin polymers (the stable fibrin clot). The stable fibrin clot subsequently undergoes degradation (Alving et al., 1995; Martinowitz & Spotnitz, 1997), as shown in Figure 2A. Figure 2B shows the manner in which commercial fibrin sealants mimic this process, and Figure 2C describes how the new fibrin sealant interjects into the process of stable clot formation. An interesting point is that additional factors such as pH, fibronectin, and temperature also influence the formation of this fibrin sealant network (Spotnitz & Prabhu, 2005).

HISTORY

The use of human plasma compounds to provide local hemostasis has been known since 1909, when Bergel published the first clinical report of the use of fibrin compounds. Grey (1915) reported its application in cerebral bleeding. A fibrin sealants historical review showed that bandages of fibrin were used to control the bleeding in parenchymatous organs in World War I (Brennan, 1991). The combination of autologous fibrinogen and a thrombin solution was reported first in 1940, when Young and Medawar described the use of plasmatic products in peripheral nerve anastomosis in

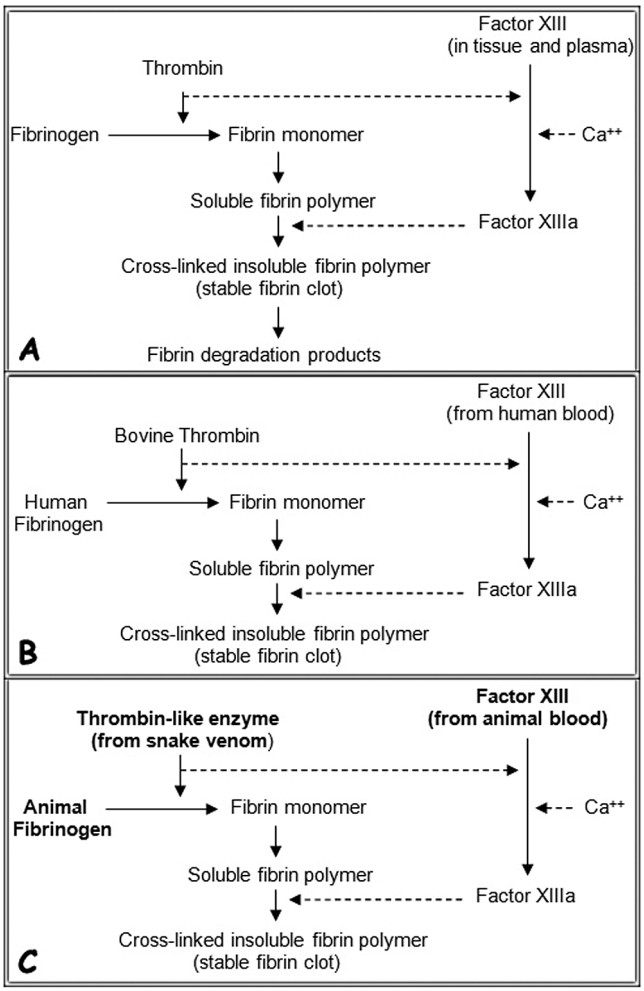

FIGURE 2. Final steps of the coagulation cascade leading directly to the formation of a stable clot: (A) final physiological steps in coagulation cascade; (B) commercial fibrin sealants mimic final steps; (C) new fibrin sealant derived from snake venom mimics the final steps.

animals. However, the results were unsatisfactory due to little stabilization and low adhesion of these products, associated with undeveloped surgical techniques. Seddon and Medawar (1942) reported similar effects and results in humans.

Cronkite et al. (1944) effectively mixed fibrinogen and thrombin to bind cutaneous grafts. Tidrick and Warner (1944) used a mixture of thrombin and human fibrinogen at the bleeding site during surgical procedures to improve hemostasis. However, the concentration was inadequate to promote stable adhesion. During the 1960s the methods to obtain a cryoprecipitate with large amounts of fibrinogen improved and surgical use increased (Alving et al., 1995). During the 1970s, the use of fibrin sealants aroused interest when production of solutions with high concentrations of fibrinogen was synthesized (Thomazini-Santos, 2001).

Matras et al. (1972) described the applicability of the fibrin sealant in peripheral nerves in rabbits. The results were positive with respect to adhesion and healing, thus encouraging the use in surgical procedures. Staindl (1979) successfully used fibrin sealant from a pool of human plasma and bovine thrombin.

In 1982, Matras applied fibrin sealant in bone cavities, facial nerves anastomosis, and bucomaxillofacial and vascular surgeries. The use of autologous fibrin sealant was ignored until 1983, when Gestring and Lemer described cryopreciptation methods to produce concentrated fibrinogen, as

further developed by Sidentrop et al. (1989) and Spotnitz et al. (1987), which would avoid infectious disease transmission (Lerner & Binur, 1990).

Finally, the first fibrin sealant was released in Europe in the 1980s. In the United States, the Food and Drug Administration prohibited its use until the 1990s because of the risk of transmitting infectious diseases, since this sealant was synthesized from human and animal blood.

TOXICITY, INFECTIOUS RISKS, AND ADVERSE EVENTS

The commercially available fibrin sealant products are synthesized as derived from bovine thrombin and human fibrinogen. Unfavorable results from application of fibrin sealants can be divided into those related to the product ingredients themselves or those related to adverse effects following clinical use. As indicated previously, there were risks of viral transmissions with documented cases (Wilson et al., 1991).

Hino et al. (2000) reported three cases of iatrogenic parvovirus B19 transmission associated with commercial fibrin sealants. This was attributed to the use of dry-heat viral inactivation, which appears to be ineffective against parvovirus. Bovine thrombin was used to lower costs, but some patients developed antibodies against bovine thrombin (Banninger et al., 1993; Israels & Israels, 1994; Rapaport et al., 1992). There were reports of flap necrosis and seroma formation following improper use of fibrin sealants during face-lift procedures (Flemming, 1992; Grossman et al., 2001). This is the results of a non-homogeneous application of fibrinogen and thrombin, as well as due to an excessively thick sealant layer. Ischemia occurs as the thick layer of the sealant begins to act as a mechanical barrier, preventing capillary revascularization under the skin flap.

Table 1 shows the unique fibrin sealants approved for commercial use by the Food and Drugs Administration in the United States (Spotnitz & Prabhu, 2005).

HISTORY OF BRAZILIAN PRODUCT DEVELOPMENT

Based on this information, the notion of producing a new fibrin sealant was proposed in 1989 by a group of researchers from the Center for the Study of Venoms and Venomous Animals (CEVAP), Sao Paulo State University, Botucatu, Brazil. The main purpose of this project was to produce a fibrin sealant that did not use human blood components. Thus, a cryoprecipitate extracted from large animals was produced to substitute for human fibrinogen. A fraction of gyroxin complex from the venom of *Crotalus durissus terrificus* was used rather than bovine thrombin.

TABLE 1. Fibrin Sealants Approved for Commercial Uses by Food and Drug Administration (FDA) in the United States

Trade name (company)	FDA approval	On label use	Components	Anti-fibrinolytic	Procedures for viral inactivation
Tisseel (Baxter)	June 1998	Cardiopulmonary bypass Splenic injury Closure of colostomies	Human fibrinogen and thrombin	Bovine aprotinin	Cryoprecipitation Freeze-drying Vapor heating Adsorption (thrombin only)
Vitagel (Orthovita)	July 2000	Anastomosis bleeding Bone cancer bleeding Capillary bed bleeding	Autologous fibrinogen Bovine collagen and thrombin	None	Collagen: acid/base treatment Thrombin: chromatographic separation
Crosseal (Ethicon, J&J)	March 2003	Liver resection surgery	Human fibrinogen and thrombin	Tranexamic acid	Cryoprecipitation Solvent detergent cleansing Pasteurization (fibrinogen) Nanofiltration (thrombin)

Note. Adapted from Spotnitz and Prabhu (2005).

Gyroxin, a neurotoxic component isolated from the *Crotalus durissus terrificus* venom, belongs to the group of thrombin-like enzymes and is an important source of serine proteinase. The isolation was initially described by Barrio (1961) and then purified by Alexander et al. (1988).

The snake venoms, mainly in the Viperidae family, contain abundant quantities of proteolytic enzymes and are divided into two groups: serine proteinases and metalloproteinases, which both affect the hemostatic system in a variety of mechanisms (Jia et al., 2003; Ferreira et al., 2006; Costa et al., 2009). Serine proteinases constitute 20% of the total proteins in the venom and are responsible for several biological functions involved in digestion, complement system activation, cellular differentiation, and hemostasis (Serrano & Maroun, 2005).

These enzymes are not lethal by themselves, but contribute to the adverse effects of the venom when associated with other venom proteins. These enzymes affect many steps in the blood coagulation cascade, either nonspecifically, by proteolytic degradation, or selectively, by activating or inhibiting specific blood factor involved in platelet aggregation, coagulation and fibrinolysis (Braud et al., 2000). The enzymes that affect the hemostatic system are divided into coagulant and procoagulant; anti-coagulant; platelet function inhibitors; and fibrinolitic system activators (Marsh, 1994; Marsh & Williams, 2005; Markland, 1998; Matsui et al., 2000; Esmon, 2000).

There are several serine proteinases, such as the kallikrein-like proteinases, that exert a hypotensive action, releasing bradykinin and the thrombin-like enzyme responsible for the formation of a fibrin clot at the end of the coagulation cascade. The isolation of thrombin-like enzymes is of interest because in development of diagnostic reagents and for treatment of hemostatic alterations, which is important in the production of fibrin sealants (Marsh & Williams, 2005).

The majority of the thrombin-like enzymes previously studied are derived from *Agkistrodon*, *Trimeresurus*, *Crotalus*, and *Bothrops* genera (Pirkle, 1998). Raw et al. (1986), using ammonium sulfate precipitation followed by gel filtration on Sephadex G-75 and affinity chromatography with sepharose-1,4-butanediol-diglycil-*p*-aminobenzamide, observed that gyroxin corresponds to a molecular mass of 34 kD, with optimal coagulation of human fibrinogen at pH 8 and a proteolytic activity in the alpha-chain of fibrinogen. Gyroxin possessed amidase activity on L-arginine-*p*-nitroanilide and L-arginine-7-amido-4-methyl-coumarin amino terminal blocked peptides and esterolytic activity on N-alpha-tosyl-L-arginine-methyl ester.

This neurotoxin is not affected by freezing and thawing or by treatment at 40°C for 15 min (Seki et al., 1980), demonstrating its stability. Gyroxin is a nonlethal toxin that produces a rotation around the long axis in animals known as barrel rotation (Barrio, 1961). Gyroxin acts on the human and animal fibrinogen cleaving the fibrin peptide A of the alpha-chain near N-terminal. The resulting fibrin monomers polymerize into a clot that differ from the one produced by thrombin. These clots are easily dissolved as the clots are more susceptible to the action of the fibrinolytic agents (Koh et al., 2001).

The applicability of this new fibrin sealant composed of fibrinogen extracted from large animals and thrombin-like enzyme extracted from *Crotalus durissus terrificus* was tested in animals and humans.

Animal Studies

Viterbo et al. (1993) examined the efficacy of the fibrin sealant in the repair of sciatic nerve of Wistar rats using bubaline, equine, bovine, and human fibrinogens. These products were compared to the commercial fibrin sealant (Tyssucol). Viterbo et al. (1983) observed effective hemostatic and adhesive properties, as well as a satisfactory regeneration of the sealed nerves. Results thus indicated that bubaline fibrinogen was a major alternative for repair of peripheral nerves.

Iuan et al. (1995) used this new fibrin sealant in the repair of Wistar rat peripheral nerves and observed effective hemostatic and adhesive properties, besides reliable regeneration of the glued nerves. Sartori et al. (1998) compared the advantages and disadvantages of this sealant and conventional suture in testicular biopsy of rams. Data demonstrated that the product was easily applied, showed fast and effective healing, and reduced the postoperative morbidity of the animals. Thomazini-Santos et al. (1998) compared the levels of fibrinogen in cryoprecipitate that was bovine, equine, ovine, bubaline, and extracted from human beings. The results demonstrated that

the bubaline cryoprecipitate had the highest level of fibrinogen when compared with the other samples.

The efficacy of a fibrin sealant made up of snake venom and bubaline fibrinogen was evaluated by rat colon anastomosis (Leite et al., 2000). Eighty rats were randomly assigned into two experimental groups: G1 control (anastomosis with extra mucous interrupted suture) and G2 (repair suture + fibrin sealant). The animals were studied at the following 4 time points: T0: preoperative, T1: d 7 postoperative, T2: d 14 postoperative, and T3: d 21 postoperative. The macroscopic characteristics of the intestinal segment open and closed anastomosis and bursting strength of the anastomosed segments were observed at each of the above times. The results showed that the anastomosed segments were coapted and there were no marked differences in the bursting strength values between the two groups. There was a decrease in the bursting strength values up until d 7 postoperative in both groups, with a progressive increase at other times. Leite et al. (2000) concluded that although important experimental studies using large animals are needed for a better evaluation of tissue repair processes, this fibrin sealant may have the potential to become a valuable tool for use in anastomosis.

In 2000, Reis examined two methods for peripheral nerve repair in rats using autologous nerve grafting: (1) coaptation with the fibrin sealant versus (2) epineural terminal lateral neurorrhaphy. The possibility of regenerating axons from an intact nerve (vagus nerve) to grow into a nerve graft (fibularis nerve) was studied. The grafts were harvested after 8 and 12 wk postsurgery and processed for electron microscopy. The regeneration rate was higher in fibrin sealant coapted grafts.

Thomazini-Santos (2001) added antifibrinolytic agents to fibrin sealant and evaluated the effect on wound edge coaptation in rats. The agents studied consisted of e-aminocaproic acid, the tranexamic acid, and the aprotinine. Statistically significant differences were not observed between experimental and conventional suture groups, but the tensile strength was higher when fibrin sealant was used. Histopathological analysis demonstrated that fibrin sealant and the tranexamic acid showed the best wound edge coaptation.

Moraes et al. (2004), examined regeneration strength of the nonpregnant adult dog uterus when the new fibrin sealant was used to reinforce hysterorrhaphy. The maximal limit and rigidity were analyzed. Twenty uterine horns from 10 bitches were hysterotomized, distributed into 2 equal groups, and hysterorrhaphy was performed using Shimieden–Cushing double-layer suture. In one group, animals received the new fibrin sealant as reinforcement. Although neither variable was significantly different, our results showed higher rigidity values in the fibrin sealant group. This may be attributed to the adhesive's effect on organ elasticity or to more granulation tissue formed in the uterine scar.

Rahal et al. (2004), analyzed the effect of the new fibrin sealant on split-thickness autologous skin graft in dogs. The glued grafts had statistically higher graft viability than sutured grafts. Histologically, tissue repair in the glued grafts was more accentuated than in sutured grafts. Evidence indicated that fibrin sealant increased the survival of autogenous split-thickness skin graft. Ferraro et al. (2005a, 2005b) determined the influence of this new fibrin sealant on tendon healing in dogs. The deep digital flexor tendon of the 5th digit of 24 thoracic limbs was partially sectioned for adhesive application. The biomechanical evaluation showed that, over time, tendon healing gained progressive resistance for maximum traction and permanent deformations with satisfactory results. In a continuous study the tendon of the 2nd digit of 30 thoracic limbs of dogs was partially sectioned for glue application. Biopsies were performed 7, 15, and 30 d postsurgery for clinical and morphological study of tendons. The morphological analysis revealed a typical tendon healing process with a lower level of inflammation in the acute phase, facilitating the cicatricial maturation phase (Ferraro et al., 2005a; 2005b).

Sampaio et al. (2007) used fibrin sealant to treat deep corneal ulcers. Twenty-one dogs were divided into seven groups of three animals each. Six experimental groups were periodically evaluated and collection was carried out on postoperative d 1, 3, 7, 15, 30, and 60, whereas one control group was evaluated throughout the experiment. Analyses were comprised of clinical evaluation and histopathology. The results indicated that fibrin sealant was efficient in repairing keratectomy wounds in dogs and contributed to an earlier healing, avoiding edema formation and keeping

corneal clearness. The new fibrin sealant was found to be cheaper, easy to apply, and feasible to use in animal models.

Further, Vicente et al. (2007) compared the effectiveness of the coaptation with fibrin sealant derived from snake venom in the repair of the peroneal nerve. Ten Wistar rats had their left peroneal nerve sectioned and repaired immediately with the fibrin sealant while the right nerve served as control. In the nerves that were repaired with the sealant, the nerves were myelinated and unmyelinated nerve fibers, with a great amount of connective tissue in the extracellular space. This process showed a stoppage of bleeding and functional recovery of the structure of the nerve. The functional recovery of the sciatic nerve repaired with fibrin sealant was evaluated by means of isolated indirect contractions applied to the myoneural complex, sciatic nerve–extensor muscle of the long finger (EMLF). Wistar rats were divided into two groups: control (no lesion) and glue (sciatic nerve injures and repaired with fibrin sealant). After 10 wk, the myoneural complex was collected and stimulated. The EMLF of the animals of the fibrin sealant group responded with contractions after the stimulus of the motor nerve. Data suggest that nerves repaired with fibrin sealant were capable of conducting nerve impulses to skeletal muscle (Vicente et al., 2008).

Human Studies

In 1999, Stolf performed a pioneer study in humans to evaluate the efficacy of the fibrin sealant in skin surgery and compared the results with conventional sutures. Twenty-one patients with basal cellular carcinoma in the nasal region participated in this study. After tumor removal, the excised areas were covered with skin removed from the right and left nasolabial folds. Skin grafting of the left nasolabial fold was glued and the right was sutured. The comparative study of both areas in the same patient showed erythema and edemas on the sutured areas, dehiscence, and serum-hemorrhagic exudation on the glued area 48 h postsurgery. The cosmetic evaluation of scar formation was superior for the glued area compared to good for the sutured area. Patients did not show any local or systemic adverse effects. Fibrin sealant had a complete adhesion in 71.4%, thus making it a valuable alternative in skin surgery (Figures 3 and 4).

The new fibrin sealant was evaluated for periodontal surgery. The grafts were made on contralateral mandibular bicuspids of 15 patients, so that each subject received one treatment of each type. Clinical analysis included measurements of probing and vertical dimension of grafts at 30, 60, and 90 d postoperatively, as well as photographic follow-up at 7, 14, 30, 60, and 90 d. The patients were submitted to a questionnaire regarding postoperative signs and symptoms. Five biopsies from each side (control and tested) were collected at 7, 14, and 45 d of healing, and were histomorphometrically analyzed for relative volume density of the connective tissue in different components and epithelium/connective tissue interface. Fibrin sealant grafts showed better clinical appearance and tissue organization on histological examination during the first 14 d postoperative. Morphometrical observation demonstrated that the control side presented a statistically higher density of inflammatory cells at 7 d and a tendency toward a lower density of collagen fibers, and less developed epithelium/connective tissue interface. The fibrin sealant was thus suitable for use in free gingival grafts, presenting outcomes comparable to conventional sutures, with the following advantages: ease of application, smaller graft dimensional change, and better postoperative healing (Oliveira, 2002).

A study by Chiquito (2007) compared suture and this new fibrin sealant for fixation of connective tissue graft to evaluate the postoperative characteristics of exposed root surfaces treated with connective tissue graft by using two fixation techniques. Forty-two patients were randomly divided into 2 groups of 21 individuals each. In the first group, graft was fixed to exposed root surfaces by new fibrin sealant; in the second group, conventional suture procedures were employed. The patients ranged in age from 19 and 49 yr and they presented a gingival defect known as marginal tissue recession. Clinical parameters such as surgical and hemostasis time, erythema, breath and taste alterations, root coverage degree, recession height, vestibular probing depth, clinical insertion level, plaque index, gingival index, amount of attached gingiva, keratinized tissue, and aesthetics were assessed during the preoperative and transoperative periods, and 7, 14, 21, 30, 60, and 90 d postoperative. Comparative analysis between the initial and final stage of each group showed statistically significant results for all variables evaluated, mainly at the end of the study. Comparison of

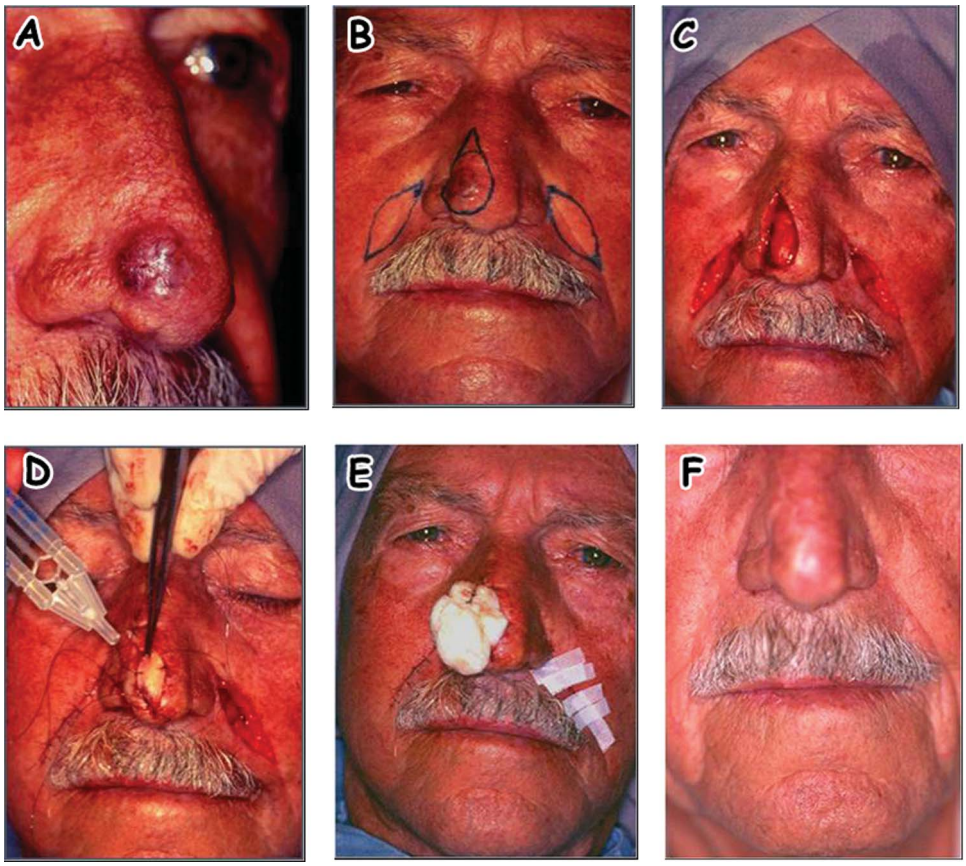

FIGURE 3. (A) and (B) Representative use of fibrin sealant in patient number 1 with nasal basal cellular carcinoma; (C) excised areas; (D) covered with the skin removed from the right and left nasolabial folds; (E) and (F) postoperative skin grafting of the left nasolabial fold glued and the right sutured.

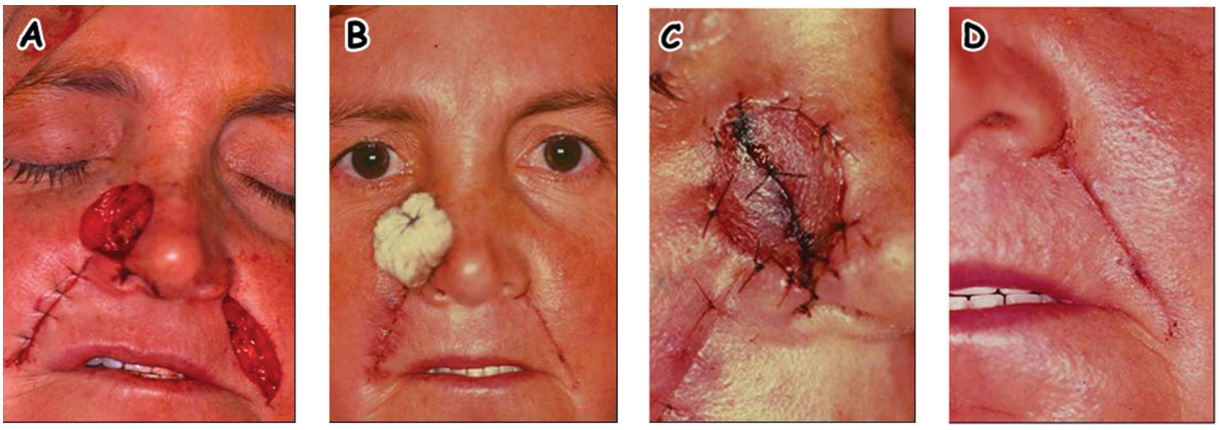

FIGURE 4. (A) Representative use of fibrin sealant in patient number 2 with nasal basal cellular carcinoma; (B) postoperative skin grafting of the right nasolabial fold glued and the left sutured; (C) sutured side and(D) glued side.

groups demonstrated statistically significant results for surgical time, amount of attached gingiva, and vestibular probing depth when test group presented better results. No significant differences were observed for the other variables. The new fibrin sealant showed favorable characteristics when applied in periodontal surgery for root coverage in marginal tissue recessions, and was an efficient substitute for conventional suture.

Barbosa et al. (2007) evaluated the applicability of the fibrin sealant in periodontal surgery. The free gingival grafts that were sutured were compared with others immobilized with the new sealant. A receptor site was randomly selected for each patient as the test area, where the graft was immobilized with the fibrin glue. The contralateral site was used as a control, where the grafts were sutured. After 1 wk, the patients answered a questionnaire regarding pain, edema, and bleeding. The grafts of the experimental group presented better appearance during the first 14 postoperative days and pain was observed more often in the suture group. Further, Barbosa et al. (2008) published more data regarding gingival grafts where the fibrin sealant was used. Five biopsies of each group were collected at 7, 14, and 45 d after healing, and were histologically and morphologically analyzed as to relative volume density of different connective tissue components. The sites in the control group presented a higher inflammatory cell density at 7 d and lower collagen density. In the experimental group, the grafts demonstrated a healing appearance.

Since the new fibrin sealant promoted a reduction of infection and edema, as well as bleeding control and pain decrease, it was used to treat venous leg ulcers. Twenty-four patients were divided into a control group (11 patients) and a treatment group (13 patients) that received fibrin sealant. The results revealed that the patients treated with the fibrin glue showed satisfactory healing when compared with controls. This new treatment may serve as an appropriate alternative to treat ulcers with several advantages: easy application, less pain, early hospital discharge, and lower cost (Gatti, 2009).

CURRENT CLINICAL AND SURGICAL APPLICATIONS

The most common applications of fibrin sealant are in different surgery areas, specially cardiovascular and thoracic surgeries, which in Europe correspond to about 30–45% of total indications (Radosevich et al., 1997). Fibrin sealant is also effective in patients undergoing surgery presenting coagulopathies. The most significant applications are described in Table 2.

Surgical

Cardiovascular The use of fibrin sealant during cardiovascular surgeries was originally developed in 1982, when the sealant was applied 413 times in a group of 340 patients when the conventional suture seemed impossible, difficult or even dangerous, with a success rate of 95%. Although it took some time, the sealing was perfect and became a routine procedure, reducing blood loss and subsequently saving patients (Borst et al., 1982).

Different methodologies have been used to develop an animal model to compare these fibrin sealants with sutures in vascular surgeries. Aorta anastomosis in rabbits utilizing four and eight sutures together with fibrin sealant is efficient to promote hemostasis (Kheirabadi et al., 2001). According to Padubidri et al. (1993), the conventional methods in microvascular anastomosis using sutures may produce vessel narrowing, foreign-body reactions, and intravascular thrombosis. The fibrin sealant was examined in association with two surgical stitches in anastomosis of rat femoral vessels, showing that this is an atraumatic technique, easy to perform, and inexpensive. The association of fibrin sealant with sutures was also performed in epigastric artery anastomosis in rodents, showing fast and easy application (Moskovitz et al., 1994).

In spite of advances in the management of bleeding associated with cardiac surgery, hemorrhages remain a cause of distress, particularly in complex cases and high-risk patients. In order to minimize these problems, White et al. (2000) utilized the fibrin sealant together with suture in dogs to determine the effectiveness in controlling bleeding from punctures in coronary arteries. Data demonstrated the successful applicability to prevent bleeding during anastomosis. The use of the

TABLE 2. Most Significant Applications of Fibrin Sealants, Showing the Tissue-Specific Uses and References

Tissue specific	Uses	References
Cardiovascular	Surgery	Borst et al., 1982; Kheirabadi et al., 2001; Padubidri et al., 1996; Moskovitz et al., 1994; White et al., 2000; Seguin et al., 1992; Endo and Kurosawa, 2004; Kjaergard and Trumbull, 2000.
Thoracic	Surgery	Nomori et al., 2000; Glover et al., 1987; Antonelli et al., 1991; Hansen et al., 1989; Hillerdal et al., 1995; Okereke et al., 2005; Brega Massone et al., 2003.
Gastrointestinal	Surgery	Ishitani et al., 1989; Schwartz et al., 2004; Tanaka et al., 2002; Heneghan et al., 2002; Cintron et al., 2000.
Neurosurgery	Surgery	Shaffrey et al., 1990; Kassam et al., 2003; Nakamura et al., 2005; Lee et al., 1991.
Ophthalmologic	Surgery	Lagoutte et al., 1989; Sharma et al., 2003; Biedner and Rosenthal, 1996.
Urologic	Surgery	Kram et al., 1989; Levinson et al., 1991; Martinowitz et al., 1992; Evans et al., 2003; Sharma et al., 2005.
Gynecologic	Surgery	Bar-Hava et al., 1999; Sciscione et al., 2001; Kiilhoma et al., 1995.
Orthopedic	Surgery	Ambacher et al., 2001; Wang et al., 2003; Levy et al., 1999.
Dental and oral surgery	Surgery	Rackocz et al., 1993; Halfpenny et al., 2001; Yucel et al., 2003.
Plastic and reconstructive surgery	Surgery	Fezza et al., 2002; Oliver et al., 2001; Nervi et al., 2001; McGill et al., 1997.
Kinetics of antibiotics from the fibrin clots	Drug delivery	Greco et al., 1991; Kram et al., 1991; Redl et al., 1983; Thompson and Davis, 1997.
Chemotherapeutic agents	Drug delivery	Kitazawa et al., 1997; Miura *et al*., 1995.
Neurotrophic factor in nerve regeneration	Drug delivery	Yin et al., 2001; Iwaya et al., 1999.
Endothelial and cell growth factor	Drug delivery	Hashimoto et al., 1992.

fibrin sealant was also described in ventricular septal repairs in three patients. During surgery for acquired ventricular septal defects, fibrin sealant reinforced the necrotic and fragile tissues by reducing perioperative bleeding (Seguin et al., 1992). In a group of 32 patients who had experienced postinfarction left ventricular free wall rupture, 23 were treated surgically and 9 were treated with fibrin sealant. Only one of the 11 deaths occurred in the fibrin sealant group, thus reaffirming the effectiveness of the product (Endo & Kurosawa, 2004).

A study comparing the fibrin sealant applied to one side of the sternum with no treatment applied to the other side was performed in 30 patients undergoing cardiac surgery. The results showed that complete hemostasis was achieved on 24 of 30 sides treated with fibrin sealant (hemostasis average time was 43 s), compared with 4 of 30 untreated sides (hemostasis average time was 180 s) (Kjaergard & Trumbull, 2000).

Thoracic The use of fibrin sealant in pulmonary resections remains controversial. This is an extremely difficult environment for surgical adhesives. Sealing the parenchyma of the lung requires a material that is both elastic and strongly adherent to the parenchymal surface. To strengthen the sealing effect of fibrin sealant for pulmonary air leakage, investigators mixed different concentrations of collagen with the glue and the effect was examined on a plastic cap with pinholes and on swine lung. Elasticity and adhesion were also measured. The mixing demonstrated more effectively sealed pulmonary air leakage due to increased elasticity of the glue (Nomori et al., 2000). Glover et al. (1987) used the fibrin sealant to close bronchopleural fistulas. Fibrin sealant was applied through a flexible fiberoptic bronchoscope. The advantages of this method include the avoidance of general anesthesia and thoracotomy. The sealant was also used to close tracheoesophageal fistulas following the same procedure (Antonelli et al., 1991).

There is a report on prophylactic treatment for application of fibrin sealant to the pulmonary surface during thoracoscopy in idiopathic spontaneous pneumothorax in patients. This treatment reduced the need for thoracotomy, minimizing hospital stay, and did not change the normal pleuro-pulmonary anatomy (Hansen et al., 1989). The fibrin sealant was also used in a study of five patients with poor lung function and large emphysematous bullae. Fibrin sealant was introduced

into the bullae through a thoracoscope. The outcome was significant without postoperative complications (Hillerdal et al., 1995).

The main function of the fibrin sealant in thoracic surgery is in the prevention or decrease in the risk of pulmonary air leakage. This problem is also associated with increase in morbidity, infection, and bronchopleural fistulas (Okereke et al., 2005). Thus, in a case-control study, 100 patients who had undergone a precision resection for lung metastases were treated with fibrin sealant or cauterization (Brega Massone et al., 2003). The sealant substantially reduced air leak time, complications, drain time, and hospitalization.

Gastrointestinal The main applications of the fibrin sealant in gastrointestinal (GIT) surgeries are in intestinal anastomosis, liver biopsies, and controlling bleeding following resection and transplantation (Radosevich et al., 1997). The feasibility of applying fibrin sealant was investigated in liver injuries using laparoscopic guidance, and microscopic changes were observed. These studies indicated a rapid control of hepatic bleeding and no evidence of hepatic toxicity (Ishitani et al., 1989).

In a prospective, randomized study of 121 patients undergoing liver resection, bleeding was controlled with standard topical hemostatic agents in 63 patients and with fibrin sealant in 58 patients (Schwartz et al., 2004). The mean time to hemostasis was 282 s with fibrin sealant and 468 s with the standard agents. Hemostasis was achieved within 10 min in 53 patients (91.4%) treated with fibrin sealant, compared with 44 patients (69.8%) treated with standard topical hemostatic agents. In addition, there were fewer postoperative complications among the patients treated with fibrin sealant. In a retrospective study of 363 patients who underwent hepatic resections without biliary reconstruction for liver cancer, fibrin sealant was used effectively to control bile leakage (Tanaka et al., 2002).

An optimal treatment for gastric variceal bleeding remains to be determined, and the use of conventional sclerosing agents is associated with high rates of recurrent bleeding. In order to control bleeding, the fibrin sealant was injected into gastric varices of 10 patients, and immediate hemostasis was achieved in 7 out of 10 (70%) with a single injection. At a median follow-up of 8 mo, there was no recorded episode of recurrent bleeding from gastric varices (Heneghan et al., 2002).

The fibrin sealant has been successfully used to treat fistulas-in-ano. A prospective, nonrandomized clinical trial with 79 patients was performed in which fibrin sealant was used to repair these fistulas. Twenty-six patients were treated with autologous fibrin sealant synthesized from their own blood, and 53 patients were treated with commercial fibrin sealant. Fourteen of 26 (54%) patients treated with autologous fibrin sealant had complete closure of their fistulas, whereas 34 out of 53 (64%) patients treated with the commercial fibrin sealant had closure of their fistulas. The fibrin sealant offered a surgically less invasive mode of managing fistulas-in-ano (Cintron et al., 2000).

Neurosurgery The potent tissue sealing properties and biocompatibility of fibrin sealants enable them to be particularly suitable for use in neurosurgery. Fibrin sealants have been used successfully to prevent leakage of cerebrospinal fluid, not only to prevent but also to treat fistulas. In a wide variety of neurosurgical procedures in 134 patients, fibrin sealant was used as an adjunct to dural closure. The success in preventing cerebrospinal fluid (CSF) leakage was 90% (121 of 134 patients), showing that the sealant is a valuable clinical tool for the neurosurgeon (Shaffrey et al., 1990).

The determination of clinical efficacy and cost-effectiveness of fibrin sealant was evaluated in patients with intracranial pathological lesions. The incidences of cerebrospinal fluid (CSF) leaks in matched groups treated with fibrin sealant or without it were compared. The costs of the fibrin sealant use were compared with the costs of postoperative management, spinal drainage, and occasionally surgical reexploration. The patients who received the fibrin sealant exhibited nondetectable postoperative CSF leaks. Patients who did not receive the fibrin sealant treatment demonstrated 4–16% incidences of postoperative leaks, whose costs exceeded the ones of using the fibrin sealant. This study indicated that the fibrin sealant reduced not only the incidence of postoperative CSF leaks but also overall management costs (Kassam et al., 2003).

The fibrin sealant was used in a comparative study of 39 patients divided into three groups: (1) dural closure alone, (2) use of autologous fibrin sealant after dural closure, and (3) use of commercial

fibrin sealant after dural closure when the patient had undergone a dural closure after spinal-cord surgery. The results demonstrated that volume of drainage fluid was significantly lower in the group with autologous fibrin sealant compared to the group without sealant. The use of autologous fibrin sealant was superior to that of commercial fibrin sealant in cost (Nakamura et al., 2005).

The applicability of fibrin sealant was demonstrated in 26 patients with various neurosurgical problems, such as repair of CSF leaks, sealing of the vascular anastomosis sites, reinforcements of aneurismal clipping, and hemostasis after resection of brain tumors. This experience with fibrin sealant suggested that it was a valuable adjuvant to various microneurosurgical procedures (Lee et al., 1991).

Ophthalmologic The idea of using fibrin sealant in the treatment of perforated or preperforated ulcers is not new. As early as the 1960s, surgical sealants and their various applications were proposed (Ellis & Levine, 1963). A study described eight cases of corneal perforations that were treated with commercial fibrin sealant. The patients had a better prognosis and reported an improvement of visual acuity in eyes treated without infection and inflammation. The use of the sealant was well tolerated, the sealant was degraded by physiological and noninflammatory mechanism of fibrinolysis, and the sealant helped corneal healing (Lagoutte et al., 1989).

The efficacy of fibrin sealant was tested in 41 patients with corneal perforations up to 3 mm in diameter and randomly assigned to 2 groups. Group 1 comprised 19 eyes treated with fibrin sealant, and group 2 comprised 22 eyes treated with cyanoacrylate. The number of the eyes with successful healing, time required for healing, status of corneal vascularization, and complications were compared in these two groups. The fibrin sealant and cyanoacrylate tissue adhesive were both effective in the closure of corneal perforation. Fibrin sealant provided faster healing and induced significantly less corneal vascularization (Sharma et al., 2003).

The incisions of six patients who underwent a bilateral symmetric strabismus surgery were closed with suture in one eye and fibrin sealant in the other. The conjunctival closure with the suture resulted in increasing discomfort and inflammation during the early postoperative period (Biedner & Rosenthal, 1996).

Urologic Several studies showed the efficacy of fibrin sealant for hemostasis during renal surgery. Kram et al. (1989) used the fibrin sealant to treat renal trauma in 14 patients. The sealant was applied over the lacerated area, and for deep penetrating injuries the fibrin sealant was injected into the wound. There was no renal loss, infection, delayed bleeding or urinary leakage showing that fibrin sealant was effective for hemostasis (Kram et al., 1989). The fibrin sealant was also used for partial nephrectomy in 7 patients. There was no delayed hemorrhage or infection, and the sealant was effective in controlling venous bleeding from the cut surface of the kidney which resulted in effective hemostasis (Levinson et al., 1991). In a study comprising 10 patients with hemophilia who had undergone circumcision, a topical fibrin sealant was applied for hemostasis, resulting in a decreased blood transfusion rate (Martinowitz et al., 1992).

A topical fibrin sealant was used in 19 patients for iatrogenic urinary-tract injury during gynecological or general surgical procedures (7), complex urinary fistulas (5), and urological surgical complications (7). Successful resolution of the injury, fistulas, or complication was attained after a single application of fibrin sealant in 18 patients (94.7%) when a direct injection technique was used. It seems to be safe and prudent to use a fibrin sealant in urological damage control from trauma, fistulas, or surgical complications (Evans et al., 2003).

Urinary-tract fistulas present unique clinical challenges often requiring open surgical excisions with interposition of healthy tissue. Advances in retrograde instrumentation have enabled endourologists to employ minimally invasive approaches to urologic diseases, including fistulas. In this study eight patients had an endoscopic injection of fibrin sealant for the treatment of urinary-tract pathology. This technique was successful in six cases (75%) with a single injection of fibrin sealant. Two (33%) of the successfully treated patients required two injections. The endoscopic injection of fibrin sealant offered a safe, minimally invasive approach that may avoid the morbidity occurring in open surgery (Sharma et al., 2005).

Gynecologic Pregnancy enhancement and embryo adherence issues have been addressed using fibrin sealant, which improves pregnancy rates in women at advanced reproductive age and

for whom in vitro fertilization attempts have repeatedly failed. A case-control study was reported to evaluate the possible contribution of the fibrin sealant to the embryo transfer with in vitro fertilization. All women who underwent the embryo transfer with the aid of fibrin sealant were compared to those who underwent standard embryo transfer. This method showed an increased embryo adherence to the endometrium (Bar-Hava et al., 1999).

The fibrin sealant was also used in 12 women with preterm premature rupture of the membranes at approximately 24 wk of gestation. The sealant was applied intracervically and there was a decrease of the amniotic liquid leakage (Sciscione et al., 2001).

Endoscopic colposuspension using fibrin sealant as a substitute for sutures was performed in 17 women suffering from stress urinary incontinence. The fibrin sealant was applied to both sides of the urethrovesical area and the urethrovesical junction was pressed for 5 min against the retropubic periost. The procedure lasted approximately 20 min and all patients were discharged on the first postoperative day. Fibrin sealant seems to be a promising new approach to correcting stress incontinence in women as shown by these experiments (Kiilhoma et al., 1995).

Orthopedic Treatment of Achilles tendon rupture has been widely discussed. Because of the tendency to a higher rate of re-ruptures and worsening of functional results following conventional treatment, some investigators from Germany evaluated the functional outcome after Achilles tendon ruptures treated with fibrin sealant to perform reactive force measurements and motion analysis in 30 patients. Patient comfort was increased with fibrin sealant through rapid healing for Achilles tendon ruptures, where postoperative leisure sports activities was similar to that prior to surgery (Ambacher et al., 2001).

Major orthopedic procedures can benefit from fibrin sealant due to an effective hemostasis to reduce blood loss and associated complications. Patients undergoing total hip replacement received either standard of care and fibrin glue (38 patients) or standard of care alone (43 patients) to evaluate the efficacy of the fibrin sealant. Blood loss was markedly reduced in the fibrin glue group by 197 ml or 23.5%, affirming that the fibrin glue was an effective hemostatic agent (Wang et al., 2003).

Total knee arthroplasty is associated with major postoperative blood loss of approximately 800 to 1200 ml, and blood transfusion is frequently required. The best procedure is to reduce blood loss during and after surgery. Levy et al. (1999) designed a study to evaluate the hemostatic efficacy of the use of fibrin sealant in patients with total knee arthroplasty. The patients were divided into two groups: a control group, in which the standard means of hemostasis were applied, and a treatment group, in which the standard means to control local bleeding were applied and a fibrin sealant was used before skin closure. The apparent postoperative blood loss was determined by measuring the volume in the suction-drain bottles and all blood transfusions were recorded. The mean apparent postoperative blood loss in the treatment group was 360 ml, compared with 878 ml in controls.. The decrease in the level of hemoglobin was 25 g/L in the treatment group, compared with 37 g/L in controls. Sixteen patients (55%) in the control group required a blood transfusion and only 5 (17%) in the treatment group. The use of fibrin sealant in this procedure was effective and safe, thus reducing blood loss and blood transfusion requirements (Levy et al., 1999).

Dental and Oral Surgery Most of the dental surgery literature described fibrin sealant use in the patients with hemophilia and other coagulopathies, but this has been expanded to the general patient population in recent years. In a study carried out on 80 patients with bleeding disorders that had undergone 135 dental extractions without previous hematologic therapy, the fibrin sealant was used as control to localized hemostasis. Secondary bleeding occurred in 9 of 12 patients with severe hemophilia when the concentration of aprotinine in fibrin sealant was 1,000 KIU/mL. When it was increased to 10,000 KIU/ml only 3 of 25 hemophilia patients presented secondary bleeding. None of the 43 patients with coagulopathies other than severe hemophilia suffered bleeding after extractions. The local use of fibrin sealant was a safe and cost-effective tool to treat patients with severe bleeding disorders (Rakocz et al., 1993).

Patients taking anticoagulants were divided into two groups to evaluate the fibrin sealant in prevention of post-extraction hemorrhage. A control group of 26 patients using suture and a study group of 20 patients using fibrin sealant presented no difference in the postoperative outcome as to

hemorrhage. Postoperative pain was reported more frequently in the control group and only one patient had significant postoperative bleeding, showing that the fibrin sealant was an effective agent to prevent post-extraction hemorrhage (Halfpenny et al., 2001).

Healing after oral cavity surgery may be problematic in some cases because it is a contaminated cavity. A study aimed at investigating the effect of fibrin sealant on healing after surgical procedures in the oral cavity of rats. The first molars were extracted with some cortical bone. The exposed cavities were filled with fibrin sealant after hemostasis in the study group and a suture was used in the control group. The rats were sacrificed after 2, 4, or 6 wk and a histologic analysis was performed. The healing process was better in the study group. Foreign-body reaction was lower in the study group (1/24 or 4.1%) than in the control group (6/18 or 33.3%). Abscess scores were better in the study (3/24 or 12.5%) than in the control group (10/18 or 55.5%).The last significant difference was in necrosis, and better results were obtained in the study (2/24 or 8.3%) than in the control group (10/18 or 55.5%).The use of fibrin sealant on wound healing in the oral cavity has a positive effect when compared with traditional suture techniques (Yucel et al., 2003).

Plastic and Reconstructive Surgery Plastic surgery is also a major user of fibrin sealants, and they are effective in hemostasis, grafts, and face-lift surgery. In order to demonstrate the applicability of the fibrin sealant in face-lift surgery, 48 patients were divided into two groups. The first 24 patients underwent face lifts without glue and the next 24 patients with the use of fibrin sealant. All face lifts used the same technique and drains were only used in those patients who did not receive the fibrin sealant. The amount of bruising and edema was compared in the two groups. The patients in whom the sealant was used had a significantly less bruising and swelling, with faster healing response. There were no bruises when the fibrin sealant was used but bruises were noted when the glue was not used (8.3%). Another benefit was that drains were not needed when fibrin glue was used. The operating times were shorter by 13.3 min with the use of fibrin sealant (Fezza et al., 2002).

In a prospective study of fibrin sealant in 20 patients who had undergone bilateral face-lift surgery, the fibrin sealant was applied just to one side. Total drainage was recorded on each side for 24 h before drains were removed. The side treated with the fibrin sealant had a median drainage of 10 ml and the control side 30 ml. The reduction in postoperative drainage could also reduce pain and bruising, increasing patient satisfaction with this procedure (Oliver et al., 2001).

Current surgical management of deep partial-thickness and full-thickness burn wounds involves early excision and grafting. Blood loss during these procedures may be large, thus prompting the use of topical hemostatic agents to control and minimize hemorrhage during grafting. A comparative clinical trial evaluated the use of fibrin sealant in burn patients undergoing skin graft procedures. Each patient served as his or her own control in this randomized, unblended study of the effect of hemostasis time at donor sites treated with the investigational fibrin sealant. A significant difference was demonstrated in the mean time to hemostasis between the fibrin sealant-treated donor sites compared painwise to the control sites. There were no adverse events associated with the use of the fibrin sealant. This investigation showed a significant decrease in the time to hemotasis at the donor skin harvest site (Nervi et al., 2001).

In a comparative study involving thermally injured patients, 34 patients received fibrin sealant while 61 did not. The proportion of total body surface areas burned was 10% in the study patients versus 10.9% in the control group. The fibrin sealant group did not receive packed red blood cell transfusions, albumin infusion, or topical bovine thrombin during excision and grafting procedures. The estimated blood loss/graft ratio was 0.46 ml/cm^2 for the study group and 0.56 ml/cm^2 for the control group, confirming that fibrin sealant reduced the need for blood transfusion (McGill et al., 1997).

Drug Delivery Fibrin sealant, as a result of its drug delivery capacity and biologic structure, can serve as an effective mechanism of delivering a variety of different medications. Determination of kinetics of antibiotics from fibrin clots showed that all antibiotics were almost completely released within 96 h. The delivered amount of each drug was sufficient to maintain the minimal inhibitory concentration (MIC) until d 4 of culture for most of antibiotics, thus resulting in a prolonged release of the drug (Greco et al., 1991).

Another in vitro study investigated the duration of the action and antibacterial effects of the fibrin sealant combined with antibiotics. The effect of fibrin sealant alone on bacterial growth was also examined. The addition of antibiotics to the fibrin sealant clots resulted in a continuous diffusion of antibiotics for up to 7 d. The antibacterial effects of fibrin sealant clots with antibiotics were significantly greater compared to without antibiotics. In addition, the presence of fibrin sealant clots resulted in a reduction in bacterial growth (Kram et al., 1991).

A combination of gentamicin, neomycin, and polymyxin E with fibrin sealant resulted in prolonged clotting time. The drug release from the clots was similar for all three antibiotics tested and mainly dependent on the concentration gradient between clot and its environment. Under the conditions of this study, about 85% of the antibiotic content of fibrin sealant clots was released within 72 h (Redl et al., 1983).

The fibrin sealant is composed of two separate solutions of fibrinogen and thrombin, and when mixed together, these solutions mimic the final stages of the clotting cascade to form a fibrin clot. The resulting fibrin patch is an effective medium for microbial growth, and the addition of antibiotics to one of these components of the fibrin sealant was shown to reduce postoperative infections. Seventeen different antibiotics were investigated in vitro. Of the 17, cefotaxime, mezlocillin, gentamicin, neomycin, and polymixin B, when added to fibrin sealant decreased the rate of microbial growth (Thompson & Davis, 1997).

A study was designed to assess a local drug delivery system of anticancer agent doxorubicin (DOX) using fibrin sealant as a drug carrier. In vitro release of the agent from fibrin sealant was examined by a dialysis method in the presence of sodium alginate. The fibrin sealant containing the anticancer agent and sodium alginate was applied on the surface of a tumor on the back of rats. The local application to the tumor resulted in an advantage of the site-specific delivery of the anticancer agent using fibrin sealant with sodium alginate as to the extent and duration of drug concentration in the tumor extracellular fluid (Kitazawa et al., 1997).

A drug delivery system for the anticancer agent cisplatin (cis-diamminedichloroplatinum, CDDP), a platinum-based chemotherapy drug, mixed with fibrin sealant was examined using a rat osteosarcoma model. This material was directly implanted into the tumors or subcutaneous tissue of rats, and the inhibitory effects on tumor growth and lung metastasis were evaluated. Data on in vitro kinetics of CDDP release by fibrin sealant showed the procedure to be effective, as evidenced by inhibition of tumor growth and metastasis; thus, this acts as a useful adjuvant to conventional chemotherapy (Miura et al., 1995).

Little is known about the role of neurotrophic factors in peripheral nerve regeneration following nerve injury. To investigate this, 48 rats underwent left sciatic nerve transaction and immediate repair. The fibrin sealant mixed with neutrophin-4 (NT-4) was injected around the nerve repair site and nerve regeneration was assessed both functionally and histomorphometrically. The results showed that the fibrin sealant mixed with NT-4 displayed a significant increase compared to the control in the regeneration distance in 5 d. The sciatic function index was significantly higher in the treated group from 40 to 60 d after nerve repair. Morphometric analysis revealed that nerves treated with the mixture showed a significant improvement in the number of regenerated axons, axonal diameter, and myelin thickness. These results suggest that the mixture of fibrin glue with NT-4 was a potent product improving rat sciatic nerve regeneration (Yin et al., 2001).

The effect of fibrin sealant and endothelial cell growth factor on the healing of defects in the avascular portion of canine menisci was investigated in 30 menisci of 15 adult mongrel dogs. The defects were treated in one of these three ways: Group 1, the defect was left empty; Group 2, the defect was filled with fibrin sealant; and Group 3, the defect was filled with fibrin sealant and endothelial cell growth factor. The healing process was evaluated macroscopically and histologically at intervals of 1, 2, 6, 12, and 24 wk. The average proportion of each defect that was filled with connective tissue was 5% in Group 1, 76.6% in Group 2, and 89.4% in Group 3. The combination of fibrin sealant and endothelial cell growth factor enhanced the revascularization and formation of granulation tissue, which accounted for the increase healing level in the avascular portion of the meniscus (Hashimoto et al., 1992).

In another study, rats received intraspinal implants of fibrin sealant containing neurotrophic agents into left dorsal quadrant cavities aspirated from lumbar enlargement. The transected L5 dorsal root stump was placed at the bottom of the lesion cavity and was secured between the fibrin sealant and spinal cord. The results indicated that neurotrophic agents enhanced dorsal root regeneration into spinal cord and that fibrin sealant was an effective medium for intraspinal delivery of neurotrophic agents (Iwaya et al., 1999).

CONCLUSIONS

The new fibrin sealant derived from snake venom and animal fibrinogen is a useful tool in medical practice due to its rapid, easy, and cheap production process, as well as the flexibility and diversity of its applications, especially in bleeding disorders. Fibrin sealant is a biodegradable biological product that does not (1) produce marked adverse reactions, (2) use human blood, (3) transmit infectious diseases, or (4) present toxicology risks during internal uses. Further, fibrin sealant (5) reduces surgical time, (6) improves postoperative recovery, (7) is highly adhesive, and (8) may be used as an adjuvant in conventional suture procedures and drug delivery.

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