The use of single-donor fibrin glue prepared by recycled cryoprecipitation in experimental liver surgery

BELA BALINT1,2,* , IBOLJA ČERNAK3 , MARIJANA PETAKOV2, DIANA BUGARSKI2, ŽIVORAD MALIĆEVIĆ3, SLAVKA MANDIĆ-RADIĆ1 and JOVAN TASESKI1

1 Institute of Transfusiology of MMA, Belgrade, Yugoslavia
2 Institute for Medical Research, Belgrade, Yugoslavia
3 Institute for Medical Research of MMA, Belgrade, Yugoslavia

Abstract—The purpose of the study was to evaluate the hemostatic effectiveness of fibrin glue (FG) prepared by a modification of cryoprecipitation technique in experimental rat liver surgery. FG component I was prepared by triple or ‘recycled’ cryoprecipitation method from single-donor plasma. Rats subjected to liver incision, partial and total lobectomy were treated with FG on the surgical cut surface or underwent standard surgical technique. The efficacy of FG-treatment was evaluated on the basis of the 24-hour survival ratio and peripheral blood hematological parameters. The mean values of fibrinogen, FXIII, fibronectin and horizontal tensile strength of FG were 54.2 ± 19.9 g/l, 13.5 ± 3.6 IU/ml, 3103.1 ± 148.9 mg/l, and 1.076 ± 0.18 N/cm², respectively. The survival of FG-treated rats subjected to partial and total lobectomy was significantly higher in comparison to the FG-nontreated animals, accompanied with higher values of red blood cell counts, hemoglobin concentration and hematocrit. When liver incision was performed, although there were no differences in survival rate, FG-treated animals had significantly higher values of the tested hematological parameters. The presented results demonstrated that by using ‘recycled’ cryoprecipitation it is possible to obtain high quality single-donor FG with successful hemostatic therapeutical effects, as confirmed in the experimental rat model of liver surgery.

Key words: Cryoprecipitation; fibrin glue; single-donor; rat.

INTRODUCTION

Fibrin glue (FG) is a two-component biological system with adhesive, sealing and topical hemostatic effects and it appears to promote angiogenesis and local tissue repair [1–5]. The FG component I consists of fibrinogen, FXIII, fibronectin, which
is activated by the addition of component 2 containing thrombin, ionized calcium and certain antifibrinolytic agent [1–5]. The effect of FG imitates the final step of coagulation cascade when fibrinogen is converted to fibrin by thrombin and subsequently in the presence of FXIII and ionized calcium fibrin polymerizes to a stable fibrin clot. FG has been used to promote therapeutic effects in a variety of clinical settings [6–12]. Recently, FG was also utilized as biocompatible delivery system for drugs and growth factors [13–17] and as a ‘matrix’ during ex vivo cultivation of endothelial cells [18].

Fibrin glue products are available as commercially prepared from pooled human blood, as well as prepared by precipitation from single-donor plasma, since the application of single-donor FG reduces the risk of virus transmission equal to the risk of a use of single blood/plasma unit [1–3]. Preparation of FG component 1 from single-donor plasma using different precipitation methods (cryoprecipitation, precipitation with ammonium sulfate or with polyethylene glycol) has been reported [19–24]. Cryoprecipitation is a simple and cost-effective procedure, but the main limitation of this method is variable and relatively low fibrinogen concentration in comparison to commercially FG products [1–3]. However, it has been demonstrated that using some modifications of basic cryoprecipitation, high quality single-donor FG component 1 can be prepared [25, 26].

In this study, we report a modification of the cryoprecipitation technique, a method named triple or ‘recycled cryoprecipitation’, for preparation of FG component 1 with acceptable fibrinogen yield. The final product quality control was evaluated through biochemical and biophysical assays. Furthermore, the effectiveness of the obtained FG was estimated in an experimental rat model of liver surgery. We hypothesized that the application of FG on surgical cut surface will result in superior local hemostasis and subsequently lower blood loss in rats, as compared to FG-untreated animals.

MATERIAL AND METHODS

FG preparation procedure

Blood was taken into CPDA-1 quadruple blood bags (Terumo Corporation, Tokyo, Japan) from 40 random blood donors, nonreactive to transmissive disease markers (hepatitis, AIDS and lues), with appropriate clinical and laboratory findings and no history of coagulopathies. CPDA-blood was centrifuged at 5080 g for 15 min at 4 ± 2°C (Hettich, Tuttlingen, Germany) and, upon separation, plasma (252 ± 38 ml in average) was frozen at −80 ± 5°C and stored for at least 12 hours. Plasma was thawed at 4 ± 2°C during around 16 hours, and centrifuged at 3890 g for 10 min at 4 ± 2°C. The cryoprecipitation procedure was repeated twice in a manner described above. After the third centrifugation, supernatant plasma was removed and the cryoprecipitate containing concentrated fibrinogen, FXIII and fibronectin (i.e. FG component 1) was stored at −30 ± 3°C till therapeutic application, when