

ORAL PRESENTATION

Preparation and Characterization of Autologous Fibrin Glue

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Author Block: G. Rock, D. Neurath, M. Harvey, M. Freedman, The Ottawa Hospital, Ottawa, ON, Canada.

Abstract Body:

Background: Cryoprecipitate is frequently combined with thrombin to produce a fibrin glue to enhance hemostasis during surgical procedures. Commercial preparations have a known fibrinogen composition but are expensive and usually provided in small volumes. Bovine thrombin has the disadvantage of causing immunological reactions in 15% of patients. We recently reported on the development of a computerized device which produces cryoprecipitate in 55 minutes. We have now modified the procedure to permit simultaneous production of an autologous thrombin. When used together, these products can produce fibrin glue entirely from autologous components.

Methods: 250 ml of plasma was processed in the automated Thermogenesis Cryoseal®. Cryoprecipitate was made within 50 minutes using the CP-3 chamber with two freeze-thaw cycles and recovered in 4x3 mL syringes. Thrombin was generated using the attached Thrombin Activation Device in which a solution of 66% ethyl alcohol and 25 mM calcium chloride and glass beads were used to produce thrombin and separate the bulk of the plasma proteins. The thrombin was harvested into 4x3 mL syringes and both components were frozen and stored separately at - 30°C. The products were assayed for fibrinogen, Factor VIII, von Willebrand Factor and thrombin activity using the Dade-Behring BCS coagulation system; total protein was determined by spectrophotometer. After combining the two products, the rate of clot initiation (R) and strength was measured using the thromboelastogram (TEG).

Results: Cryoprecipitate was produced with an average total fibrinogen of 122±13 mg or 22±7.7 g/L representing 20±2% recovery, Factor VIII was 91.2±48 U with a concentration of 14.2±3.3 U/mL, von Willebrand Factor was 123.4±53.1 U or 19.9±5.2 U/mL, and total protein was 0.44±0.11 g. The thrombin preparation contained a total of 447 U or 45.8±7.8 U/mL of thrombin. SDS gel electrophoresis showed 10 bands in the thrombin preparation with a major band at 400 kd. Bovine thrombin gave similar bands. The TEG curve obtained using the two autologous products was similar to that seen with standard cryoprecipitate and bovine thrombin (R = 18.7±7.1).

Conclusion: An autologous human thrombin can be made during automated cryoprecipitate production. This thrombin is in sufficient concentration to initiate clotting and crosslinking of cryoprecipitate for use as an entirely autologous fibrin glue.