CROSSEAL® Fibrin Sealant (Human)

WARNING

CROSSEAL® MUST NOT BE USED IN CONTACT WITH CSF OR DURA MATER

DESCRIPTION

Crosseal™, Fibrin Sealant (Human) is a single use kit consisting of two packages: (1) one containing one vial each of frozen sterile solutions of Biological Active Component (BAC) and Thrombin and (2) one containing a sterile spray application device. The two components are mixed and applied topically as described in the Dosage and Administration Section.

The BAC and Thrombin components appear as white to slightly yellowish opaque masses when frozen and as clear to slightly opalescent and colorless to slightly yellowish solutions when thawed. The components contain no preservatives.

BAC is a sterile solution, pH 6.9-7.3, consisting mainly of a concentrate of human fibrinogen. Fibrinogen is a protein from human blood that forms a clot when combined with thrombin.

The composition of the BAC solution is:

Active ingredient:
Concentrate of human fibrinogen (40-60 mg/ml)

Other Ingredients:
Tranexamic acid
Arginine hydrochloride
Glycine
Sodium chloride
Sodium citrate
Calcium chloride
Water for injection (WFI)

Thrombin is a sterile solution, pH 6.8-7.2, containing highly purified human thrombin and calcium chloride for activation of clotting of the final combined product. Thrombin is a highly specific protease that transforms the fibrinogen contained in BAC into fibrin. The composition of the Thrombin solution is:

Active Ingredients:
Human thrombin (800-1200 IU/ml)
Calcium chloride (5.6-6.2 mg/ml)

Other Ingredients:
Human albumin
Mannitol
Sodium acetate
Water for injection (WFI)

Cryoprecipitate, the starting material for BAC, and cryo-poor plasma, the starting material for the production of Thrombin are made from pooled human plasma obtained from US licensed plasma collection centers.

Each individual plasma unit obtained for production of Crosseal™ is tested by the supplier according to FDA regulations (21 CFR 640.60) and memoranda concerning testing of individual donation units for HBsAg, HIV-1/2 Ab, HIV p24Ag, HCV Ab.

In addition, the plasma is tested in mini-pools by OMRIX using a test procedure referred to as Nucleic Acid Testing (NAT) using Polymerase Chain Reaction (PCR) Technology and must be found negative for HAV, HBV, HCV, and HIV-1. While NAT for HCV is approved by FDA, investigational testing is still being performed with HIV-1, HAV and HBV to determine the effectiveness of NAT to detect low levels of viral material. The significance of a negative result for these viruses is unknown since the effectiveness of the test has not been established. NAT for Parvovirus B19 is also performed and the level of contamination is not permitted to exceed 10,000 copies/ml. This limit is applied to restrict the viral load of Parvovirus B19 in the starting plasma pool.

The cryoprecipitate is treated with aluminum hydroxide gel to adsorb the Vitamin K dependent clotting factors and then incubated with a solvent detergent (SD) mixture consisting of 1% tri-n-butyl phosphate and 1% Triton X-100 for inactivation of enveloped viruses (first virus inactivation step).

The solvent detergent (SD) reagents are removed by castor oil extraction and reverse phase chromatography (C-18 column) and the preparation is subsequently treated by pasteurization (second virus inactivation step).

Prior to pasteurization, sucrose (1.8 g/g column filtrate) and glycine (0.11 g/g) are added as stabilizers and the mixture is warmed to 37 °C under stirring. The pH is adjusted to 6.8 – 7.4. The solution is heated to 60 ± 0.5 °C and maintained at that temperature for 10 hours.

After pasteurization, the stabilizers used for heat treatment are removed by diafiltration and the product is concentrated by ultrafiltration. For final formulation, tranexamic acid is added to the BAC as a stabilizer prior to sterile filtration. The filtered solution is then filled aseptically in 1 ml, 2 ml or 5 ml aliquots, frozen at ≤ - 60 °C and then stored at -30 ± 5 °C until distribution.

Cryo-poor plasma, the starting material for the production of Thrombin, is applied to an anion exchange column for binding of prothrombin and activation into thrombin. The resultant thrombin does not bind to the column and is eluted with calcium chloride.

Thrombin undergoes SD treatment for 6-6.5 hours at 26 ± 1°C.

The SD reagents are removed by cation exchange chromatography. Mannitol (as a 15% solution), and human albumin are added to the product as stabilizers to a final concentration of 2% (w/w) and 0.2% (w/w), respectively. The stabilized solution is passed through a nanofiltration module, which is the second viral clearance step.

The filtrate is formulated with calcium chloride to 40 mM and the concentration of human albumin is adjusted to 0.6%.

The Thrombin bulk solution is sterile filtered and aseptically filled in 1 ml, 2 ml or 5 ml aliquots, frozen at
≤ - 60 °C and then stored at -30 ± 5° C.

Although the BAC and Thrombin components undergo two distinct independent virus inactivation/removal steps shown to be capable of significant viral reduction, no procedure has been shown to be completely effective in removing viral infectivity from derivatives of human plasma (see Clinical Pharmacology and Warnings).

CLINICAL PHARMACOLOGY

Thrombin is a highly specific protease that transforms the fibrinogen contained in BAC into fibrin. Thrombin is partly adsorbed by the fibrin so formed. Excess thrombin, if any, is inactivated by protease inhibitors in the blood. In a study to evaluate the blood levels of ^{125}I thrombin and ^{3}H tranexamic acid after application of Crosseal™ to hepatic wounds in rabbits, it was determined that excess thrombin is quickly complexed with anti-thrombin and its total elimination and inactivation is rapid. The study results verify that systemic exposure to thrombin is approximately equivalent to that generated by a minor hemorrhage and poses no increased risk of thromboembolism**.** BAC is stabilized with tranexamic acid. With regard to tranexamic acid, the study shows that it was absorbed very quickly with total elimination from plasma in all animals within ten hours of treatment. The study results show that both tranexamic acid and thrombin are absorbed systemically when administered directly to a hepatic wound, but the systemic exposure is minimal.

Intracerebral application of Crosseal™ to rabbits at a dose equivalent to the human dose resulted in neurological symptoms including, but not limited to, trembling, involuntary head movements, and clonic contractions. Doses of 4- to 33-fold greater than the human dose resulted in additional severe neurological symptoms [including tachypnea, hypertony, hyperextension (neck), hyperexcitability, and convulsions] and death. These symptoms were not observed in the absence of tranexamic acid.

VIRAL CLEARANCE

The manufacturing procedure for Crosseal™ includes processing steps designed to reduce the risk of viral transmission. In particular, both BAC and Thrombin undergo two discrete virus inactivation/removal steps, which can be summarized as follows:

Step	Component	
	BAC	**Thrombin**
1	Solvent detergent treatment (1%TNBP, 1% Triton X-100) for 4 hours at 30°C	Solvent detergent treatment (1%TNBP, 1% Triton X-100) for 6 hours at 26°C
2	Pasteurization (10 hours at 60°C)	Nanofiltration

The efficacy of these procedures in inactivating a range of viruses has been assessed. The viruses used for validation studies were selected to give a range of physico-chemical characteristics and are summarized in the following table:

Virus	Family	Genus	Genome	Envelope	Size (nm)	Resistance
HIV-1	Retro	Lentivirus	RNA	Yes	80-130	Low
Sindbis	Toga	Alphavirus	RNA	Yes	60-70	Low
BVDV	Flavi	Pestivirus	RNA	Yes	60-70	Low
PRV	Herpes	Varicelloviridae	DNA	Yes	150-200	Medium
EMCV	Picorna	Cardiovirus	RNA	No	28-30	Medium
HAV	Picorna	Hepatovirus	RNA	No	28-30	High
Canine Parvovirus	Parvo	Parvovirus	DNA	No	18-26	Very high

- Human Immunodeficiency Virus 1 (HIV-1) is recommended in the CPMP "Note for Guidance on Plasma Derived medicinal Products" (CPMP/BWP/269/95) to be used directly in validation studies, it also serves as a model for HIV-2.
- Sindbis is an RNA negative strand enveloped virus and is a model for Hepatitis C virus. It is not possible to use Hepatitis C virus itself, as it cannot be propagated. This model virus has been employed in many cases to demonstrate the efficacy of solvents and non-ionic detergents e.g. TnBP/Triton X-100 to break down the envelope structure of the virus.
- Bovine Viral Diarrhea Virus (BVDV) is an RNA positive strand enveloped virus of the *Flaviviridae* family, *Pestivirus* genus. HCV, a hepacivirus, is also a member of this family. For this reason BVDV is regarded as a good model for HCV. Virions are 60-70nm in diameter.
- Pseudorabies (PRV) is a Herpes virus that can serve as a model for human Herpes viruses which may be found in blood (e.g. EBV, CMV and HHV6) as all Herpes viruses have a similar structure and are morphologically indistinguishable. Herpes viruses are large viruses between 150-200nm. Pseudorabies can also serve as a model for other DNA enveloped viruses. Pseudorabies is particularly suitable for viral spiking studies as it can be obtained in high titres (approx. 9 logs) and an accurate quantitative plaque assay is available.
- Encephalomyocarditis virus (EMCV) is a small (28-30nm) non-enveloped RNA virus, which is relatively resistant to physico-chemical treatments. EMCV is a Picornavirus and can be considered a model for Hepatitis A virus, which is also a Picornavirus and is the same size.
- Hepatitis A Virus (HAV) is a small non-enveloped virus that may be transmitted by blood products. It is therefore considered to be a relevant virus.
- Canine Parvovirus is a small (18-26nm) non-enveloped DNA positive strand virus, which is resistant to the actions of chemical treatments. It can be considered a model for the human Parvovirus B19 on the basis of morphology and structure. This model virus has been used as a negative control for the Solvent/Detergent steps and to determine the ability of the other steps in the process to eliminate non-enveloped virus.

The results of viral removal/inactivation validation studies are summarized in the following tables:

a) BAC

Virus	HIV-1	BVDV	PRV	EMCV	HAV	CPV
	Reduction factor (log_{10})					
SD Treatment	>4.42	>4.39	>3.96	Not Done	Not Done	0.0
Pasteurization	>4.39	>5.46	Not Done	3.69	2.66	1.33
Global Reduction Factor	**>8.81**	**>9.85**	**>3.96**	**3.69**	**2.66**	**1.33**

b) Thrombin

Virus	HIV-1	SBV	BVDV	PRV	EMCV	HAV	CPV
	Reduction factor (log_{10})						
SD Treatment	>5.82	>5.31	>4.74	>4.25	Not Done	Not Done	0.0
Nanofiltration	>4.36	>5.32	Not Done	>5.47	6.37	6.95	5.85
Global Reduction Factor	**>10.18**	**>10.63**	**>4.74**	**>9.72**	**6.37**	**6.95**	**5.85**

The above validated process steps are designed to reduce the risk of viral transmission when used in conjunction with plasma screening. In addition to the screening of source plasma as previously described, each manufacturing pool is tested for HBsAg, HIV-1/2 Ab, HCV Ab, and for NAT of HCV. However, this pool testing is of a lower sensitivity than the individual unit testing.

In clinical studies of Crosseal™ involving 216 patients, there have been no proven reports of seroconversion. In addition, over 7,000 patients have been treated with Crosseal™ since non-US marketing was initiated in 1997 and there have been no spontaneous reports of seroconversion.

CLINICAL STUDIES

Crosseal™ was evaluated in a pivotal Phase III single-blind, randomized, parallel-group, multi-center study against FDA-approved control topical hemostats in 121 patients undergoing liver resection at 15 centers. Patients were randomized (stratified by surgeon) at the conclusion of the liver resection surgery if general oozing was present that could not be controlled by further surgical methods and a topical hemostat was needed to control the bleeding from the liver surface. For the primary endpoint, time to hemostasis, Crosseal™ was shown to be statistically significantly superior to the control hemostatic agents **(p=0.011 one-sided).**
Center effects are to be expected in multicenter studies, particularly in surgical indications. Data from one center, which used a specific control agent, made a major contribution to this result. However, of the sixteen surgeons who treated more than one patient in this study, ten found the time to hemostasis to be equivalent to, or shorter than that achieved with the specific control agent used.

Primary Endpoint: Time To Reach Hemostasis	
Crosseal™	FDA-Approved Control Topical Hemostats
5.3 minutes	7.7 minutes
Intent-to-treat analysis; one sided: p = 0.011	

INDICATIONS AND USAGE

Crosseal™ is indicated as an adjunct to hemostasis in patients undergoing liver surgery, when control of bleeding by conventional surgical techniques, including suture, ligature and cautery is ineffective or impractical. Crosseal™ is not indicated for the treatment of massive and brisk arterial bleeding.

CONTRAINDICATIONS

Crosseal™ is contraindicated in individuals known to have anaphylactic or severe systemic reaction to human blood products. The physician must weigh the potential benefit of treatment with Crosseal™ against the potential for hypersensitivity reactions.

Do not inject Crosseal™ directly into the circulatory system or tissue.

Crosseal™ must not be used in surgical operations where contact with the CSF or dura mater could occur.

WARNINGS

Because this product is made from human plasma, it may carry a risk of transmitting infectious agents, e.g., viruses, and theoretically, the Creutzfeldt-Jakob disease (CJD) agent. The risk of transmitting an infectious agent has been reduced by screening plasma donors for prior exposure to certain viruses, by testing for the presence of certain current virus infections, and by inactivating and removing certain viruses (see Clinical Pharmacology). Despite these measures, such products can still potentially transmit disease. There is also the possibility that unknown infectious agents may be present in such products. All infections thought by a physician to have been possibly transmitted by this product should be reported by the physician or other healthcare provider to the American Red Cross at 1-800-293-5023. The physician should discuss the risks and benefits of this product with the patient.

PRECAUTIONS

General

Crosseal™ must only be administered topically.

Information to Patients

Some viruses such as hepatitis A virus and parvovirus B19 are particularly difficult to remove or inactivate. Parvovirus B19 most seriously affects pregnant women, or immune-compromised individuals. Symptoms of parvovirus B19 infection include fever, drowsiness, chills, and runny nose followed about two weeks later by a rash and joint pain. Evidence of hepatitis A may include several days to weeks of poor appetite, tiredness and low-grade fever followed by nausea, vomiting and abdominal pain. Dark urine and a yellowed complexion are also common symptoms. Patients should be encouraged to consult their physician if such symptoms appear.

Carcinogenesis, Mutagenesis, Impairment of Fertility

Long-term animal studies have not been performed to evaluate the carcinogenic potential of Crosseal™ due to the human origin of both thrombin and fibrinogen contents. The effect of Crosseal™ on fertility has not been evaluated.

Studies performed in bacteria to determine mutagenicity were negative for Thrombin alone, BAC (containing fibrinogen, citrate, glycine, tranexamic acid, and arginine hydrochloride), TnBP alone, and Triton X-100 alone at all concentrations tested. All concentrations of the combination of TnBP and Triton X-100 also tested negative in assays performed to determine mammalian cell mutagenicity, chromosomal aberrations and micronuclei induction.

Pregnancy Category C

Adequate and well-controlled studies in pregnant women have not been performed. Crosseal™ should be used in pregnancy only if the potential benefit to the pregnant woman justifies the potential risk to the fetus. Studies to evaluate the potential reproductive/developmental toxicity of Crosseal™ have not been performed due to the human origin of both the thrombin and fibrinogen components.

The IV injection of the combination of TnBP and Triton X-100 into pregnant rats at doses up to approximately 600-fold (TnBP, 900 µg/kg/day) and 3000-fold (Triton X-100, 4500 µg/kg/day) the human dose, resulted in increased post-implantation loss and in an increased number of late resorptions. No embryo-fetal adverse effects were observed at doses up to 200-fold (TnBP, 300 µg/kg/day) and 1000-fold (Triton X-100, 1500 µg/kg/day) the human dose. Pregnant rabbits IV injected with the combination of TnBP at doses approximately 300-fold (TnBP, 450 µg/kg/day) and 1500-fold (Triton X-100, 2250 µg/kg/day) the human dose had increased resorption rates, decreased fetal body weights, and an increased number of runts. No embryo-fetal adverse effects were observed at doses up to 100-fold (TnBP, 150 µg/kg/day) and 500-fold (Triton X-100, 750 µg/kg/day) the human dose.

Pediatric Use

Of the 216 patients treated in adequate and well-controlled studies of Crosseal™ in liver surgery, eight were pediatric patients. Of these, five were less than 2 years old and three were between 2 and 12 years old. An additional 92 patients under the age of 18 have received Crosseal™ during liver surgery in the UK. Use of Crosseal™ in pediatric patients is supported by these data and by extrapolation of findings for safety and efficacy in adults.

Geriatric Use

Of the total number of subjects in clinical trials of Crosseal™ in liver surgery, 24 were over 65 years of age. Although no overall differences in safety or effectiveness were observed between the elderly and younger patients, greater susceptibility of some older patients to adverse reactions cannot be ruled out.

ADVERSE REACTIONS

As with any plasma derivative, anaphylactic reactions may occur in rare cases. No adverse events of this type were reported during the conduct of the clinical trials.

Mild reactions can be managed with anti-histamines. Severe hypotensive reactions require immediate intervention using current principles of shock therapy.

INTERACTIONS

None known.

DOSAGE AND ADMINISTRATION

FOR TOPICAL USE ONLY- DO NOT INJECT.

Crosseal™ should be sprayed or dripped onto the tissue in short bursts (0.1 - 0.2 ml) to produce a thin, even layer. If the hemostatic effect is not complete, a second layer should be applied. The amount of Crosseal™ required depends upon the area of tissue to be treated and the method of application. As an approximate guide, if a layer of 1 mm thickness is produced by spraying Crosseal™, the surface areas that can be covered by each of the kit sizes are given in the following table:

Crosseal™ Package Size	Area of Coverage with Layer of 1 mm Thickness
1.0 mL	20 cm^2
2.0 mL	40 cm^2
5.0 mL	100 cm^2

Standard surgical techniques for hemorrhagic control, including suture, ligature and cautery, should be used prior to the application of Crosseal™. Excess blood should be removed from the site of application if possible, although a dry field is not essential, and Crosseal™ should then be applied with the application device supplied. This device allows for the simultaneous application of equal amounts of the two components and ensures mixing, which is essential for the sealant to achieve optimal efficacy. Crosseal™ forms a transparent layer on application through which specific bleeding points may be observed, in which case these may be sutured or electrocauterized through the layer of Crosseal™.

Instructions for Use

1. Thawing of BAC and Thrombin solutions.

   Thaw the BAC and Thrombin solutions in one of the following ways:
   - 2°C to 8°C (refrigerator) vials thaw within 1 day; or
   - 20°C to 25°C (room temperature) vials thaw within 1 hour.

2. Preparation

The circulating nurse should open the package and transfer the BAC and Thrombin to the sterile field. The scrub nurse should prepare the assembly immediately prior to use, as follows:

a) Draw the contents of the two vials into the two sterile syringes (see diagram enclosed in the application device package).

b) Both syringes should be filled with equal volumes and should not contain air bubbles.

c) Caution should be taken when twisting vials off. It should be a gentle maneuver to ensure valve engagement.

3. Method of Application by Dripping

Keeping the tip of the applicator as close to the tissue surface as possible, but without touching the tissue during application, apply individual drops to the area to be treated. The drops should be allowed to separate from each other and from the tip of the applicator. If the applicator tip becomes blocked, the catheter tip can be cut back in 0.5 cm increments.

4. Method of Application by Spraying (2ml and 5ml kits only)
   a) Connect the short air tube on the application device to the male luer-lock end of the long air tube.
   b) Connect the female luer-lock of the air tube (with the 0.2 µm filter) to an air regulator capable of delivering between 35-45 psi of pressure.
   c) The air regulator should be used in accordance with the manufacturer's instructions.
   d) An air pressure of 35-45 psi (measured by air flow) should be used for spraying.

e) The distance between the nozzle and the tissue surface should ideally be between 10 and 15 cm during spraying.

HOW SUPPLIED

Crosseal™, is supplied as a kit consisting of two separate packages: (1) a package containing one vial each of BAC (40-60 mg/ml fibrinogen) and Thrombin (800-1200 IU/ml human thrombin, 5.6-6.2 mg/ml calcium chloride) frozen solutions and (2) a spray application device. The kit is supplied as 1 ml, 2 ml or 5 ml dosages.

The frozen solutions consist of a white to slightly yellowish opaque mass. When thawed the solutions are clear to slightly opalescent and colorless to slightly yellowish.

The application device package contains an applicator and an air tube with a 0.2µm filter. The application devices are sterile and for single use only.

Special Handling and Storage Conditions

- **Long Term Storage**
  Store frozen vials at -18°C or colder.
- **Short Term Storage**
  Unopened vials can be stored at 2°C to 8°C for up to 30 days.

Both components of Crosseal™ have been shown to be stable for up to 24 hours at room temperature.

The device package should be stored separately at room temperature.

Do not use after the expiration date stated on the box, or after 30 days if stored at 2°C to 8°C after thawing.

Do not re-freeze BAC or Thrombin once they are thawed.

Do not mix BAC or Thrombin with any other product.

Discard if the packaging of any of the components of Crosseal™ is damaged.

Manufactured by:
OMRIX Biopharmaceuticals, Ltd.
MDA blood bank,
Sheba Hospital, Ramat-Gan
POB 888, Kiryat Ono 55000
ISRAEL

Manufactured for:
(logo)American Red Cross
Blood Services
Washington, DC 20006 USA

U.S. License No. 1603

Issued March 2003