

## PRODUCT INFORMATION

### Horseradish Peroxidase Labeled Lectins

**Catalog Number:** H-2001-1

**Description:** Pure *Ricinus communis* - I lectin (RCA-I) from castor bean, Horseradish Peroxidase conjugated.

**Lot Number:**

**Protein Concentration:** 1 mg purified RCA-I Horseradish Peroxidase / 1 ml Buffer.  
(Based on OD280)

**Carbohydrate Specificity:**  $\beta$ -D-Galactopyranoside.

**Inhibitory Carbohydrate:** Lactose >> Galactose.

**Activity:** Less than 1  $\mu$ g/ml will agglutinate human type O erythrocytes.

**Buffer:** 0.01M Phosphate - 0.15M NaCl, pH 7.2 - 7.4.

**Chemical Used for Conjugation:** Horseradish Peroxidase.

**Storage:** Store liquid material frozen in aliquots in amber vials or covered with foil. Avoid freeze thaw cycles. Clarify by centrifugation. No preservatives have been added. Sodium azide will inactivate the enzyme, peroxidase.

**Stability:** The liquid material is stable for at least one year when stored frozen in aliquots.

**Caution:** Refer to the enclosed MSDS for information regarding lectins. The aluminum seals have sharp edges and the vial itself may have cracks which can cause lacerations. Use caution when opening the vial.

**RCA-I is a toxic protein. Refer to MSDS before opening vial !! Do not allow to dry. Use 10% bleach for all clean-up. Soak all glassware in bleach after use. Wear mask and gloves when handling. For Research and in vitro use only. E Y is not liable for any other unauthorized uses**

**Procedure for Use:** See reverse side.

**Remarks:** Lactose is added to prevent denaturing of the protein during shipping. Dialyze or pass through a desalting gel prior to use.

**References:**

1. Saltvedt, E. (1976) Biochim. Biophys. Acta. **451** : 536-548.
2. Olsnes, S., et al. (1974) J. Biol. Chem. **249** : 803-810.
3. Wu, A. M., et al. (1982). Abstracts, Society Complex Carbohydrates, No. 76.
4. Battacharyya, L. and Brewer, C. F. (1988) Arch. Biochem. Biophys. **262** : 605-608.
5. Olsnes, S. and Pihl, A. (1973) Biochemistry. **12** : 3121-3126.

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## PRODUCT INFORMATION

### Horseradish Peroxidase Enzyme Activity Assay

**Chemical Principle:** Peroxidase +  $H_2O_2 \rightarrow$  Complex  
Complex +  $AH_2$  (donor)  $\rightarrow$  Peroxidase +  $H_2O$  + A (colored)

**Assay Reagents:** BUFFER: 0.01M Sodium phosphate, pH 6.0.  
ENZYME: Dilute with Buffer. Acceptable dilution: 1-2  $\mu$ g/ml.  
DYE: 1% o-dianisidine in methanol prepared fresh daily. Store in amber bottle or wrapped in foil.  
SUBSTRATE: Prepare 0.3%  $H_2O_2$  solution in deionized or distilled water from stock  $H_2O_2$  solution Prior to use dilute to a final concentration 0.003% in Buffer.

**Procedure:**

1. Add 0.05 ml of DYE to 6.0 ml of SUBSTRATE. Add 2.9 ml to Reaction test tube and 2.9 ml to Control test tube.
2. At time=0, add 100 $\mu$ l of diluted ENZYME to Reaction tube and 100 $\mu$ l PBS to Control tube. Mix thoroughly.
3. Measure and record optical density at 460nm (OD460) every 15 seconds for 3 minutes, or take the end point reading after 3 minutes by stopping the reaction with 100 $\mu$ l of concentrated  $NaN_3$ .
4. Use this value to determine the rate of change in absorbance per minute.

**Enzyme Activity Calculations:** One unit of peroxidase activity is that amount of enzyme decomposing 1  $\mu$ mole of peroxide/minute at 25°C.  $11.3 \times 10^3 \text{ cm}^{-1}$  is the molar absorbance of  $H_2O_2$ .

$$OD460 / \text{min} = \frac{OD460 / 3\text{min} - OD \text{ Control} / 3\text{minutes}}{3\text{minutes}}$$

$$\text{mg enzyme} / \text{ml reaction mixture} = \frac{[\text{enzyme dilution}]}{30}$$

$$\text{units} / \text{mg} = \frac{OD460 / \text{min}}{11.3 \times \text{mg enzyme} / \text{ml reaction mixture}}$$

**Caution:** Due to inhibitory sugar present in the conjugates solution, to dilute the Conjugate 50-100 times with buffer before assay.

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# "TOXIC" MATERIAL SAFETY DATA SHEET

Effective Date: March 31, 2006

Revision 6

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## PRODUCT IDENTIFICATION

Name: Ricin Alpha Chain and Ricin Beta Chain. Crude and purified Ricinus communis, Viscum album, Abrus precatorius. Also, these proteins conjugated to fluorescein isothiocyanate, tetramethylrhodamine isothiocyanate, Texas Red®, D-Biotin, Horseradish Peroxidase, Alkaline Phosphatase, and Ferritin.

Catalog Number (s): AAL-2001, AL-2001, AL-2003, BA-2001, BA-2002, BA-4001, BA-5401, BAL-2001, F-2001, F-2002, F-4001, F-5401, FAL-2001, GP-2001, GP-2002, GP-4001, GP-5401, H-2001, H-2002, H-4001, H-5401, HAL-2001, I-2001, I-2002, I-4001, I-5401, L-2000, L-2001, L-2002, L-4000, L-4001, L-5400, L-5401, LA-2001, LA-2002, LA-4001, LA-5401, MB-2001, PB-2001, R-2001, R-2002, R-4001, R-5401, RAL-2001, T-2001, T-2002, T-4001, T-5401, TAL-2001, A-2001, A-2002, A-4001, A-5401, AK-2001, AK-2002, AK-4001, AK-5401, LGK-001, LGK-001A, LGK-002, LGK-002A, LGK-003, RAC-1, RBC-1.

Formula: Complex polypeptides.

Synonyms: RCA-I, RCA-II, RCA120, RCA60, castor bean lectin, ricin, ricin toxin, VAA, mistletoe lectin, viscumin, APA, abrin, jequirity bean lectin. These proteins may be coupled to various fluorescent dyes, enzymes, ferritin, or D-biotin. CAS Number: 1393-62-0 (for Abrus precatorius).

## EMERGENCY INFORMATION

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**EMERGENCY PHONE:**  
**650-342-3296**

## HAZARDOUS COMPONENTS

Specific protein (s) as listed on the vial label. These proteins are always supplied in solution, never in dried form. The concentration of the specific protein in solution generally will be greater than 0.1% (w/v, 1mg/1ml) and less than 1% (w/v, 10mg/ml) unless otherwise indicated on the vial label and the product information sheet. These proteins are toxins which function by inhibiting protein synthesis. Extreme caution should be used when handling these products. The fluorescent molecules fluorescein and tetramethylrhodamine isothiocyanate, as well as Texas Red® are possible carcinogens. Although the complete properties of these molecules have not been fully investigated, similar compounds are known to be chemically reactive with proteins and other biomolecules. Ferritin may be tumorigenic and should not be allowed to come in contact with skin. Alkaline Phosphatase, Horseradish Peroxidase, and D-Biotin may be harmful if ingested, inhaled, or allowed to contact the skin. Some individuals may exhibit a stronger adverse reaction to these chemicals than other individuals. All of the above toxins and related products should be handled only by qualified personnel and should be used in a facility isolated from the common work area. Sodium azide is included in most solutions as a preservative at a concentration of 0.02-1%.

## HEALTH HAZARD INFORMATION

EXPOSURE LIMITS: No exposure limits have been established, however, no exposure is considered safe. Ricin (RCA-II, RCA60) has been tested for toxicity. The LD50 (iv) in mice is 2µg/100 grams body weight. The other toxins are less potent but specific toxicity levels have not been determined.

EFFECTS OF OVEREXPOSURE: Any contact with these toxins may result in nausea, cramps, fatigue, and other flu-like symptoms. Chronic or high dose exposure to the toxins may result in serious illness or death. These toxins function by inhibiting protein synthesis. Any living tissue may be affected by these toxins.

ROUTES OF EXPOSURE: Contact with skin and mucous membranes are the primary routes of exposure. Care should be taken to avoid the formation of aerosols when handling any of the solutions. The use of glass equipment is discouraged since sharp edges may puncture protective equipment and directly inject toxic material.

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TOXIC MSDS for Crude and Purified Proteins and Enzymes Continued - page 2 of 2.

## PHYSICAL CHARACTERISTICS

APPEARANCE: Liquids will be clear to dark purple.  
SOLUBILITY: All liquids are completely miscible in water and buffers.

## FIRE AND EXPLOSION HAZARDS

Not considered to be a fire hazard. High heat completely will destroy any toxic activity of the proteins.  
EXTINGUISHING MEDIA: Water spray or CO<sub>2</sub>.  
SPECIAL FIRE FIGHTING PRECAUTIONS: As required for the surrounding fire.

NOTE: Most solutions contain 0.02-1% sodium azide as a preservative. Azide may react with lead and copper plumbing to form explosive metal azides. Flush with copious amounts of water when disposing material in the sink.

## REACTIVITY DATA

STABILITY: Stable. Decomposition products are not known to be hazardous.  
HAZARDOUS POLYMERIZATION: Will NOT occur.  
INCOMPATIBILITY: None known. (Lead and copper may react with sodium azide).

## SPILL / LEAK PROCEDURES

MATERIAL RELEASE / SPILL: Avoid contact with liquid. Wear a self-contained breathing apparatus and a full protective suit when cleaning up large spills. Flush contaminated area with a 0.5-5% solution of hypochlorite (household bleach diluted 1:5 with water is sufficient. Clean up spill with a paper towel soaked in household bleach. Do not allow solutions to dry on environmental surfaces. Wash affected area with detergent after the area has been treated with bleach.

WASTE DISPOSAL: Incinerate, autoclave, or dispose of paper waste in accordance with all Local, State, and Federal regulations. All laboratory equipment and supplies that contact these toxins must be soaked in bleach for at least 1 hour or autoclaved. All of these proteins are fully biodegradable. These proteins are generally not considered environmental hazards once treated with bleach or autoclaved.

## EMERGENCY FIRST AID PROCEDURES

May be harmful if swallowed, inhaled, or allowed to absorb through the skin. Wash contacted area with soap and water for 15 minutes. If inhaled remove to fresh air. Report exposure to the appropriate safety official. Consult a physician immediately.

## SPECIAL HANDLING PRECAUTIONS

VENTILATION: Appropriate ventilation to prevent the formation and dispersion of aerosols is required. When possible these materials should be handled in a designated biohazard work area that is segregated from common laboratory work areas.

EYE PROTECTION: Required. Goggles or Safety glasses with side shields are recommended.

RESPIRATORY PROTECTION: Required. An approved respirator designed for use with toxic liquids is necessary to prevent inhalation of aerosols.

PROTECTIVE GLOVES: Required when handling any of these materials.

ADDITIONAL: A full length laboratory gown with sleeves is required to prevent contact of any spilled liquids with clothing or exposed skin.

## SPECIAL PRECAUTIONS

This material is for research and experimental application only. It is not intended for food, drug, household, agricultural, or cosmetic use. All materials should be handled only by technically qualified individuals experienced with working with potentially hazardous chemicals. The above information is correct to the best of our knowledge. The user should make independent decisions regarding completeness of the information, based on all sources available. EY Laboratories, Inc. shall not be held liable for any damage resulting from handling or contact with the above product.

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