

**PRODUCT INFORMATION**  
**Affinity Purified Antibodies and Their Conjugates**

EY Laboratories' **AF** series antibodies are prepared from antisera by using affinity chromatography. The finished products may contain a low percentage of denatured protein due to unfavorable conditions during elution of the antibodies from the affinity column. Some cross reactivity studies have been done, contact Technical Service to request specific information.

Affinity purified antibodies and their conjugates are specifically prepared for laboratories who are involved with basic or diagnostic research. Purified antibodies give the highest possible ratio of the conjugate/antibody. This is an important factor for quantitative analysis of antigens. EY Laboratories' conjugated antibodies are designed for use in immunodiffusion, immunoelectrophoresis, fluorescent microscopy, light and electron microscopy.

The technology used in preparing the antigen specific and affinity purified antibody minimizes interference from other complex forming components in the antisera. Bovine serum albumin is used to coat vials containing affinity purified antibodies or their conjugates. This is to prevent loss of the antibody through adherence to the glass surface.

**Catalog Number:** HAF-531-1

**Description:** Horseradish Peroxidase Conjugated Rabbit Affinity Purified Antibody to Equine IgG, 1mL

**Lot Number:**

**Expiration Date:** 1 year from date of manufacture

**Protein Concentration:**  
(Based on OD280)

**Chemical Used for Conjugation:**  
(where applicable) **Horseradish Peroxidase**

**Buffer:** 0.01M Phosphate - 0.15M NaCl, pH 7.2-7.4. 0.05% sodium azide is added as a preservative EXCEPT for peroxidase conjugates and alkaline phosphatase conjugates.

**Storage:** Store liquid frozen in aliquots EXCEPT for Ferritin and Alkaline Phosphatase conjugates which must be refrigerated, not frozen. Alkaline Phosphatase conjugates contain up to 50% glycerol.

**Stability:** The liquid material is stable for several years when stored in aliquots with 0.05% sodium azide added as a preservative.

NOTE: DO NOT add sodium azide to peroxidase conjugates.  
Usage: Dilute 1% BSA in PBS at least 100 x before use.

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

**For Research and Laboratory Use Only.**

**EY LABORATORIES, INC.**  
107 North Amphlett Blvd.  
San Mateo, CA 94401

Tel: 650-342-3296  
Fax: 650-342-2648  
Orders: 1-800-821-0044  
(Outside CA only)

**Product Information**  
**Horseradish Peroxidase Enzyme Activity Assay**

**Chemical Principle:** Peroxidase + H<sub>2</sub>O<sub>2</sub> → Complex  
Complex + AH<sub>2</sub> (donor) → Peroxidase + H<sub>2</sub>O + A (colored)

**Assay Reagents:** BUFFER: 0.01M Sodium phosphate, pH 6.0.

ENZYME: Dilute with Buffer. Acceptable dilution: 1-2 µg/ml.

DYE: 1% o-dianisidine in methanol prepared fresh daily. Store in amber bottle or wrapped in foil.

SUBSTRATE: Prepare 0.3% H<sub>2</sub>O<sub>2</sub> solution in deionized or distilled water from stock H<sub>2</sub>O<sub>2</sub> 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 100µl of diluted ENZYME to Reaction tube and 100µl 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 100µl of concentrated NaN<sub>3</sub>.
  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 µmole of peroxide/minute at 25°C. 11.3 x 10<sup>3</sup> cm<sup>-1</sup> is the molar absorbance of H<sub>2</sub>O<sub>2</sub>.

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

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

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

**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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**MATERIAL SAFETY DATA SHEET**

Effective Date: March 31, 2006

Revision 4

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**PRODUCT IDENTIFICATION**

Name: Purified proteins or biotin labeled with Horseradish Peroxidase or Alkaline Phosphatase.  
 Catalog: HP-02, BA-104, BA-105, BA-108, BA-109, H-1102 to H-9000, LA-1104 to LA-9000,  
 Number (s): PA-2100 to PA-2701, AA-2100 to AA-2701, HAF-001 to HAF-2354, AAF-001 to AAF-2354, HA-01 to HA-013, AA-01 to AA-013, HAL-1104 to HAL-4701, AAL-1104 to AAL-4701.  
 Synonyms: Protein A, Avidin (egg white), Biotin, Lectins, Secondary Antibodies labeled with Horseradish Peroxidase or Alkaline Phosphatase.

**EMERGENCY INFORMATION**

EY Laboratories, Inc.  
 107 North Amphlett Blvd.  
 San Mateo, CA 94401

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

**HAZARDOUS COMPONENTS**

Specific protein(s) as listed on the vial label. Solutions are at a concentration generally greater than 0.5mg protein/ ml. Biological activity of these labeled proteins will vary. Horseradish Peroxidase and Alkaline Phosphatase are both potent enzymes which may be harmful if ingested, inhaled, or allowed to absorb through the skin. Both enzymes are known to cause allergic responses in sensitive individuals.

**HEALTH HAZARD INFORMATION**

EXPOSURE LIMITS: None established. The toxicological properties of these products have not been thoroughly investigated. Care should be taken when handling any of these materials.

EFFECTS OF OVEREXPOSURE: May causes localized eye, skin, or mucous membrane irritation. Some sensitive individuals may develop a chronic allergic reaction with exposure.  
 ROUTES OF EXPOSURE: Inhalation of powders and skin contact with liquids are the primary routes of exposure. Care should be taken to avoid the formation of aerosols when handling any of the solutions.

**PHYSICAL CHARACTERISTICS**

APPEARANCE: Powders are a light brown. Solutions will be light to dark brown.

SOLUBILITY: Powders are completely soluble in many biological buffers and water. All liquids are completely miscible in water and biological buffers.

**FIRE AND EXPLOSION HAZARDS**

Not considered to be a fire hazard.

EXTINGUISHING MEDIA: Water spray or CO<sub>2</sub>.

SPECIAL FIRE FIGHTING: None required.

**PRECAUTIONS:**

NOTE: Alkaline Phosphatase conjugates contain less than 0.05% 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. The nature of any decomposition products are not known. They are not believed 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 powder or liquid. 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. Due to the small quantities of material involved these products are generally not considered to be environmental hazards. All of these proteins are fully biodegradable.

**EMERGENCY FIRST AID PROCEDURES**

May be harmful if swallowed, inhaled, or allowed to absorb through the skin. Wash contacted area with water for 15 minutes. If inhaled remove to fresh air. Report exposure to the appropriate safety official. Consult a physician if irritation occurs or if there is any indication of an allergic response, such as watering eyes, sneezing, or difficulty breathing. Any eye contact should be reported to a physician immediately

**SPECIAL HANDLING PRECAUTIONS**

VENTILATION: No special ventilation is required but it is recommended to handle these reagents in a fume hood when possible.  
 EYE PROTECTION: Required. Goggles or safety glasses with a side shield are recommended.

RESPIRATORY PROTECTION: Recommended as a safety precaution, specifically when working with powders. An approved respirator may be required for those individuals already known to be sensitive to these materials.

PROTECTIVE GLOVES: Required when handling any of these materials.

**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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