

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: RAF-512-1

Description: TRITC Conjugated Rabbit Affinity Purified Antibody to Bovine IgG, Fc Specific Antibody, 1mL

Lot Number:

Expiration Date: 1 year from date of manufacture

Protein Concentration: OD550/280=
(Based on OD280)

Chemical Used for Conjugation: TRITC
(where applicable)

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

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

General Procedure Fluorescent Labeled Antibody

The following is a general Procedure and Trouble-Shooting Guide. The information is provided only for your convenience. The success of your experiments are not guaranteed by EY Laboratories, Inc.

Tissue Sections

1. Wash and block tissue section. Do not use serum products, they contain glycoproteins which may lead to high levels of non specific background. After blocking, rinse briefly with Buffer.
2. Dilute **Fluorescent Labeled Antibody** to desired concentration 20-100 µg/ml using Buffer.
3. Incubate tissue section with Fluorescent Labeled Antibody for 30 minutes in a moist chamber.
4. Wash tissue section with Buffer three times.
5. Examine tissue section with Fluorescent microscope. Use appropriate filter.
Ref. M. Imbar et. al., (1973). Intl. Journal of Cancer, **12**, 93-99

Cell Suspension

1. Wash cells with Buffer.
2. Collect cells by centrifugation.
3. Dilute **Fluorescent Labeled Antibody** to 100 µg/ml using Buffer.
4. Incubate approximately 1x10⁶ cells with 1 ml diluted Fluorescent labeled Antibody for 15 minutes at room temperature or in a 37°C water bath.
5. Wash cells with Buffer three times using centrifugation.
6. Examine cells, with or without fixation with Fluorescent microscope. Use appropriate filter.
Ref. K. Phiss. (1977). Experimental Pathology, **14**, S15

Fluorochromes must be protected from light. Perform incubation, when practical, in a dark room or covered in foil.

Absorption and Emission

| | Absorption/Excitation Rate | Emission Max. |
|------------|----------------------------|---------------|
| FITC | 492 nm | 517 nm |
| TRITC | 554 nm | 570 nm |
| Texas Red™ | 596 nm | 615 nm |

TROUBLE SHOOTING GUIDE

| Problem | Cause | Solution |
|-----------------------------|------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Weak or no Staining | 1. Low concentration of antibody conjugate. 2. Insufficient incubation time. 3. Photobleaching | Causes #1 - #2 a. Increase incubation time. b. Increase concentration conjugate. a. Avoid exposure to light. |
| High Background | 1. Antibody conjugate is too concentrated. 2. Insufficient washing. 3. Autofluorescent sample. | a. Decrease concentration of Antibody conjugate. b. Shorten incubation times. a. Perform multiple washings and prolong washing time. a. Use fluorochrome with different excitation and emission spectrum. b. Use a different antibody conjugate (enzyme or colloidal gold). |
| Unexpected Staining Pattern | Multiple causes | a. Perform control reactions. b. Use other cytochemical technique to prove or disprove the findings. |

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MATERIAL SAFETY DATA SHEET

Effective Date: March 31, 2006

Revision 4

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PRODUCT IDENTIFICATION

Name: Purified proteins labeled with fluorescein isothiocyanate (FITC), tetramethylrhodamine isothiocyanate (TRITC), or Texas Red a trademark of Molecular Probes for the sulfonyl chloride derivative of sulforhodamine 101

Catalog Number (s): FP-01, RP-01, TP-01, F-1102 to F-9000, R-1102 to R-9000, T-1102 to T-9000, FA-2100 to FA-2701, RA-2100 to RA-2701, TA-2100 to TA-2701, FAF-001 to FAF-2354, RAF-001 to RAF-2354, TAF-001 to TAF-2354, FAL-1104 to FAL-4701, RAL-1104 to RAL-4701, TAL-1104 to TAL-4701, FA-01 to FA-013, TA-01 to TA-013, DM1011F to DM1064F, FNP-01 to FNP-05, BA-101, BA-102, BA-612.

Synonyms: Protein A, Avidin (egg white), Glycosylated Bovine Serum Albumin, Lectins, Secondary and Monoclonal Antibodies labeled with FITC, TRITC, or Texas Red®

EMERGENCY INFORMATION

EY Laboratories, Inc.
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**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. FITC, TRITC, and Texas Red® are possible carcinogens in their pure form. Compounds with similar chemical structures are known to be reactive with proteins and other biomolecules. The complete properties of the dyes after labeling have not been evaluated. These compounds should be treated as potentially hazardous. All solutions contain less than 0.05% sodium azide as a preservative.

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: Causes localized eye, skin, or mucous membrane irritation. Some sensitive individuals may develop a chronic allergic reaction with exposure. The known effects are due to the protein. No specific effects of the bound dye are known at this time.

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 orange. Solutions will be yellow to dark purple.

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. At high concentrations the chemicals may emit toxic fumes. Such high concentrations are not normally found in a research laboratory.

EXTINGUISHING MEDIA: Dry chemical powder or CO₂.

SPECIAL FIRE FIGHTING PRECAUTIONS: Wear self-contained breathing apparatus and protective clothing to prevent contact with skin and eyes.

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MSDS for Fluorescent Labeled Proteins Continued - page 2 of 2.

NOTE: Most solutions 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. Decomposition products are not known to be hazardous.

HAZARDOUS POLYMERIZATION: Will NOT occur.

INCOMPATIBILITY: Alcohols, strong bases and acids, strong oxidizing agents, and heat. (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.

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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