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-532-1	
Description:	TRITC Conjugated Rabbit Affinity Purified Antibody to Equine IgG, Fc	
Lot Number:	Specific, 1mL	
Expiration Date:	1 year from date of manufacture	
Protein Concentration: (Based on OD280)	OD550/280=	
Chemical Used for Conjugation: (where applicable)	TRITC	
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	
Storage:	added as a preservative EXCEPT for peroxidase conjugates and alkaline phosphatase conjugates. 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. Use: Dilute 1% BSA in PBS at least 100 x before use.	
Caution:	Reference 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.	
XIX		

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.
- Examine tissue section with Fluorescent microscope. Use appropriate filter. Ref. M. Immbar et. al., (1973). Intnl. 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.
- 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.
- 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	 Low concentration of antibody conjugate. Insufficient incubation time. 	Causes #1 - #2 a. Increase incubation time. b. Increase concentration conjugate.
Ũ	3. Photobleaching	a. Avoid exposure to light.
High Background	 Antibody conjugate is too concentrated. Insufficient washing. 	 a. Decrease concentration of Antibody conjugate. b. Shorten incubation times. a. Perform multiple washings and prolong washing time.
	3. Autofluorescent sample.	 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.



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

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MATERIAL SAFETY DATA SHEET

Effective Date: March 31, 2006 Revision 4 Page 1 of 2

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
 FP-01, RP-01, TP-01, F-1102 to F-9000, R-1102 to R-9000, T-1102 to T-9000, R-1102 to F-9000, R-1102 to F-9000, T-1102 to F-9010, TAF-001 to FAF-2354, RAF-001 to RA-2701, TA-2100 to TAF-2354, FAL-1104 to FAL-4701, RAL-1104 to RAL-4701, TAL-1104 to TAL-4701, RAL-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. 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. 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	Causes localized eye, skin, or mucous membrane irritation. Some sensitive
OVEREXPOSURE:	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.
	nationing 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 the head of the concentrations the chemicals may emit toxic fumes. Such high concentrations are per manally found in a research laboratory.

EXTINGUISHING MEDIA: SPECIAL FIRE FIGHTING PRECAUTIONS:

Dry chemical powder or CO₂. Wear self-contained breathing apparatus and protective clothing to prevent contact with skin and eyes.

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) 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:

HAZARDOUS POLYMERIZATION: INCOMPATIBILITY: Stable. Decomposition products are not known to be hazardous. Will NOT occur. Alcohols, strong bases and acids, strong oxidizing agents, and heat. (Lead and copper may react with sodium azide).

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

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

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.

hazards. All of these proteins are fully biodegradable.

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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