

PRODUCT INSERT**REF****FluoroQuench™ Stain/Quench Reagent**

Catalog #s FQAE100, FQAE100, FQAE500, FQAE2000

FluoroQuench™ Plus Stain/Quench Reagent

Catalog #s FQZAE100, FQZAE500, FQZAE2000

For General Laboratory Use.**INTENDED USE**

FluoroQuench™ products are used to fluorescently stain and fix lymphocytes in the microcytotoxicity assay.

SUMMARY AND EXPLANATION

FluoroQuench™ products include a mixture of reagents to facilitate the microscopic evaluation cells and to extend storage of trays for later evaluation.

PRINCIPLE(S)

In HLA testing, a quenching reagent can significantly reduce background fluorescence. EDTA is used to stop complement-dependent cell lysis. Ethidium Bromide (EB) is used to stain dead cells. Acridine Orange (AO) is used to stain live cells. The FluoroQuench Plus™ contains an additional fixative used to prevent EB from entering live cells, thus preserving the live cells for extended reading.

REAGENTS**A. Identification**Common or Established Name: FluoroQuench™ and FluoroQuench™ Plus; catalog IDs are listed above.Primary Ingredients: Bovine Hemoglobin, Ethylenediaminetetraacetic acid (EDTA), Ethidium Bromide (EB), Acridine Orange (AO), plus several proprietary reagents.**B. Warning or Caution**

1. Refer to the Material Safety Data Sheet for detailed information.
2. Caution: To avoid contamination, do not return unused product to stock bottle.

C. Instructions for Reconstitution or Preparation for Use

Shake well before use.

**D. Storage Instructions**

Store reagents at temperature indicated on package. Use before printed expiration date.

E. Purification or Treatment Required for Use

None.

F. Instability Indications

Do not use if precipitate is observed. Red fluorescent clumps are an indication of bacterial contamination.

INSTRUMENT REQUIREMENTS

No special instruments are required.

SPECIMEN COLLECTION AND PREPARATION

For the microcytotoxicity assay, refer to appropriate tissue typing tray product insert. For cell isolation procedures, see product inserts for One Lambda cell isolation products, such as FluoroBeads® and LymphoKwik®.

PROCEDURE

A. Materials Provided

FluoroQuench™ or FluoroQuench™ Plus Stain/Quench Reagent

B. Materials Required, But Not Provided

5 µl dispenser

Cover slides for tray. Recommended: Insta-Seal™ Cover Slides (OLI Cat. ID#TIS250U) or FluoroSeal™ Cover Slides (OLI Cat. ID#TF250U)

C. Directions for Use

Important: Mix well before use. To avoid contamination, do not return unused FQAE to stock bottle.

1. Use your current methods to isolate and label your test samples.
2. After complement incubation, dispense 5µl of FluoroQuench “gently” on top of mineral oil into each well.
3. Place Terasaki Insta-Seal™ (OLI Cat. #TIS250U) cover slide or Terasaki Fluoro-Seal™ (OLI Cat. #TF250U) cover slide on the tray.
4. Let trays stand at 20 - 25° C for approximately 15 minutes before microscopic evaluation.
5. For FluoroQuench™: Trays can be stored at 2 - 5° C for 2-3 days in the dark
For FluoroQuench™ Plus: Trays can be stored at 2 - 5° C for 7 - 10 days in the dark

RESULTS

See “Specific Performance Characteristics” below.

LIMITATIONS OF THE PROCEDURE

Cell isolation difficulties and contamination of the lymphocyte preparation with red cells, yeast, monocytes, platelets, or granulocytes may cause erroneous results. Additionally, erroneous results may occur when cell concentrations are above or below the acceptable level.

EXPECTED VALUES

Not applicable.

SPECIFIC PERFORMANCE CHARACTERISTICS

Nonspecific fluorescent background will be reduced, and complement-dependent cell lysis will be stopped. Dead cells will fluoresce red while viable cells will fluoresce green.

BIBLIOGRAPHY

Dombrausky, Monahan, “Comparison of 10% Hemoglobin/5% EDTA Quench vs. FluoroQuench”. HLA Technical Workshop, 1994.

REVISION HISTORY

Revision	Date	Revision Description
2	2007/09	REAGENTS, B.1.2, delete warnings no longer applicable to the product. Update EC REP address.

EC REP EUROPEAN AUTHORIZED REPRESENTATIVE

MDSS GmbH, [Schiffgraben 41, D-30175](#), Hannover, Germany

