DELFIA® Buffers Guide

DELFIA is a heterogeneous time-resolved fluorometric assay method in which an enhancement step assures high sensitivity and wide response range. To achieve the best results in a DELFIA assay, the optimal buffer composition should be chosen. A number of ready-to-use buffer products are available as catalog items, or users may prepare their own formulations based on the following guidelines.

Assay Buffers for DELFIA

The use of a Tris-based buffer is recommended. Phosphate buffers can be used with N1-chelates, but lower signals might be gained compared to Tris-based buffers. For storage purposes, phosphate buffers must not be used due to their chelating nature.

To avoid non-specific binding the buffer should contain a blocking agent such as bovine serum albumin (BSA). There are many different grades of BSA and some of these contain a considerable amount of heavy metals that will eventually show as high levels of background in the assay. Using purified BSA is highly recommended; alternatively a high grade of casein or ovalbumin can be used to block the non-specific binding.

A detergent such as Tween 20 or Tween 40 is also needed in the buffer to further prevent the non-specific binding to the plate.

To keep the fluorescence background low as well as maintain good precision the assay buffer should contain low concentrations of a chelator such as diethylenetriaminepentaacetic acid (DTPA) or ethylenediaminetetraacetic acid (EDTA). It is, however, essential to remember that the presence of too much chelator in the assay buffer will eventually start competing for the lanthanide chelate and will destroy the assay. As a general rule, no more than 50 µmol/L of chelator should be used in the assay buffer when working with compounds labeled with N1-chelate.

An example of an assay buffer composition for a DELFIA assay could be 50 mM Tris-HCl, pH 7.5-8, containing 0.9% NaCl, 0.2-0.5% of purified BSA, 0.01-0.1% Tween (20 or 40) and 20 µM EDTA.

Hybridization Assays

Hybridization assays require additional sodium chloride in the buffer. To achieve successful hybridization, a standard assay buffer (as described above) can be used, but should be supplemented with 0.5-1 M NaCl. On the other hand, with sticky oligos, additional reagent to prevent non-specific binding is needed. Reagents such as polyacrylic acid (up to 1 mg/mL) or polyvinylpyrrolidone, PVP (0.05-0.2%, MW approximately 360,000 g/mol) are recommended in this case.

Catalog items:

Cat. No.	Product	Description	Size
1244-106	DELFIA Assay Buffer. Ready to use.	Tris-HCl buffered (pH 7.8) salt solution with BSA, bovine globulin, Tween 40, an inert red dye, and < 0.1% sodium azide as preservative.	50 mL
1244-111	DELFIA Assay Buffer. Ready to use.		250 mL
4002-0010	DELFIA Assay Buffer. Ready to use.		1000 mL
4006-0010	Hybridization Buffer. Ready to use.	Tris-HCl buffered solution (pH 7.8), containing 1 mol/L NaCl, < 0.1% sodium azide, BSA, bovine gamma globulins, Tween 20, DTPA and an inert red dye.	50 mL
CR86-100	Research Buffer Set, 2X concentrate.	BSA and detergent in separate vials.	250 mL
CR85-100	DELFIA Assay Buffer w/o detergents, 5X concentrate.	Contains BSA and bovine gamma globulins.	250 mL
CR84-100	Stabilizer	7.5% BSA solution (purified).	50 mL

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DELFIA Assay Buffer
We recommend our ready-to-use DELFIA Assay Buffers (Prod. No. 1244-106, 1244-111 and 4002-0010) which are optimized for DELFIA binding assays to keep the fluorescence background low and prevent non-specific binding to the plate.

Hybridization Buffer
The Hybridization Buffer (Prod. No. 4006-0010) is a buffered protein and detergent solution with high salt concentration. It is intended for use as a diluent for Eu/Sm/Tb-labeled oligonucleotide probes in hybridization reactions.

Research Buffer Set
Research Buffer Set (Prod. No. CR86-100) is ideal if the assay has some restrictions on the buffer. It contains basic buffer concentrate that can be modified by the user by adding assay specific blocking proteins or detergents. Bovine serum albumin (BSA) and detergent are included in separate vials.

DELFIA Assay Buffer Without Detergents
DELFIA Assay Buffer without detergents (Prod. No. CR85-100) is five times concentrated and intended for use in assays where detergents may interfere.

Stabilizer
Stabilizer (Prod. No. CR84-100) is a BSA purified from trace amounts of heavy metals for use in coating of plates and as a blocking agent in assay buffer. The Stabilizer can be used also in the storage buffer of lanthanide labeled reagents.


DELFIA Eu-GTP Binding Kit

Wash Buffers for DELFIA

To enable the high sensitivity of the DELFIA assays, automated plate washers should be used, with typically 4-6 wash cycles prior to addition of the Enhancement Solution. To avoid dissociation of the lanthanide during washes, neutral buffered solutions like Tris-HCl (pH 7.5-8) with detergents are recommended.

Catalog items:

Cat. No.	Product	Description	Size
1244-114	DELFIA Wash concentrate	25-fold concentration of Tris-HCl buffered (pH 7.8) salt	250 mL
4010-0010	DELFIA Wash concentrate	solution with Tween 20. Contains Germall II as preservative.	1000 mL

DELFIA Wash Concentrate
DELFIA Wash Concentrate is supplied at 25-fold concentration and should be diluted in water prior to use.

Storage Buffer for DELFIA Lanthanide Labeled Compounds

Labeled proteins and peptides should be stored at a high concentration and in the absence of chelators or competing metals in the buffer. Do not store diluted reagents. In most cases, 50 mmol/L Tris-HCl buffered saline solution (pH 7.5-8.0) containing 0.1-0.5% purified BSA will ensure the stability of the labeled compound during storage. If the labeled protein requires storage at +4°C, it is advisable to add a bacteriostatic agent such as sodium azide (NaN_3) at concentration of 0.05-0.1%. Neither DELFIA Assay Buffer (Prod. No. 1244-106, 1244-111, 4002-0010) nor phosphate buffers are suitable for storage of labeled proteins or peptides. If during storage the background level of the assay tends to increase due to aggregation formation, the labeled compound should be filtered through a 0.2 µm membrane.

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