



Technology: HTRF®

Manual

Protein-Protein Interaction

HTRF Anti-VHH-Eu

Part Numbers	610VHHKLA	610VHHKLB
Test Size	5,000 tests	20,000 tests

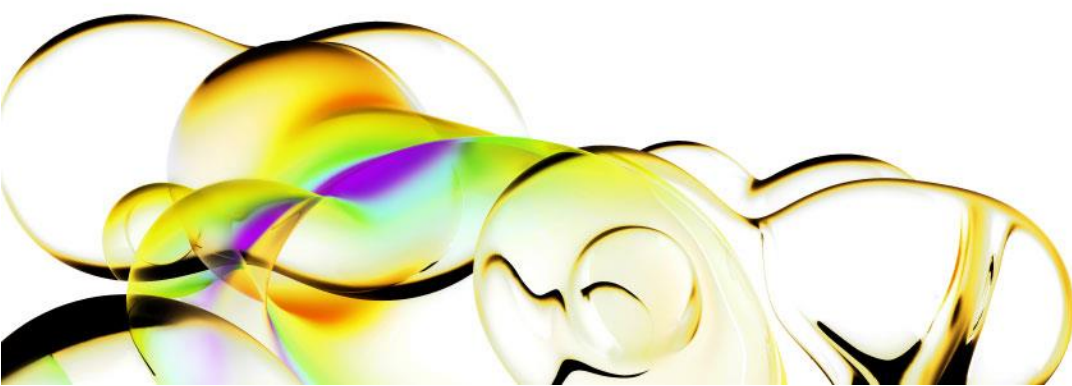
Storage : 2-8°C

Assay volume : 20 µL

Version: 01

Revision date: June 2023

For research only. Not for use in diagnostic procedures.



REAGENT DESCRIPTION

In an HTRF protein:protein interaction assay, one protein is labeled (directly or indirectly) with the donor, and the other protein is labeled (directly or indirectly) with the acceptor. When the two proteins interact, the donor molecule is brought into proximity with the acceptor molecule. Excitation of the donor will result in signal generation proportional to the binding of proteins.

Monoclonal Anti-VHH antibody was labeled with Eu³⁺ cryptate. Anti-VHH antibody binds to the native form of both alpaca and llama VHH and recombinant nanobodies. The binding is rapid and stable, making it an ideal choice for use in a variety of assays.

MATERIALS

REAGENT	5,000 TESTS	20,000 TESTS
HTRF Anti-VHH-Eu cryptate Frozen in phosphate buffer pH 7.0 containing protease free bovine serum albumin	1 vial - 0,25 mL	1 vial - 1 mL

REVVITY REAGENTS NOT PROVIDED	PART#
PPI - Europium detection buffer	Cat # 61DB9RDF* *200 mL - ready-to-use
Plates - HTRF 96-well low volume plate	66PL96001

For HTRF microplate recommendations, and for a list of HTRF® - compatible readers and set-up recommendations, please visit our website.

For reading, an HTRF® - compatible reader is needed. Make sure to use the appropriate set-up.

STORAGE AND STABILITY

- Store the reagent at 2-8°C.
- Under appropriate storage conditions, reagents are stable until the expiry date indicated on the label.
- Once thawed, stock solutions are stable for two days at 2-8°C. They can be refrozen (at ≤ -16°C) and thawed once only. Do not repeat freezing and thawing.

ASSAY FORMAT

When used as suggested, one vial from the two available sizes will provide sufficient reagent for 5,000 and 20,000 tests respectively, using a 20 µL final assay volume.

	VOLUME
Other assay components	10 µL
Acceptor (d2 or XL665) conjugate	5 µL
Donor (Eu or Tb Cryptate) conjugate	5 µL
Final volume*	20 µL

*Assay volumes can be adjusted proportionally to run the assay in 96- or 1536-well microplates.

REAGENT HANDLING

Buffers

Revvity PPI - Europium detection buffer Cat # 61DB9RDF* has been optimized for maximum performance.

It is mandatory to use the same buffer to prepare the donor and the acceptor (d2 or XL665) conjugates.

When using specific in-house buffers for the preparation of working solutions, we recommend a basic buffer such as PBS (Phosphate Buffered Saline) or Hepes with a pH maintained between 5.5 and 8.5.

It can be supplemented with BSA (0.1%), and detergents such as Tween 20, Triton X100 or CHAPS (up to 0.5%) to prevent reagent coating. Avoid SDS, due to its denaturing effect on XL665.

Please note that a phosphate-free buffer must be used in biochemical kinase assays to prevent interferences pertaining to the binding of anti-phospho specific antibodies.

Use of Europium toolbox conjugates requires a final KF concentration between 100 mM and 400 mM.

Conjugates

Allow each vial of frozen conjugate to warm up at room temperature. Reconstitute the lyophilizate following the instructions below:

Mix gently.

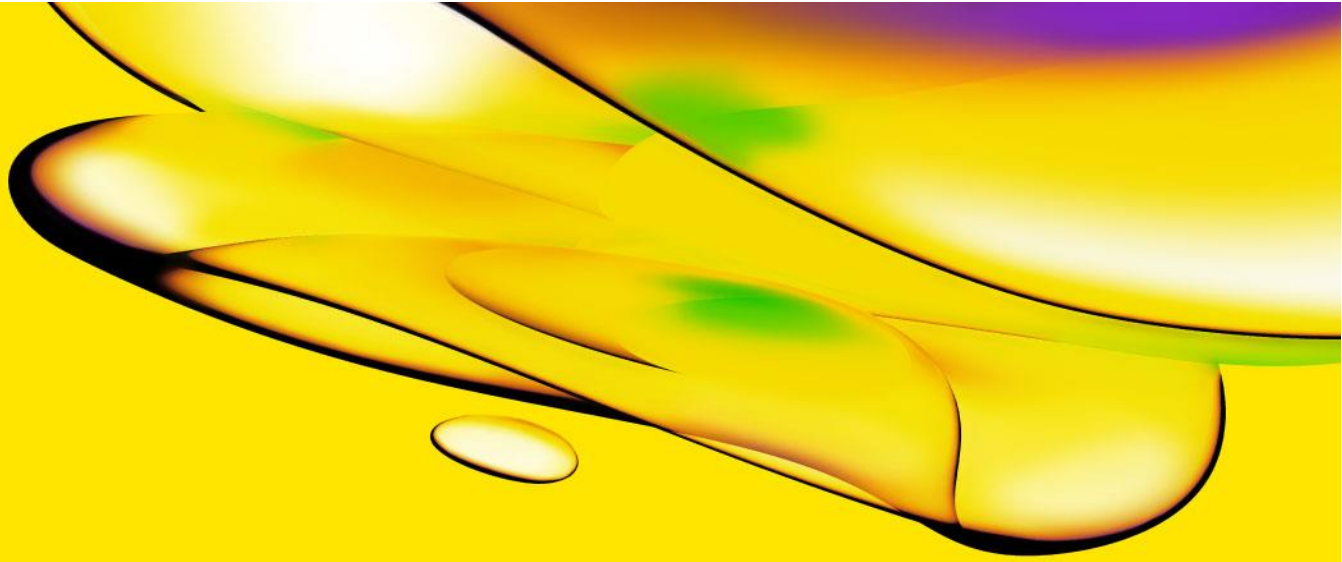
HTRF Anti-VHH-Eu CRYPTATE	STOCK SOLUTION PREPARATION	WORKING SOLUTION PREPARATON (SEE ASSAY FORMAT ABOVE)
5,000 tests	0,25 mL Mix gently	Dilute 100-fold the stock souldion in PPI detection buffer Mix gently E.g. Add 24.75 mL PPI detection buffer to 0.25 mL of stock solution
20,000 tests	1 mL Mix gently	Dilute 100-fold the stock souldion in PPI detection buffer Mix gently E.g. Add 99 mL PPI detection buffer to 1 mL of stock solution

Additional info is included in the batch information provided with the reagent.

The optimal amount per well will be dependent on assay conditions. For additional information on assay optimization, please refer to the technical note: Guidelines for optimizing protein : protein interaction assays using HTRF PPI reagents.

Make sure to prepare stock and working solutions according to the instructions that correspond to the packaging you have purchased (number of tests).

This product contains material of biological origin. Use for research purposes only. Do not use in humans or for diagnostic purposes. The purchaser assumes all risk and responsibility concerning reception, handling and storage.



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