Certificate of Analysis LanthaScreen® TR-FRET PPAR Delta Competitive Binding Assay Kit, 1 Kit



Part Number: PV4893 Lot Number: 2082216

Immediate Storage: Refer to Component's Storage Temperature.

Shipping Conditions: dry ice

5781 Van Allen Way Carlsbad, CA 92008 Phone: 760.603.7200 www.thermofisher.com

Components:

Description	Composition	Quantity	Part Number	Lot Number
PPAR delta - LBD (GST)	Recombinant Human Peroxisome Proliferator Activated Receptor delta ligand binding domain in 50 mM Tris–HCl (pH 8.0), 150 mM KCl, 0.5 mM EDTA, 0.03% NP–40, 5 mM DTT and 20% Glycerol.	10 μg	PV4693*	2022384C
Fluormone™ Pan-PPAR Green	2 μM in 95% ethanol/water.	200 μL	PV4896	1906504D
LanthaScreen® Tb-anti-GST (Goat) Antibody	10 mM HEPES 137 mM NaCl, 2.7 mM KCl (HEPES buffered saline).	25 µg	PV3550*	2068723B
TR-FRET PPAR Assay Buffer	Proprietary Buffer (pH 7.5).	2 × 25 mL	PV4895	2044018A
DTT Solution	1M DTT in water.	1 mL	P2325	2033225E

^{*}See individual COA

Storage and Handling: The performance of this product is guaranteed for 6 months from the date of purchase if stored and handled properly.

Description	Storage and Handling	
PPAR delta - LBD (GST)	If properly stored in its original container at −80°C, this product is guaranteed for 6 months from date of purchase. Mix gently, do not vortex.	
Fluormone™ Pan-PPAR Green	-80°C. Vortex prior to use. Note: To completely remove Fluormone™ from the glass insert tube, a 10 µL pipette tip with a very narrow long tip, such as Rainin GPSL10S, is required.	
LanthaScreen® Tb-anti-GST (Goat) Antibody	If properly stored at -20°C, this product is guaranteed for 6 months from date of purchase.	
TR-FRET PPAR Assay Buffer	Store at 4°C. Thaw upon receipt.	
DTT Solution	–20°C	

QUALITY ASSURANCE

Functional Testing:

Kit components are functionally tested for performance in the assay.

 $\label{lambda} LanthaScreen^{\circledR} \mbox{ is a registered trademark of Life Technologies Corporation.} \\ Invitrogen^{\intercal M} \mbox{ is a trademark of Life Technologies Corporation.}$

Certificate of Analysis PPAR delta - LBD (GST), 10 µg



Peroxisome Proliferator Activated Receptor delta Ligand Binding Domain, GST-tagged

Part Number: PV4693 Lot Number: 2022384C Immediate Storage: -80°C Shipping Conditions: dry ice

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Description:

Recombinant human Peroxisome Proliferator Activated Receptor delta ligand binding domain (amino acids 165-442; NP_006229), GST-tagged, expressed in E. *coli* cells.

Concentration:

 $0.51\ mg/mL$ total protein as measured using the Bradford protein assay with BSA as a standard.

Calculated 8,510 nM

Assay Concentration(s):

2 nM PPAR delta - LBD (GST) is the final concentration recommended for the LanthaScreen® TR-FRET PPAR delta Competitive Binding Assay (PV4893).

5 nM PPAR delta - LBD (GST) is the final concentration recommended for the LanthaScreen® TR-FRET PPAR delta Coactivator Assay (PV4685).

Aliases:

PPARD, NR1C2

Storage and Handling:

If properly stored in its original container at -80° C, this product is guaranteed for 6 months from date of purchase. **Mix gently, do not vortex.**

Do NOT make single use aliquots.

Storage Buffer:

50 mM Tris-HCl (pH 8.0), 150 mM KCl, 0.5 mM EDTA, 0.03% NP-40, 5 mM DTT and 20% Glycerol.

Note: Effective April 1, 2008 the storage buffer for this protein was modified to contain 0.03% NP-40 as a stabilizing agent. The TR-FRET assay performance is unaffected.

QUALITY ASSURANCE

Functional Testing:

The performance of each lot of PPAR delta - LBD (GST) is confirmed in the following assay(s):

LanthaScreen® TR-FRET PPAR delta Competitive Binding Assay (PV4893). The ligand GW0742 was shown to competitively displace Fluormone™ Pan-PPAR Green (PV4896) from PPAR delta - LBD (GST).

LanthaScreen® TR-FRET PPAR delta Coactivator Assay (PV4685). The ligand GW501516 was shown to recruit Fluorescein-C33 coactivator peptide (PV4606) HVEMHPLLMGLLMESQWGA to bind with PPAR delta - LBD (GST).

Gel Information for PPAR delta - LBD (GST)

Page Description: The SDS-PAGE and/or Native PAGE were run on 4-20% Tris-Glycine Novex® gels (Catalog #: EC6025BOX).

Lane 1: Invitrogen™ BenchMark™ Protein Ladder (Catalog #: 10747-012).

Lane 2: 0.4 µg PPAR delta -LBD (GST)

Lane 3: 0.8 µg PPAR delta -LBD (GST)

Lane 4: 2.0 µg PPAR delta -LBD (GST)

Lane 5: 4.0 µg PPAR delta -LBD (GST)

KDa 220 — 160 — 120 — 120 — 150 — 90 — 90 — 970 — 100

Purity:

75% as determined by a Coomassie® blue stained SDS-PAGE gel.

Molecular Weight:

59.9 kDa. Calculated from the protein sequence(s).

Protein sequence alignment with reference sequence(s)

GenBank Accession Number: NP_006229.1

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MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVL GST
MSPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAEISMLEGAVL IVGN PPAR delta
101 DIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSSKYIA
101 DIRYGVSRIAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPQIDKYLKSSKYIA
201 WPLQGWQATFGGGDHPPKSDLVPR
201 WPLQGWQATFGGGDHPPKSDLVPRPWSNQTSLYKKAGSAAAPFTGSQYNPQVADLKAFSKHIYNAYLKNFNMTKKKARSILTGKASHTAPFVIHDIETLW
165 -----GSQYNPQVADLKAFSKHIYNAYLKNFNMTKKKARSILTGKASHTAPFVIHDIETLW
       QAEKGLVWKQLVNGLPPYKEISVHVFYRCQCTTVETVRELTEFAKSIPSFSSLFLNDQVTLLKYGVHEAIFAMLASIVNKDGLLVANGSGFVTREFLRSL
QAEKGLVWKQLVNGLPPYKEISVHVFYRCQCTTVETVRELTEFAKSIPSFSSLFLNDQVTLLKYGVHEAIFAMLASIVNKDGLLVANGSGFVTREFLRSL
401 RKPFSDIIEPKFEFAVKFNALELDDSDLALFIAAIILCGDRPGLMNVPRVEAIQDTILRALEFHLQANHPDAQYLFPKLLQKMADLRQLVTEHAQMMQRI
321 RKPFSDIIEPKFEFAVKFNALELDDSDLALFIAAIILCGDRPGLMNVPRVEAIQDTILRALEFHLQANHPDAQYLFPKLLQKMADLRQLVTEHAQMMQRI
224
501 KKTETETSLHPLLQEIYKDMY.
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Anita Targosz, Associate Director

Date: 08/Oct/2018

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Invitrogen™ is a trademark of Life Technologies Corporation.

BenchMark™ is a trademark of Life Technologies Corporation.

Coomassie® is a registered trademark of Imperial Chemical Industries.

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^{*} highlighted residues denote differences from the reference protein sequence(s)

Certificate of Analysis LanthaScreen® Tb-anti-GST (Goat) Antibody, 25 μg



Part Number: PV3550 Lot Number: 2068723B Immediate Storage: -20°C Shipping Conditions: dry ice

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Description:

Tb-anti-GST Antibody used for TR-FRET Assays.

Concentration:

0.54 mg/mL as determined spectrophotometrically at A280. Calculated **3,600 nM** based on absorbance.

Molecular Weight:

150 kDa. The weight of the Tb label is negligible compared to the weight of the antibody.

Chelate to Antibody Ratio:

4:1 Determined spectrophotometrically.

Storage and Handling:

If properly stored in its original container at -20°C, this product is guaranteed for 6 months from date of purchase.

Spinning the antibody is important to reduce noise in the assay readout. Upon thaw of stock or aliquot, mix antibody by inversion or pipetting and centrifuge stock vial or aliquot of antibody at ~10,000 rpm for 5 minutes prior to use. After centrifugation, pipet the quantity of antibody needed for the assay from the top of the liquid, thereby minimizing the potential mixing of material that has been spun to the bottom of the vial. To reduce the number of freeze/thaws aliquots may be prepared. However, to facilitate spinning and pipetting, aliquots below 50 µL are not recommended.

Storage Buffer:

10 mM HEPES 137 mM NaCl, 2.7 mM KCl (HEPES buffered saline).

Instrument Settings:

LanthaScreen® Tb-anti-GST (Goat) Antibody, used in a TR-FRET assay, requires a fluorescence plate reader capable of reading TR-FRET and filters in the **green** range. For instrument specific set-up information go to www.thermofisher.com/instrumentsetup.

Equivalent Usage:

The LanthaScreen® Tb-anti-GST (Rabbit) Antibody and the LanthaScreen® Tb-anti-GST (Goat) Antibody are INTERCHANGEABLE in any protocol requiring a LanthaScreen® Tb-anti-GST antibody.

QUALITY ASSURANCE

Functional Testing:

The performance of each lot of LanthaScreen® Tb-anti-GST (Goat) Antibody is confirmed by the LanthaScreen® TR-FRET LXR alpha Coactivator Assay (PV4655) using a titration of the agonist T0901317 (Caymen Chemicals 71810).

Anita Targosz, Associate Director

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Date: 29/Jan/2019

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