

Certificate of Analysis

LanthaScreen® TR-FRET PPAR gamma Coactivator Assay Kit, 1 Kit



Part Number: PV4548
Lot Number: 1664434
Immediate Storage: -80°C
Shipping Conditions: dry ice

5791 Van Allen Way
Carlsbad, CA 92008
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Fax: 760.602.6500
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Components:

Description	Composition	Quantity	Part Number	Lot Number
PPAR Gamma-LBD (GST)	Recombinant Human Peroxisome Proliferator Activated Receptor Gamma Ligand Binding Domain (amino acids 201-475; NP_005028), GST-tagged, in 50 mM Tris-HCl (pH 8.0), 150 mM KCl, 0.5 mM EDTA, 0.5% Big CHAP, 2 mM DTT and 20% Glycerol.	10 µg	PV4545*	1223242F
Fluorescein – TRAP220/DRIP-2 Coactivator Peptide	100 µM Fluorescein-NTKNHPMLMNLKDNPAQD in 50 mM HEPES (pH 7.5), 2554 Da.	100 µL	PV4549	1249902D
LanthaScreen® Tb-anti-GST (Goat) Antibody	10 mM HEPES 137 mM NaCl, 2.7 mM KCl (HEPES buffered saline).	25 µg	PV3550*	1366221C
Nuclear Receptor Buffer F	Proprietary Buffer (pH 7.5)	2 × 25 mL	PV4547	1030334F
DTT Solution	1M DTT in water.	1 mL	P2325	1597615A

*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 Gamma-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.
Fluorescein – TRAP220/DRIP-2 Coactivator Peptide	-20°C. Vortex prior to each use.
LanthaScreen® Tb-anti-GST (Goat) Antibody	If properly stored at -20°C, this product is guaranteed for 6 months from date of purchase.
Nuclear Receptor Buffer F	20–30°C. Mix prior to usage.
DTT Solution	-20°C

QUALITY ASSURANCE

Functional Testing:

Kit components are functionally tested for performance in the assay using a titration of a known agonist or antagonist.

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For questions, please contact our Technical Support Team

N. Am Ph#: 800-955-6288 or INTL Ph#: 760-603-7200 Select option 5, ext. 40266 Email: drugdiscoverytech@lifetech.com

Certificate of Analysis

PPAR Gamma-LBD (GST), 10 µg

Recombinant Human Peroxisome Proliferator Activated Receptor Gamma Ligand Binding Domain, GST-tagged



Part Number: PV4545
Lot Number: 1223242F
Immediate Storage: -80°C
Shipping Conditions: dry ice

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Description:

Recombinant Human Peroxisome Proliferator Activated Receptor Gamma Ligand Binding Domain (amino acids 201-475; NP_005028), GST-tagged, expressed in insect cells.

Concentration:

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

Calculated **10,200 nM**.

Assay Concentration(s):

34 nM PPAR Gamma-LBD (GST) is the final concentration recommended for the PolarScreen™ PPARγ Competitor Assay, Green (PV6136).

0.5 nM PPAR Gamma-LBD (GST) is the final concentration recommended for the LanthaScreen® TR-FRET PPAR gamma Competitive Binding Assay (PV4894).

5 nM PPAR Gamma-LBD (GST) is the final concentration recommended for the LanthaScreen® TR-FRET PPAR gamma Coactivator Assay (PV4548).

Aliases:

NR1C3

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.**

Stable through 5 freeze-thaw cycles.

Storage Buffer:

50 mM Tris-HCl (pH 8.0), 150 mM KCl, 0.5 mM EDTA, 0.5% Big CHAP, 2 mM DTT and 20% Glycerol.

QUALITY ASSURANCE

Functional Testing:

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

LanthaScreen® TR-FRET PPAR Gamma Competitive Binding Assay (PV4894). The ligand GW1929 was shown to competitively displace Fluormone™ Pan-PPAR Green (PV4896) from PPAR Gamma-LBD (GST).

LanthaScreen® TR-FRET PPAR Gamma Coactivator Assay (PV4548). The ligand GW1929 was shown to recruit Fluorescein-TRAP220/DRIP-2 peptide (PV4549) NTKNHPMLMNLKDNPAQD to bind with PPAR Gamma-LBD (GST).

PolarScreen™ PPAR Gamma Competitor Assay, Green (PV6136). The ligand rosiglitazone was shown to displace Fluormone™ PPAR Gamma Green (PV3356) from PPAR Gamma-LBD (GST).

Gel Information for PPAR Gamma-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: 1 µg PPAR gamma LBD (GST)

Lane 3: 2 µg PPAR gamma LBD (GST)

Lane 4: 5 µg PPAR gamma LBD (GST)

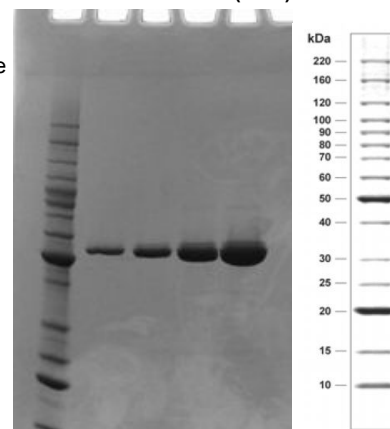
Lane 5: 10 µg PPAR gamma LBD (GST)

Purity:

95% as determined by a Coomassie® blue stained SDS-PAGE gel.

Molecular Weight:

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



Protein sequence alignment with reference sequence(s)

1 MAPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAIEISMLEGAVL GST TAG
1 MAPILGYWKIKGLVQPTRLLLEYLEEKYEEHLYERDEGDKWRNKKFELGLEFPNLPYYIDGDVKLTQSMAIIRYIADKHNMLGGCPKERAIEISMLEGAVL IVGN
201 ----- NP_005028

101 DIRYGVSR IAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPIQIDKYLKSSKYIA
101 DIRYGVSR IAYSKDFETLKVDFLSKLPEMLKMFEDRLCHKTYLNGDHVTHPDFMLYDALDVVLYMDPMCLDAFPKLVCFKKRIEAIPIQIDKYLKSSKYIA
201 -----

201 WPLQGWQATFGGGDHPKSDLVPR
201 WPLQGWQATFGGGDHPKSDLVPRHNQTSLYKKAGTMQLNPESADLRALAKHLYDSYIKSFPLTKAKARAILTGKTTDKSPFVIYDMNSLMMGEDKIKFK
201 -----QLNPESADLRALAKHLYDSYIKSFPLTKAKARAILTGKTTDKSPFVIYDMNSLMMGEDKIKFK

224
301 HITPLQEQSKEVAIRIFQGCQFRSVEAVQEI TEYAKSIPGFVNLDLNQVTL LKYGVHEIIYTMLASLMNKDGV LISEGQGFMTREFL KSLRKPFGDFME
264 HITPLQEQSKEVAIRIFQGCQFRSVEAVQEI TEYAKSIPGFVNLDLNQVTL LKYGVHEIIYTMLASLMNKDGV LISEGQGFMTREFL KSLRKPFGDFME

224
401 PKFEFAVKFNALELDDSDLAIFIAVIILSGDRPGLLNKVP IEDIQDNLLQALELQLKLNHPESSQLFAKLLQKMTDLRQIVTEHVQLLQVIKKTETDMSL
364 PKFEFAVKFNALELDDSDLAIFIAVIILSGDRPGLLNKVP IEDIQDNLLQALELQLKLNHPESSQLFAKLLQKMTDLRQIVTEHVQLLQVIKKTETDMSL

224
501 HPLLQEIKDLY.
464 HPLLQEIKDLY

* highlighted residues denote differences from the reference protein sequence(s).



Becky. Baker, QA Engineer III

Date: 14/Mar/2014

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For Research Use Only. Not for use in diagnostic procedures.

Certificate of Analysis

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



Part Number: PV3550
Lot Number: 1366221C
Immediate Storage: -20°C
Shipping Conditions: dry ice

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Carlsbad, CA 92008
Phone: 760.603.7200
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Description:

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

Concentration:

0.50 mg/mL as determined spectrophotometrically at A280.
Calculated **3,300 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:

9:1 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.lifetechnologies.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).

Tony Goossens, Engineer II, QA/QC

Date: 10/Apr/2013

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For questions, please contact our Technical Support Team

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