Certificate of Analysis LanthaScreen $^{\otimes}$ TR-FRET AR Coactivator Assay, 800 X 20 μL

Part Number: A15878 Lot Number: 3133714 Immediate Storage: -80°C Shipping Conditions: dry ice

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

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SCIENTIFIC

Components:

Description	Composition	Quantity	Part Number	Lot Number
AR - LBD (GST)	Proprietary Buffer (pH 7.5) containing protein stabalizing agents and Glycerol.	100 µg	A15676*	2753687C
Fluorescein-D11-FxxLF Coactivator Peptide	100 μM Fluorescein-VESGSSRFMQLFMANDLLT in 50 mM HEPES (pH 7.5), 2506 Da.	100 µL	PV4382	2319552E
LanthaScreen® Tb-anti-GST (Goat) Antibody	10 mM HEPES 137 mM NaCl, 2.7 mM KCl (HEPES buffered saline).	25 µg	PV3550*	2845081C
TR-FRET Coregulator Buffer A	Proprietary Buffer (pH 7.5)	2 × 25 mL	PV4384	2678045A
DTT Solution	1M DTT in water.	1 mL	P2325	2488866F

*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		
AR - 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-D11-FxxLF 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.		
TR-FRET Coregulator Buffer A	4°C. Mix prior to use.		
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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Certificate of Analysis AR - LBD (GST), 100 μg

Androgen Receptor Ligand Binding Domain, GSTtagged

Recombinant Rat Androgen Receptor ligand binding domain (amino acids 606-902, NP_036634), GST-tagged, expressed in insect cells.

0.54 mg/mL total protein as measured using the Bradford protein assay

Note: As of November 2012 we no longer perform radioligand binding assays to determine the active nM concentration and specific activity. The reported nM concentration is based on the Bradford protein assay and calculated using the molecular weight.

406 nM AR - LBD (GST) is the final concentration recommended for the PolarScreen [™] AR Competitor Assay Kit, Green (A15880). **28 nM** AR - LBD (GST) is the final concentration recommended for the

Part Number: A15676 Lot Number: 2753687C

Immediate Storage: -80°C

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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.

Stable through at least 8 freeze-thaw cycles. Store on ice when thawed and refreeze promptly after use.

Storage Buffer:

Proprietary Buffer (pH 7.5) containing protein stabilizing agents and glycerol.

LanthaScreen™ TR-FRET AR Coactivator Assay (A15878).

Description:

Concentration:

with BSA as a standard.

Assay Concentration(s):

Calculated 8,390 nM.

Aliases:

NR3C4, dihydrotestosterone receptor (DHTR)

QUALITY ASSURANCE

Functional Testing:

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

LanthaScreen[™] TR-FRET AR Coactivator Assay (A15878). The ligand dihydrotestosterone (DHT) was shown to recruit Fluorescein-D11FxxLF peptide (PV4382) VESGSSRFMQLFMANDLLT to bind with AR - LBD (GST).

PolarScreen [™] AR Competitor Assay Kit, Green (A15880). The ligand dihydrotestosterone (DHT) was shown to displace Fluormone AL Green from AR - LBD (GST).

Gel Information for AR - LBD (GST)

		•	,		
Page Description: The SDS- PAGE and/or Native PAGE were run on 4-20% Tris-Glycine Novex™ gels (Catalog #: EC6025BOX).				kDa 220 — 160 — 120 —	111
EC0025BOX).				90 —	
				80	
Lane 1: Invitrogen™				60	
BenchMark™ Protein Ladder (Catalog #: 10747-012).		_	_	50 — 40 —	-
	1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1			40 -	
Lane 2: 0.3 µg AR - LBD (GST)				30 —	
				25 —	
Lane 3: 0.6 µg AR - LBD (GST)					
				20 —	-
Lane 4: 1.5 µg AR - LBD (GST)					
				15 —	
Lane 5: 3.0 µg AR - LBD (GST)				10 —	_

Purity:

90% as determined by a SDS-PAGE gel stained with SimplyBlue ${}^{\rm TM}$ SafeStain.

Molecular Weight:

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

Certificate of Analysis

Protein sequence alignment with reference sequence(s)

GenBank Accession Number: NP_036634

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

Chevohn Joseph, Director, Quality

Date: 21/Jul/2023

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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: 2845081C Immediate Storage: -20°C Shipping Conditions: dry ice

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

Calculated 3,300 nM based on absorbance.

0.50 mg/mL as determined spectrophotometrically at A280.

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

Description:

Concentration:

Molecular Weight:

Chelate to Antibody Ratio:

7:1 Determined spectrophotometrically.

the antibody.

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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).

Chevohn Joseph, Director, Quality

Date: 05/Mar/2024

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