Certificate of Analysis PolarScreen™ GR Competitor Assay, Green, 400 x 20 µL



Part Number: A15897 Lot Number: 1628594 Immediate Storage: -80°C Shipping Conditions: dry ice 5791 Van Allen Way Carlsbad, CA 92008 Phone: 760.603.7200 Fax: 760.602.6500 www.lifetechnologies.com

Components:

Description	Composition	Quantity	Part Number	Lot Number
GR Full Length	10 mM potassium phosphate (pH 7.4), 5 mM DTT, 0.1 mM EDTA, 10 mM sodium molybdate and 10% glycerol. This storage buffer is yellow in color due to the formation of DTT/Molybdate complex.	8.3 mg	A15663*	1424618B
Fluormone™ GS1 Green	500 nM in 20 mM Tris, 90% Methanol	50 μL	P2813	1220342C
GR Stabilizing Peptide, 10X	1 mM in 10 mM potassium phosphate (pH 7.4)	3 × 1 mL	P2815	401833Z
GR Screening Buffer, 10X	100 mM potassium phosphate (pH 7.4), 200 mM Na ₂ MoO ₄ , 1 mM EDTA and 20% DMSO. This GR storage buffer is yellow in color due to the formation of DTT/Molybdate complex.	3 × 1 mL	P2814	1354922A
DTT Solution	1M DTT in water.	1 mL	P2325	1484398A

^{*}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
	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.
	Store at -20°C. Vortex the Fluormone™ before 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.
GR Stabilizing Peptide, 10X	−20°C or −80°C. Vortex prior to use.
GR Screening Buffer, 10X	20-30°C. Mix prior to use.
DTT Solution	−20°C

QUALITY ASSURANCE

Functional Testing:

Assay Specifications:

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

Using the conditions described in the kit protocol and keeping the plates in the dark and covered to prevent evaporation this kit has the following specification(s):

Specification	Value
ΔmP (EC _{aa})	≥ 120 mP

Manufacturing Specifications:

The concentration of Fluormone $\ensuremath{^{\text{TM}}}$ GS1 was determined by measuring its absorbance.

Note: The method for determining the concentration of Fluormone $^{\text{TM}}$ GS1 has been changed from fluorescent intensity to absorbance. Please refer to the protocol or contact Technical Support at 760-603-7200 extension 40266.

Fluormone™ is a trademark of Life Technologies Corporation.

PolarScreen™ is a trademark of Life Technologies Corporation.

Certificate of Analysis GR Full Length, 8.3 mg

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Part Number: A15663 Lot Number: 1424618B Immediate Storage: -80°C Shipping Conditions: dry ice

Description:

Full-length human Glucocorticoid Receptor (GR) expressed in insect cells, partially purified in order to maintain high ligand binding activity. Further purification results in decreased activity, presumably due to loss of endogenous accessory proteins. Studies have shown that GR must be associated with a complex of chaperone proteins for ligand activation.

Concentration

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

Calculated 124,000 nM.

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.

Assay Concentration(s):

1312 nM GR Full Length is the final concentration recommended for the PolarScreen™ GR Competitor Assay, Green (A15897).

3232 nM GR Full Length is the final concentration recommended for the PolarScreen™ GR Competitor Assay, Red (A15898).

Note: GR Full Length is partially purified to retain activity. Only a small percentage of the total protein in the tube is active GR. The nM concentration quoted here for use in the assays is not the nM concentration of active GR and is not representative of the assay's sensitivity to determine binding constants of test compounds.

Aliases:

NR3C1

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 freeze on dry ice as product may be sensitive to pH shift from exposure to CO_2 .

Storage Buffer:

10 mM potassium phosphate (pH 7.4), 5 mM DTT, 0.1 mM EDTA, 10 mM sodium molybdate and 10% glycerol. This storage buffer is yellow in color due to the formation of DTT/Molybdate complex.

QUALITY ASSURANCE

Functional Testing:

The performance of each lot of GR Full Length is confirmed in the following assay(s):

PolarScreen™ GR Competitor Assay, Green (A15897). The ligand dexamethasone was shown to displace Fluormone™ GS1 from GR Full Length.

PolarScreen™ GR Competitor Assay, Red (A15898). The ligand dexamethasone was shown to displace Fluormone™ GS Red from GR Full Length

Molecular Weight:

85.7 kDa.

Purity:

Partially purified to retain activity.

Part Number: A15663, Lot Number: 1424618B

Protein sequence alignment with reference sequence(s)

GenBank Accession Number: NP_000167

1 MDSKESLTPG REENPSSVLA QERGDVMDFY KTLRGGATVK VSASSPSLAV ASQSDSKQRR LLVDFPKGSV SNAQQPDLSK AVSLSMGLYM GETETKVMGN GR Full Length MDSKESLTPG REENPSSVLA QERGDVMDFY KTLRGGATVK VSASSPSLAV ASQSDSKQRR LLVDFPKGSV SNAQQPDLSK AVSLSMGLYM GETETKVMGN GR Full Length DLGFPQQQQI SLSSGETDLK LLEESIANLN RSTSVPENPK SSASTAVSAA PTEKEFPKTH SDVSSEQQHL KGQTGTNGGN VKLYTTDQST FDILQDLEFS SCASTAVSAA PTEKEFPKTH SDVSSEQQHL KGQTGTNGGN VKLYTTDQST FDILQDLEFS SCASTAVACA PTEKEFPKTH SDVSSEQQH SA

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Becky, Baker, QA Engineer III

Date: 07/Mar/2014

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Coomassie[®] is a registered trademark of Imperial Chemical Industries.

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

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