

RECOMBINANT HUMAN ADENOSINE A_{2A} RECEPTOR
2017
Protein information

Target Name	Adenosine A _{2A} receptor (A _{2A} receptor)
Catalogue Number	PP1
Class	GPCR Class A
Sequence	Full-length, wildtype sequence, with a N-terminus Strep tag II , 8xHis-tag , and TEV protease cleavage site : MWSHPQFEKHHHHHHHHENLYFQG PIMGSSVYITVELAIAVLAILGNVLVCWAVWLNSNLQNVNTNY FVVSLLAAADIAVGVLAIPIFAITISTGFCAACHGCLFIACFVLVLTQSSIFSLLAIAIDRYIAIRIPLRYNGL VTGTRAKGIIAICWVLSFAIGLTPMLGWNNCGQPKEGKNHSQGC GEGQVACL FEDV VPMNYMVYF NFFACVLVPLLLMLGVYLRIFLAARRQLKQMESQPLPGERARSTLQKEVHAAKSLAIIVGLFALCWLPL HIINCFTFFCPDCSHAPLWLMYLAIVLSHTNSVWNPFIYAYRIREFRQTFRKIIRSHVLRQQEPFKAAG TSARVLAAHGSDGEQVSLRLNGHPPGVWANGSAPHPERRPNGYALGLVSGGSAQESQGNTGLPDV ELLSHELKGVCEPPGLDDPLAQDGAGVS
Affinity Tag	His/Strep (both N-terminal)
Origin	Human (Homo sapiens)
Theor. MW	47,7kDa
Accession #	P29274 (UniProt)

Protein production

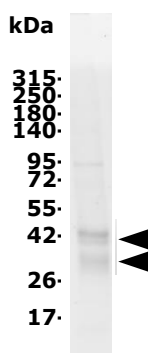
Expression system	<i>Sf9</i> insect cells (baculovirus)
Purification	Immobilized Metal Affinity Chromatography
Purity	>90%
Activity	Confirmed by radiobinding assay
Concentration	Up to 5mg/ml
Sample Buffer	50mM Hepes pH 7.4, 200mM NaCl, 0.05%/0.006% DDM/CHS
Available quantity	From 10µg up to mg scale
References	1- Igonet S et al. Stabilization of native and functional Adenosine receptor (in preparation). 2- Jawhari A. Towards Native and Stable GPCRs for Conformational Antibody Development. <i>Discovery on Target</i> , Boston 2015. 3- Desuzinges Mandon E. et al. Novel systematic detergent screening method for membrane proteins solubilization. <i>Anal Biochem.</i> 2017 Jan 15;517:40-49.

Miscellaneous

Shipment Temperature	Dry ice
Storage conditions	Store at -80°C



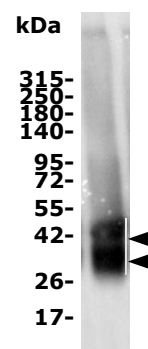
Quality Controls (Purity and Activity):



SDS-PAGE, 4-15% acrylamide gel
Bio-rad Stain-Free™ detection

SDS-PAGE, Stain-Free detection

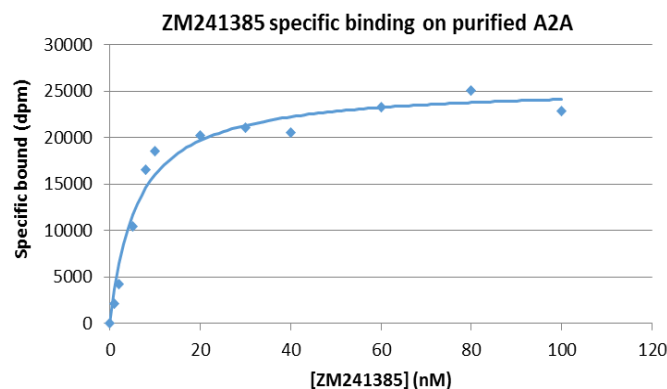
Purified A_{2A} was migrated on a 4-15% Tris-glycine SDS-PAGE and the total proteins were Stain-Free detected. Black arrows indicate the target. Upper arrows indicate full-length A_{2A}. Lower arrow indicate shorter A_{2A} resulting from partial cleavage of C-term end.



SDS-PAGE, 4-15% acrylamide gel
WB Anti-ICL3 A_{2A} antibody (7F6-G5-A2)

SDS-PAGE, western blotting

Purified A_{2A} was migrated on a 4-15% Tris-glycine SDS-PAGE, transferred to pvdf membrane and immunodetected with a monoclonal anti- ICL3-A_{2A} (7F6-G5-A₂, SCBT). Black arrows indicate the target.



QC: Activity measured by radiobinding assay

Binding of [3H]ZM241385 was measured on purified A_{2A}. A K_D of 6nM was determined for ZM241385.

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