

Overview

Synonyms	RBP4; Retinol-Binding Protein 4
Description	The properties of retinol binding protein is the transport carrier of vitamin A in the plasma. Human-retinol binding protein is a single-chain polypeptide with a molecular weight of approximately 21000 and one binding site for retinol and other forms of vitamin A. In addition, compounds related to retinol, such as retinal, retinoic acid, retinyl esters and geometric isomers of retinol and of retinal were evaluated for their ability to bind to this protein. In plasma, RBP4-retinol forms a complex with transthyretin (TTR), also known as thyroxine-binding protein and prealbumin. Defects in RBP4 cause retinol-binding protein deficiency, which affects night vision. iXCells rhRBP4 with C-terminal Histidine tag has a molecular mass of 22 kDa analyzed by reducing SDS-PAGE and is obtained by chromatographic techniques.
Source	<i>HEK 293</i> Measured by its ability to bind all-trans retinoic acid. The binding of retinoic acid results in the quenching of Trp fluorescence in RBP4. >1.0 µM all-trans retinoic acid is bound under the described conditions. Assay Protocol: 1. Dilute rhRBP4 to 49 µg/mL in Assay Buffer. 2. Make serial dilutions of retinoic acid in 95% ethanol at 100, 30, 10, 3 and 1 µM. 3. The formation of Protein/Retinol binding in microtubes: Biological Activity 3.1. Mix 112.5 µL of 49 µg/mL rhRBP4 and 12.5 µL of retinoic acid serial dilutions in microtubes. 3.2. For a blank, mix 112.5 µL of 49 µg/mL rhRBP4 and 12.5 µL of 95% ethanol in a microtube. 4. Incubate reaction tubes at room temperature for 30 minutes. 5. In a plate load 100 µL of the reaction mixtures and blank. 6. Read at excitation and emission wavelengths of 280 nm and 340 nm (top read), respectively, in endpoint mode. 7. Calculate concentration at which 50% quenching of rhRBP4 is achieved by plotting raw RFUs vs. concentration of retinoic acid with 4-PL fitting. Use this value to estimate the concentration of retinoic acid fully bound by the rhRBP4.
Sequence	ERDCRVSSFR VKENFDKARF SGTWYAMAKK DPEGLFLQDN IVAEFVSDET GQMSATAKGR VRLLNWDVC ADMVGTFTDT EDPAKFKMKY WGVASFQKG NDDHWIVDTD YDTYAVQYSC RLLNLDGTCA DSYSFVFSRD PNGLPPEAQK IVRQRQEELC LARQYRLIVH NGYCDGRSER NLLHHHHHH

Properties

Measured Molecular Weight	22kDa, observed by reducing SDS-PAGE.
Purity	> 97% as analyzed by reducing SDS-PAGE&RP-HPLC.
Formulation	Lyophilized from a 0.2 µm filtered solution in 50mM Tris-HCl, 150mM NaCl, pH 7.5.
Reconstitution	Reconstituted in ddH ₂ O or PBS at 100 µg/ml.
Endotoxin Level	<0.2 EU/µg, determined by LAL method.
Storage	Lyophilized recombinant Human RBP4 remains stable up to 6 months at lower than -70°C from date of receipt. Upon reconstitution, Human RBP4 should be stable up to 1 week at 4°C or up to 3 months at -20°C. For long term storage it is recommended that a carrier protein (example 0.1% BSA) be added. Avoid repeated freeze-thaw cycles.

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