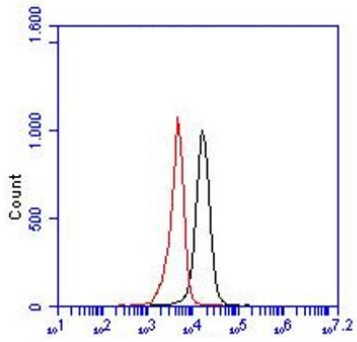
CERTIFICATE OF ANALYSIS – TECHNICAL DATA SHEET

Product name	EPCR, Human, clone RCR-252		
Catalog number	HM2145-100UG		
Lot number	-	**Expiry date**	-
Volume	1 ml	**Amount**	100 µg
Formulation	0.2 µm filtered in PBS+0.1%BSA	**Concentration**	100 µg/ml
Host Species	Rat IgG1	**Conjugate**	None
Endotoxin	<24 EU/mg	**Purification**	Protein G
Storage	4°C		

Application notes

	IHC-F	IHC-P	IF	FC	FS	IA	IP	W
Reference #				1,2	1-4		5	5
Yes				●	●		●	●
No								
N.D.	●	●	●			●		

N.D.= Not Determined; IHC = Immuno histochemistry; F = Frozen sections; P = Paraffin sections; IF = Immuno Fluorescence; FC = Flow Cytometry; FS = Functional Studies; IA = Immuno Assays; IP = Immuno Precipitation; W = Western blot



FC: Flow cytometry: detection of EPCR in HUVEC cells. Red and black line represent the isotype control and HM2145 with a concentration of 20 µg/ml, respectively.

Dilutions to be used depend on detection system applied. It is recommended that users test the reagent and determine their own optimal dilutions. The typical starting working dilution is 1:50.

- FC: Antibody RCR-252 stains the extracellular domain of human EPCR. As positive control RE-1 cells were used and as negative control N1 cells. (Ref.1)
- W: A non-reduced sample treatment was used. The observed band size is/ 50 kDa (Ref.5).
- FS: Antibody RCR-252 functions as blocking antbody. It ablates APC binding to EPCR and blocks APC-mediated signaling. The antibody was functionally tested by pretreatment of cells with 10-25 µg/ml RCR-252 (Ref.1-4).
- Positive control: RE-1 (rat 3Y1 fibroblasts stably transfected with human EPCR); Negative control: N1 cells

General Information

Description

The monoclonal antibody RCR-252 recognizes human endothelial protein C receptor (EPCR), a highly glycosylated type I transmembrane protein of 221-amino-acids. These amino acids comprise an extracellular domain, a 25-aa transmembrane domain, and a short (3 aa) intracytoplasmic sequence coding for an ~46 kDa protein. Deglycosylation will reduce the protein mass to 25 kDa. EPCR is expressed strongly on the endothelial cells of arteries and veins in heart and lung, less intensely in capillaries in the lung and skin, and not at all in the endothelium of small vessels of the liver and kidney. EPCR is the receptor for protein C, a key player in the anticoagulation pathway. The protein C anticoagulant pathway serves as a major system for controling thrombosis, limiting inflammatory responses, and potentially decreasing endothelial cell apoptosis in response to inflammatory cytokines and ischemia. The essential components of the pathway include thrombin, thrombomodulin, the endothelial cell protein C receptor (EPCR), protein C and protein S. The pathway is initiated when thrombin binds to thrombomodulin on the surface of endothelium. EPCR

augments protein C activation by binding protein C and presenting it to the thrombin-thrombomodulin activation complex. Activated protein C (aPC) retains its ability to bind EPCR, and this complex appears to be involved in some of the cellular signaling mechanisms that down-regulate inflammatory cytokine formation (TNF, IL-6). EPCR is shed from the vasculature by inflammatory mediators and thrombin. EPCR binds to activated neutrophils in a process that involves proteinase 3 and Mac-1. Furthermore, EPCR can undergo translocation from the plasma membrane to the nucleus. EPCR can be cleaved to release a soluble form (sEPCR) in the circulation. This sEPCR is detected as a single species of 43 kDa, resulting from shedding of membrane EPCR by the action of a metalloprotease, which is stimulated by thrombin and by some inflammatory mediators. Soluble EPCR binds PC and aPC with similar affinity, but its binding to aPC inhibits the anticoagulant activity of aPC by blocking its binding to phospholipids and by abrogating its ability to inactivate factor Va. sEPCR can be detected in plasma. In normal persons, sEPCR is present in levels of 83.6 +/- 17.2 ng/ml. Elevated levels of sEPCR are positively correlated to a higher risk for thrombosis. Furthermore, a haplotype (A3 allele) has been linked to elevated levels of sEPCR (264 +/-174 ng/ml).

Immunogen Human EPCR-positive RE-1 cells.

Aliases Endothelial Cell Protein C Receptor, Activated protein C receptor, CD201.

Gene Gene name: PROCR

References

1. Ye, X et al; The endothelial cell protein C receptor (EPCR) functions as a primary receptor for protein C activation on endothelial cells in arteries, veins, and capillaries. Biochem biophys res commun 1999, *259*: 671
2. Dömötör, E et al; Activated protein C alters cytosolic calcium flux in human brain endothelium via binding to endothelial protein C receptor and activation of protease activated receptor-1. Blood 2003, *101*: 4797
3. Sturn, D et al; Expression and function of the endothelial protein C receptor in human neutrophils. Blood 2003, *102*: 1499
4. Finigan, J et al; Activated protein C mediates novel lung endothelial barriere enhancement. J Biol Chem 2005, *280*: 17286
5. Bae J et al; Receptors of the protein C activation and activated protein C signaling pathways are colocalized in lipid rafts of endothelial cells. PNAS 2006, *104*: 2867

Storage&stability Product should be stored at 4°C. Under recommended storage conditions, product is stable for at least one year.

Precautions For research use only. Not for use in or on humans or animals or for diagnostics. It is the responsibility of the user to comply with all local/state and federal rules in the use of this product. Hycult Biotech is not responsible for any patent infringements that might result from the use or derivation of this product.

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Approved by Manager of QC	Date
Brenda Teunissen	02/12/2019

Do you have any questions or comments regarding this product? Please contact us via support@hycultbiotech.com.