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Catalogue# MCA-S128: Mouse Monoclonal Antibody to Visual Arrestin (a.k.a. Arrestin-1 and S-antigen): SAG

Immunogen: The arrestin proteins are a family of regulators of cell signaling of G protein-coupled receptors (GPCR). Visual arrestin was first discovered as a result of the experimental model of human uveitis, an autoimmune disease of the eye. In this model, called experimental allergic uveitis, animals were injected with extracts made from the retina of the same species mixed with Freund's complete adjuvant. The animals mounted a strong immune response to the extract, and the antibody response was used to identify several immunogenic retinal proteins. One of these was called S-antigen, for soluble antigen. The protein was found to be abundant in retina, about 48 kDa in molecular weight, and localized in the outer segments of the photoreceptors (1, 2).

Several years later, Hermann Kühn and colleagues discovered that this protein binds to phosphorylated rhodopsin and prevents this protein from activating transducin (3, 4, 5). Transducin is a typical heterotrimeric G protein, composed of α and βγ subunits. Rhodopsin phosphorylation is mediated by Rhodopsin kinase (a.k.a. GRK1), the prototypic member of a family of GPCR kinases. Since the S-antigen protein arrested the activity of rhodopsin it was renamed arrestin, and became the prototypic member of the arrestin protein family.

Subsequently, Robert Lefkowitz and colleagues discovered a related protein which bound to phosphorylated β-adrenergic GPCRs and prevented these proteins from activating their specific heterotrimeric G proteins (6). Because of this relationship to the β-adrenergic receptor and functional and structural similarities to visual arrestin this protein was named β-arrestin. The β-adrenergic receptor was phosphorylated by the β-adrenergic receptor kinase (a.k.a. GRK2), an enzyme belonging to GPCR kinase family.

Studies of visual transduction therefore aided greatly in understanding other kinds of GPCR signaling. In mammals, there are four arrestin isoforms; Visual arrestin (a.k.a. S-antigen and arrestin-1) and cone arrestin (a.k.a. arrestin-4) are largely confined to photoreceptors. β-arrestin 1 (a.k.a. arrestin 2) and β-arrestin-2 (a.k.a. arrestin-3) are ubiquitous and regulate non-visual GPCRs.

MCA-S128 was raised against recombinant bovine arrestin-1 with the first 20 amino acids of the C-terminus truncated. The antibody does not bind the other three arrestin molecules. In the retina, MCA-S128 binds to rod cell bodies and rod outer segments. The HGNC name for this protein is SAG.

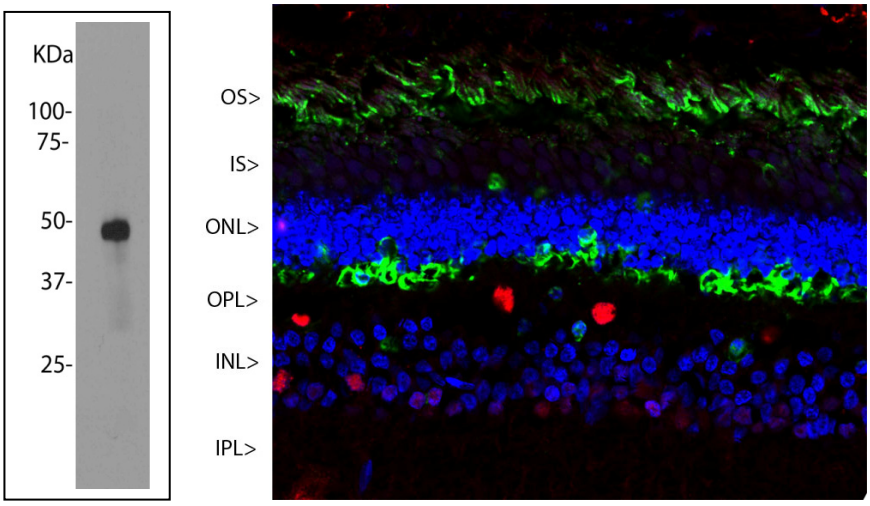

Figure: Left: Blot of bovine retinal extracts probed with MCA-S128. The antibody stains a band corresponding to retinal arrestin at about 48 kDa. **Right:** Confocal image of a pig retina stained with MCA-S128 (green). Visual arrestin is most abundant in the outer segments (OS) and inner surface of the outer nuclear layer (ONL), and

can be used to identify components of rod photoreceptor cells. (Cone photoreceptors have a different arrestin isotype). Other retinal layers are inner segments (IS), outer plexiform layer (OPL), inner nuclear layer (INL) and inner plexiform layer (IPL). The red stain is Fox2, an RNA binding nuclear protein related to Fox3/NeuN, which stains nuclei of horizontal neurons and some other neurons in the INL and IPL. Nuclear DNA was revealed with DAPI (blue).

Antibody Characteristics: MCA-S128 is a mouse IgG1 class antibody with a κ light chain. MCA-S128 is known to react with visual arrestin from human, bovine, mouse, pig and rat. This antibody was affinity purified on Protein G at 1 mg/mL and was diluted in PBS containing 10 mM sodium azide as a preservative.

Suggestions for use: Use at dilutions of ~1:1,000 for immunofluorescence. For western blots Use at 1:5,000. A suitable control tissue is retinal homogenate. The arrestin protein runs at about ~48 kDa on SDS-PAGE gels.

Storage Instructions: Shipped on ice. Please store at 4°C for regular uses. For long term storage, please leave frozen at -20°C and avoid freeze/thaw cycles.

Limitations: This product is for research use only and is not approved for use in humans or in clinical diagnosis.

References:

1. Dorey C, Faure JP. [Isolation and characterization of a retinal antigen inducing experimental autoimmune uveo-retinitis]. [Article in French] Ann Immunol (Paris). 128:229-32 (1977).

2. Wacker WB, Donoso LA, Kalsow CM, Yankeelov JA Jr, Organisciak DT. Experimental allergic uveitis. Isolation, characterization, and localization of a soluble uveitopathogenic antigen from bovine retina. J Immunol. 119:1949-58 (1977).

3. Kühn H, Hall SW, Wilden U. Light-induced binding of 48-kDa protein to photoreceptor membranes is highly enhanced by phosphorylation of rhodopsin. FEBS Lett. 176:473-8 (1984).

4. Pfister C, Chabre M, Plouet J, Tuyen VV, De Kozak Y, Faure JP, Kühn H. Retinal S antigen identified as the 48K protein regulating light-dependent phosphodiesterase in rods. Science 228:891-3 (1985).

5. Wilden U, Hall SW, Kühn H. Phosphodiesterase activation by photoexcited rhodopsin is quenched when rhodopsin is phosphorylated and binds the intrinsic 48-kDa protein of rod outer segments. Proc Natl Acad Sci USA 83:1174-8 (1986).

6. Lohse MJ, Benovic JL, Codina J, Caron MG, Lefkowitz RJ. β-arrestin: a protein that regulates beta-adrenergic receptor function. Science 248: 1547-1550 (1990).

7. Smith WC, Mc Dowell JH, Dugger DR, Miller R, Arendt A, Popp MP, Hargrave PA. Identification of regions of arrestin that bind to rhodopsin. Biochemistry 38:2752-61 (1999).

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