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Exosomal Marker Antibody Sampler Kit

1 Kit (8 x 20 microliters)



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For Research Use Only. Not for Use in Diagnostic Procedures.

Product Includes	Product #	Quantity	Mol. Wt	Isotype/Source
Alix (3A9) Mouse mAb	2171	20 µl	95 kDa	Mouse IgG1
Annexin V Antibody	8555	20 µl	30 kDa	Rabbit
CD54/ICAM-1 (E3Q9N) XP® Rabbit mAb	67836	20 µl	89, 92 kDa	Rabbit IgG
CD9 (D8O1A) Rabbit mAb	13174	20 µl	22, 24, 35 kDa	Rabbit IgG
GM130 (D6B1) XP® Rabbit mAb	12480	20 µl	140 kDa	Rabbit IgG
EpCAM (D1B3) Rabbit mAb	2626	20 µl	40 kDa	Rabbit IgG
HSP70 (D69) Antibody	4876	20 µl	70 kDa	Rabbit
Flotillin-1 (D2V7J) XP® Rabbit mAb	18634	20 µl	49 kDa	Rabbit IgG
Anti-rabbit IgG, HRP-linked Antibody	7074	100 µl		Goat
Anti-mouse IgG, HRP-linked Antibody	7076	100 µl		Horse

Please visit cellsignal.com for individual component applications, species cross-reactivity, dilutions, protocols, and additional product information.

Description

The Exosomal Marker Antibody Sampler Kit provides an economical means to evaluate the presence of exosomal markers. The kit includes enough primary antibody to perform two western blot experiments for each target.

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibodies.

Background

Exosomes are small membrane-bound vesicles that in recent years have emerged as important molecules for inter-cellular communication. Exosomes are produced during both normal and pathophysiological conditions, and cancer cells have been shown to secrete exosomes in greater amounts than normal cells (reviewed in 1). The exosomal markers contained in this kit are Alix, Annexin V, ICAM-1, CD9, GM130, EpCAM, flotillin, and HSP70.

Alix, a cytosolic scaffold protein, regulates many cellular processes including endocytic membrane trafficking, cell adhesion through interactions with ESCRT (endosomal sorting complex required for transport) proteins, endophilins, and CIN85 (Cbl-Interacting protein of 85 kDa) (2, 3).

Annexin V is a ~30 kDa protein that binds to phospho-lipids in a calcium-dependent manner (4). All annexins contain a putative PKC binding site, but only annexin V has been identified as an inhibitor of this pathway (5).

Intracellular cell adhesion molecule-1 (CD54 or ICAM-1) is a cell surface glycoprotein that belongs to the immunoglobulin superfamily (IgSF) of adhesion molecules. CD54 is expressed at low levels in diverse cell types, and is induced by cytokines (TNF-alpha, interleukin-1) and bacterial lipopolysaccharides (6). Apical localization on endothelial cells (or basolateral localization on epithelial cells) is a prerequisite for leukocyte trafficking through the endothelial (or epithelial) barrier (6).

The CD9 antigen belongs to the tetraspanin family of cell surface glycoproteins. Tetraspanins interact with a variety of cell surface proteins and intracellular signaling molecules in specialized tetraspanin-enriched microdomains (TEMs), where they mediate a range of processes including adhesion, motility, membrane organization, and signal transduction (7). Additional research identified CD9 as an abundant component of exosomes, and may play a role in the fusion of these secreted membrane vesicles with recipient cells (8). GM130 is required for membrane fusion events that mediate ribbon formation during Golgi assembly (9). The Golgi apparatus functions in the modification, organization, and transport of proteins and membrane targeted to other parts of the cell, such as the plasma membrane, lysosomes, and endosomes. This regulated transport is important for appropriate protein localization, secretion, and signal transduction (reviewed in 10).

Epithelial cell adhesion and activation molecule (EpCAM/CD326) is a transmembrane glycoprotein that mediates calcium-independent, hemophilic adhesions on the basolateral surface of most epithelial cells (11). One of the first tumor-associated antigens discovered, EpCAM has long been a marker of epithelial and tumor tissue. Research studies have shown that EpCAM is highly expressed in cancer cells and can be used as a biomarker for the detection of tumor-derived exosomes (reviewed in 1, 12, 13).

Flotillins belong to a family of lipid raft-associated integral membrane proteins that are ubiquitously expressed and located to lipid rafts on the cell plasma membrane where they support signal transduction and regulate lipid raft motility and localization (14-17). In addition to its colocalization with lipid rafts on the plasma membrane, flotillin-1 also has been found at compartments of the endocytic and autophagosomal pathways, such as recycling/ late endosomes, the Golgi complex, as well as the nucleus (18, 19). HSP70 is a molecular chaperone expressed constitutively under normal conditions to maintain protein homeostasis and is induced upon environmental stress (20). HSP70 is able to interact with unfolded proteins to prevent irreversible aggregation and catalyze the refolding of their substrates in an ATP and co-chaperone dependent manner (21). An immune response is elicited upon excretion of heat shock proteins from tumor exosomes (reviewed in 1).

Background References

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