SM5062

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Monoclonal Antibody to TER ATPase / VCP - Ascites

Alternate names: 15S Mg(2+)-ATPase p97 subunit, Transitional endoplasmic reticulum ATPase, Valosin-containing protein

Catalog No.: SM5062

Quantity: 0.1 ml

Background: Valosin-containing protein (VCP or p97) belongs to the AAA (ATPase associated activities) family and acts as a molecular chaperone to a wide variety of cellular activities. Some of these activities include the alteration of both nuclear and mitotic golgi membranes, ubiquitin-proteasome dependent protein degradation, regulation of the NF-kappa b pathway, and extraction of membrane proteins.
VCP has been shown to contain a substrate binding domain (N) and two conserved ATPase domains (D1 and D2). The three dimensional structure of VCP resembles that of a cylinder with the D1 and D2 stacked upon one another in a homo-hexameric ring formation.

Uniprot ID: P55072

NCBI: NP_009057.1

GeneID: 7415

Host / Isotype: Mouse / IgG2a

Clone: 5

Immunogen: Synthetic Peptide: C G(792) G S V Y T E D N D D D L Y G (806) corresponding to amino acid residues 792-806 from Mouse VCP.
Remarks: This peptide (SM5062CP) is available for use in Neutralization and Control experiments.

Format: **State:** Liquid ascites
Preservatives: 0.05% Sodium Azide

Applications: **Flow Cytometry:** 1/400.
Immunoprecipitation.
Immunofluorescence: 1000.
Western Blot: 1/2000, detects an ~97 kDa protein representing VCP from total lysate of cultured Human B cells.
Immunohistochemistry on Frozen Sections: 1/500.
Immunohistochemistry on Paraffin Embedded Sections: 1/500.
Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.

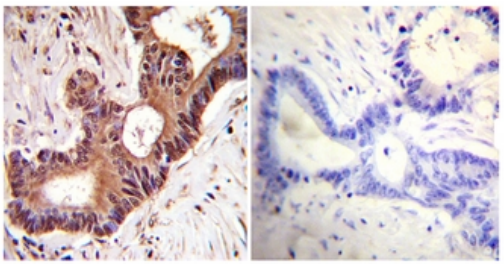
Specificity: This antibody detects VCP protein in Human, Mouse, and Rat samples.

Storage: Store undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer.
Avoid repeated freezing and thawing.
Shelf life: one year from despatch.

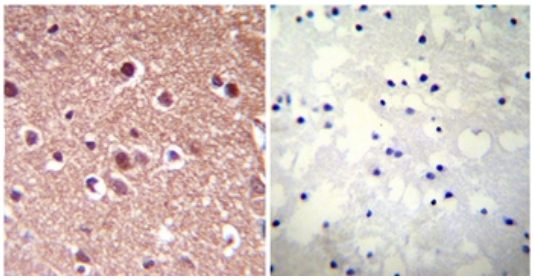
General Readings:

1. J. Biol. Chem., Jun 2006; 281: 17369-17378.
2. JBC 2006 manuscript, M600509200v1.
3. J Immunol. 2007 Feb 15;178(4):2535-41.
4. Autophagy. 2010 Feb;6(2):217-27.
5. Reprod Biol Endocrinol. 2011 Aug 19;9():117.
6. Muscle Nerve. 2007 Oct;36(4):447-54.

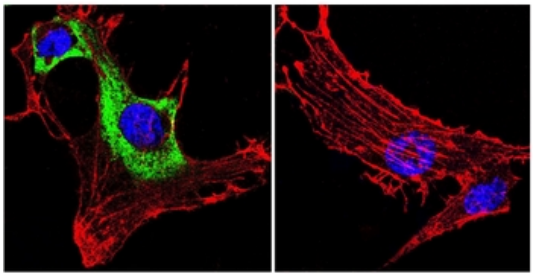
Pictures:



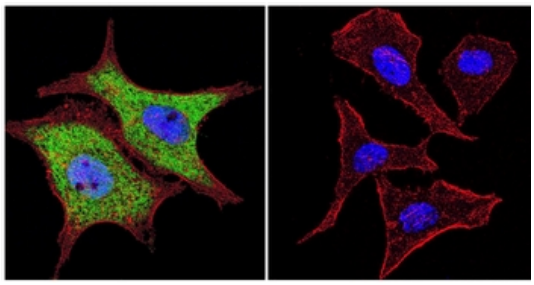
Immunohistochemistry was performed on cancer biopsies of deparaffinized Human colon carcinoma tissues. To expose target proteins, heat induced antigen retrieval was performed using 10mM sodium citrate (pH6.0) buffer, microwaved for 8-15 minutes. Following antigen retrieval tissues were blocked in 3% BSA-PBS for 30 minutes at room temperature. Tissues were then probed at a dilution of 1/1000 with SM5062 or with primary antibody (Negative Control) overnight at 4°C. in humidified chamber. Tissues were washed extensively with PBST and endogenous peroxidase activity was quenched with a peroxidase suppressor. Detection was performed using a biotin-conjugated secondary antibody and SA-HRP followed by colorimetric detection using DAB. Tissues were counterstained with hematoxylin and prepped for mounting.



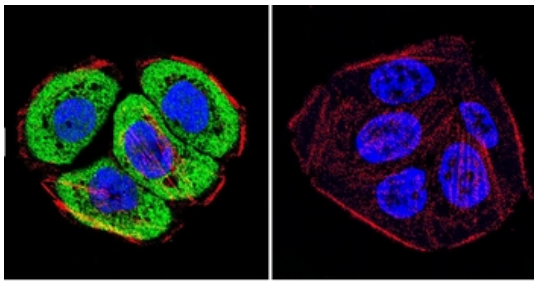
Immunohistochemistry was performed on both normal and cancer biopsies of deparaffinized Human brain tissue tissues. To expose target proteins heat induced antigen retrieval was performed using 10mM sodium citrate (pH6.0) buffer microwaved for 8-15 minutes. Following antigen retrieval tissues were blocked in 3% BSA-PBS for 30 minutes at room temperature. Tissues were then probed at a dilution of 1/1000 with a mouse monoclonal antibody recognizing Anti-VCP (*Cat.-No* SM5062) or without primary antibody (negative control) overnight at 4°C in a humidified chamber. Tissues were washed extensively with PBST and endogenous peroxidase activity was quenched with a peroxidase suppressor. Detection was performed using a biotin-conjugated secondary antibody and SA-HRP followed by colorimetric detection using DAB. Tissues were counterstained with hematoxylin and prepped for mounting.



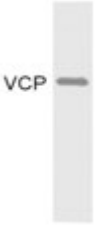
Immunofluorescent analysis of VCP using VCP Monoclonal antibody (5) (*Cat.-No* SM5062) shows staining in C6 glioma cells. VCP staining (green), F-Actin staining with Phalloidin (red) and nuclei with DAPI (blue) is shown. Cells were grown on chamber slides and fixed with formaldehyde prior to staining. Cells were probed without (control) or with or an antibody recognizing VCP (*Cat.-No* SM5062) at a 1/20-1/200 overnight at 4°C, washed with PBS and incubated with a Dylight-488 conjugated secondary antibody.



Immunofluorescent analysis of VCP using VCP Monoclonal antibody (5) (*Cat.-No* SM5062) shows staining in Hela cells. F-Actin staining with Phalloidin (red) and nuclei with DAPI (blue) is shown. Cells were grown on chamber slides and fixed with formaldehyde prior to staining. Cells were probed without (control) or with or an antibody recognizing VCP (*Cat.-No* SM5062) at a 1/20-1/200 overnight at 4°C, washed with PBS and incubated with a Dylight-488 conjugated secondary antibody.



Immunofluorescent analysis of VCP using VCP Monoclonal antibody (5) (‹em›Cat.-No‹/em› SM5062) shows staining in ViDr colon carcinoma cells. F-Actin staining with Phalloidin (red) and nuclei with DAPI (blue) is shown. Cells were grown on chamber slides and fixed with formaldehyde prior to staining. Cells were probed without (control) or with or an antibody recognizing VCP (‹em›Cat.-No‹/em› SM5062) at a 1/20-1/200 overnight at 4°C, washed with PBS and incubated with a Dylight-488 conjugated secondary antibody.



Western blot of VCP from CA46 cell lysate using SM5062.