

Background

Cellular morphology, adhesion, and motility occur through dynamic reorganization of actin-based superstructures. Actin-binding proteins are critical for regulating actin polymerization and superstructure formation. The Arp2/3 complex is an actin polymerization-inducing complex that includes Arp2, Arp3, p41-Arc, p34-Arc, p21-Arc, p20-Arc, and p16-Arc. Several nucleation promoting factors, such as WASP and coronin, regulate the activity of the Arp2/3 complex. In addition, the Arp2/3 complex may be regulated by phosphorylation of specific subunits in the complex. Arp2 has two phosphosites, Thr-237 and Thr-238, that are evolutionarily conserved, and are phosphorylated along with Tyr-202 in response to growth factor stimulation. These phosphorylation events may regulate binding to the pointed end of actin filaments, and alanine substitutions of these Arp2 phosphosites inhibit membrane protrusions. Thus, phosphorylation may be another mode of Arp2/3 complex regulation in addition to the activity of nucleation-promoting factors.

Background References

Kelleher, J.F. et al. (1995). *J Cell Biol.* 131(2):385.
 LeClaire, L.L. et al (2008). *J Cell Biol.* 182(4):647.
 Soderling, S.H. (2009). *Sci Signal.* 2(55):pe5 (Review).

Product Citations

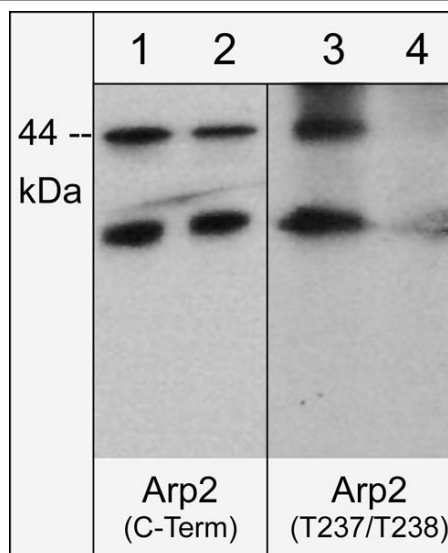
Machlus, K.R. et al. (2016) *Blood.* 127(11):1468.
WB: mouse megakaryocytes

Osma-Garcia, I.C. et al. (2015) *Eur J Immunol.* doi: 10.1002
WB: mouse macrophages

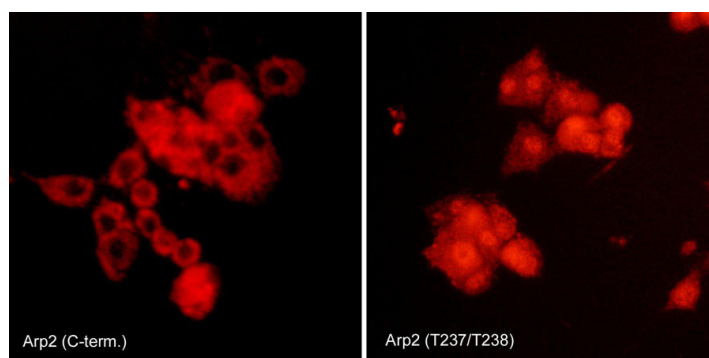
Park, M. et al. (2013) *J Biol Chem* 288:33324.
WB: human brain microvascular endothelial cells hCMEC/D3

Spillane, M. et al. (2012) *J Neurosci.* 32(49):17671.
ICC: chick embryonic neuron

Kalwa, H. & Michel, T. (2011) *J Biol Chem.* 286(3):2320.
WB: bovine aortic endothelial cells



Western blot of human A431 cells treated with Calyculin A (100 nM) for 30 min. Blot lanes were untreated (lanes 1 & 3) or treated with lambda phosphatase (lanes 2 & 4) then probed with anti-Arp2 (C-terminal) (lanes 1 & 2) or anti-Arp2 (Thr-237/Thr-238) (lanes 3 & 4).



Immunocytochemical labeling of Arp2 phosphorylation in rat PC12 cells differentiated with NGF. The cells were probed with Arp2 (C-terminal region) and Arp2 (Thr-237/Thr-238) rabbit polyclonal antibodies, then the antibodies were detected using appropriate secondary antibody conjugated to Cy3.

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Immunogen**Uniprot ID: P61160**

Arp2 (C-terminal region) synthetic peptide (coupled to KLH) corresponding to amino acid residues in the C-terminal region of human Arp2. This peptide sequence is highly conserved in rat, mouse, chicken, and frog Arp2 proteins.

Buffer and Storage

Rabbit polyclonal, affinity-purified antibody is supplied in 100 l phosphate-buffered saline, 50% glycerol, 1 mg/ml BSA, and 0.05% sodium azide. Store at -20°C. Do not aliquot. Stable for 1 year.

Applications

WB	1:1000
ELISA	1:2000
ICC	1:100
IP	1:50

Species Reactivity

Hu, Rt, Ms, Ck, Fr

End user should determine optimal dilution for their particular applications and experiments.

Western blot membranes were incubated with diluted antibody in 5% non-fat milk, Tris buffer, 0.04% Tween20 for 1 hour at room temperature.

Abbreviations: E = ELISA, ICC = immunocytochemistry, IHC = immunohistochemistry, IP = immunoprecipitation, MS = mass spectrometry, WB = western blot
Hu = Human, Ms = Mouse, Rt = Rat, Ck = Chicken, F = Frog, B = Bovine

Specificity

This antibody was affinity purified using Arp2 (C-terminal region) peptide (without carrier). The antibody detects 44 and 32 kDa* proteins corresponding to the molecular mass of Arp2 on SDS-PAGE immunoblots of human A431, HUVEC, HeLa, and Jurkat cells as well as rat PC12, mouse C2C12, and rabbit fibroblasts.

*All molecular weights (MW) are confirmed by comparison to MW standards and to western blot mobilities of known proteins with similar MW.

"Native" western blot utilizes non-reducing sample buffer (no mercaptoethanol or SDS), normal SDS-PAGE gel electrophoresis, and no methanol in transfer buffers.

Related Products

AP3871 Arp2 (Thr-237/Thr-238), phospho-specific Rabbit Polyclonal

AX3875 phospho-Arp2 (Thr-237/Thr-238) Blocking Peptide

AK6060 Actin & Tubulin Antibody Sampler Kit

AP1671 Actin (Tyr-53), phospho-specific Rabbit Polyclonal

WK6110 N-WASP Phospho-Regulation Antibody Sampler Kit

CK6180 Coronin-1B Phospho-Regulation Antibody Sampler Kit

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