Background: Caspase-9 (ICE-LAP6, Mch6) is an important member of the cysteine aspartic acid protease (caspase) family (1,2). Upon apoptotic stimulation, cytochrome c released from mitochondria associates with the 47 kDa pro-caspase-9/Apaf 1. Apaf-1 mediated activation of caspase-9 involves intrinsic proteolytic processing resulting in cleavage at Asp315 and producing a p35 subunit. Another cleavage occurs at Asp330 producing a p37 subunit that can serve to amplify the apoptotic response (3-6). Cleaved caspase-9 further processes other caspase members, including caspase-3 and caspase-7, to initiate a caspase cascade, which leads to apoptosis (7-10).

Specificity/Sensitivity: Cleaved Caspase-9 (Asp315) (D8I9E) Rabbit mAb recognizes endogenous levels of caspase-9 protein only when cleaved at Asp315. Non-specific proteins that are induced by apoptosis under certain conditions may be detected.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Asp315 of human caspase-9 protein.

Recommended Antibody Dilutions:
Western blotting 1:1000
Immunoprecipitation 1:50
Immunofluorescence (IF-IC) 1:800

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com.

Background References:
Confocal immunofluorescent analysis of HeLa cells, serum-starved (left) or treated with Staurosporine #9963 (1 μM, 3 hr; right), using Cleaved Caspase-9 (Asp315) (D8I9E) Rabbit mAb (green). Actin filaments were labeled with Dylight™ 554 Phalloidin #13054 (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).

Flow cytometric analysis of Jurkat cells, untreated (blue) or treated with Etoposide #2200 (25 μM, overnight; green), using Cleaved Caspase-9 (Asp315) (D8I9E) Rabbit mAb. Anti-rabbit IgG (H+L), F(ab’)2 fragment (Alexa Fluor® 488 conjugate) #4412 was used as a secondary antibody.