BACKGROUND

In eukaryotes, the ubiquitin-mediated proteolysis by the 26S proteosome is one of the most fundamental regulatory mechanisms controlling cell cycle progression, transcriptional regulation, signal transduction, and many other cellular processes. The ubiquitination of targets for degradation is a three-enzyme process, in which the E1 ubiquitin-activating enzyme and E2 ubiquitin-conjugating enzyme are relatively nonspecific, whereas different E3 ubiquitin ligases recognize specific substrates. Two distinct ubiquitin conjugation pathways mediate cell division by affecting the transition from G1 to S phase, the separation of sister chromatids during anaphase, and the exit from mitosis. The first event in G1/S requires the ubiquitin-conjugating enzyme Cdc34 (or Ubc3) and a ubiquitin-protein ligase complex termed SCF complex (Skp1-Cullin-F-box protein) in order to activate DNA replication. The two mitotic events involve a large ubiquitin-ligase complex called the anaphase-promoting complex-cyclosome (APC/C) in combination with one of two distinct ubiquitin-conjugating enzymes (Ubc10 or Ubc4). APC/C regulates mitosis by affecting chromosome and spindle dynamics and by regulating the activity of mitotic cyclin-dependent kinase (Cdks).[1]

Skp1 (S-phase kinase-associated protein 1) is a core component of SCF ubiquitin ligases and mediates protein degradation, thereby regulating eukaryotic fundamental processes. SCF typically contain at least four subunits: Skp1, a cullin, a RING finger protein (Rbx1, also called Hrt1 or Roc1), and a member of the large family of F-box adaptor protein which acts as a receptor for target proteins and directly involved in the specific substrates recruitment. The F-box is a degenerated sequence of about 70 amino acids required but not sufficient for the interaction between a given F-box protein and Skp1. Skp1 is an adaptor between one of the variable F-box proteins and Cullin. Structure-function studies in yeast and mammals have demonstrated that the cullin functions as a scaffold in assembling the different subunits of the SCF complex. The cullin interacts at its carboxyl terminus with the RING domain protein Rbx1 to form the catalytic domain, and at its amino terminus end with Skp1. Ubiquitin is transferred from a ubiquitin-conjugating enzyme (E2) onto substrates recruited by the F-box protein. After poly-ubiquitination, the substrate is recognized by the proteasome and degraded. Protein degradation mediated by the SCF complexes has been shown to influence a variety of cellular processes such as the cell cycle, signal transduction and gene expression.[2]

There is only one known functional Skp1 protein in human and yeasts. Nevertheless, this unique protein is able to interact with different F-box proteins to ubiquinate different substrates. The human Skp1, for example, was originally identified as a protein that associates the cyclin A-CDK2 (cyclin-dependent kinase 2) complex with the F-box protein Skp2 and the SCFskp2 complex can ubiquitinate the p27Kip1 inhibitor of CDK, allowing CDK activity to drive cells into S phase. Another well-studied SCF complex in human, SCFFWD1, targets the NF-κB pathway and beta-catenin, a downstream signaling factor in the Wnt pathway of development and proliferation. Additionally, It has been demonstrated that Skp1 plays a critical role in the preservation of genetic stability. Inhibition of Skp1 functions in cells lead to caused the formation of multinucleated cells, defects in centrosomes and mitotic spindles, impaired chromosome segregation, and chromosomal instability and also resulted in neoplastic transformation with high penetrance.[3] Beside the canonical SCF, it appears that in some cases, Skp1 and F-box proteins may function in non-SCF complexes and that some F-box proteins have functions on their own. Studies have also characterized Skp1 as an RNA polymerase II elongation factor.[4]

References:
1. Vodermaier, H.C.:Curr. Biol. 14:R787-R796, 2004
2. Zheng, N. et al: Nature 416:703-9, 2002
3. Piva, R. et al: Mol. Cell. Biol. 22: 8375-87, 2002
4. Conaway, R.C. et al: Science 296:1254-8, 2002

TECHNICAL INFORMATION

Source:
SKP1 Antibody is a mouse monoclonal antibody raised against recombinant human SKP1 fragments expressed in *E. coli*.

Specificity and Sensitivity:
This antibody detects SKP1 proteins in various cell lysate.

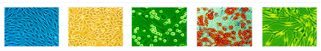
Storage Buffer: PBS and 30% glycerol

Storage:
Store at -20°C for at least one year. Store at 4°C for frequent use. Avoid repeated freeze-thaw cycles.

APPLICATIONS

Application:	*Dilution:
WB	1:1000
IP	1:50-100
IHC (Paraffin)	1:50-200
ICC	1:50-200
FACS	1:50-200

**Optimal dilutions must be determined by end user.*

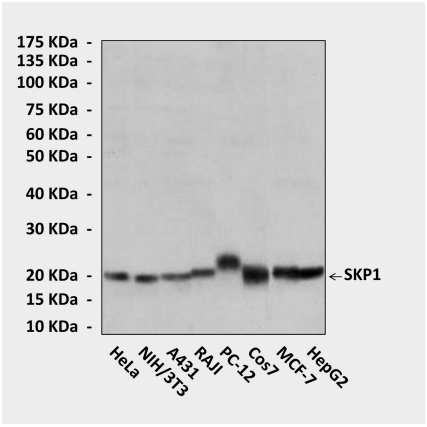


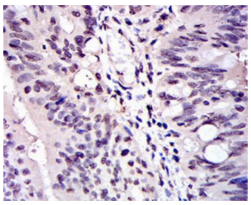
CELL APPLICATIONS, INC.

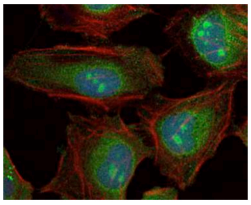
SKP1 Antibody
Cat. No. CP10434

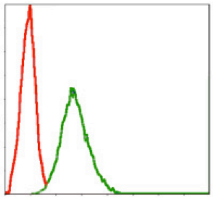
Applications:	**WB, IP, IHC, ICC, FACS**
Detected MW:	**19 kDa**
Species & Reactivity:	**Human, Mouse, Rat**
Isotype:	**Mouse IgG1**

QUALITY CONTROL DATA









Top: Western blot detection of SKP1 proteins in various cell lysates using SKP1 Antibody. **Middle Upper:** This antibody stains HeLa cells in confocal immunofluorescent testing (SKP1 Antibody: Green; Actin filaments: Red; DRAQ5 DNA dye: Blue). **Middle Lower:** It also stains paraffin-embedded human rectum cancer tissue in IHC analysis. **Bottom:** This antibody also specifically detects SKP1 proteins in NIH3T3 cells in FACS assay (SKP1 Antibody: Green; control mouse IgG: Red).