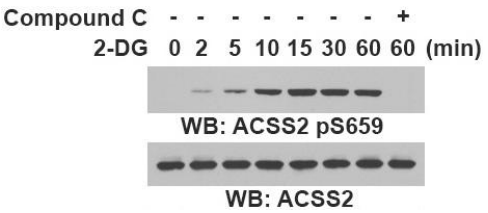
ACSS2 (Phospho-Ser659) Antibody

#58003

Number: 58003-1, 58003-2
Amount: 50μg/50μl, 100μg/100μl
Swiss-Prot No. :Q9NR19
Form of Antibody: Rabbit IgG in phosphate buffered saline (without Mg2+ and Ca2+), pH 7.4, 150mM NaCl,0.02% sodium azide and 50% glycerol.
Storage/Stability: Store at -20℃/1 year
Immunogen: The antiserum was produced against synthesized phosphopeptide derived from Human ACSS2 around the phosphorylation site of serine 659 .
Purification: The antibody was affinity-purified from rabbit antiserum by affinity-chromatography using epitope-specific phosphopeptide. The antibody against non-phosphopeptide was removed by chromatography using non-phosphopeptide corresponding to the phosphorylation site.
Specificity/Sensitivity:ACSS2 (phospho-Ser659) antibody detects endogenous levels of ACSS2 only when phosphorylated at serine 659.
Reactivity: Human,Mouse
Applications:
Predicted MW: 78kd
WB :1:500~1:2000



ACSS2 S659 Phosphorylation in U87 Cells

Background :
Overcoming metabolic stress is a critical step in tumor growth. Acetyl coenzyme A (acetyl-CoA) generated from glucose and acetate uptake is important for his-tone acetylation and gene expression. However, how acetyl-CoA is produced under nutritional stress is un-clear. We demonstrate here that glucose deprivation results in AMP-activated protein kinase (AMPK)-medi-ated acetyl-CoA synthetase 2 (ACSS2) phosphoryla-tion at S659, which exposed the nuclear localization signal of ACSS2 for importin a5 binding and nuclear translocation. In the nucleus, ACSS2 binds to tran-scription factor EB and translocates to lysosomal and autophagy gene promoter regions, where ACSS2 incorporates acetate generated from histone acetylation turnover to locally produce acetyl-CoA for histone H3 acetylation in these regions and pro-mote lysosomal biogenesis, autophagy, cell survival, and brain tumorigenesis. In addition, ACSS2 S659 phosphorylation positively correlates with AMPK ac-tivity in glioma specimens and grades of glioma malig-nancy. These results underscore the significance of nuclear ACSS2-mediated histone acetylation in main-taining cell homeostasis and tumor development.

References:
Li et al., 2017, Molecular Cell 66, 1–14 June 1, 2017 ª 2017 Elsevier Inc.
http://dx.doi.org/10.1016/j.molcel.2017.04.026

Application in this Article

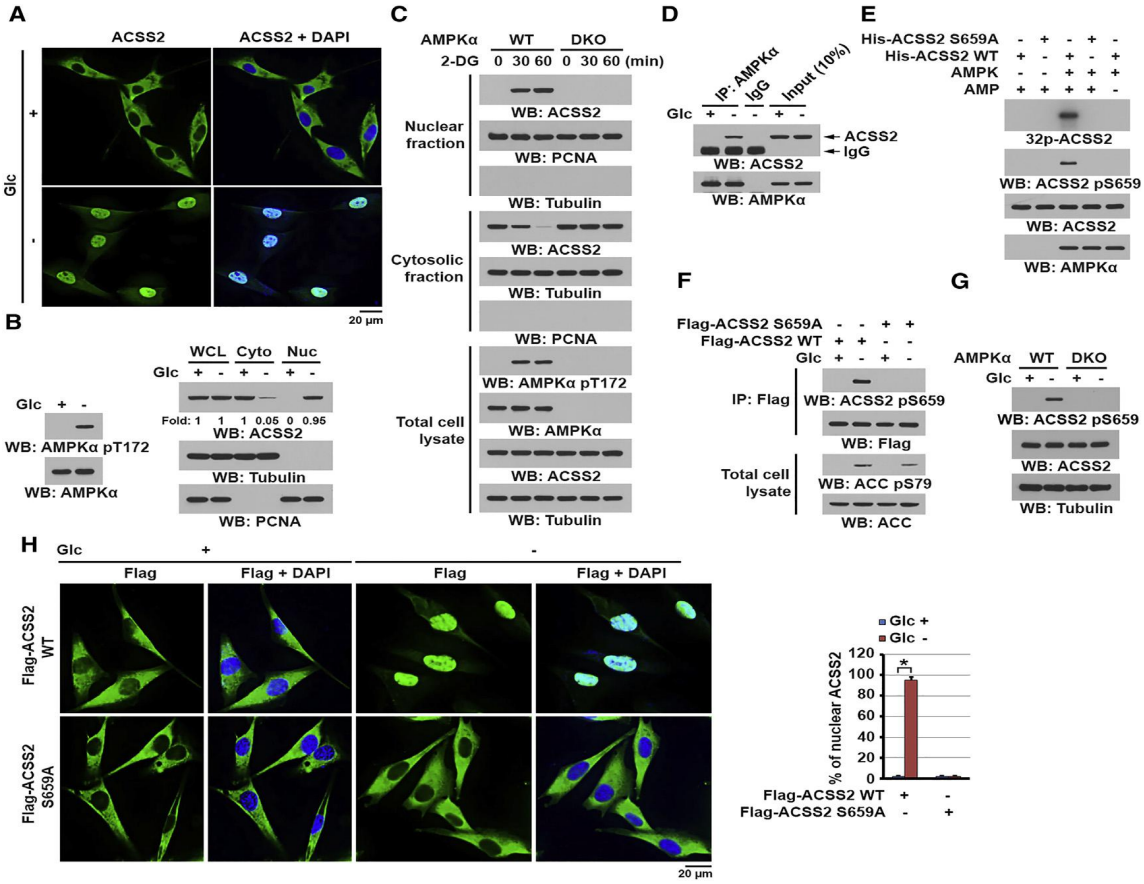


Figure 1. ACSS2 S659 Phosphorylation by AMPK Induces Nuclear Translocation of ACSS2

(B–G) Immunoblot analyses were performed with the indicated antibodies.

(A) U87 cells were deprived of glucose for 1 hr. Immunofluorescent analyses were performed with an anti-ACSS2 antibody. DAPI (blue) was used to stain the nuclei.

(B) U87 cells were deprived of glucose for 1 hr. Total cell lysates and cytosolic and nuclear fractions were prepared. Activation of AMPK is reflected by AMPKa T172 phosphorylation.

(C) WT and AMPKa1 and AMPKa2 double-knockout (DKO) MEFs were treated with 2-DG (25 mM) for the indicated periods. Total cell lysates and cytosolic and nuclear fractions were prepared.

(D) U87 cells were deprived of glucose for 10 min. Immunoprecipitation with an anti-AMPKa antibody was performed.

(E) Purified AMPK was mixed with the indicated bacterially purified His-ACSS2 proteins and [g-^{32}P] ATP in the presence or absence of AMP (100 mM). Auto-radiographic analysis was performed.

(F) U87 cells expressing the indicated FLAG-ACSS2 proteins were deprived of glucose for 1 hr. Immunoprecipitation with an anti-FLAG antibody was performed. Activation of AMPK is reflected by its substrate acetyl-CoA carboxylase (ACC) S79 phosphorylation.

(G) WT and AMPKa1/2 DKO MEFs were deprived of glucose for 1 hr. Total cell lysates were prepared.

(H) U87 cells expressing the indicated FLAG-ACSS2 proteins were deprived of glucose for 1 hr. Immunofluorescent analyses were performed with an anti-FLAG antibody. The percentage of nuclear ACSS in 20 cells in each group was quantitated (right) using ImageJ. A two-tailed Student's t test was used. *p < 0.001.