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**#4185** Store at -20C

## Phospho-AMPK $\alpha$ 1 (Ser485)/AMPK $\alpha$ 2 (Ser491) Antibody

**For Research Use Only. Not for Use in Diagnostic Procedures.**

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
W	H M R Mk	Endogenous	62	Rabbit	#Q13131, #P54646	5562, 5563

### Product Usage Information

#### Application

Western Blotting

#### Dilution

1:1000

### Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100  $\mu$ g/ml BSA and 50% glycerol. Store at -20°C. Do not aliquot the antibody.

### Specificity/Sensitivity

Phospho-AMPK $\alpha$ 1 (Ser485)/AMPK $\alpha$ 2 (Ser491) Antibody detects endogenous levels of AMPK $\alpha$ 1/ $\alpha$ 2 only when phosphorylated at serine 485 or serine 491. This antibody does not cross-react with other related proteins.

### Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser491 of human AMPK $\alpha$ 2. Antibodies are purified by protein A and peptide affinity chromatography.

### Background

AMP-activated protein kinase (AMPK) is highly conserved from yeast to plants and animals and plays a key role in the regulation of energy homeostasis (1). AMPK is a heterotrimeric complex composed of a catalytic  $\alpha$  subunit and regulatory  $\beta$  and  $\gamma$  subunits, each of which is encoded by two or three distinct genes ( $\alpha$ 1, 2;  $\beta$ 1, 2;  $\gamma$ 1, 2, 3) (2). The kinase is activated by an elevated AMP/ATP ratio due to cellular and environmental stress, such as heat shock, hypoxia, and ischemia (1). The tumor suppressor LKB1, in association with accessory proteins STRAD and MO25, phosphorylates AMPK $\alpha$  at Thr172 in the activation loop, and this phosphorylation is required for AMPK activation (3-5). AMPK $\alpha$  is also phosphorylated at Thr258 and Ser485 (for  $\alpha$ 1; Ser491 for  $\alpha$ 2). The upstream kinase and the biological significance of these phosphorylation events have yet to be elucidated (6). The  $\beta$ 1 subunit is post-translationally modified by myristoylation and multi-site phosphorylation including Ser24/25, Ser96, Ser101, Ser108, and Ser182 (6,7). Phosphorylation at Ser108 of the  $\beta$ 1 subunit seems to be required for AMPK activation, while phosphorylation at Ser24/25 and Ser182 affects AMPK localization (7). Several mutations in AMPK $\gamma$  subunits have been identified, most of which are located in the putative AMP/ATP binding sites (CBS or Bateman domains). Mutations at these sites lead to reduction of AMPK activity and cause glycogen accumulation in heart or skeletal muscle (1,2). Accumulating evidence indicates that AMPK not only regulates the metabolism of fatty acids and glycogen, but also modulates protein synthesis and cell growth through EF2 and TSC2/mTOR pathways, as well as blood flow via eNOS/nNOS (1).

### Background References

1. Hardie, D.G. (2004) *J Cell Sci* 117, 5479-87.
2. Carling, D. (2004) *Trends Biochem Sci* 29, 18-24.
3. Hawley, S.A. et al. (1996) *J Biol Chem* 271, 27879-87.
4. Lizcano, J.M. et al. (2004) *EMBO J* 23, 833-43.
5. Shaw, R.J. et al. (2004) *Proc Natl Acad Sci USA* 101, 3329-35.
6. Woods, A. et al. (2003) *J Biol Chem* 278, 28434-42.
7. Warden, S.M. et al. (2001) *Biochem J* 354, 275-83.

### Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

### Western Blot Buffer

**IMPORTANT:** For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

### Applications Key

**W:** Western Blotting

### Cross-Reactivity Key

**H:** Human **M:** Mouse **R:** Rat **Mk:** Monkey

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